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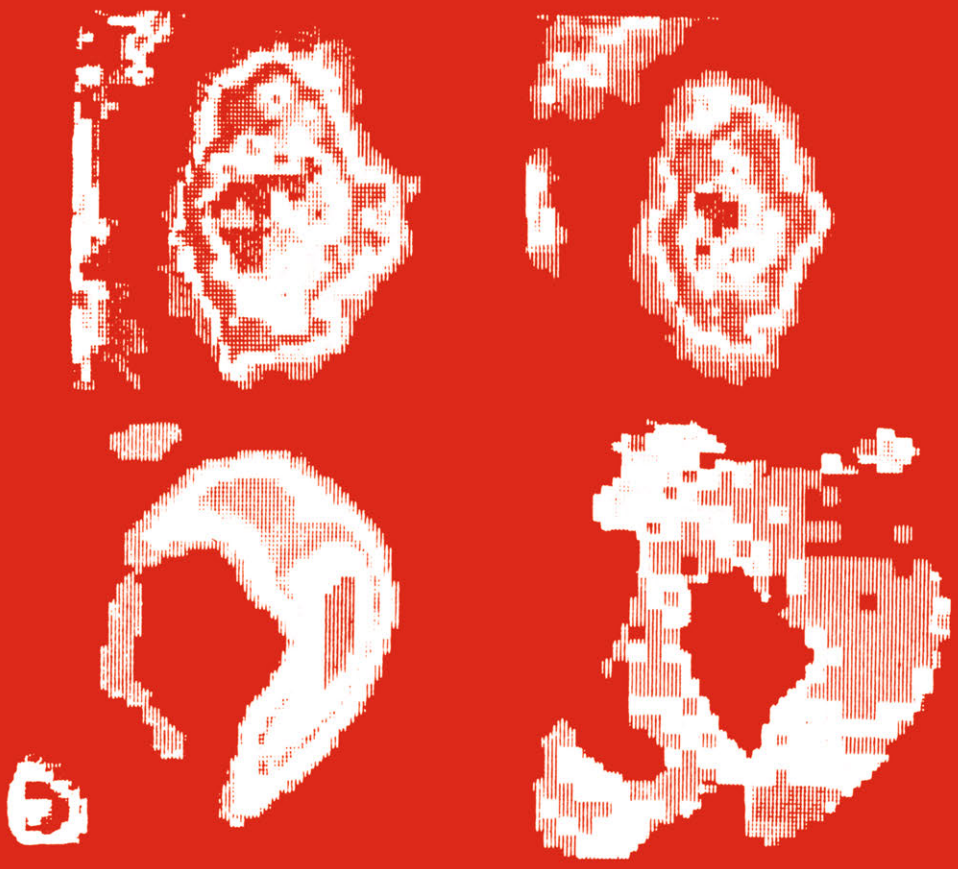
Cardiology

An International Perspective



VOLUME 1

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PREFACE

Once in four years, cardiologists of the world united into the International Society and Federation of Cardiology come together to discuss the most pressing problems of cardiovascular pathology, sum up the accomplishments of the intervening years, and set directions for future research and exploitation of the existing knowledge. Not too much time passed since the I Paris Congress of International Foundation of Cardiology in 1950, but since then we have been witnessing a real information explosion. Extraordinary amounts of new knowledge, accumulated during the past three decades, has revolutionized our understanding of major cardiovascular diseases as well as approach to their treatment and diagnosis.

The IX World Congress of Cardiology, held in Moscow in June 20-26, gathered 5,099 delegates from 78 countries. In the course of the Congress, prominent scientists presenting 21 lectures on topical problems of theoretical and practical cardiology; 900 papers were heard at 37 Symposia and 58 Free Communication Sessions. The papers and discussions demonstrated an increased contribution of fundamental research to clinical arsenal. Another feature of modern biomedical research is that it makes use of the latest accomplishments in other fields of science and technology: physics, chemistry, electronics, etc. Of late, a new research discipline, called molecular cardiology, has formed. By elucidating the most subtle basic mechanisms whereby pathological processes develop, it is expected to afford ultimate control of cardiovascular diseases.

The Congress demonstrated that new knowledge about the diagnosis and treatment is being successfully introduced into clinical practice. New, mainly non-invasive, sensitive diagnostic techniques to study the heart and vessels function have appeared. Already existing angiographic and nuclear techniques are being further elaborated. Taking together the growing amount of relevant information and the development of new devices, techniques and effective drugs, the chances of successful treatment and reduction of cardiovascular mortality have been greatly improved. Considerable progress has been made by preventive cardiology through intensive investigation of prevalence and changes in the incidence of cardiovascular

diseases as well as factors that may influence these two. Large-scale cooperative programs to control arterial hypertension and coronary heart disease are being developed and implemented.

On the whole, the IX Congress in Moscow showed that modern cardiology is a rapidly advancing science with great potential and feasible prospects.

Academician Eugeni I Chazov

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LECTURES

ARE THERE MARKERS FOR THE PHYSIOPATHOLOGY OF ESSENTIAL HYPERTENSION?

Alberto Zanchetti

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INTRODUCTION

Widespread interest for the physiopathology of hypertension has undoubtedly been raised by the hope of understanding the error or the errors of regulation by which higher blood pressure values are attained, but this cognitive interest has often been coupled with the hope that improved physiopathologic understanding might result in improved management of hypertension. This practical interest is linked with the belief that qualitatively or, at least, quantitatively different mechanisms underlie the rise in blood pressure in different patients. The hope of learning the best individual treatment for a given physiopathological profile requires a process of simplification, that is the identification of simple but meaningful indices, or markers, from which a more complex functional pattern can be described. The question mark in the title of this lecture "Are there markers for the physiopathology of essential hypertension?" has been placed to signify the uncertainty as to whether any of the markers that have been proposed and used really measures the function we assume that it measures, whether other functional variables have a fixed relation with the supposed marker and can be inferred from measurement of the latter, and finally whether useful therapeutic guidelines can be derived from its measurement. We shall see that it is not easy to get rid of these question marks.

MARKERS OF SYMPATHETIC ACTIVITY

Measuring sympathetic activity in man has been, and still is, one of the most elusive undertakings in the clinical physiology of hypertension. Two main approaches have been followed, 1) that of assessing cardiovascular reflexes tonically regulating sympathetic activity in order to identify a primary or secondary disturbance in sympathetic regulation, and 2) that of measuring plasma catecholamines as an index of a possible increase in transmitter release from sympathetic postganglionic endings.

Cardiovascular Reflexes and Hypertension

Interest has been concentrated upon reflexes originating from low pressure and high pressure cardiovascular receptors. Limited information is available about low pressure receptor reflexes, but it has been suggested[1] that an exaggerated sympathetic vasoconstrictor response to withdrawal of the tonic inhibition from low pressure cardiopulmonary receptors occurs in borderline hypertension. A greater amount of attention has been concentrated on high pressure, or arterial (sino-aortic), receptor reflexes. Sleight[2], by studying the bradycardic and tachycardic reflex responses to injection of pressor and depressor drugs, has introduced the concept of baroreflex resetting accompanying human hypertension, resetting being characterized by a raised threshold and a diminished sensitivity. My group[3,4] has explored pressor and depressor responses to carotid sinus manipulation by the neck pressure chamber. Figure 1 illustrates the differences in the baroreflex responses we observed in a group of normotensive subjects and in two groups of moderate and severe hypertensives. The pressor response to reduction in carotid transmural pressure (that means baroreflex deactivation) was greatest in the normotensive group, decreased in moderate hypertensives, and was lowest in severe hypertensives. On the contrary, the depressor response to increased carotid transmural pressure (i.e., baroreflex stimulation) was lowest in normotensives, intermediate in moderate hypertensives and greatest in severe hypertensives. This resetting of baroreflexes, marking the progressive rise in arterial pressure, consists in an upward shift of the baroreceptor threshold without a real loss in reflex sensitivity, and allows maintenance of sympathetic activity, as well as its modulation, in hypertension. Available evidence, however, is that baroreflex resetting is secondary to the rise in pressure[5], and cannot be used as a marker of primary changes characterizing the development of hypertension. We have recently shown[6] that resetting of the carotid sinus reflex is identical in patients with renovascular hypertension and in patients with essential hypertension. Occurrence of secondary resetting does not rule out, however, that there might also be a primary component, and indeed there is some evidence[7] that primary baroreceptor resetting might occur in spontaneous hypertensive rats.

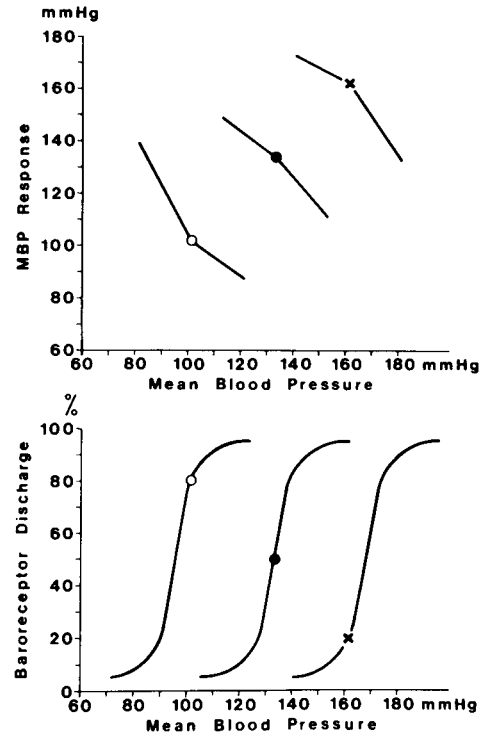

Fig. 1. Above: changes in mean arterial pressure responses (ordinates) to changes in carotid sinus transmural pressure in normotensives (hollow circle), moderate hypertensives (filled circle) and severe hypertensives (cross). Below: schematic drawing of the stimulus-response curves relating arterial blood pressure with carotid baroreceptor firing. The set-point of the reflex (i.e., the point corresponding to the stimulus provided by the existing blood pressure) may be located nearby baroreceptor saturation in normotensive subjects (line to the left) and migrate progressively to the baroreceptor threshold in moderate and severe hypertension (modified from Mancia et al.[4]).

Plasma Catecholamines in Hypertension

The knowledge that plasma noradrenaline represents the spill-over of transmitter released by postganglionic sympathetic endings has suggested its use as a marker of sympathetic activity, and as a test of the hypothesis that essential hypertension may be maintained by enhanced sympathetic activity. Goldstein[8] has recently reviewed 32 studies comparing plasma noradrenaline concentrations

in hypertensive and normotensive groups (Figure 2). Pooling all the data together, relating to 1085 normotensives and 1496 hypertensives, there is a slight but statistically significant difference in plasma noradrenaline between normotensives and hypertensives, the average concentration being somewhat higher in hypertensives (285 vs 231 pg/ml). In the whole, 28 out of 32 studies have found a higher noradrenaline concentration in hypertensives, but in only 13 studies the hypertensive-normotensive difference was statistically significant. Furthermore, there are reasonable doubts that plasma noradrenaline, being only an indirect reflection of transmitter released by sympathetic endings, is a sufficiently sensitive index of that mild increase in tonic sympathetic activity which might occur in essential hypertension, or in a few hypertensive subjects. For instance, we have shown[9] that the increase and decrease in blood pressure (and sympathetic activity) which follow baroreflex deactivation and stimulation by the neck pressure chamber is accompanied by negligible changes in plasma noradrenaline. Plasma noradrenaline might also be a poor marker of sympathetic overactivity in hypertension as there is evidence that plasma noradrenaline correlates with sympathetic activity to muscle blood vessels[10] while there is no excessive muscle vasoconstriction in hypertension[11]. Plasma adrenaline might perhaps represent a better marker of enhanced splanchnic sympathetic activity, and it is interesting that there are reports[12,13] of increased plasma adrenaline concentration in essential hypertension. Interest to this direction is added by recent evidence[14] that adrenaline can enhance noradrenergic sympathetic transmission by a positive feedback action on pre-junctional beta-adrenergic receptors.

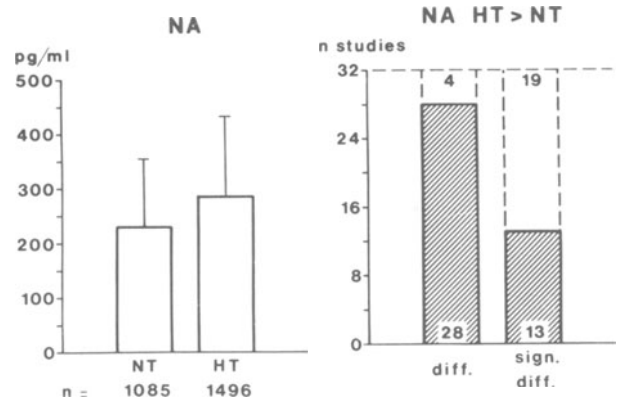

Fig. 2. Left: means and SEM of plasma noradrenaline (NA) in 1085 normotensive (NT) subjects and 1495 hypertensive (HT) subjects in 32 different studies. Right: number of studies in which NA was greater in HT than in NT, and number of studies in which this difference was statistically significant (from data of Golstein[8]).

ABNORMAL ION TRANSPORT ACROSS CELL MEMBRANES

The greatest interest has recently concentrated on abnormal ion transport across cell membranes[15,16] as a possible marker of essential hypertension and, even more importantly, of genetic predisposition to hypertension. Abnormal transport of sodium and/or potassium ions across membranes of erythrocytes[17-20], leucocytes[21] and lymphocytes[22] have been described in essential hypertensives and in normotensive children of hypertensive parents, and the hypothesis has been advanced that reduced cellular sodium extrusion might lead to hypertension, in the presence of excessive sodium intake, by increasing intracellular sodium and calcium concentration.

Several considerations do not yet allow us to use membrane abnormalities as safe markers of hypertension or of predisposition to hypertension. Firstly, there is no general agreement about which of the abnormalities described by the various investigators (of the ouabain-sensitive sodium-potassium pump[21], of the furosemide-sensitive sodium-potassium co-transport[23,24], and of the sodium-sodium (or sodium-lithium) counter-transport[20]) is the physiopathologic important marker. Secondly, the initial report[23,24] that some of these abnormalities would occur only in essential hypertension and in children of hypertensive parents, has not been confirmed by other investigators[25,26], who have found a considerable overlap between the distribution of these abnormalities in essential hypertensives, in normotensives with a family history of hypertension, and in normotensives without a family history of hypertension. Thirdly, there is no general agreement as to whether these abnormalities actually lead to increased intracellular concentrations of sodium[15], and, fourthly, it is largely unknown whether the abnormalities found in blood cells also occur elsewhere, and whether they might lead to hypertension by increasing vascular smooth muscle contractility, or by increasing peripheral or central excitability or rather by interfering with renal sodium excretion. Finally, it has recently been maintained[27] that these ion flux abnormalities would not be primary markers of a diffuse genetic membrane alteration, but would rather be secondary consequences of the action of a so-called natriuretic hormone, the secretion of which would be the consequence of a primary impairment of renal sodium excretion.

THE RENIN-ANGIOTENSION SYSTEM AND BODY FLUID VOLUMES

Plasma Renin Activity in Hypertension

It is agreed that plasma renin activity can vary widely under physiological conditions, and that patients with arterial hypertension may have different renin levels and can be classified as having low, normal and high renin hypertension[28]. The meaning of the classification of hypertensive patients in separate groups

according to the renin/sodium index is still the object of considerable debate, however. The Glasgow group[29] has made the point that both in normotensives and in hypertensives renin values are distributed as a single uninterrupted frequency distribution curve. It has also been remarked by several authors[29,30] that low renin essential hypertension cannot be taken as an invariable marker of increased volume. For the time being, plasma renin is a useful diagnostic index for secondary hypertension, but its use as a marker of subtypes of essential hypertension is of quite dubious significance.

Body Fluid Volumes in Hypertension

Plasma volume, extracellular fluid volume, exchangeable sodium have been measured in hypertensive subjects by several authors, all of whom have failed to identify any group of volume expanded patients in essential hypertension. Recently, Beretta-Piccoli et al.[31] have further explored the problem providing data of considerable interest. Subnormal exchangeable sodium and the absence of a relation of exchangeable sodium and blood pressure in younger hypertensive patients suggests that excess sodium intake or excessive fluid volume are not particularly important as pathogenetic mechanisms in the early stages of essential hypertension, although a positive relation between exchangeable sodium and blood pressure in older hypertensives suggest that sodium and expanded body fluid volumes might play a greater role in hypertension with advancing age.

MARKERS OF HYPERTENSION AS GUIDELINES TO TREATMENT AND PREVENTION

Plasma Renin Activity and Treatment of Hypertension

Plasma renin activity has been the most popular marker and guideline for predicting the success of either beta-blockers or diuretics according to a well-known theory expounded by John Laragh and his group[32] who suggested that beta-blockers were particularly effective in high and normal renin hypertensives, and diuretics in low renin hypertensives. However, the various arguments discussed above should caution against an indiscriminate use of these indices as a sufficiently precise profile of the hypertensive patient and as an approach to the pharmacologic management of hypertension[30,33]. Several of the inferences that have been made in recent years have not resisted criticism, but caution should also be used against translating sound criticism into an uncritical and generalized denial. It is certainly true that recent research[30,33] has not supported the opinion that hypotension induced by beta-blockers results solely or mainly from renin suppression, and that the renin/sodium index can effectively screen patients responsive from those unresponsive to beta-blocking therapy. This does not mean, however,

that the renin-suppressive activity of beta-blockers is always and necessarily free of hypotensive consequences. There is evidence that renin may play some pressor role in a number of cases of hypertension, especially in those patients whose renin is stimulated by treatment with diuretics and vasodilators. Figure 3 shows that in those patients in whom an exaggerated increase in renin due to diuretic therapy blunted the hypotensive action of the diuretic, addition of either an angiotensin II-antagonist, saralasin, or of a beta-blocker can remove the counteraction of excessive renin and produce a hypotensive effect.

Membrane Abnormalities and Prevention of Hypertension

The discovery of membrane abnormalities in the cells of hypertensives and of normotensives with family history of hypertension has suggested that these abnormalities could be used to identify the subjects genetically prone to the development of hypertension if exposed to excessive dietary salt intake. Several investigators are preaching to drastically reduce dietary salt, and are waving the attractive image of the bon sauvage, the simple savage, who does not eat salt and is free from hypertension[34]. I have listed above the

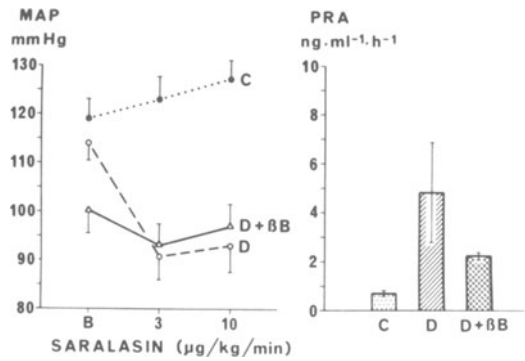

Fig. 3. Effects of associating the beta-blocker, propranolol (ßB) 160 mg/day for 1 week with the diuretic, chlorthalidone (D), in 12 patients whose plasma renin activity rose above 2.0 $nmol.^{-1}.h^{-1}$ after administration of the diuretic alone. Left: effects on mean arterial pressure (MAP) of saralasin infusion starting from a sitting baseline (B) in control untreated conditions (C, filled circles and dotted line), after two week's diuretic therapy (D, hollow circles and interrupted line) and after one week's combined therapy with diuretic and beta-blocker (ßB, hollow triangles and continuous line). Right: effects on supine plasma renin activity (PRA) (from Zanchetti et al.[33]).

many unanswered questions about the nature and the meaning of membrane abnormalities related to hypertension, and I would like to end by stressing that clinical and preventive application of these concepts must wait until an answer is provided to the many questions unsolved. Until more is learnt of the physiopathology of hypertension, extrapolation of hypotheses to the practice of medicine and, particularly to preventive medicine, is likely to bring about more disadvantages than benefits[35].

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CONTROL AND TREATMENT OF ARTERIAL HYPERTENSION

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Control of hypertension can be interpreted in two ways. According to the WHO Technical Report "Arterial Hypertension" (1978), "the term 'hypertension control' includes all measures for health protection and promotion related to high blood pressure", and "control programs" refer to public health actions aimed at hypertension control. The other meaning applies to the efficacy of therapy in the sense that high blood pressure of the patient should be kept under control. In other words, aspects of community programs of hypertension and of individual antihypertensive treatment have to be considered when we speak of hypertension control. This lecture will mainly deal with effective drug treatment of high blood pressure. However, in view of the large number of subjects who have elevated blood-pressure levels and the possible public health and economic consequences that may arise if drug therapy is generally recommended, it is advisable to discuss, at the beginning, some of the large controlled studies in which the significance of effective blood-pressure control for the community was investigated.

The first reliable data on the effectiveness of antihypertensive drugs in reducing morbidity and mortality caused by high blood pressure have been presented in the Veterans Administration Cooperative Study (1967, 1970). It was demonstrated that in patients suffering from severe hypertension, with diastolic pressures between 115 and 129 mm Hg, and in those with moderate hypertension, having an average diastolic pressure of between 105 and 114 mm Hg, complications of hypertension, such as cerebrovascular events and congestive heart failure, were definitely reduced, whereas statistical significance was not reached for the diminution of myocardial infarction. In subjects with diastolic pressures ranging from 90 to 104 mm Hg, no statistically significant reduction in morbidity and mortality due to

cardiovascular events was obtained in the treated group as compared with the placebo control group (1970). Since then, various controlled trials of the treatment of mild hypertension have been undertaken and, with the exception of that organised by the British Medical Research Council (1981), been finished and evaluated.

An interim analysis of the four completed trials has recently been published (WHO/ISH Mild Hypertension Liaison Committee, 1982) which compares the design of the projects and their results. It also refers to the trials which are still in course, mainly that of the Medical Research Council (MRC) in Britain and that of the European Working Party on High Blood Pressure in the Elderly (EWPHE) (Amery and De Schaepdryver, 1981). The results of the various studies differ: the positive outcome of the Australian National Blood Pressure Study (ANBPS) (1980) and of the American Hypertension Detection and Follow-up Program (HDFP) (1979) has not been confirmed in the US Public Health Service Study (USPHS) (McFate Smith et al., 1979) and the Oslo Study (Helgeland 1980) (Table 1). The reason for these discrepancies is mainly the insufficient sample size in the two latter trials, which failed to reveal a positive effect of treatment on the frequency of cardiovascular events. However, care is also indicated with respect to the interpretation of the two positive studies, of which the American HDFP was not a placebo-controlled trial, but a comparison between two ways of treatment - either in special clinics (stepped care) or with the usual medical care in the community (referred care).

From the available data it is obvious that the annual risk in individuals with mild hypertension is small. In the untreated control groups, mortality of all causes (not only cardiovascular) was 0.5% in the Australian Study, the Oslo Study, and the MRC trial. It was higher in the HDFP Study, in which patients with mild hypertension had a 1.1% mortality rate per year under referred care, but only 0.7% per year under the stepped-care management (WHO/ISH Mild Hypertension Liaison Committee, 1982). In the ANBPS, fatal ischaemic heart disease was less frequent in the treated group than in the placebo group, but the absolute numbers were small (2 vs 8 patients), and for non-fatal myocardial infarction no similar difference was observed (18 vs 17 patients). In addition, the follow-up of the placebo group revealed that, after the three years of the study, the blood pressure fell further in 48% and rose in 12%, whereas it remained in the mild hypertension range in 32% (Report by the Management Committee of the Australian Therapeutic Trial in Mild Hypertension, 1982). From these data it may be concluded that a considerable percentage of subjects with mild hypertension does not need drug treatment, but that it suffices to observe them closely and to watch the course of their blood pressure. Nevertheless, it has to be kept in mind that, in a population control study of middle-aged male civil servants in London, a blood-pressure dependent rise in mortality has been calculated, the cause of death being either coronary heart disease or stroke (Rose, 1981).

Table 1. Crude Mortality Rates (per 1000 person-years) in Treated Subjects and Controls* in A.N.B.P.S., H.D.F.P., (Stratum I), and the Oslo Trial, and in Controls in the M.R.C.Trial

Trial	Diastolic blood-pressure (mm Hg)	Age range (yr)	Person years	Mortality									
				All causes		All cardio-vascular		All cerebro-vascular		Coronary heart disease		cardio-vascular	
				No.	Rate	No.	Rate	No.	Rate	No.	Rate	No.	Rate
A.N.B.P.S.													
Treated	} 95-109	30-69	6991	25	3.6	8	1.1	3	0.4	5	0.7	17	2.4
Controls			6868	35	5.1	18	2.6	6	0.9	11	1.6	17	2.5
H.D.F.P.													
Stepped care	} 90-104	30-69	19115	231	12.1	122	6.4	17	0.9	86	4.5	109	5.7
Referred care			19063	291	15.3	165	8.7	31	1.6	107	5.6	126	6.6
Oslo													
Treated	} <110	40-49	2233	10	4.5	7	3.1	0	0.0	6	2.7	3	1.3
Controls			2088	9	4.3	6	2.9	2	1.0	2	1.0	3	1.4
M.R.C.													
Controls	90-109	35-64	16415	83	5.1	46	2.8	7	0.4	35	2.1	37	2.3

*Stepped care and referred care for H.D.F.P.

So far, no indicators are available that would predict whether a patient will benefit from drug treatment of mild hypertension. In the ANBPS, analysis of the placebo controls revealed that one fourth of the subjects who had a diastolic blood pressure below 95 mm Hg on the first three visits would be hypertensive three years later, if they were left without treatment, whereas the remaining 75% would still have pressures below 95 mm Hg (Report by the Management Committee of the Australian Therapeutic Trial in Mild Hypertension, 1982). This, however, means that a substantial group would receive unnecessary treatment, if antihypertensive drugs were automatically prescribed to subjects with mild hypertension. Hence, the decision to treat mild hypertension should not only be based on the values of diastolic blood pressure, but has to take into account other factors as well that are characteristic for the individual, such as age, sex, body weight, and possible risk factors - for instance high serum lipids, cigarette smoking, and others - which may be of greater significance for the manifestation of cardiovascular complications, especially ischaemic heart disease, than a slight elevation in blood pressure (Bauer and Hunyor, 1978).

In connection with the observations made in the HDFP Study and the demonstration that stepped care in special clinics may produce better results - not only with respect to cardiovascular complications of blood pressure - it should be mentioned that, also in uncontrolled studies, care of hypertensive patients was somewhat more effective in hospital out-patient departments than in general practice. However, after hospital treatment, discharge back to general practitioners may result in a satisfactory control (Bulpitt et al., 1982).

The fact that it is possible to lower the risk of fatal and non-fatal myocardial infarction by the treatment of mild hypertension should not let us forget that the number of those who benefit from preventive treatment is only a fraction of those who are treated. The mass approach to preventing complications of mild hypertension means little benefit for the individual, or, as has been stated recently: "A measure that brings large benefits to the community offers little to each participating individual" (Rose, 1982). There is the danger that, on the basis of positive findings in epidemiological studies, generalizing recommendations will be made to apply drug treatment to all individuals with mild hypertension instead of making a more careful selection considering additional criteria. Our efforts should aim at singling out those who are at a higher risk than at undifferentiated mass treatment. Keeping this in mind, we may now turn to the various possibilities of effective treatment.

GENERAL MEASURES

At the beginning of active treatment, especially in patients with mild to moderate hypertension, general therapeutic measures

are advisable. These include weight reduction by means of appropriate dietary regimens, restriction of salt intake to 4-6 g daily, cessation of cigarette smoking, and moderation in alcohol consumption. The modest reduction in salt intake is generally recommended, but few reliable data are available so far showing that it really affects high blood pressure (Morgan et al., 1979). Further controlled studies on the effect of a limited dietary salt restriction are necessary, particularly in comparison with the administration of low doses of diuretics. Physical activity has been claimed to diminish high blood pressure, but there is little evidence to support such a statement; there is at least no indication that physical inactivity enhances the risk of developing high blood pressure. Psychological stress is said to contribute to chronic blood-pressure elevation, but here again no convincing data are in hand, and for this reason the ill-defined term and the corresponding claim should be avoided. Similarly, various behavioral procedures have been said to be effective in lowering high blood pressure such as biofeedback methods, yoga, relaxation, transcendental meditation, and psychotherapy (Patel, 1975a, b). All these practices take a long time, are expensive, and may at best bring transitory relief - they are certainly not suitable for use on a large scale (Andrews et al., 1982). Hence, taken together, the effects of general measures on high blood pressure remain limited and will in most cases assist drug treatment rather than replace it.

ANTIHYPERTENSIVE DRUGS - THE PRESENT SITUATION

Today, an abundance of active drugs is available for the treatment of hypertension, and practising physicians are often confused about the various pharmacological possibilities of interfering with the regulation of blood pressure. The drugs differ with respect to their pharmacodynamic profiles, their mechanisms of action, their efficacy and potency, their pharmacokinetics and unwanted effects. To a certain degree, this holds also true for the various representatives within a class of drugs, and, consequently, not all diuretics, all β-blockers, or all vasodilators have identical profiles of activity and are interchangeable. Admittedly, minor differences are occasionally overstated, mainly for promotional reasons, although they are of little, if any practical significance. On the other hand, by far not all the diversities are trivial, and quite a few make it possible to adjust treatment better to the needs of the individual patient.

According to their mode of action, four large groups of antihypertensive agents can be distinguished (Table 2). Among the drugs which act on the efferent sympathetic system, a further differentiation is possible between those which act on centrally located adrenergic receptors and those which act preferentially in the periphery, mainly on the heart and the resistance vessels.

Table 2. Antihypertensive Agents Grouped According to their Mode of Action

1. Drugs that interfere with the efferent sympathetic system, including the α- and β-adrenoceptors
2. Drugs acting directly on arteriolar smooth muscle (vasodilators)
3. Drugs affecting electrolyte-water balance, and, secondarily, total peripheral resistance
4. Drugs that interfere with the renin-angiotensin system

Monotherapy

According to the recommendations in the WHO Technical Report on Arterial Hypertension (1978), treatment should usually start with one drug in the form of monotherapy, either with a β-adrenoceptor blocker or with a diuretic (Table 3). Only subsequently, if no satisfactory response has been achieved with a single drug, combinations of various types of drug should be given. This principle has been accepted by national organisations, such as the national societies or leagues against hypertension, but it is obvious that not in all countries, for instance the Federal Republic of Germany, this suggestion is observed. Many physicians here prefer the use of fixed combinations right from the beginning instead of trying to achieve a satisfactory result with the minimum of drugs. In favor of such a policy it is often argued that the simultaneous administration of various drugs makes it possible to reduce the dosages of the individual components, which have not only an additive, but often also a synergistic action. Consequently, besides a more marked blood-pressure lowering effect, the incidence and severity of side effects and adverse reactions may be diminished. However, there is also the danger to introduce unwanted effects together with an additional drug, and therefore the principal requirement - to administer as few drugs as possible - remains valid.

The debate about the criteria for the first choice in monotherapy still goes on (Withworth and Kincaid-Smith, 1982). It has been claimed that in younger patients β-adrenoceptor blocking drugs are preferable, and that in older patients or in those in whom the blood pressure has been elevated for a long time the treatment may start with a diuretic. However, it is difficult to draw a dividing line between older and younger individuals, and there are also quite a few exemptions from such a generalizing rule. Some years ago, the activity of the renin-angiotensin system was also suggested as selection criterion, patients with high or normal plasma renin activity being candidates for β-blocker therapy and those with suppressed activity for a diuretic (Bühler et al., 1972); but also here, too

Table 3. Drugs to be used for Monotherapy

Standard drugs:
β-adrenoceptor blockers
relatively cardioselective ($\beta_1 > \beta_2$), non-selective ($\beta_1 \sim \beta_2$)
or
diuretics:
thiazides, indapamide
thiazides + potassium-sparing drugs
Possible alternatives:
α-adrenoceptor blocker: prazosin (indoramin)
α- and β-adrenoceptor blocking: labetalol
converting enzyme inhibitor: captopril

many exceptions from the rule have been found besides the inherent difficulties of the test method (Report on Round Table, 1975, Morgan, 1976). The assessment of the haemodynamic situation may provide more helpful guidance but in many cases the trial-and-error principle has to be applied, and the choice is left to the personal experience and preference of the doctor (Waal-Manning, 1976; Gross, 1982a). This holds also true for the selection of one of the numerous representatives that are in hand in the two classes of drugs. All the available evidence indicates that the blood-pressure lowering potential of all β-adrenoceptro blockers is similar, provided they are given in the correct dosage, and the same holds true for the thiazides and related diuretics (Gross, 1982b).

β-Adrenoceptor blockers as well as diuretics cause only a moderate fall in blood pressure, which rarely exceeds 25 mm Hg systolic and 15 mm Hg diastolic, the maximum response being generally achieved after one or a few weeks of treatment. A positive correlation has been found between the height of diastolic pressure at the beginning of diuretic therapy and the reduction of pressure obtained after several weeks (Turner, 1977). Whereas diuretics are usually prescribed in relatively small doses, corresponding to 25 or, at the most, 50 mg of hydrochlorothiazide per day, the β-adrenoceptor blockers are, as a rule, given in higher doses than for the treatment of angina or tachyarrhythmias. A slow increase in dosage may result in a better response of blood pressure, but the dosage range is variable, and the dose-response curve for the blood-pressure lowering effect is shallow and does not follow closely that for antagonizing the effects of isoprenaline on the blood pressure. This holds also true for the delayed onset and for the duration of action, which exceeds that derived from measuring plasma concentrations.

Various arguments have been brought forward in favor of using relatively cardioselective β-adrenoceptor blockers in the treatment of hypertension. However, no convincing data have been presented which support a better efficacy or tolerability of drugs which act preferentially on β_1-adrenoceptors. The attempt to distinguish between various generations of β-adrenoceptor blocking drugs is a promotional gag rather than a scientific reasoning. The intrinsic sympathetic activity and the duration of action may be of greater significance for the selection of the drug, the former being rather unwanted in the treatment of high blood pressure, the latter such that it permits the administration of one dose every 24 hours. Recently, it has been shown that blockade of β-adrenoceptors may be responsible for changes in plasma lipid concentrations, an increase in total and very low-density lipoprotein (VLDL) triglycerides, and a decrease in high-density lipoprotein (HDL) cholesterol and free fatty acid concentrations (Waal-Manning, 1976; Day et al., 1982; Leren et al., 1980). These changes in plasma lipid concentrations are ascribed to a marked stimulation of α-adrenoceptors, which results in an inhibition of lipoprotein lipase and a subsequent rise in plasma triglycerides. Excessive α-adrenoceptor stimulation has also been claimed to impair the production of HDL cholesterol in patients who had already a low HDL cholesterol concentration before the administration of β-adrenoceptor blockers (Day et al., 1982). However, it has not been proven that these changes in plasma lipids are of clinical significance, even if they are maintained for some time.

Special mentioning merit β-adrenoceptor blocking drugs such as labetalol, which, besides its affinity to β_1- and β_2-adrenoceptors, has an affinity to α_1-adrenoceptors, the ratio of β- to α-blocking activity being about 4:1. The lower affinity to α_1-adrenoceptors, however, suffices to induce a more marked blood-pressure lowering effect than is achieved with exclusive blockade of β-adrenoceptors and which may even result in occasional overshooting orthostatism (Prichard and Richards, 1982). So far, labetalol is the only marketed representative of drugs which bind to both types of adrenoceptors, but others may be expected which have more favorable pharmacokinetics and a longer duration of action than labetalol. In any case, this type of adrenoceptor blocking drug is suitable for monotherapy and may be administered at the beginning of treatment.

In this context, the question arises whether a selective α_1-adrenoceptor blocking drug, such as prazosin, is suitable for monotherapy. The drug has continuously gained acceptance after problems of orthostatic responses, produced by the first administration, had been overcome by the lowering of the initial dose (Brogden et al., 1977). Although prazosin may be used alone, it is preferably given together with a diuretic to keep the dosage down also during prolonged treatment and to avoid sodium fluid retention (Bolli et al., 1980). The lack of tachycardic response as well as the reduction in cardiac preload are haemodynamic advantages of prazosin, but the

danger of initial orthostatic reactions, the frequently observed development of tolerance, and the subsequent increase in dose, necessary to obtain a reliable reduction in blood pressure, make it advisable to use the drug carefully. Hence, it is not suitable for initial monotherapy.

Among the diuretics, it is especially hydrochlorothiazide, bendrofluazide, and the long-acting chlorthalidone, which are the standard drugs in this kind of monotherapy. The question arises whether they should be given routinely together with potassium supplements, as is widely done in the USA and the UK, or whether it would be preferable to add a potassium-retaining diuretic to them. In general, potassium loss is limited, and serum potassium concentrations rarely fall below 3.5 mEq/l, provided the doses remain in the low range, corresponding to 25 mg hydrochlorothiazide or even less of chlorthalidone. The fact that supplements of potassium chloride are much more frequently administered in the USA than in Germany or other European countries may be explained by the generally higher doses of the diuretics that are given in America as compared to Europe. It has, however, to be kept in mind that potassium chloride, administered together with a diuretic, is excreted rapidly and does not cause a rise in the serum concentration of potassium (Lowe et al., 1979). In the majority of cases, there is no need for potassium supplements, provided that dietary potassium supplementation is adequate. The same holds true for the combination of two diuretics, a thiazide derivative or a related drug together with a potassium-conserving drug such as triamterene or amiloride. The occurrence of hypokalaemia may be reduced by such a combination, but it has also been demonstrated that, in aequi-effective antihypertensive doses, the addition of a potassium-conserving drug may have no special effect on serum potassium concentration (Anavekar et al., 1979).

Indapamide merits special mentioning, since it differs chemically from the thiazides and has in addition peculiar pharmacokinetic features, which are responsible for a prolonged duration of action. Besides the diuretic activity, a vasodilatory effect is claimed for indapamide, which may be demonstrable in low doses affecting electrolyte and water excretion little (Anavekar et al., 1979; Passeron et al., 1981). Hence, the request to give low doses of diuretics in the treatment of hypertension may be more easily realized with indapamide than with other diuretics owing to its vasodilatory component, its high lipophilicity, and its slow excretion (Wheeley et al., 1982).

Like the β-adrenoceptor blockers, also the diuretics may be responsible for slight changes in the lipoprotein pattern of the plasma. A mild increase in VLDL lipoproteins has been shown simultaneously with a reduction in HDL cholesterol, but it remains doubtful whether any clinical significance may be attributed to these variations. The diuretics have now been established in the treatment of

hypertension for 25 years and may be considered one of the best-tolerated groups of drugs, being nearly free from severe adverse reactions.

In the future, a third group of drugs may be more widely used in the monotherapy of hypertension, namely the vasodilators which act by interfering with calcium transport in the smooth-muscle cells of the arterioles (Leonetti et al., 1981; Muiesan et al., 1981). These drugs, also called calcium antagonists, have been widely used in the symptomatic treatment of angina, but they also lower total peripheral resistance. Usually, they cause an increase in heart rate by stimulating the baroreceptors. An exception is verapamil, which reduces heart rate by direct action on the atrioventricular conductivity. Besides verapamil, nifedipine and diltiazem have been used in the treatment of hypertension, but further derivatives, with possibly a longer duration of action, may be expected soon (Hulthén, U.L. et al., 1982; Krebs et al., 1982; Bühler et al., 1982 in press). As long as reflex tachycardia will be marked, the calcium-channel blockers will probably not be used in monotherapy, but only together with β-adrenoceptor blocking drugs. However, as soon as derivatives will arrive which do not increase heart rate or only transitorily so, these drugs may find a place in the initial treatment of high blood pressure.

Combined Treatment

In the step-wise treatment of hypertension, monotherapy is succeeded by the simultaneous administration of two or more drugs which act by different mechanisms on the regulation of blood pressure. The most obvious combination is that of β-adrenoceptor blockers and diuretics, which have a partly additive effect on blood pressure and in most cases cause a more pronounced lowering than either drug alone. However, since treatment has usually been started with either a β-blocker or a diuretic, it could also be that the response to the added component is more marked, and that this second drug might be more suitable for monotherapy than the drug used primarily. To make sure that it is really necessary to give two drugs, the other component would have to be tested alone, before being joined to the drug first used. In countries where drug combinations are preferred to single drugs, such as in the Federal Republic of Germany, already numerous fixed-ratio combinations of diuretics and β-adrenoceptor blockers are available; in other countries, such as Sweden, there is none so far, and the two types of drug have to be administered separately, with the advantage that they can be dosed individually allowing a greater flexibility.

The large number of available antihypertensive agents allows numerous combinations, but it has to be requested that the two or more drugs given together should not only differ with respect to

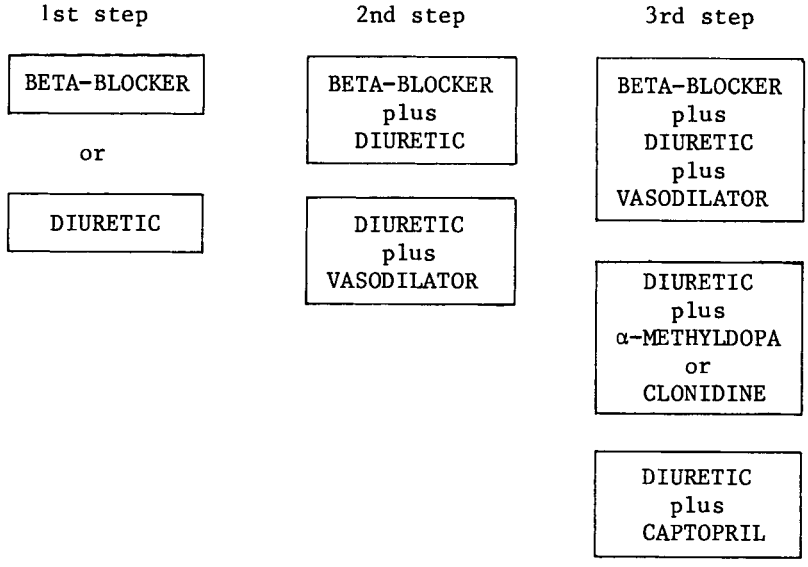
their mechanisms of action, but also act synergistically and correspond to one another with regard to their duration of action. In most cases, combination therapy includes a diuretic in low dose, which generally enhances the action of the other component(s). Recently, there has been the tendency to add a combination of two diuretics, including a potassium-conserving one, but in most cases this is unnecessary, especially in fixed-ratio combinations of antihypertensives, provided the diuretic dosage is kept low.

Not all the available and possible combinations can be mentioned, but only a few examples shall be given. If the simultaneous administration of a diuretic and a ß-blocker results in an unsatisfactory lowering of blood pressure, a vasodilator may be added - either one of the hydralazine type, or a blocker of the slow calcium-channels, or an α_1-adrenoceptor blocker (Table 4). Such a triple combination has the advantage of a peripheral mechanism of action and, provided the doses of the individual components may be varied, will allow adequate control in 75 to 80% of the cases.

Further possibilities are drugs that stimulate centrally located α_2-adrenoceptors, such as clonidine and α-methylnoradrenaline, the active principle of α-methyldopa (Table 4). New interest has arisen in clonidine, because it has been found to be active in smaller doses than often recommended previously, i.e. in the order of 50 to 100 mcg, and causes therefore fewer side effects. Guanfacine is very similar to clonidine, and the same is the case with guanabenz. Guanethidine, which, twenty years ago, was a definite progress in antihypertensive therapy as the first specific peripheral adrenergic neurone blocker is hardly used any more, since it is poorly absorbed, causes orthostatic reactions and other unwanted effects. All these drugs are hardly given alone, but together with at least a diuretic and mostly with a further drug in addition.

Quite recently, captopril, which interferes with the renin-angiotensin system by inhibiting the angiotensin I converting enzyme, responsible for the formation of the active octapeptide angiotensin II, has been increasingly used in the treatment of severe hypertension, often in patients who had been resistant to other types of drug treatment (Atkinson et al., 1980; White et al., 1980). The mechanism by which captopril and other drugs, which inhibit the activity of the converting enzyme or kininase II similarly, interfere with the blood-pressure regulation is not quite clear. The activity of kininase II is not completely blocked by captopril, and some angiotensin II is still present, but the main difficulty in understanding the antihypertensive action of the drug is the fact that in essential hypertension the renin-angiotensin system is of little, if any, pathogenic significance. Nevertheless, the success of captopril has stimulated research in this area, and similar drugs, characterized as converting enzyme inhibitors, may be expected soon. Of these, MK 421 or enalapril, which has a longer duration of action

Table 4. Stepwise Treatment of Hypertension



than captopril, is already quite advanced in clinical trials. Captopril itself and probably other converting enzyme inhibitors have no marked antihypertensive activity, unless the renin-angiotensin system is stimulated. By giving a diuretic simultaneously, preferably a loop-diuretic, such as furosemide, the sensitivity to captopril is enhanced, and this may be a suitable means to obtain a satisfactory response. On the other hand, the combination of captopril with a β-blocker does not enhance the blood-pressure lowering effect in essential hypertension in a similar way as the addition of a diuretic (MacGregor et al., 1982). This is a good example demonstrating that combinations of antihypertensive drugs have to be selected carefully. Even less clear than the antihypertensive mechanism is the beneficial haemodynamic effect of captopril in congestive heart failure. Here, not only afterload is reduced by a reduction in total peripheral resistance, but also preload, since the capacitance vessels are also dilated under the influence of the drug. The onset of action is quite prompt, and it seems that in most cases it is not necessary to add a diuretic. It will, however, take some more time until the place of this type of drug in the treatment of congestive heart failure is definitely established.

Treatment of Refractory Hypertension

In the relatively small number of patients in whom the usual stepped-care regimen (including a diuretic, a β-adrenoceptor blocker,

and a vasodilator) has not resulted in a satisfactory control of blood-pressure, various drugs have been tried that have a more drastic blood-pressure lowering effect. In a recent article, three of these drugs - diazoxide, minoxidil, captopril - and a combination including prazosin have been comparatively studied in the treatment of refractory hypertension (Swales et al., 1982). In such patients, care has to be taken not to lower the blood pressure too abruptly, since cerebral hypoxia with all its consequences or myocardial infarction may occur, if the blood pressure falls suddenly to levels that are far beyond those to which the perfusion of the tissues is adapted (Ledingham and Rajagopalan, 1979). Minoxidil is certainly the most powerful vasodilator, but it has to be given together with a diuretic and a β-blocker (Figure 1) (Tenschert et al., 1980); captopril has a lower efficacy, which, however, can be increased by the addition of a diuretic. The quadruple therapy, which consists of the addition of prazosin to the standard regimen of a diuretic, a β-blocker, and hydralazine, is considered quite complex and may only be suitable in the hospital, since problems of compliance may arise at home. Nevertheless, by suitable use of the drugs available today nearly all hypertensives can be satisfactorily controlled and the progress of cardiovascular complications stopped. Although such complex treatment regimens are anything but ideal and may include adverse reactions, one should keep in mind that, 25 years ago, these patients would not have survived, but would have died of their disease within a few months or years.

WHAT ARE THE PERSPECTIVES?

To come back to the beginning, one has to consider the prospects for prevention and treatment of high blood pressure. Since we have numerous active drugs in hand which enable the control of high blood pressure - admittedly not ideally, but with a reasonable benefit/risk as well as benefit/cost ratio - it would be more important to concentrate on developing preventive measures. However, prevention of a chronic disease, of which the causes are unknown, is much more difficult to realize than treatment and may be effective only if substantial and continuous efforts are made which may not be feasible economically. Furthermore, despite the fact that preventive interventions are generally suggested - such as weight control, reduced salt intake, physical activity, behavioral education, and the avoidance or elimination of adverse psychological and social factors - hardly any convincing data are available which would justify definite recommendations for the time being. Many more studies have to be undertaken to answer at least part of the unsolved questions and to provide a more solid basis for proposing changes in behavior, dietary habits, or professional activities. We have to concede that there is more belief than facts in supporting general measures as being effective in the control of high blood-pressure. This unsatisfactory situation makes it urgent to start well planned long-term studies to provide the urgently needed data.

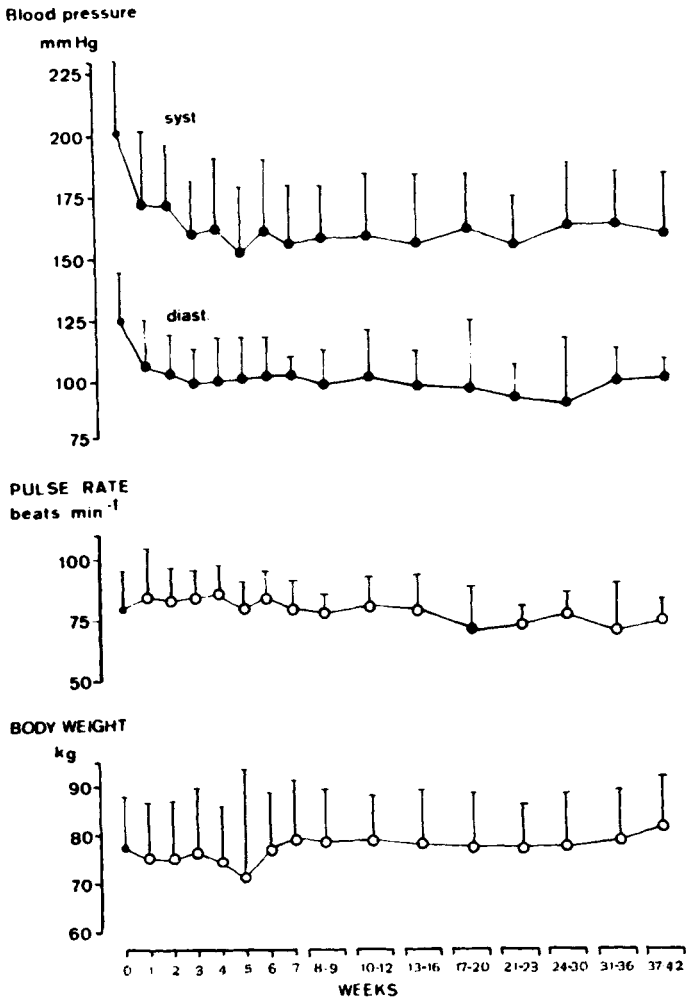


Fig. 1. Supine systolic and diastolic blood pressure, pulse rate and body weight in 22 patients with severe hypertension. Solid circles (●) indicate significant differences ($p < 0.05 - < 0.001$) to initial values, open circles (○) non-significant changes during the observed period of 42 weeks.

The easier way will be to develop further drugs for the treatment of hypertension, and here we shall see quite a few more come up. Since most possibilities of interfering pharmacologically with the regulation of blood pressure have already been extensively studied and since corresponding drugs are in hand, it may hardly be expected that there will be principally new types of antihypertensives. One of the new possibilities might be the interference with

the central serotoninergic system, and the development of ketanserin, a blocker of serotonin$_2$-receptors ($5HT_2$) could be a step in this direction. The drug is said to have a specific affinity to $5HT_2$-receptors, but it has recently been demonstrated that it binds also to α_1-adrenoceptors (Van Nueten et al., 1981; Fozard, 1981). A blood-pressure lowering effect has been obtained in hypertensive patients (DeCree et al., 1981; Wenting et al., 1982) but it remains to be shown that it is due to blockade of $5HT_2$ receptors and not mainly to blockade of α_1-adrenoceptors. Of course, continuous, more or less conspicuous improvements of already existing pharmacological principles will be made, with the result that the number of antihypertensive drugs will increase within the forthcoming years. This will not necessarily mean that the new drugs will be definitely better than those in hand today; they may have marginal advantages in one or the other direction, but it is hard to imagine that there will be a breakthrough comparable to that during the period from 1953 to 1962, when effective control of high blood pressure became possible for the first time.

If we consider the requirements for an ideal antihypertensive drug, as they have been defined many years ago (Table 5), we may say that most of them are satisfactorily met with the drugs available today. However, there still remains the request for improved tolerability, although we have to admit that quite a few of the drugs now in hand are remarkably well tolerated. We must also make it clear that there will be no ideal drug, and that all antihypertensives, those which are at our disposal and those to come, will have some unwanted effects and will have to be carefully studied in that direction.

The main problem to be settled is the control of mild hypertension, by far the most important from the community point of view. Should mild hypertension be treated in all subjects in whom it has been diagnosed, and should the elevated blood pressure be lowered by regular long-term intake of drugs? It is reassuring to know that effective treatment of severe and moderate hypertension is possible

Table 5. Requirements for an Antihypertensive Drug

1.	Decrease in blood pressure: slow onset, not too pronounced, similar in supine and standing positions
2.	Duration of action: 12 to 24 hours
3.	Active by mouth
4.	No negative inotropic effect on the heart, no tachycardia
5.	No tolerance
6.	No or only mild adverse reactions or side effects, no reduction of physical capacity and mental alertness
7.	Suitable for long-term treatment

today, and that cardiovascular complications may be prevented to a substantial degree. However, it is at least as important to avoid unnecessary treatment and not to expose all those who have a mild rise in blood pressure to drugs that may cause some risk without providing any benefit. More research is needed along these lines to improve further the control and treatment of hypertension.

In concluding, a few rules for the management and control of hypertension should be given (Table 6). It is not to be expected that there will be a change in these general rules in the future, and this holds true not only for the treatment with the drugs we have in hand, but also for the preparations that may be available tomorrow.

Table 6. Management of Hypertension, General Rules for Drug Treatment

1. Gradual and slow lowering of blood pressure
2. Special care with old-age people; avoid abrupt changes, no strong-acting diuretics
3. Lowering of blood pressure to be achieved with minimum number of drugs
4. Application of the stepped-care program
5. One or two doses per day
6. Preference for peripherally acting drugs

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BIOCHEMICAL MECHANISMS OF ALTERED METABOLISM IN ISCHEMIC HEART

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Oxygen deficiency in heart muscle is most commonly induced by reduction in coronary flow, referred to as ischemia. As contrasted to hypoxia or anoxia in which flow of blood with low or zero oxygen tension is maintained, ischemia leads to accumulation of metabolic products that further modify rates of biochemical reactions. After periods of severe ischemia ranging from 30 minutes to 1 hour or more, irreversible damage occurs.[1] Damage of this severity is characterized by disruption of the plasma membrane that is preceded by swelling of both the cell and mitochondria. Concurrently, the myofibrils and intercellular junctions are disrupted, and there is margination of nuclear chromatin. Reperfusion of an irreversibly-injured cell leads to accumulation of Ca^{++} within the mitochondria and failure to recover contractile activity.

In discussing the biochemical events leading to ischemic damage, attention will be directed to two aspects of cell metabolism. The first of these is energy metabolism. In oxygen-deficient muscle, formation of high-energy phosphates depends upon efficient utilization of the oxygen that remains by mitochondrial oxidative phosphorylation, and upon generation of ATP via the glycolytic pathway. As will be discussed, generation of high-energy phosphates by both of these pathways may be impaired by accumulation of metabolites. The second aspect of deranged metabolism in ischemic muscle to be discussed will be destruction of cellular organelles by hydrolytic enzymes. Activation of the lysosomal system has been observed frequently in ischemic tissue, including muscle.[2-7] Lysosomal enzymes possess the potential to hydrolyze macromolecules, such as proteins, nucleic acids and polysaccharides, and small molecular weight substances, such as phospholipids and metabolic intermediates. The possibility that

accumulation of metabolites may modify the hydrolytic activity of lysosomal enzymes and that a link may exist between depletion of high-energy phosphates and accelerated hydrolysis of cell constituents by lysosomal enzymes will be discussed.

Energy Metabolism of Ischemic Muscle. Metabolism of Glucose in Ischemic Hearts

In aerobic cells supplied with normal plasma levels of glucose and fatty acids, glycolysis makes a small contribution to generation of ATP. Approximately 20% of myocardial oxygen consumption is accounted for by oxidation of glucose; about 1% of ATP synthesis occurs in the passage of these glucose residues through the glycolytic pathway.[8] The steps restricting glycolysis in aerobic and ischemic muscle are shown in Table 1. The major steps restraining glycolysis in aerobic muscle are glucose transport, hexokinase, phosphofructokinase, and pyruvate dehydrogenase. The major oxidative substrate of heart muscle is free fatty acid. Oxidation of free fatty acid results in restraint of glycolysis at the transport, phosphofructokinase, and pyruvate dehydrogenase steps. The mechanism of the inhibition of transport is unknown, while phosphofructokinase is restrained by accumulation of citrate. Pyruvate dehydrogenase is inhibited by higher tissue levels of CoA and NADH, and by conversion of the enzyme to the inactive phosphorylated form. Inhibition of these steps accounts for preferential utilization of fatty acids by heart muscle.

Addition of insulin accelerates glucose transport as much as 30-fold, and shifts the major restraint on glycolytic rate to glucose phosphorylation. In aerobic hearts supplied only glucose, phosphofructokinase is the major reaction limiting glycolytic rate. Addition of fatty acids inhibits this reaction, as well as pyruvate dehydrogenase, and restrains glycolytic rate despite the presence of insulin.

So long as coronary flow is maintained, oxygen deficiency accelerates glycolysis, as a result of faster rates of glucose transport, hexokinase, and phosphofructokinase. The mechanism of acceleration of glucose transport is poorly understood, but appears to involve greater sensitivity of the transport process to stimulation by insulin, and perhaps a more direct effect, resulting from the fall in energy levels. Acceleration of hexokinase results from a reduction in tissue levels of glucose-6-P, a potent product inhibitor of the enzyme, and an increased tissue level of P_i, an activator. The fall in glucose-6-P is a consequence of an acceleration of phosphofructokinase due to decreased tissue levels of ATP, an inhibitor of the enzyme, and increased levels of AMP and P_i, activators of phosphofructokinase. In anoxic hearts, glycolytic rate is limited by flux through the glyceraldehyde-3-P dehydrogenase reaction. Activity of the enzyme is increased by the higher levels of the substrates, glyceraldehyde-3-P and Pi, but restrained by higher tissue concentrations of the product, NADH. NADH produced by glycolysis

Table 1. Rate-controlling reactions in glycolysis

Reaction	Effector	Direction of change
Glucose transport: extracellular glucose -------- intracellular glucose	insulin	increase
	anoxia	increase
	pressure development	increase
	fatty acid	decrease
Hexokinase: glucose + Mg ATP→ glucose-6-P + MgADP	ATP, ADP, P_i	increase
	glucose-6-P	decrease
Phosphofructokinase: fructose-6-P + MgATP → fructose-1,6 diP + MgADP	acid pH	decrease
	fructose-6-P	increase
	ATP, citrate	decrease
	P_i, AMP. ADP	increase
	fructose-1, 6-diP	increase
Glyceraldehyde-3-P dehydrogenase: glyceraldyhyde-3-P + NAD^+ + P_i--------1,3-diP glycerate + NADH + H^+	NADH	decrease
	1,3-diP-glycerate	decrease
Pyruvate dehydrogenase: pyruvate + CoA + NAD^+ → acetyl CoA + NADH + H^+ + CO_2	phosphorylation	decrease
	acetyl CoA, NADH	decrease

must be oxidized either by transport of reducing equivalents into the mitochondria via the malate-aspartate shuttle[9] or by conversion of pyruvate to lactate. When glycolysis is increased, production of NADH by glyceraldehyde-3-P dehydrogenase accelerates. Oxidation of this extra NADH by either conversion of oxaloacetate to malate or pyruvate to lactate requires increased levels of NADH to accelerate these reactions. The level of NADH thus increases in proportion to glycolytic rate and eventually reaches a concentration that is inhibitory to glyceraldehyde-3-P dehydrogenase.[10] Although ATP production via glycolysis may be increased 10-fold, inhibition of oxidative phosphorylation leads to as much as a 90% inhibition of overall ATP production. In association, contractile activity of anoxic hearts is severely impaired, as are the enzymatic reactions dependent upon the availability of ATP and other high-energy compounds.

In ischemic heart, energy depletion is complicated by accumulation of metabolites, particularly lactate and hydrogen ions. Glycolysis is inhibited, as compared to anoxic hearts. Rates of glucose transport, hexokinase, and phosphofructokinase are sufficiently rapid to provide saturating levels of substrate for glyceraldehyde-

3-P dehydrogenase. Flux through this enzyme is reduced however, by a fall in intracellular pH together with higher levels of NADH.[11] The pH optimum of glyceraldehyde-3-P dehydrogenase is above pH 8. In ischemic muscle, the average intracellular pH was about 6.8 as compared to a pH of 7.0 in aerobic hearts.[12] Accumulation of lactate interferes with oxidation of NADH, causing the nucleotide to accumulate and glyceraldehyde-3-P dehydrogenase to be inhibited. These observations emphasize the importance of the accumulation of metabolites in producing the biochemical defects in glycolytic regulation found in ischemic hearts and indicate that low rates of flux through glyceraldehyde-3-P dehydrogenase in ischemic hearts are due to higher tissue levels of NADH and hydrogen ions.

Metabolism of Free Fatty Acids in Ischemic Hearts

Plasma levels of free fatty acids (FFA) are frequently elevated in the first 1 or 2 hours following a myocardial infarction and this increase has been implicated in frequency of serious ventricular arrythmias.[13] The free acid is the principal form that is utilized by heart muscle (for review[14]). The majority of serum FFA is bound to albumin. The amount of acid that is free in solution is small and is determined by the FFA/albumin molar ratio. The unbound pool of FFA is in equilibrium with a tissue pool of FFA (Fig. 1). The nature of the tissue pool is unknown, but it may be composed of FFA in the cytoplasm as well as FFA bound to intracellular membranes and soluble proteins. The first step in the cellular metabolism of fatty acids is their conversion to long-chain fatty acyl-CoA esters (FACoA). This process, referred to as activation, is catalyzed by long-chain acyl-CoA synthetases that are located on the outer mitochondrial membrane. The tissue levels of fatty acid and ATP are normally well above the K_m of the synthetases for these substrates. However, the concentration of free CoA (CoASH) in the cytosol is unknown, due to the fact that only 5-10% of total CoA is in the cytosol and is distributed among fatty acyl-CoA, acetyl-CoA and free CoA. The activity of the synthetases is inhibited by the products of the reaction, fatty acyl-CoA, AMP and PP_i.

After fatty acyl-CoA is formed, the fatty acyl moiety undergoes reactions which function to move the acyl group from the site of activation on the outer mitochondrial membrane to the mitochondrial matrix where it is oxidized. The first reaction is the transfer of the acyl group from CoA to carnitine (Carn). The second reaction(s) is transport of the acyl moiety across the inner mitochondrial membrane. The transport of acyl-carnitine (FACarn) involves an exchange reaction in which acyl-carnitine moves across the mitochondrial membrane in exchange for carnitine. This appears to be a 1:1 exchange which would mean that the content of carnitine on both sides of the membrane would remain constant under physiological conditions. The third reaction in this segment is the transfer of the acyl group

from carnitine to matrix CoA, forming fatty acyl-CoA that can be used for β-oxidation. Heart muscle has a high carnitine/CoA ratio in the cytosol, about 175, which would favor transfer of acyl units to carnitine, maintain low levels of fatty acyl-CoA in the cytosol, and direct acyl units away from lipid synthesis toward oxidation.

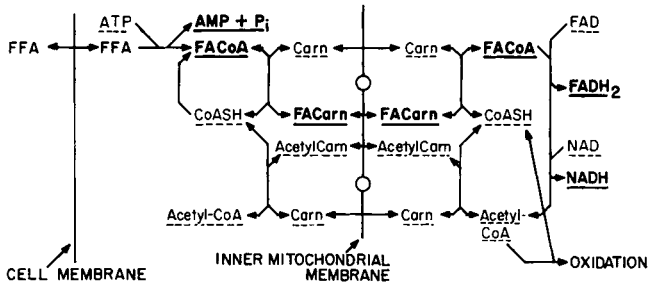

Fig. 1. Pathway for fatty acid oxidation in heart muscle illustrating intermediates that are elevated during ischemia in hearts perfused in the presence of fatty acid. This figure illustrates the two transferase systems for acyl units, each located on the inner and outer surfaces of the inner mitochondrial membrane. It includes compartmentation of CoA in two non-exchangeable pools (cytosolic and mitochondrial matrix); carnitine translocase is shown on the inner mitochondrial membrane between the two transferase systems. Metabolites that increase in ischemic hearts are shown with dark letters and are under-lined with a solid line; those that decrease are shown with light letters and are underscored with a broken line.

Following production of fatty acyl-CoA in the matrix, the next major sequence of reactions involves the β-oxidation system. Four separate enzymes are involved, and the overall reaction results in conversion of a two-carbon unit of fatty acyl-CoA to acetyl-CoA along with the reduction of NAD and FAD (1 mole each) to NADH and $FADH_2$. In order for the rate of β-oxidation to be sustained, the reduced nucleotides must be oxidized by electron transport, while acetyl-CoA is oxidized by the citric acid cycle.

In heart muscle, acetyl-CoA produced by β-oxidation can be used at appreciable rates only for oxidation by the citric acid cycle. Flux through the citric acid cycle is geared to the rate of oxidative phosphorylation through feedback control by changes in the levels of high energy phosphates and NADH. The only alternative route for the

disposal of mitochondrial acetyl-CoA is transfer of the acetyl unit across the mitochondrial membrane to cytosolic carnitine where it is stored as acetyl-carnitine and provides a buffer against large changes in mitochondrial acetyl-CoA.

Under oxygen-deficient conditions such as ischemia or anoxia, the amount of oxygen available to support oxidation by the citric acid cycle is reduced (Figure 1). The levels of $FADH_2$ and NADH increase, and β-oxidation becomes inhibited. Levels of acetyl-CoA fall while the long-chain acyl derivatives of CoA and carnitine increase to very high levels. Since 95% of the total CoA is mitochondrial, most of the increase in acyl CoA must occur in this organelle. On the other hand, 95% of the total carnitine is cytosolic and most of the acyl carnitine must accumulate on the outside of the inner mitochondrial membrane. The uptake and activation of fatty acid is reduced. High levels of fatty acyl-CoA have been shown to inhibit the acyl-CoA synthetases[15-18] and adenine nucleotide translocase[19-20] in a specific manner, and many other enzymes in a non-specific way.[21] Free fatty acids[20] and long-chain acyl-carnitine,[23] in addition to having a detergent effect at high concentration, have been shown to inhibit Na^+-K^+ ATPase. Myocardial ischemia, therefore, in addition to resulting in a reduced capacity for ATP synthesis results in accumulation of compounds which are potentially detrimental to myocardial function and metabolism.

The Lysosomal System and Protein Turnover in Ischemic Hearts

As noted in the introduction to this paper, there is biochemical and morphological evidence of activation of the lysosomal system in ischemic hearts. Release of lysosomal enzymes into the cytoplasm or accelerated formation of autophagic vacuoles could lead to damage of cellular constituents, including the plasma membrane, sarcoplasmic reticulum and mitochondria, and lead to irreversible injury. Proteins, nucleic acids and phospholipids could be attacked by the appropriate hydrolases. At present, only the effects of ischemia on protein turnover have been investigated in detail.

If protein synthesis were inhibited more markedly than proteolysis in oxygen-deficient muscle, proteins with rapid rates of turnover could be lost, and marked metabolic derangements would ensue. Both synthesis and degradation of protein are energy-dependent processes[7,24-26] but these processes are modified to varying degrees by anoxia or ischemia.

In anoxic as compared to ischemic hearts, nitrogen balance was more negative in the absence of insulin (Figure 2). Protein degradation was more severely inhibited in ischemic than anoxic hearts, both in the presence and absence of insulin. Factors that may account for the more profound inhibition of protein degradation in

ischemic muscle include accumulation of lactate and hydrogen ions.[7] In this regard, addition of a high concentration of lactate (50mM) or a reduction in perfusate pH to 6.8 reduced proteolysis in aerobic hearts by about 25%. When both lactate was added and pH reduced, proteolysis was inhibited about 50%. These findings indicate that inhibition of proteolysis in energy-poor muscle is due both to a fall in high energy phosphate stores and to accumulation of metabolites.

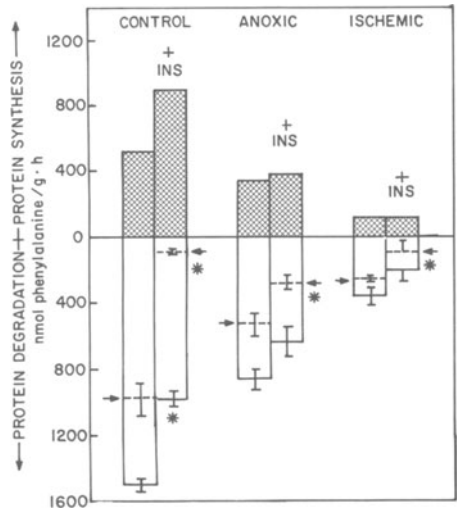

Fig. 2. Effects of ischemia and anoxia on protein synthesis, protein degradation and nitrogen balance. Protein degradation (plotted downward from the zero line) was calculated by measuring release of phenylalanine in the presence of cycloheximide - nitrogen balance was measured by assessing phenylalanine release in the absence of cycloheximide (shown by the broken lines and arrow heads). Protein synthesis was calculated by subtracting nitrogen balance from protein degradation (plotted upward from zero line; cross-hatched bars). Six hearts were perfused in each group with buffer containing 15mM glucose, normal plasma levels of 19 amino acids and 0.01mM phenylalanine. Insulin (25 mu/ml) was added as indicated. Data represent the mean ± S.E. *$p<0.05$ versus no insulin.

Changes in the size of lysosomes or in the total activity of lysosomal enzymes have been found to accompany a number of physiological and pathological modifications in the myocardium.[27] An increase in protein degradation in aerobic hearts perfused in the absence of insulin was associated with a decrease in the latency of cathepsin D.[28] These changes were associated with an increase

in the size of autophagic vacuoles within myocardial muscle cells.[29] When insulin was present, rates of proteolysis were reduced by 50%, and latency of lysosomal enzymes was increased.

Although accelerated rates of proteolysis was associated with decreased latency of lysosomal enzymes and appearance of autophagic vacuoles in aerobic hearts, this correlation was lost in oxygen-deficient tissue. As found in studies of liver lysosomes[2] activities of cathepsin-D were higher in homogenates of anoxic and ischemic hearts, even though the rate of protein degradation was inhibited.[7] The effects of anoxia and ischemia on enzyme activity are thought to reflect the greater fragility of autophagic vacuoles found within ischemic hearts.[6] Although ultrastructural changes suggest that tissue damage involved hydrolysis of cellular components by lysosomal enzymes, proteolysis was not accelerated. The latter finding indicates that the energy requirement involves initial steps in the proteolytic pathway and that anoxia and ischemia do not lead to accumulation of the products of proteolysis, peptides and free amino acids, within the heart.[7,30] These findings indicate that protein breakdown is inhibited in ischemic muscle, and that damage to this cellular component is unlikely to account for irreversible injury.

Concluding Remarks

In concluding this description of metabolism in the ischemic heart, an overview of the changes will be presented in an attempt to suggest common features and possible mechanisms of damage. Decreased coronary flow impairs oxygen delivery and removal of metabolic products. As a result, ATP is converted to ADP and AMP. Loss of total adenine nucleotide ensues, presumably due to hydrolysis of AMP by 5'-nucleotidase. Decreased levels of ATP interfere with ion pumping, Ca^{++} sequestration, and many other energy-dependent reactions. Reperfusion of ischemic hearts results in rapid resynthesis of creatine phosphate, but ATP levels remain depressed. Irreversible damage may result when ATP levels are too low to support critical cellular functions such as contraction, volume control and substrate activation. Metabolic intermediates in the pathways of glycolysis and fatty acid oxidation accumulate and inhibit further flux of substrate through these pathways. Inhibition of glycolysis impairs even further the capacity for ATP generation, while accumulation of fatty acyl-carnitine blocks the activity of Na^{+} -K^{+} ATPase and contributes to the difficulty in regulation of cell volume. The decrease in intracellular pH probably accounts for the initial fall in contractility through interference with the calcium cycle, and contributes to the fall in glycolytic rate. In some, as yet unknown manner, the lysosomal system is activated and may be involved in disruption of sarcolemmal, mitochondrial, and other membrane systems.

This damage does not involve destruction of cellular proteins. The fall in intracellular pH and accumulation of compounds with detergent activity may enhance membrane damage. Future work could profitably be focussed on determining the nature of the damage that results from lysosomal activation and in identifying the link between energy-depletion and activation of this system.

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CALCIUM AND CARDIOVASCULAR DISEASE

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INTRODUCTION

When the calcium-antagonist agents were initially used by Fleckenstein's group, it was found that beta-adrenoceptor agonists opposed the specific action of high dose verapamil (10^{-5}M) in inhibiting myocardial contractility (Figure 3 and 7 in Fleckenstein, 1971). The restorative effect of isoproterenol was very similar to that of an increased extracellular calcium (Figure 6 in Fleckenstein, 1971). Thus early thinking saw calcium-antagonists and beta-antagonists as having opposite effects on trans-sarcolemmal calcium flux. Some even argued that the calcium-antagonists had beta-blocking qualities, an argument that was laid to rest when it was found that verapamil was unable to inhibit an isoproterenol-induced tachycardia or inotropic response (Nayler et al, 1968). The explanation was that verapamil could not prevent catecholamine-induced increases in the tissue level of cyclic AMP nor the beta-mediated activation of adenyl cyclase; rather, verapamil blocked the trans-sarcolemmal calcium influx provoked in K^+ depolarized hearts by either catecholamines or by an increased external calcium (Watanabe et al, 1974).

This paper reviews the properties of calcium antagonist agents and evaluates a new hypothesis that trans-sarcolemmal calcium fluxes may play a role in provoking ventricular fibrillation (Thandroyen, 1982). This hypothesis is tested by examining the effects of calcium-antagonists on an isolated rat heart model. Previous data have suggested that beta-adrenergic stimulation can decrease ventricular fibrillation threshold in models of myocardial infarction (Lubbe et al, 1978).

Before evaluating this hypothesis, it is relevant to examine current concepts of the cellular modes of action of the beta-adrenoceptor antagonists and calcium-antagonists.

CONTRASTING PROPERTIES OF BETA-ADRENOCEPTOR BLOCKERS CALCIUM-ANTAGONISTS

Mode of action of beta-adrenoceptor antagonists

A beta-adrenoceptor antagonist is an agent that interacts with the beta-receptor to provoke a characteristic series of events which include, in the case of the myocardium, a positive chronotropic and inotropic stimulation. Many of the effects of beta-stimulation are thought to be mediated by cyclic AMP. Hence because beta-adrenoceptor antagonists competitively interfere with the binding between the beta-antagonist and its receptor, beta-antagonists should decrease the tissue cyclic AMP response to beta-antagonists.

Mode of action of calcium-antagonists

In contrast to this rather clear sequence of events, the definition of a calcium-antagonist is somewhat obscure. It is best to go back to the original descriptions by Fleckenstein (Fleckenstein, 1971). In his basic experiment, acute contractile failure of the guinea-pig heart was produced by a large over-dose (1 mg/kg) of verapamil. There was an abrupt rise in the venous pressure and fall in the arterial pressure which was reversed by the injection of calcium chloride. Thus the first property of the calcium-antagonist drugs is that they selectively abolish "the contractile response of the guinea-pig papillary muscle in a low concentration without any significant change in the single fiber action potentials". This effect is antagonized by the calcium ions (Fleckenstein, 1971). Each molecule of verapamil antagonizes approximately 200 calcium ions whereas in the case of other antagonists such as the compound D-600 or nifedipine, 1 molecule antagonizes up to several thousand calcium ions. The second quality of the calcium-antagonists is that although they appear to affect the action potential in no significant way, when voltage clamp studies are undertaken, they can be shown to "block the transmembrane calcium influx into the excited heart muscle fibers but do not affect the simultaneous sodium movements which are connected with the action potential" (Fleckenstein, 1971). This inhibitory effect of calcium antagonists on the calcium slow inward current is also opposed by the addition of excess external calcium.

Properties of calcium channel

To control the vast difference in the concentration between calcium in the extracellular fluid and the cytosol (a concentration gradient of about 10^3) requires regulation of the calcium transmission across the sarcolemma. There are several possible modes of calcium entry. First, calcium can enter via the calcium channel or calcium channels (which have not been characterized). The sodium channel has been the subject of numerous models, including recent modifications of the classical Hodgkin-Huxley concept of the three "m" activation gates and the single "h" inactivation gate (Weld et al, 1982). Such data and models are not available for the calcium channel although a similar pattern of interaction is sometimes postulated.

Therefore, at the moment, the understanding of the calcium channel is largely descriptive. Some of the properties of calcium channels are:

1. They are "opened" by a voltage stimulus as the resting potential depolarizes to about -40 mV (Noble, 1979).
2. They "close" as calcium influx ceases and potassium efflux is enhanced (Bassingthwaighte et al, 1976).
3. They "open" in the presence of sodium and calcium ions externally (Schneider et al, 1975).
4. They can be "opened" by beta-stimulation (Reuter, 1974; Reuter & Scholtz, 1977). It is not sure in the case of the myocardium whether these receptor-operated channels are the same as the channels which respond to the influx of sodium ions. In the case of vascular smooth muscle clear arguments have been made (Bolton, 1979; Towart, 1981; Van Breemen et al, 1982) for the differentiation between DOC (depolarization operated channels) and ROC (receptor operated channels). In the case of the myocardium, K^+-depolarization inhibits the sodium channel so that an increased voltage stimulation plus beta-stimulation may be required to "open" the calcium channel (Schneider et al, 1975); hence the concept of DOC's for the myocardium seem invalid.
5. The channels are "closed" or "blocked" by calcium-antagonist group of drugs which include verapamil, nifedipine and diltiazem. It is not clear whether these drugs have similar or different modes of action on the calcium channel. It is also not clear whether these drugs act predominantly on the outside of the sarcolemma, as suggested in the case of verapamil (Langer et al, 1975); more recent evidence proposes that verapamil is concentrated many-fold within the heart (Lullman et al, 1979) cell and may act deeply in the membrane or on the inner surface of the sarcolemma (Payet et al, 1980).

This emphasis of the effects of calcium-antagonists on the trans-sarcolemma should not obscure the growing evidence that there is also an intracellular site of action, for example on calmodulin (Boström et al, 1981; Hidaka et al, 1979).

Functionally different calcium channels

Not only is the nature of the calcium channel not clear, but there are apparent differences between the calcium channels in various tissues. Although an increased extracellular calcium concentration antagonizes the effect of high-dose verapamil in depressing myocardial contractility (Hamm et al, 1982), calcium has no such effect when it comes to the inhibitory action of verapamil on the sinus node; in that case externally added calcium does not "antagonize" verapamil but rather leaves unchanged its effect in inhibition of conduction (Hariman et al, 1979). That difference provides a valuable practical approach to the problem of depressed myocardial function sometimes encountered when patients are given verapamil intravenously - injected calcium salts should restore myocardial contractility without impairing the therapeutic effect of verapamil on the atrioventricular node.

There is also very recent evidence that the calcium channel of vascular smooth muscle is stimulated by alpha-2 antagonists such as clonidine and inhibited by alpha-2 antagonists such as yohimbine (Van Meel et al, 1981). Hence the postjunctional alpha-2 receptors appear to be intimately linked to calcium transport in vascular smooth muscle. No such data exist for the myocardium.

Thus, from the practical point of view, the calcium channels in nodal tissue, in myocardial tissue and in vascular smooth muscle, all appear to have somewhat different characteristics although they share the property of being the site of action of the calcium-antagonist agents.

Ca^{2+}-ANTAGONISTS AND VENTRICULAR FIBRILLATION

We have proposed the hypothesis that calcium ions are involved in the genesis of ventricular fibrillation (Thandroyen, 1982). The first step in the evolution of the hypothesis was the recognition that elevation of tissue cyclic AMP in ischemic tissue could be linked to the onset of ventricular fibrillation (Lubbe et al, 1978; Opie et al, 1979; Opie et al, 1980). Calcium was seen as the further "messenger" of cyclic AMP (Opie et al, 1982). Recent data emphasize the role of calcium rather than of cyclic AMP. We tested the hypothesis by examining whether the three first generation Ca^{2+}-antagonist agents (verapamil, nifedipine, diltiazem) could inhibit ventricular fibrillation in the isolated rat heart model with coronary ligation (Thandoryen, 1982). All the agents were used in doses just below those causing atrioventricular block, and all elevated the ventricular fibrillation threshold of the isolated coronary ligated perfused rat heart. The antifibrillatory effects of the Ca^{2+}-antagonists were considerably greater than those of d&-propranolol (Opie et al, 1982). All the Ca^{2+}-antagonists tended to reduce cyclic AMP, although again not to the same extent as high-dose d&-propranolol. This "antifibrillatory" effect of the calcium-antagonist agents could be a non-specific phenomenon, as in the case of d&-propranolol (Lubbe et al, 1981).

Thus the effect of verapamil (1.5×10^{-7}M) is no more specific for the Ca^{2+}-antagonist than for any Na^{+}-antagonist effects of verapamil, because the ℓ-isomer has a similar effect to the d-isomer. Hence, in an isolated heart not subject to external catecholamine stimulation, both sodium and calcium channels are involved in ventricular fibrillation. Our recent data show that in conditions of adrenaline-stimulation (adrenaline 5×10^{-7}M) ℓ-verapamil caused a marked fall in fibrillation threshold whereas d-verapamil had virtually no effect. In theory, the ℓ-isomer inhibits the slow calcium channel (Bayer et al, 1975) whereas the d-isomer inhibits the fast sodium channel (Kohlhardt et al, 1978), hence the dℓ-isomer has local anesthetic properties (Bondi, 1978). Consequently, the inhibition of adrenaline-mediated effects by ℓ-verapamil rather than by d-verapamil favours the view that the calcium antagonist effects of verapamil are involved in these conditions of stimulation by external catecholamine. These data suggest that calcium ions are involved in the production of increased sensitivity to stimulated ventricular fibrillation occurring after the combination of coronary ligation and added beta-stimulation.

SUMMARY

The properties of calcium-channel antagonist agents are described. Because these agents render the isolated heart less sensitive to ventricular fibrillation a role for calcium ions in the genesis of ventricular fibrillation is proposed. The data obtained with verapamil isomers in isolated hearts with coronary ligation suggest (i) a role for both calcium and sodium ions in the fall of the fibrillation threshold following coronary artery ligation, and (ii) a role only for calcium ions when coronary ligation is combined with catecholamine stimulation.

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CELLULAR AND MOLECULAR MECHANISMS OF ATHEROSCLEROSIS

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INTRODUCTION

The progress of science and technology, the development of fundamental studies in cardiology during the last decade have opened new frontiers for the exploration of mechanisms of cardiovascular diseases. Today the major thrust in atherosclerosis research is aimed at developing a molecular-cellular theory, which would explain molecular and cellular processes that give rise to morphological and clinical manifestations of the disease. The essence of the research boils down to the integral assessment of factors in pathogenesis of atherosclerosis. From the general to the particular and from the particular to the general - that is the way of scientific research. It is especially important to abide by this principle when studying such complex diseases as atherosclerosis.

Complexity of the disease is mainly manifested by dislipoproteinemia. The mechanism of lipid metabolism disorders during the atherosclerosis has been lately specified. In most cases the disorders proved to be localized in the system of transport, targeted delivery and removal of cholesterol, its esters, and phospholipids. Plasma lipoproteins play a key role in the system. Now it is becoming clear what defects lipoprotein synthesis, processing and catabolism lead to dislipoproteinemia in man.

STRUCTURE OF LIPOPROTEIN AND ATHEROSCLEROSIS

In this field the main effort is concentrated on deciphering the mechanism responsible for cholesterol and phospholipids donor-acceptor functions of plasma lipoproteins. This direction seems to

be promising for investigation of atherosclerosis since in the course of epidemiological studies a negative correlation was found between HDL cholesterol level and the incidence of ischemic heart disease in men after 40 years of age.

On the other hand, the experiments on cell cultures and a perfused vessel demonstrated that HDL, particularly HDL_3, are capable of uptaking cholesterol from cellular membranes and the vessel wall.[1,2] The population of particles with HDL_2 density transports cholesterol into the liver for excretion from the organism. It is HDL_2 level that is reduced in IHD patients. That is why we took interest in the characteristics of chemical composition of HDL and their main subfractions (HDL_2 and HDL_3) in patients with documented coronary atherosclerosis. (Figure 1).

It is assumed that the main acceptor characteristics of HDL are determined by the phospholipid layer of lipoproteins. HDL phospholipids of healthy subjects consist of 70% lecithin and 12% sphingomyelin. It was shown in model systems that a lecithin increase in HDL enhances their cholesterol acceptor capability and vice versa.[3]

These data served as a requisite for elucidation of the role of HDL phospholipid composition in realization of their antiatherogenic properties in patients suffering from coronary atherosclerosis.

Firstly, we were interested whether in patients with different manifestation of coronary atherosclerosis: 1) lecithin/sphingomyelin ratio of plasma HDL_2 and HDL_3 is changed, and if so, to what extent; 2) apo B/apo A ratio is changed at different lipoprotein spectrum, assuming that cholesterol is transported into the vessel wall via LDL and removed via HDL.

Two of these indices were studied in the plasma of patients with different manifestations of coronary atherosclerosis by selective angiography data. The area of total atherosclerotic lesion was calculated in arbitrary units according to a special formula, which took into account the extent of occlusion, localization and spread of the lesion.

At normal lipoprotein spectrum the area of atherosclerotic lesion in CHD patients is smaller than that at dyslipoproteinemia-hyperlipoproteinemia or at a low HDL cholesterol level. Apo B/apo A_1 ratio in all CHD patients, even those with normal lipoprotein spectrum, was higher than in the control group. The highest (Figure 2) values of such ratio were observed at low HDL cholesterol. We assume that an increase of this ratio above one showed that cholesterol inflow to the arterial wall exceeded its outflow. It was still unclear what accounted for the reduction of cholesterol outflow from the arterial wall; a decrease in number of HDL particles, alterations of their characteristics, or both.

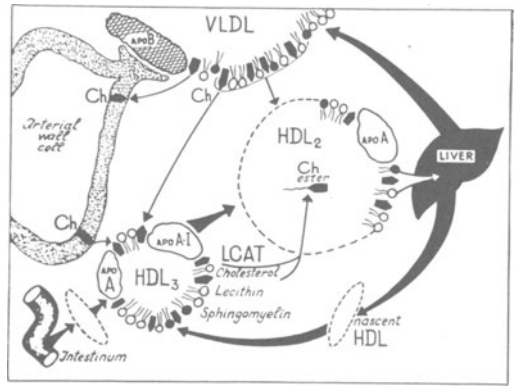

Fig. 1. Schematic representation of metabolism of VLDL and HDL. VLDL-very high density lipoprotein. HDL-high density lipoprotein. LCAT-lecithincholesterolacyltransferase.

To elucidate the question phospholipid composition and molar ratio of lecithin to sphingomyelin have been studied in HDL_2 and HDL_3. This ratio to a considerable degree determines the fluidity of surface monolayer and cholesterol-acceptor function of these particles.

A decrease of lecithin/sphingomyelin ratio was observed both in HDL_2 and HDL_3 of patients with coronary atherosclerosis in comparison with the control group. The greatest decrease was found in patients with low HDL cholesterol (Figure 3).

These facts indirectly prove the assumption that relative lecithin content largely determines the fluidity and cholesterol-acceptor function of HDL phospholipids. In our experiment a reduction of lecithin/sphingomyelin ratio in HDL_3 resulted in the decrease of biolayer fluidity and ability to accept cholesterol from cellular membranes or VLDL.

It is also quite probable that a possibility of LCAT (lecithin-cholesterolacyltransferase)-mediated transformation into HDL_2 is somewhat limited for HDL_3 with a low lecithin level. Under normal conditions this enzyme catalyzes cholesterol esters formation from free lecithin and cholesterol. While esterifying, cholesterol moves from biolayer into a hydrophobic "core". Lecithin is a substrate for this reaction. The decrease of LCAT-reaction leads to a re-

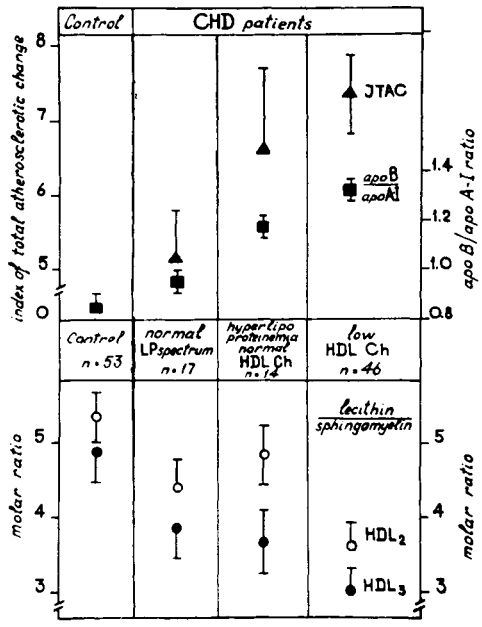

Fig. 2. HDL phospholipid composition and apo-B/apo-A ratio in healthy group and CHD patients. The left top ordinate-the degree of morphological involvement according to selective angiographic data. The right top ordinate- apo-B/apo-A ratio. The bottom ordinate- lecithin/sphingomyelin ratio. O- for HDL_2. O- for HDL_3.

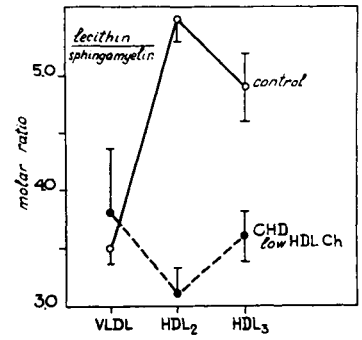

Fig. 3. VLDL,HDL_2 and HDL_3 phospholipid composition in healthy group and CHD patients with low HDL.

duction of cholesterol esters formation and thus decreases the formation from HDL_3 particles of HDL_2 which transport cholesterol to the liver for excretion.

Thus, the decrease of lecithin/sphingomyelin ratio in the phospholipid monolayer of lipoproteins of this class in patients with coronary atherosclerosis results both in lowering of the ability to accept cholesterol from membranes and expel it from the body.

The data on the lecithin/sphingomyelin ratio in three groups of particles, namely, VLDL, HDL_2 and HDL_3 are important with respect to investigation of mechanisms of atherosclerosis. Cholesterol inflow to and outflow from the membranes and its excretion from the organism are closely connected with cholesterol exchange between these particles. It was found that lecithin/sphingomyelin ratio in HDL_2 and HDL_3 of the control group was higher than in very low density lipoproteins. Naturally, that facilitates cholesterol inflow from VLDL to HDL_3 and HDL_2 in the control group of subjects. On the contrary, in patients with coronary atherosclerosis lecithin/sphingomyelin ratio did not exceed the values observed in VLDL. Naturally, under these conditions adequate amounts of cholesterol are not transferred from VLDL to HDL_3 and HDL_2. Thus, it is accumulated in these lipoproteins and together with them is transported into the vessel wall in larger quantities (Figure 3).

To confirm this assumption cholesterol content was calculated per a weight unit of apo B and apo A_1. In the group of patients with coronary atherosclerosis cholesterol share per a weight unit of apo B in VLDL was increased in comparison with the control group (Figure 4).

All these data indicate the decrease in cholesterol-acceptor and cholesterol-transport functions of HDL during coronary atherosclerosis which, in our view, is a possible mechanism of a tissue lipoidosis. We particularly stress the importance of changes in the molecular organization of HDL surface monolayer for this process. The obtained data lay the foundation not only for profound investigation of molecular basis of atherosclerotic process but, taking into account a possibility of changing antiatherosclerotic lipoprotein properties by affecting its phospholipid monolayer, open new prospects for the development of prevention techniques.

Along with lipoprotein structure the particles behavior in the organism is conditioned by a whole hierarchy of factors, which manifest themselves at different levels - from a single cell to the whole organism.

Surely, dislipoproteinemia is the most important sign of atherosclerosis. However, cells from different parts of the vascular system react quite differently to this metabolic disorder. Segmented

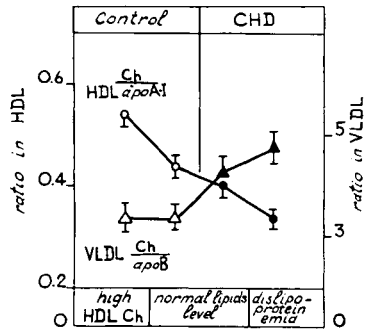

Fig. 4. Cho/apo-B ratio in plasma HDL and VLDL of control group and CHD patients.

character of atherosclerotic lesions in arteries and existence of "predilected sites" for plaque formation prove that the intrinsic characteristics of the vessel wall are really important for lesion development. This is why we think it equally important to study along with lipoproteins the structural and functional characteristics of vessel wall cells.

VESSEL WALL IN CULTURE AND ATHEROSCLEROSIS

At present most of the studies are concentrated on cellular manifestations of atherosclerosis at early and late stages: functional and morphological endothelial injuries, migration and proliferation of intimal cells; dedifferentiation and redifferentiation of medial smooth muscle cells; formation of foam cells and lipid-laden cells; production of extracellular matrix components by the endothelium and SMC. Cell-cell interactions are being intensively studied: endothelial cells - SMC, blood born cells - intimal cells. Special attention is paid to the adhesion of platelets and monocytes (macrophages) in zones of endothelial damage. The latter event plays a key role both in atherogenesis and thrombogenesis.

Cellular aspects of human atherosclerosis mostly remain obscure. The bulk of information on the problem has been obtained on the basis of histological analysis of autopsy material which does not allow to precisely evaluate cellular dynamics in atherosclerotic lesion zones. Only recently the methods of cellular biology, specifically a technique of culturing human vascular cells open new possibilities for the study of human atherosclerosis.

Nowadays cell culture may be figuratively called a "highway" in the investigation of human atherosclerosis. Actually, so far this is the only possible kind of experiment on man. It is of im-

portance that for a certain period of time cells in primary culture retain their intrinsic in situ properties.[4,5] Main manifestations of atherosclerosis at a cellular level (endothelial monolayer damage and repair, SMS proliferation, lipoidosis, connective tissue matrix formation) can be reproduced in culture; the same simple chemical conditions make it possible to reproduce the early stages of thrombi formation: adhesion, platelet spreading and formation of aggregates and thrombi of platelets. Finally, the primary culture makes it possible to study the effect of different drugs on cellular manifestations of human atherosclerosis.

Therefore, the experiments on cell cultures performed in our Center are rather multipurpose.

The Center was the first to develop original techniques of obtaining endothelial and SMC cultures both from uninvolved and atherosclerotic areas of human aorta. We obtained a significantly high cell yield from tissue (50-90%), while 90% of cells retained viability and attached to the substrate. Thus, the cultures rather adequately reflected the cellular composition of various parts of human aorta.

Endothelial cells of human atherosclerotic aorta in primary culture are heterogenous: 5-20% of the population are made up by giant multinuclear cells. They have all signs of endothelial cells (VIII factor of coagulation, Weibel-Palade bodies). Similar cells were found on the luminal surface of the in situ human aorta by scanning electron microscopy. In the vessel affected by atherosclerosis giant endothelial cells are mainly concentrated in the region of fatty streaks and plaques. 40-50% of all the lesion area in these zones is covered with such atypical monolayer (Figure 5). That gave us reason to assume that giant endothelial cells are lesion zone "markers" on the luminal surface. Morphological heterogeneity of endothelium is most prominent in fatty streak and plague regions. Therefore, the study of endothelial polymorphism in culture, specifically of the mechanism responsible for giant multinuclear cells formation, seems to be a promising direction of research. It cannot be ruled out that these cells are the "hot spots" of lipoprotein metabolism-endocytosis, transendothelial transport etc.

Primary cultures of human aortic intimal cells also proved to be morphologically heterogenous. Four main types can be distinguished among them: 1) elongated, 2) polygonal, 3) asymmetric, and 4) stellate cells (Figure 6).

Judging by morphological criteria, cells of the in situ intimal aortic layer too are heterogenous (Figure 7). To study the morphology of vascular cells in situ the vessel was prefixed with formaldehyde and placed into alcoholic-alkaline solution till complete dissociation of collagen-elastic matrix. Thus, four morphologically

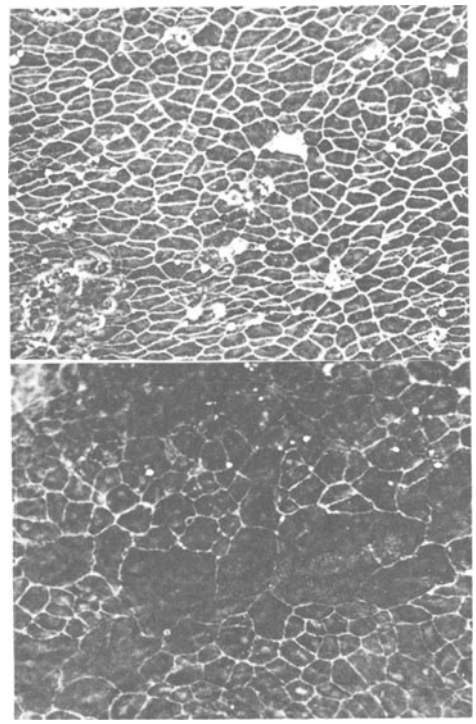

Fig. 5. Scanning electron microscopy of human aortic endothelium in situ. Vessel wall was silver stained and fixed under phisiological pressure. The upper part- homogenous endothelial surface of child aorta. The bottom part- the field of heterogenous endothelial lining of adult aorta. (x220)

different types of cells have been identified: 1) elongated, 2) elongated with side processes, 3) flattened cells of irregular shape and, 4) stellate cells. We have not yet identified the cell types found in culture with their prototypes in the vessel, but hope to do so in the nearest future. However, we already have reasons to think that polymorphism of intimal cells in culture at least in part reflects the morphological heterogeneity of the intimal layer of human aorta.

It stands to reason that the investigation of metabolic and functional characteristics of cells in primary culture is a prerequisite for the understanding of the role of each cell type in cellular manifestations of atherosclerosis. We have studied the parameters that have bearing on atherosclerosis at the cellular level: proliferation, lipoidosis and fibrosis.

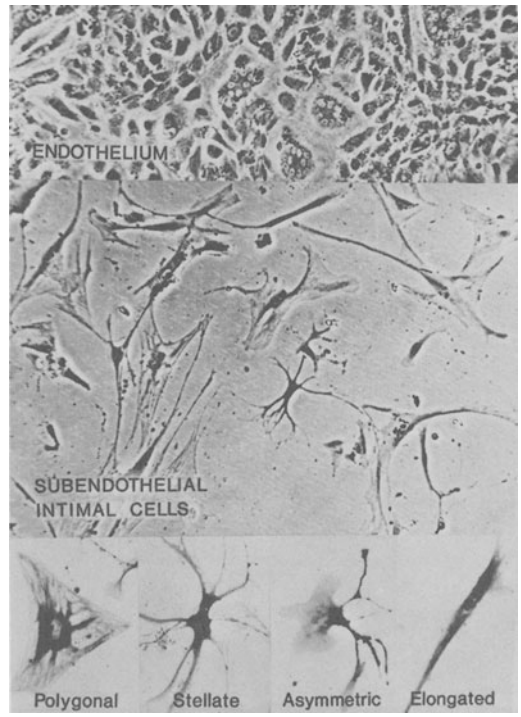

Fig. 6. Phase-contrast microscopy of primary aortic endothelial and intimal cell culture. Upper section-confluent monolayer with giant multinuclear endothelial cells. Medium section- primary intimal cell culture on the 7th day after seeding, general view. Bottom section- four main morphological types of intimal cell in culture: polygonal, stellate, asymmetric and elongated cells.

Intimal cells isolated from a normal region, fatty streak and the plaque of one aorta differ as to their proliferative activity. In cultures obtained from the zones of primary fatty infiltration the thymidine index on average exceeded the normal value by 4-fold. Cells from fatty streak divided more intensively as compared with plaque cells (Figure 8). Our results contrast with the data obtained on animals for we have not registrated intensive cell proliferation in zones of advanced atherosclerotic lesions. The loci of proliferation are mainly associated with morphologically unchanged segments of the vessel wall with minimal lipid infiltration.

A lot of cells isolated from atherosclerotic lesions retain in culture their main morphological feature - the lipid inclusions.[6,7] In appearance they resemble the so-called "foam" cells. It is

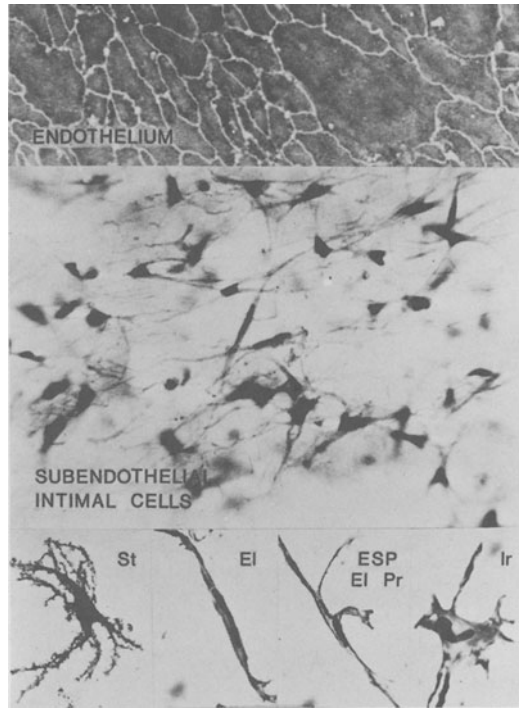

Fig. 7. Human aortic cell polymorphism in situ. The upper section- scanning electron microscopy of human aorta endothelial lining with small, medium-sized, large and giant endothelial cells. The medium section- population of intimal cell, isolated from prefixed vessel by alcoholic-alkaline dissociation, general view. The bottom section- four main morphological types of intimal cells in situ: St-stellate cells, El-elongated cells, ESP-elongated cells with side processes, Ir-flatten cells of irregular shape.

assumed that a part of foam cells may be formed from the smooth muscle cells of intima; another part of foam cells is formed of macrophages. The reasons for such a transformation are still unknown. We suppose that there are 2 possible ways of conversion into a "foam" cell. Firstly, the accumulation of lipids inside the cell may result from a dysfunction of intracellular lipid metabolism. Secondly, it can be a consequence of the excessive uptake of LDL circulating in the plasma. We have studied both possibilities.

Cells of normal and atherosclerotic aorta were compared as to their ability to synthesize lipids. For this purpose, radioactive acetate was added into culture. The newly synthesized lipids became

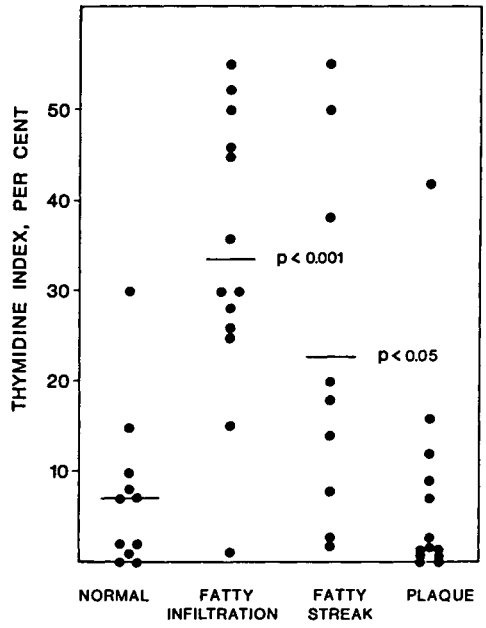

Fig. 8. ^{3}H-Thymidine incorporation into cultured intimal cells. The proliferation response of intimal cells in primary culture isolated from non-involved area and lesions. Ordinate- thymidine index in culture on the 7th day of cultivation. -- mean value for a set of measurement. On the absciss-results with primary intimal cell culture, obtained from normal, nonaffected region, fatty infiltration area, fatty streak and plaque.

labeled; they were extracted, fractionated, and radioactivity was measured in each fraction. It was shown that cells isolated from atherosclerotic lesions include more label into the fraction of phospholipids and cholesterol esters in comparison with the cells isolated from uninvolved areas. In case of cholesterol esters fraction the difference is nearly 10-fold. Since cholesterol esters are the main class of lipids accumulated in the plaque, one can assume that accumulation of cholesterol esters results a shift in intracellular metabolism towards enhanced synthesis of these lipids (Figure 9).

Another possible way of accumulation of cholesterol esters in aortic cells is the uptake of LDL circulating in blood, which contain these lipids. We have studied LDL uptake by cells of normal and atherosclerotic aorta, using fluorescence labeled lipoproteins.[8] Protein part of LDL molecule was covalently bound with rhodamine

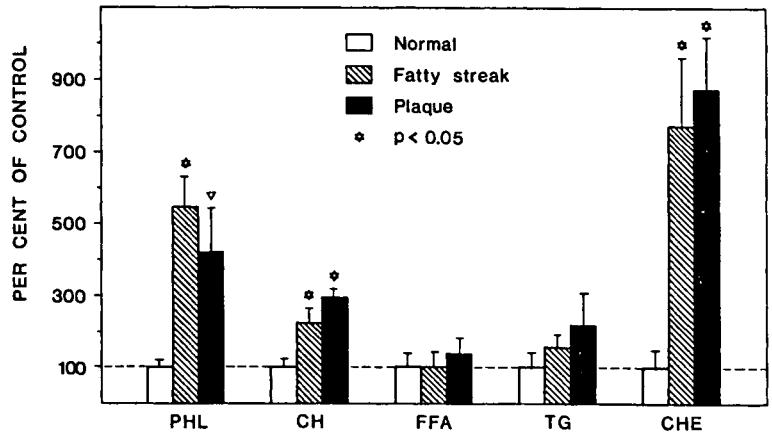

Fig. 9. Incorporation of ^{3}H-Acetate into lipid fractions of cultured intimal cells. Ordinate- mean value of radioactivity incorporation in per cent to control. Absciss-the main classes of lipids. PHL-phospholipids, CH-cholesterol, CHE-cholesteryl esters, FFA-free fatty acids, TG-triglycerides. White columns-cells of healthy regions, shaded columns-cells of fatty streaks, black columns-cells of plaques.

isothiocyanate. Fluorescence was excited, measured and recorded in a FASC-II flow cytofluorimeter. We judged of LDL uptake into aortic cells by the intensity of fluorescence. It was found that LDL uptake by atherosclerotic plaque cells exceeds that of the cells from uninvolved areas of the same vessel by 1.5-2-fold and by 4-fold in terms of LDL uptake per a surface unit. It is of interest that endocytotic activity of plaque cells also exceeds the normal values. Phagocytic activity was evaluated by the uptake of RITC-labeled E.coli bacteria. The number of bacteria internalized by a cell was measured in the flow cytofluorimeter. We have found a direct correlation between the effectiveness of LDL uptake by the plaque cells and their phagocytic activity. We assume that plaque cells have a higher level of specific and non-specific endocytosis.

It was demonstrated by cytofluorometry that the population of plaque cells falls into two subpopulations differing in the effectiveness of non-specific LDL uptake. Under the indicated conditions subpopulation A uptakes LDL several times more effectively than subpopulation B. Cells of both populations were sorted out, and it was found that they have considerable morphological differences. Cells of subpopulation A, which effectively incorporate LDL via non-specific endocytosis, have numerous lipid inclusions; many of them are typical "foam" cells. At the same time, cells of subpopulation B have low effectiveness of LDL uptake under these conditions and no lipid in-

clusions. It can be assumed that lipid transformation of plaque cells is, at least in part, a consequence of enhanced ability to uptake LDL via non-specific pathway (Figure 10).

As is known, high density lipoproteins (HDL) can discharge cells overloaded with lipid inclusions. We decided to find out whether HDL can completely discharge a human intimal cells of lipid inclusions. HDL were added into primary intimal cell culture, and in 24h the number of cells with inclusions was measured. Cultures not treated with HDL served as a control. In cultures isolated from

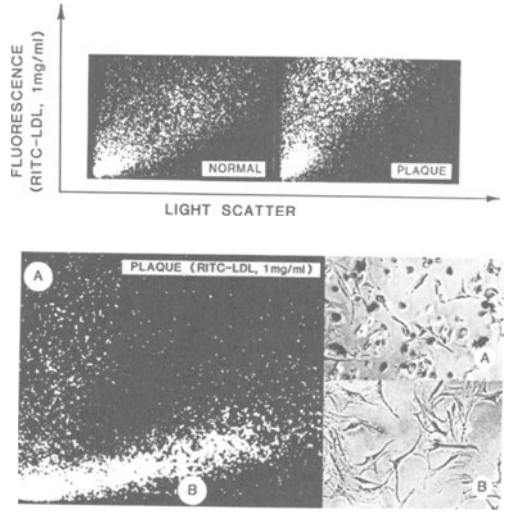

Fig. 10. Features of fluorescently-labeled LDL interaction with human aortic intimal cells in the primary culture. Scattering diagram, obtained with fluorescence activated cell sorter FACS-II is presented on the upper part of this figure. Ordinate-the level of fluorescence of single cells in arbitrary units. Absciss-light scatter. Left population of normal intimal cell from noninvolved part of aorta. The level of fluorescence is proportional to the average size of cells. Right-population of intimal cells from plaque. It is seen that plaque cells has a higher level of fluorescence. Bottom left: scattering diagram for intimal aortic cells from the plaque, incubated with RITC-LDL plus excess of non-labeled LDL. A-population with nonregulated high rate of RITC-LDL uptake. The top right part-phase-contrast microscopy of A-population of intimal cell sorted out and seeded on the plastic. B-population of intimal cell with down LDL control of RITC-LDL uptake. These cells were presented under phase microscopy to the right under A.

early atherosclerotic lesions HDL can decrease the number of cells with lipid inclusions by 1.5-2-fold. HDL did not have such an effect on cultures obtained from the plaque. Thus, the intimal cells of human plaque have the following metabolic characteristics: 1) they poorly proliferate in culture; 2) contain a lot of cholesterol esters; and 3) their intracellular lipid metabolism is shifted towards accumulation of cholesterol esters. The intimal cells isolated from fatty streaks and primary fatty infiltration have different characteristics: 1) they can actively proliferate in culture; 2) homogenous with respect to the ability to uptake LDL; 3) HDL can completely free these cells from lipid inclusions.

The differences found in the characteristics of cells localized in the plaque and fatty streak may help to explain the mosaic pattern of atherosclerotic lesion development in human aorta.

PLATELETS AND ATHEROSCLEROSIS

Along with lipoproteins, circulating in blood, platelets can also play an important role in lipid infiltration of the vessel wall. The mechanism of platelet participation in the plaque formation in case of primary endothelial injury and following thrombus formation is commonly known. In the experiments on animals platelet growth factor, which stimulates the proliferation of smooth muscle in the injury zone, has been found. It was also shown that lipid metabolism and LDL incorporation into the wall is activated in the zone of endothelial repair.

Now, the plurality of platelet functions in the regulation of lipid metabolism in the vessel wall becomes more and more evident.[9]

Thus, it was found in the *in vivo* experiments that platelets exhibit specific reversible LDL binding and, when activated, can chemically modify LDL apoprotein in such a way that specific endocytosis of the particles by macrophages is stimulated. So, in zones of endothelial injury the inflow of LDL into the wall can be accelerated due to the directed LDL transport by platelets and via the stimulation of specific endocytosis. The interaction of LDL with vessel wall cells and platelets *in situ* is more complex.

As is known, LDL of hypercholesterolemic animals increase platelet "sensitivity" to aggregation inducers and have an expressed cytotoxic effect on endothelial cells in culture. The initial phases of endothelial cell injury are accompanied by the loss of athrombogeneity and an increase of platelet adhesion to the endothelium. Such platelets can locally bind and modify LDL on the endothelial surface and stimulate lipid accumulation via specific uptake.[9]

It is common knowledge that non-damaged endothelial sheet

in situ and in culture serves as a barrier limiting the inflow of LDL. Vast de-endothelialization (denudation) of the vessel results in sharp stimulation of LDL accumulation by the wall. Unregulated accumulation of LDL takes place in the de-endothelialized zone.

It was demonstrated in our experiments with a perfused artery that HDL partially inhibited the uptake of LDL in the undamaged vessel wall area, but did not change LDL uptake in the denuded zone. One can conclude from these experiments that an integrate sheet of endothelial cells is the site of "antiatherogenic" HDL action (Figure 11).

Finally I would like to say a few words about our attempts at correcting the manifestations of atherosclerosis at a cellular and molecular level. We have seen that morphological features, metabolic characteristics and main functions of cells of normal and atherosclerotic aorta are retained in primary culture. Cells in culture are polymorphic like cells in vivo. The cells isolated from the zones of early atherosclerotic lesions have enhanced proliferative activity as compared with "normal" cells. Metabolism of lipids and lipoproteins in plaque cells is shifted toward accumulation of cholesterol esters. These facts make it possible for us to regard the primary cell culture as a suitable experimental model to testify some antiatherogenic drugs.

Intimal cells in primary culture of atherosclerotic aorta were treated with dibutiryl cAMP to decrease their proliferative activity

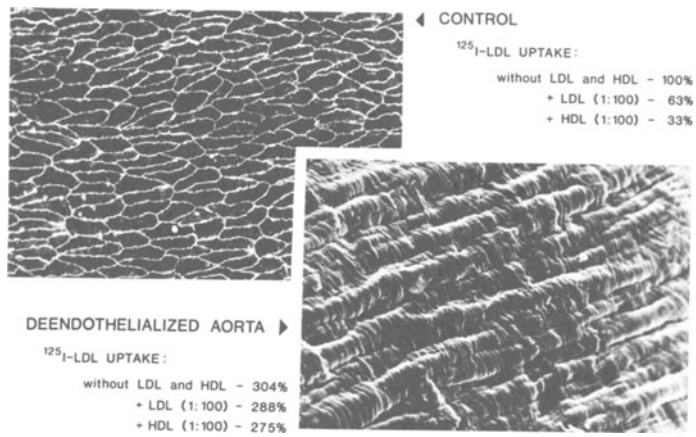

Fig. 11. Effect of HDL on LDL uptake into intact and deendothelialized rabbit aorta. HDL regulates LDL uptake into nondamaged but not into deendothelialized area of rabbit aorta. Scanning electron microscopy of normal endothelial surface (left picture) and denuded surface (right).

and the level of cholesterol esters. As is known, cAMP inhibits proliferation of many cells. Besides, this intracellular regulator activates lipolysis and controls other pathways of lipid metabolism. In our cultures dibutiryl cAMP significantly decreased the incorporation of thymidine into the cells of both normal and atherosclerotic aorta (Figure 12).

The incubation with dibutiryl cAMP resulted in a reduction of cholesterol esters level in cells isolated from the lesions of all types, but did not change their content in the cells obtained from the uninvolved intima. Thus, dibutiryl cAMP can be regarded as an agent of double effect, which normalizes at least two cellular dysfunctions: enhanced proliferative activity and high level of cholesterol esters.

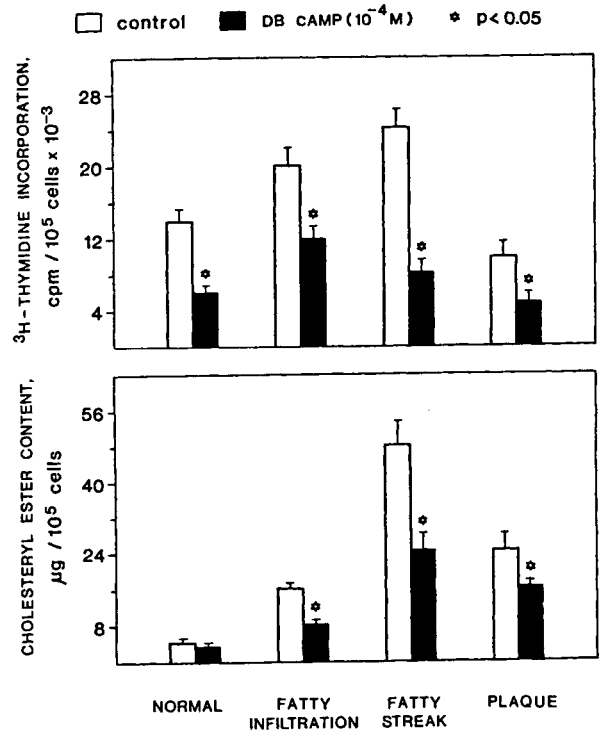

Fig. 12. Effect of dibutyryl cyclicAMP on ^{3}H-thymidine incorporation and cholesteryl ester content in cultured intimal cells. White column-cells of normal region, lesions in standard media. Black column-the same cells with db-c AMP.

Finally, I would like to emphasize the close links between the fundamental studies of atherosclerosis carried out in the USSR Cardiology Research Center and the needs of practical medicine, and primarily of cardiology.

Thus, the studies of three-dimensional spatial HDL organization are aimed at investigation of the defects in the structure and packing of particles in various types of hypercholesterolemia and in CHD patients. This can lead to the engineering of artificial HDL-like particles with "improved" antiatherogenic characteristics.

The plurality of platelet functions in the control of atherogenesis and thrombi formation makes us regard the platelet as the key target for directed pharmacological action. In the light of the data we have obtained, it is of special interest to find out the mechanisms of LDL transportation and modification in the zone of thrombi formation. We assume that platelets stuffed with antithrombogenic and antiatherogenic drugs can be used as a container targeted to the zone of the vessel wall damage.

Our studies on primary cultures of normal and atherosclerotic human arteries have shown that polymorphism, functional and metabolic heterogeneity of endothelial and intimal cells are the prerequisites for emergence of atherogenesis. It is remarkable that cellular manifestations of atherosclerosis (lipid infiltration of cells, proliferation) can be corrected using a number of pharmacological preparations. These preliminary data make it possible to hope that the primary culture of cells of atherosclerotic vessels may be useful for the screening of drugs with pronounced antiatherogenic effect.

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CORONARY ARTERY DISEASE IN CHILDREN

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Because coronary disease is such a prevalent condition and is responsible for such a large number of fatalities, it is usually associated with age. However, recent documented evidence of the existence of coronary heart disease in the young, indicates that youth does not rule out the possibility of this condition. Even myocardial infarction and/or sudden death, which are the culmination of coronary arteriosclerosis, are encountered in the young.

The natural history of the development of coronary heart disease can be divided into three stages: (1) an incubation period (ab ova), which may start in foetal life and continue into infancy, childhood, and adolescence; (2) a latent period, which is asymptomatic but in which pathological changes can already be present in young age; and (3) a clinical period, in which signs and symptoms first appear.

Autopsy examinations of young adults who died of noncardiac causes (mostly of injuries sustained in war) first drew attention to the presence of atherosclerosis in young adults, a process which must have started in childhood. These findings triggered research which resulted in the description of several histologic coronary arterial changes in infancy and childhood which were directly correlated with the extent of coronary atherosclerosis in adults[1-7]. In addition some of the risk factors associated with coronary atherosclerosis such as hypertension and hypercholesterolemia, may be present in childhood[8,9].

In addition to the factors which affect large groups of the population, various relatively rare pediatric diseases are associated

with advanced coronary atherosclerosis or other types of coronary narrowing in infancy. Among these are progeria[10]; congenital heart diseases causing elevated pressure in the ascending aorta and coronary hypertension[11,12]; and inherited disorders of metabolism[13-17].

On the basis of the data collected on these subjects it is now widely accepted that coronary atherosclerosis is a problem starting in childhood, mainly in its pathogenic and preventive aspects, and rarely also in its clinical presentation. The unusual finding of myocardial infarction in infancy and childhood may also result from many other systemic diseases and exogenous factors not associated with atherosclerosis or other forms of gradual narrowing of the arterial lumen.

Histologic Changes in the Coronary Arterial Wall in Infancy and Childhood

The coronary arterial walls undergo continuous changes throughout infancy and childhood. Since these changes have been proven to be related to atherosclerosis, only coronary arteries of foetuses may be considered completely normal. On this assumption a histologic study of coronary arteries of foetuses up to the age of 34 weeks was undertaken.

The normal foetal coronary arterial wall consists of (1) the intima, a single layer of endothelial cells; (2) the internal elastic membrane which consists of homogeneous elastic material surrounding the intima as a continuous tube with longitudinal corrugations; (3) the media, a few layers of smooth muscle cells and some elastic fibers; and (4) the adventitia, an external layer of loose connective tissue. The effect of risk factors influencing the foetal circulation was demonstrated[18].

After the 34 week gestation, the structure of the coronary arterial wall becomes more complex. A musculoelastic layer develops between the media and the intima where foci of fibrous thickening appear. The musculoelastic layer is formed by a series of degenerative and proliferative processes, mainly splitting of the internal elastic membrane, proliferation of smooth muscle cell within the split areas, and increase in the amount of the elastic fibers within the media[19]. The intimal changes include formation of intimal cushions composed of fibroblasts, elastic fibers and droplets of acid mucopolysaccharides.

In studies from our Institute, as well as from other centers, it was demonstrated that the extent of the intimal and medial changes is greater in males than in females. This sex difference was more prominent in population groups with a high prevalence and incidence of coronary heart disease in adult life[5,19].

The musculoelastic layer and the intimal thickening were prominent in ethnic groups with a high incidence of coronary artery disease such as Ashkenazy Jews, the population of the eastern part of Finland[5], and American Caucasians and Blacks[20,21]. On the other hand, the changes were minimal or absent in ethnic groups with a low prevalence and incidence of coronary heart disease such as Yemenite Jews, Bedouins from Negev Israel[7], the population of the western part of Finland[5] and Haitian Blacks[20]. These findings suggest that the coronary arterial morphologic changes in infancy and childhood underlie coronary atherosclerosis in adults. This would indicate that a genetic factor may be responsible for the structure of the coronary arteries we are born with, and partially responsible for the development of coronary atherosclerosis.

The combined results of these findings suggest that the presence of coronary atherosclerosis in adults is closely associated with the coronary arterial morphologic changes in infancy and childhood. In view of this, the presence of a genetic factor responsible for the structure of the coronary arteries we are born with is certainly indicated, and is also perhaps partially responsible for coronary atherosclerotic development in later life. The genetic disposition is probably combined with environmental factors.

Risk Factors for Coronary Atherosclerosis in Infancy and Childhood

Hyperlipidemia. Elevated levels of serum cholesterol are associated with an increased risk of coronary disease. However, only one type of hyperlipidemia associated with premature coronary heart disease presents a major risk factor in early childhood; this is hypercholesterolemia with xanthomatosis Fredrickson's type II, which is a genetically determined dominant trait with an incidence of 0.5-2%. It was shown that young people with hypercholesterolemia are more than 30 times susceptible to premature myocardial infarction than normocholesteremics of the same age group. Moreover, it has been shown that most young males sustaining myocardial infarction had hypercholesterolemia in youth. Although patients in the homozygote state are mainly affected, the heterozygote state is associated with increased incidence of coronary heart disease in patients over 30 years[22].

It is today an accepted policy in most medical centers that if a child from a high risk family has two elevated serum cholesterol measurements, lipoprotein fractions are measured to determine the distribution of total cholesterol among the lipid fractions. This information may be useful as more is learned about factors affecting cholesterol distribution among the lipoprotein fractions. Marked changes in the concentration of serum lipids and lipoproteins occur during the first year of life; a dramatic increase of serum total cholesterol, LDL and HDL begins around puberty; children tend to have higher HDL than do adults.

Recent epidemiologic investigations indicate that high levels of high density lipoprotein (HDL) cholesterol act as a protective factor against coronary artery disease and, conversely, that persons with subnormal levels of HDL cholesterol have a significantly increased risk of having arteriosclerosis.

It is premature to give prognostic significance to elevated HDL cholesterol levels in children: for the present, attention should be directed toward total serum cholesterol level and LDL cholesterol[8]. Mean HDL-C tends to be high at the areas with high mean total cholesterol, even in childhood. The mean HDL-C level could thus be a response of the metabolism to the total level, so as to assume a satisfactory HDL total level. We do know, however, that the latter varies greatly within adult populations and is inversely and strongly related to coronary risk. The relationship between dietary cholesterol and serum cholesterol is further complicated by genetic characteristics.

In spite of detailed descriptions in the literature and the fact that they are the most common known genetic diseases affecting man, the inherited hyperlipoproteinemias comprise only a small percentage of cases of hypercholesterolemia. The majority of hypercholesterolemic individuals are affected by environmental influences, dietary cholesterol intake has gained major attention and interest because it can potentially be controlled[8,23-29].

Hypertension. Hypertension has been established as a major risk factor for coronary atherosclerosis in adults[22]. It alone has been proven to produce atherosclerosis even in the absence of other risk factors[31]. The direct effect of hypertension on the coronary arteries was demonstrated by the finding of obliterative intimal hyperplasmia and medial thickening revealed by light microscopy studies performed in our Institute, and by others in patients with coarctation of the aorta or supraventricular aortic stenosis in experimental coarctation in a subhuman primate model[32].

In a recent electron microscopic study of coronary microcirculation in patients with coarctation of the aorta in our Institute[33], morphologic changes were also found in the small arterioles. These changes were more severe and widespread in young adults than in children and were directly related to age. A possible mechanism in which hypertension aggravates atherosclerosis is the stretching of the arterial wall, which increases its permeability to cholesterol and lipoproteins.

In recent years primary hypertension in children has been recognized as a public health problem[34]. Better recognition of the extent of the problem was achieved by blood pressure measurements in routine physical examination and better determination of the effect of age on blood pressure[35]. The study by Graham et al.[36]

and Londe's[37-40] studies demonstrated the importance of not using the adult standards to define hypertension in children. The incidence of persistent hypertension in children is now estimated to be 1-4%. Until recent years primary hypertension in children was considered unusual. Londe et al.[37-40], however, demonstrated that essential hypertension was much more common than previously thought. Various studies have shown that blood pressure is continuously rising from infancy, with acceleration in adolescence. On the other hand, a hypertensive child may become a normotensive adult in 35% of the cases. Multiple determinations should be performed before a child be considered hypertensive. Hypertension was found to be more frequent in siblings of hypertensive individuals than in those of normotensive individuals.

On initial evaluation (single determinations), Heyden et al.[35] in the Evans County (Georgia) Study found that 11% of 435 adolescents were hypertensive. Repeat evaluation carried out 7 years later on 30 of these 435 patients from the hypertensive group, revealed 11 to have persistent high blood pressure, while 6 had some form of vascular complication during their early adulthood, including two deaths. This indicates that not only do these children face an increased risk of CHD later in life, but they also suffer significant early morbidity and mortality.

Tobacco Smoking. The Joint Report of the Study Group on Smoking and Health in 1957 [28] proved unequivocally that tobacco smoking is a major contributing factor to the pathogenesis of atherosclerosis. These findings were confirmed by epidemiologic studies which found a direct relationship between smoking and coronary heart disease. Further data on coronary arteriography studies proved a direct relationship between the degree of cigarette consumption and the severity of coronary atherosclerosis[42-47].

The incidence of hypertension and coronary heart disease is highest and its onset earliest in those who began smoking at less than 20 years of age. Because of this the increasing trend in childhood smoking is particularly disturbing.

Inherited Disorders of Metabolism Associated with Coronary Obstructive Lesions in Infancy and Childhood

Disorders of Lipid Metabolism. In Sandhof's disease[14] there is a neural accumulation of GM_2 ganglioside and neural visceral accumulation of globoside due to deficient activity of hexosaminidase A and B. Onset occurs in the first two months of life and the clinical features include development retardation and cherry-red macula. Two cases of Sandhof's disease with luminal narrowing of the coronary arteries due to intimal proliferation of fibroblasts have been reported[14].

In Fabry's disease[13] ceramide trihexoside accumulates in the walls of small blood vessels, due to a deficiency of ceramide trihexosidase. Coronary involvement is frequent and the clinical features include anginal pain, cardiac enlargement, congestive heart failure, and myocardial infarction. GM_1 gangliosidosis is a condition in which galactosidase deficiency causes neural and visceral accumulation of GM_1 ganglioside. Cardiac lesions have been found in about 30% of the cases, but coronary atheromatous plaques containing balloon cells of foamy periodic-acid-Schniff-negative cytoplasma were reported in only one case[15].

Amino Acid Metabolism, Mucopolysaccharidoses Disorders of Protein Metabolism

Homocystinuria. Homocystinuria is a defect in serine and homocystine metabolism, presenting clinically with ocular abnormalities, long extremities, scoliosis, osteoporosis, mental retardation, and thrombotic vascular disease, including coronary occlusion[17].

Alkaptonuria. In this condition homogentisic acid is accumulated and excreted in the urine, due to a deficiency of homogentisic oxidase. The clinical features include dark urine, pigmentation of cartilage, arthritis, and mitral and aortic valvulitis. Blue-black pigmented coronary atheromatous plaques cause myocardial infarction, which is a common cause of death in this condition[16].

Mucopolysaccharidoses. The coronary arteries are affected in two of the six mucopolysaccharidoses in which the heart is involved, namely Hurler's and Hunter's syndromes. The coronary arteries are narrowed by plaques containing mucopolysaccharides. Such cells also invade the myocardium[13].

Protein Metabolism. In primary hyperoxaluria there is excessive synthesis of oxalic acid with calcium oxalate accumulation in various organs, including the coronary arteries[13]. Tangier disease is an inborn error of metabolism characterized by deficiency of high density lipoprotein in plasma and storage of cholesterol esters in many tissues, including the heart. Patients with Tangier disease and coronary heart disease due to coronary deposition of cholesterol esters have been described[13].

Congenital Heart Anomalies with Coronary Obstructive Lesions

Conditions associated with elevated pressure in the ascending aorta and coronary arteries. Coarctation of the aorta and supraventricular aortic stenosis cause elevated pressure in the ascending aorta which affects the coronary arteries[11,12]. Light microscopy studies in these patients revealed coronary luminal narrowing due

to thickening of the media, proliferation of fibroblasts in the intima, and degenerative and proliferative changes of elastic fibers. The mortality rate from cardiovascular causes in patients undergoing operative correction of coarctation of the aorta over the age of 35 years is ten times greater than the rate in patients between 15 and 34 years of age who were operated upon. This has been attributed to the coronary arterial changes which develop with age[44].

In a recent electron microscopic study in our Institute[33], age-related changes were found in the coronary microcirculation of patients with coarctation of the aorta. These changes included destruction of the normal wall of the small coronary arteries and coronary arterioles with collagenous transformation. The pre-capillary sphincters, metarterioles capillaries and venules were normal.

Single coronary artery. Single coronary artery is a congenital anomaly which may be isolated or associated with other congenital cardiac lesions. The incidence of coronary atherosclerosis in this anomaly is not increased, but the patients are more susceptible to its consequences[7].

Short main left coronary artery. It has been suggested that patients with congenitally short main left coronary artery have an increased incidence of atherosclerosis in the left coronary system[45].

Miscellaneous Conditions Associated with Coronary Obstructive Lesions in Infancy and Childhood

Progeria. Hutchinson-Gilford syndrome is a disease of extremely accelerated aging, starting in infancy and developing throughout childhood. The disease is characterized by thinning and atrophy of the skin, loss of subcutaneous fat, growth failure, osteoporosis, joint stiffness, and alopecia. Atherosclerosis begins in infancy and presents clinically in the first decade of life as angina pectoris, hypertension, congestive heart failure, and myocardial infarction. The latter is rarely present in the first year of life, but the basic disorder in the syndrome is dysplasia of mesenchymal tissue of unknown etiology[10].

Werner's syndrome. Werner's syndrome is a condition of premature aging with retardation of development, hair loss or greying, hyperkeratosis of skin, cataracts, hypogonadism and osteoporosis. Atherosclerosis and arterial thrombosis are frequent[46].

Trisomy 18. A single case of trisomy 18 with arterial changes including degeneration of the internal elastic membrane, thickening of the intima by fibroblastic proliferation, and calcification of the media has been reported[47].

Pseudoxanthoma elasticum. Pseudoxanthoma elasticum is an autosomal recessive condition in which calcification and fragmentation of elastic fibers occur in the eyes, skin and blood vessels, including the coronary arteries. Cardiovascular manifestations include anginal pain, congestive heart failure and hypertension[48].

Coronary Arteritis

Until recently the problem of coronary arteritis was limited to isolated cases of infantile periarteritis nodosa. In the last decade, however, increasing numbers of cases with Kawasaki's disease (mucocutaneous lymph node syndrome)[45] have been reported, mainly in Japan, but also in Europe and the United States. Kawasaki's disease is an acute febrile disease presenting in early childhood with conjunctical congestion, indurative edema, erythema, and desquamation of the palms and feet, strawberry tongue, dry red fissured lips, and exanthema of the trunk. Electrocardiographic changes and coronary periarteritis are frequent. In the chronic stage coronary aneurysms and myocardial infarction occur in an as yet undetermined number of the patients. The etiology of this condition is unknown. Some of the patients had aorta coronary bypass grafting. In most cases, however, the aneurysms disappear after one year and the coronary arteries are thin, with isolated obstructive lesions[49,50].

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CARDIOMYOPATHIES: PATHOLOGY AND INFECTIOUS IMMUNE MECHANISMS

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Much confusion of defining and classifying cardiomyopathies has existed in the past. As a result of the work by the Task Force set up by the World Health Organization and the International Society and Federation of Cardiology[1], this has been clarified. Cardiomyopathies are now defined as "heart muscle diseases of unknown cause" and classified into three major types: dilated, hypertrophic and restrictive. For each type characteristic morphologic features exist.

Dilated cardiomyopathy

At necropsy all chambers of the overweight hearts are usually severely dilated, the endocardium is thickened, intercavitary thrombus frequent, and coronary arteries are normal[2]. Histologically, normal arrangement of myocardial fibers is found and if dilatation has been severe, disproportion of nuclear changes - which reflect hypertrophy - and myocardial fibers which may show normal diameters, reflecting dilatation, are seen[3]. Smooth muscle hypertrophy and hyperplasia in the endocardium confirm dilatation. An increase in interstitial fibrous tissue, limited to the inner rim of the myocardial wall, may be present. The intramyocardial vessels are usually normal.

Histochemical analysis undertaken on biopsy material obtained by bioptome[4] show no distinctive patterns and merely reflect the degree of hypertrophy and to some extent duration of heart failure. Thus the various enzyme components may be increased, normal or decreased.

Ultrastructural changes are those of hypertrophy[5] often with degenerative changes, particularly in long standing cases[6].

It has been argued that in view of the fact that no specific morphologic features exist that diagnosis can only be established with certainty at necropsy. This, of course, applies to every disease affecting the human body but diagnosis of dilated cardiomyopathy can be achieved during life clinically, aided by non-invasive techniques. Cardiac and extra-cardiac causes can be excluded with certainty in a large number of cases. There is, however, a significant number of patients in whom invasive techniques have to be undertaken, foremost among which is examination of endomyocardial tissue obtained by bioptome. By this means possible involvement of the myocardium by myocarditis, sarcoidosis or infiltrative diseases can be assessed[7].

Hypertrophic cardiomyopathy

Despite the denial of the existence of this type of cardiomyopathy as a distinct entity by some workers in the past, there is abundant evidence of sufficient highly characteristic features, not only clinically but also morphologically that hypertrophic cardiomyopathy is a separate, recognizable entity[8]. Macroscopically, asymmetric hypertrophy is often a striking characteristic feature. Echocardiographically, it has been shown that if the ratio of the ventricular septum to the posterior left ventricular free wall exceeds 1.3, hypertrophic cardiomyopathy is likely to be present[9]. Subsequently it has been established that minor degrees of asymmetric hypertrophy of the septum can be present in many other conditions and histologic confirmation needs to be undertaken in such instances [10]. Frequently however, if the ratio of the free ventricular wall with the septum is calculated[11], the ratio exceeds 3 (even in the severest form of other types of hypertrophy the ratio remains unity). If such severe disproportion is found, then asymmetric hypertrophy alone permits a diagnosis of hypertrophic cardiomyopathy to be made. The bulging of the septum displaces the anterior papillary muscle which contributes to the malfunction of the mitral valve apparatus, resulting in mitral insufficiency[12]. An impression of the anterior mitral valve leaflet may be found in the form of endocardial thickening in the outflow tract of the left ventricle[13]. The coronary arteries, as in dilated cardiomyopathy, are usually normal.

The characteristic histologic features include disarray of extremely hypertrophied myocardial fibers, bizarre shaped nuclei, often surrounded by a clear zone, the so-called perinuclear halo and varying degrees of often cellular fibrous tissue. Several years ago an index assessing semiquantitatively each of the histologic features was devised[14] and if the value exceeds 50%, then

hypertrophic cardiomyopathy is present. Disarray, its distribution and severity has also been more recently undertaken[15] confirming and extending the findings previously reported[14].

Histochemical examination often shows severe accumulation of glycogen which has diagnostic importance. Other enzyme systems merely reflect severe hypertrophy[14].

Ultrastructurally, disarray of myocardial fibrils is often widespread and striking but is not diagnostically helpful[14]. Furthermore, abnormal intercellular junctions are also frequently found, at one time considered pathognomic[16], but previously and subsequently discounted[8]. Despite the overlap of ultrastructural changes with "ordinary" hypertrophy it can be concluded that if the changes of disorganization are widespread they characterize the condition and act as an adjunct to histologic and macroscopic diagnosis. The various histologic features considered in combination and the accumulation of glycogen permit a firm diagnosis[8].

Though previously the presence or absence of a systolic gradient has been emphasized, it has subsequently been established that it does not influence the natural history of the disease[17]. Similarly, at morphologic levels it has previously been found that if obstruction had been present, the abnormal features were principally concentrated in the asymmetrically thickened septum. If no obstruction was clinically present the same histologic and ultrastructural changes were seen but were widely distributed focally throughout the myocardium[18]. This distribution is, however, not always found[19].

Restrictive cardiomyopathy

Two hitherto considered separate entities are included under this heading, endomyocardial fibrosis, morphologically first described by Davies[20] and considered to be a disease limited to the tropics and Löffler's endocarditis parietalis fibroplastica (Löffler's endomyocardial disease)[21] considered to be confined to temperate zones. It has however been shown that both conditions belong to the same disease spectrum[22]. In this condition, irrespective of geographical origin, left, right or both ventricles, may be involved. The endocardium which is often several millimeters thick, is the characteristic feature. Inflow and partly outflow tract of the left ventricle, and the apex and the area beneath the tricuspid valve in the right ventricle, are the typical sites, but endocardial changes may be seen elsewhere in the ventricle as well as in the atria[23]. Thrombus is frequently superimposed, forming the obliterative "phase" of this disease.

Histologically, the endocardium is arranged in layers: superficially, beneath the thrombus a zone of loose and dense connective

tissue is present beneath which the so-called granulation tissue layer is found. This consists of numerous dilated blood vessels, embedded in loose connective tissue. Inflammatory cells including occasionally eosinophils may be found. From this zone fine septae extend into the underlying myocardium[24]. Histochemical and ultrastructural changes are those of hypertrophy.

By definition the etiology or etiologies for each of the types of cardiomyopathy is unknown. Many suggestions have been made in the past and these have been summarized previously[25].

Increasing evidence is accumulating that infectious immune mechanisms may be responsible for some types of cardiomyopathy. These will now be discussed. Before the various studies that have been undertaken are detailed, it is perhaps relevant to summarize some general remarks regarding infectious immune mechanisms. An immunological reaction is defined as a specific combination of antigens with humoral antibody or sensitized cells. An immunologic event can be defined as tissue damage resulting from the initiation of immune reactions[26]. These reactions have been classified into four major types: I immediate hypersensitivity, involving reaginic antibody; II other cytotoxic effects of antibody; III dependence on immune complexes and IV dependence on delayed cell-mediated immune reaction[27]. Modification of this classification has been suggested [26], firstly immune disorders are rarely caused by a single mechanism but involve interactions of several mechanisms and secondly it is difficult to distinguish clearly between immunologic and inflammatory events, particularly in chronic disorders.

The possible infectious immune mechanisms or immune mechanisms in the various types of cardiomyopathies will now be considered.

Dilated cardiomyopathy

By definition the etiology is unknown. The disease is often heralded by an upper respiratory infection and increasing neutralizing antibody titres against Coxsackie B_3 Echo and Herpes virus have been found in patients with this type of cardiomyopathy[28]. Significantly higher titres to B virus infections, particularly with a history lasting less than one year have also been shown in a significant number of patients when compared with an equal number of age and sex matched normal individuals[29]. The linkage between active viral infection and dilated cardiomyopathy is not yet well established, though several clinical reports testify to such relationship[30]. Heart-reactive antibodies have been demonstrated correlating with the severity of symptoms and duration of disease[31]. Studies of cell mediated immunity have also been undertaken[32] and defective T suppressor cell function in dilated cardiomyopathy have also been demonstrated[33]. In the patients investigated with high

neutralizing antibody titres against Coxsackie virus[29], endomyocardial biopsies failed to show any evidence of present or past myocarditis. The significance of the various reports is as yet unclear but the possible pathogenetic mechanisms have been summarized[30]. A significant number of patients show evidence of an immune disturbance and react in an unusual manner to common virus infection. The progressive illness, punctuated by recurrences, may be due to immune mechanisms. Virus infection may trigger production of antibodies directed at suppressor cells. These antibodies may inactivate or possibly coat the T cell receptors in such a manner that their normal regulatory role in modulating T cell function is impaired. This may result in heightened B cell activity with production of antibodies directed to self. Similarly T suppressor cell dysfunction may also affect cell-mediated immunity. Further work in these concepts is clearly indicated.

Hypertrophic cardiomyopathy

This type of cardiomyopathy has shown a human leukocyte antigen (HLA) linkage. These antigens are closely associated with the gene governing immune response (located on the sixth chromosome). It has been shown that the differences between white and black races exists (B12 and B5 antigens) and these patients were normotensive; whereas patients without these antigens, sporadic cases, were hypertensive[34].

Restrictive cardiomyopathy

The possible association of infection such as filariasis[35] has long been debated. Hypersensitivity response to streptococcal infection[36], abnormal immunologic reaction, and immunologic factors have also been previously reported[37,38]. The suggestion that endomyocardial fibrosis and Löffler's endomyocardial disease belong to the same disease spectrum, the origin of which can be traced back to the presence of eosinophils has been suggested several years ago[39]. More recently, it has been shown that eosinophils when associated with endomyocardial disease demonstrate certain abnormalities; these consist of degranulation. It has furthermore been shown that eosinophilia may result from a variety of infectious (and non-infectious) causes and it has been demonstrated that the binding capacity for complexed IgG and increased phagocytosis of eosinophils exists, with unmasking of Fc receptors. Degranulation occurs as a result of binding of IgG or C3b coated particles or parasites[40]. Electron microscopically it has been shown that the granules are in appearance consistent with cationic proteins. These proteins and possibly with combination of peroxidases result in the changes that lead to endomyocardial disease.

It can, therefore, be concluded that a variety of infectious immune mechanisms may be operative in the various types of cardiomyopathy but much work needs still to be undertaken. Examination of fresh endomyocardial tissue obtained by bioptome[4] has demonstrated several important findings, for example in dilated cardiomyopathy IgG is preferentially bound to myocardial tissue, whereas if a virus infection had been the cause IgM is positive in many patients using direct fluorescent techniques[31]. The value of endomyocardial biopsies, particularly in cardiomyopathies is without doubt[41]. In patients with unsuspected myocarditis evidence is now accumulating that sequential biopsies of patients under treatment helps to monitor therapy[42]. This invasive form of investigation is also contributing greatly in elucidating pathogenic mechanisms of cardiomyopathy.

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TREATMENT OF CARDIOMYOPATHIES

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In order to advise rational treatment it is necessary to define and to classify the conditions known as cardiomyopathies. Cardiomyopathies are defined as "Heart Muscle Diseases of Unknown Causes" to distinguish them from myocardial diseases due to disorders of other systems; the "Specific Heart Muscle Diseases". This lecture is concerned with the treatment of cardiomyopathies so defined.

Cardiomyopathies are classified into three main types according to their disorders of structure and function:

1. Hypertrophic; with and without systolic pressure gradients.
2. Dilated (congestive).
3. Restrictive/Obliterative.

(Goodwin and Oakley 1972; Goodwin 1974; 1979; 1981).

The report of the WHO/ISFC Task Force 1980 has confirmed this earlier classification with only very minor modifications.

Hypertrophic Cardiomyopathy

This condition is characterized by massive hypertrophy of the ventricular septum and free wall of the left ventricle and sometimes of the right ventricle also. The systolic volume of the left ventricle is reduced; contraction is powerful, rapid and incoordinated. Ejection fractions are in the region of 90%. The shape of the left ventricle is grossly distorted. Diastolic function is seriously impaired in a very complex way resulting in abnormal resistance to filling and relaxation. The condition is familial; death is sudden and unexpected in 50% of the patients, some families having a very

high incidence of sudden death (Malignant Hypertrophic Cardiomyopathy) (Maron et al., 1978[a]).

Treatment

Treatment aims at preventing sudden death, at improving hemodynamics, at reducing symptoms and at limiting progression of the disease.

Sudden Death

Sudden death is most likely to be due to arrhythmia (Goodwin and Krikler, 1976) for there is a significant relation between sudden death and ventricular tachycardia (McKenna et al., 1981[a,b]; Goodwin and Krikler, 1976), but other factors such as impediment to filling of the left ventricle and acute reduction in ventricular volume also play a part. There is no relation between sudden death and the presence or severity of systolic gradient.

Prevention of sudden death centers mainly around effective antiarrhythmic therapy. Neither beta adrenergic blocking agents nor calcium blocking agents have been shown by our group to reduce the incidence of arrhythmias (McKenna et al., 1980; McKenna et al., 1981[b]). Episodes of ventricular tachycardia detected by ambulatory monitoring correlate inversely with prognosis. Amiodarone which prolongs the action potential and increases the refractory period has been shown to be effective in reducing the number and frequency of ectopic rhythms and of ventricular tachycardia (McKenna et al., 1981[c]).

All patients with hypertrophic cardiomyopathy should have ambulatory ECG monitoring. If there are episodes of repeated multifocal ventricular premature contractions or runs of ventricular tachycardia then Amiodarone should be started. The dosage should be 200 mg three times daily for one week, then 200 mg twice daily; the dose may be further reduced later, if necessary by discontinuing the drug on one or two days of the week. There is a latent period of one to two weeks before the drug begins to act. Since Amiodarone affects thyroid function and can produce both hypo and hyperthyroidism, thyroid function tests should be performed before and during treatment. The effects of calcium-blocking agents may be potentiated by Amiodarone and thus Amiodarone and Verapamil should not be used together. The effects of digitalis and anticoagulants are also potentiated by Amiodarone. The action of Amiodarone persists for many months after the drug has been stopped in patients who have been taking it for long periods.

Complications of Amiodarone

Unfortunately, Amiodarone has a number of side effects. These include peripheral neuropathy, photosensitivity, headache, nausea, vertigo, but are usually mild and reversible. Vomiting may occur during the initial phase of treatment with the loading dose. Micro deposits on the cornea are regularly seen by slit lamp examination. They do not affect visual acuity though may impart a faint bluish tinge to the vision. Regular ophthalmic examination is important however. Amiodarone is rarely contraindicated in hypertrophic cardiomyopathy, but care should be taken to exclude patients with atrioventricular block or troublesome bradycardia unless a pacemaker has previously been inserted. Pulmonary fibrosis is a rare complication of Amiodarone treatment and usually reversible when the drug is stopped.

Measures to Prevent Sudden Death from Mechanical Causes

Attempts to prevent sudden death from resistance to ventricular filling center around measures to improve compliance, reduce resistance to filling and aid relaxation. Events which reduce ventricular volume add to the possibility of sudden death, and thus hypotension and hypovolemia should be avoided.

The effects of beta adrenergic blocking agents on ventricular filling and compliance are variable. In acute studies both Practolol (Webb Peploe et al., 1971) and Propranolol (Swanton et al., 1977) reduce diastolic filling pressure and increase diastolic volume. The effect of chronic oral treatment with beta adrenergic blocking agents is less consistent, but in most patients the active suction period of diastole (between the opening of the mitral valve and 0 point of the impulse cardiogram) when most of ventricular filling occurs is significantly prolonged. Filling of the left ventricle at the moment when left ventricular pressures are still falling is increased. This indicates improvement in relaxation, an effect that is independent of heart rate (Alvares, 1980). In theory, by these effects, and by slowing of heart rate, especially on effort, and by allowing more time for ventricular filling, beta blockade should help to reduce the risk of sudden death. Unfortunately, the available data apart from the series of Frank et al. 1978 who used very large doses of Propranolol, do not indicate that this is so.

Although Verapamil has been reported to improve diastolic function there is, as yet, no evidence of prolonation of life; indeed, Verapamil may shorten life because sudden death has occurred after starting Verapamil and pulmonary edema has been documented, both in our own experience and in that of Rosing et al. 1981.

However, it is encouraging that Nifedipine has been shown to improve diastolic compliance in acute studies (Lorell et al., 1982) though a disadvantage of Nifedipine would be its vasodilator effect which might reduce ventricular volume and so predispose to elimination of the cavity and arrest of blood flow.

There is no definite evidence that surgical treatment prevents sudden death, by an effect on ventricular function, except possibly by reduction in systolic pressure gradients and improvement in ventricular volume.

Treatment to Improve Hemodynamics

Hypertrophic cardiomyopathy is essentially a disease of impaired ventricular diastolic function: diastole is grossly disorganized (Alvares, 1980), relaxation and the rate of filling are impaired, both regionally and globally (Sanderson et al., 1978; Goodwin, 1982).. The massive ventricular hypertrophy, the reduced ventricular volume in systole and powerful contraction of the ventricle which expels all the contents in the first half of systole combine with the abnormalities of filling to offer serious impediment to effective left ventricular function. It is the powerful contraction of the massive left ventricular muscle that brings about apposition of the septum to the mitral valve apparatus and causes pressure gradients to develop in systole. Reduction of ventricular volume by impaired inflow and by reduced afterload both contribute to the elimination of the ventricular cavity. True obstruction does not occur (Criley et al., 1965; Goodwin, 1979; Murgo et al., 1980).

Systolic pressure gradients can be modified by beta blockade given acutely but there is no evidence that chronic beta blocking treatment is beneficial in this way. Verapamil has been reported to reduce pressure gradients (Rosing et al., 1979[a]). Reduction in the force of ventricular contraction may help to reduce cavity elimination but the most important area of impaired hemodynamics is diastole, and it is in this phase of the cycle that beta blocking agents exert their most important action. Chronic beta adrenergic blockade usually prolongs the active suction phase of diastole and improves relaxation independent of heart failure, the negative chronotropic effects of beta blocking agents is important also, as the bradycardia allows more time for filling of the ventricles.

Nifedipine improves diastolic filling acutely (Lorell, 1982) but may be disadvantageous chronically by reducing ventricular volume by vaso-dilatation and reduction of afterload.

Verapamil may improve diastolic function without vaso-dilatation. In my department the effect of Verapamil on systolic function has been studied in 18 patients with hypertrophic cardiomyopathy by

systolic time intervals. Verapamil was found to have negative inotropic effects when left ventricular systolic function was normal before treatment, but to have peripheral vaso-dilator effects when left ventricular function was impaired before treatment. Two patients in the study developed pulmonary edema (Herr, K. to be published).

The effect of Amiodarone on hemodynamics has not yet been fully worked out. Its beta blocking action may be of some value.

Surgical treatment reduces or abolishes gradients and reduces left ventricular end diastolic pressure.

Thus, to improve hemodynamics beta blockade therapy is the first line of treatment. In patients who do not respond, Verapamil may be tried but atrioventricular conduction defects or high left atrial pressures are contra-indications (Rosing et al., 1982). Treatment with Verapamil should always be started in hospital.

At present Amiodarone is not indicated for hemodynamic reasons but it may be combined with Propranolol to combine improvement in hemodynamics relief of symptoms and control of arrhythmias.

Verapamil and Amiodarone should not be given together.

Symptomatic Treatment

The principle symptom is dyspnea on exertion, due to the high left atrial pressure resulting from the stiff left ventricle and the high left ventricular end diastolic pressure which rises on effort. Angina is common, usually occurring on effort but prolonged cardiac ischemic pain can occur at rest and myocardial infarction (with normal coronary arteries) has been reported (Maron et al., 1979). The exact reasons for the angina are not known; massive oxygen demand by greatly hypertrophied muscle, impaired diastolic coronary flow in diastole due to the abnormal relaxation of the stiff left ventricle, and a metabolic fault leading to oxygen deprivation may all be considered as possibilities. Dizziness, syncope and palpitations, can all be ascribed to arrhythmia, but syncope may be due to "mechanical" factors; sudden fall in cardiac output due to reduction of left ventricular volume and resistance of the left ventricle to filling. It is doubtful if the outflow tract gradients alone contribute to syncope; they are produced by the hypertrophied walls of the left ventricle collapsing inwards when the ventricle is not distended with blood. When ventricular volume is seriously reduced the cavity of the ventricle may be virtually eliminated by the powerfully contracting hypertrophied muscle and in this way gradients are created.

The treatment of dyspnea is by beta adrenergic blocking agents or calcium blocking agents. Propranolol improves dyspnea in around 70% of patients, probably by reducing the stiffness of the left ventricle, improving relaxation and increasing the time available for filling (Webb Peploe et al., 1971; Alvares, 1980; Goodwin, 1982). In my experience a non-selective beta adrenergic blocking agent gives better results than a selective one which has less effect on outflow tract gradients (Hubner et al., 1973). Large doses, up to 300 mg a day, of Propranolol, are needed but smaller doses (40 mg a day) should be used initially.

When arrhythmias have been detected and there are also symptoms of angina and dyspnea, Amiodarone and Propranolol may be tried. This combination may prove to be superior to Verapamil.

Beta blockade does not relieve symptoms in every patient, probably because, owing to the grossly global and regional irregular patterns of relaxation and filling (Sanderson et al., 1978; Alvares, 1980), the effects vary with the extent and severity of the mal-orientated myofibrillar lesions.

Verapamil has been reported to improve symptoms. Kaltenbach et al. (1979) reported reduction in symptoms and in left ventricular muscle mass in 22 patients. Rosing et al. (1979[b]) reported improvement in symptoms in 11 of 15 patients, with increase in exercise tolerance in 6.

Adverse effects included bradycardia, sinus arrest, pulmonary edema and systemic hypotension.

Personal experience indicates that while Verapamil may relieve symptoms in some patients, the incidence of side effects and complications may be higher than with beta adrenergic blockade. It has so far not been possible to identify precisely which patients with hypertrophic cardiomyopathy are most likely to benefit from Verapamil and which are most likely to sustain untoward effects.

The first line treatment for symptoms should be beta blockade and the agent of choice should be Propranolol. If beta adrenergic blockade fails, then Verapamil should be tried cautiously starting with a small dose (20 mg three times daily) and working up to 120 mg three times daily. Patients who have heart failure or conduction defects are unsuitable. On present evidence Verapamil and Propranolol should not be given together. It must be emphasized that more precise identification of the hemodynamics in individual patients and clearer identification of sub-groups of patients is required before the exact place of Verapamil can be determined. The place of other calcium blocking agents has not yet been studied in detail; Nifedipine may prove to be helpful but care should be taken to prevent undue vaso-dilatation and hypotension.

Attacks of severe and prolonged chest pain may be intractable and difficult to treat. Frequently they do not respond to Propranolol. Verapamil may be tried in such patients, while Amiodarone has occasionally been shown to have dramatic effects for reasons that are not fully understood but perhaps because of its weak beta adrenergic blocking action.

When atrial fibrillation occurs, the cardiac output usually falls dramatically and pulmonary edema or congestive heart failure may occur because of the loss of atrial drive and tachycardia; embolism may occur. The patient should immediately be given Heparin and cardioversion carried out. If this fails the ventricular rate must be slowed and digitalis may be cautiously given, together with diuretics. There is usually little risk of precipitating an outflow tract crisis in such patients. Amiodarone, which stabilizes atrial fibrillation, may convert it to sinus rhythm and should be started, but care is necessary since Amiodarone increases digitalis blood levels, and digitalis should be reduced when the Amiodarone starts to take effect in 3-10 days. Small doses of Propranolol to slow the ventricular rate may be used as an alternative to Amiodarone, but care is needed if heart failure threatens.

Infective Endocarditis

Treatment of infective endocarditis in hypertrophic cardiomyopathy does not differ from infective endocarditis complicating other forms of heart disease. Since the mitral valve is the most usually affected valve the need for mitral valve replacement should be remembered if hemodynamic reasons or failure to control infection dictate surgical treatment.

Congestive Heart Failure

Heart failure usually indicates a severe and late form of the disease. It should be treated cautiously with diuretics.

Digitalis in Hypertrophic Cardiomyopathy

The dire effects ascribed to digitalis in exacerbating gradients and causing sudden death have not been confirmed by subsequent experience. However, the positive inotropic effects of digitalis make it likely that it will further reduce ventricular volume and since there is no impairment of systolic function from the disease until it becomes far advanced, digitalis should be avoided except where atrial fibrillation or congestive heart failure are present. The inconstant effects reported for digitalis may be explained by the potentially adverse effects of the positive

inotropic action on the one hand and the potentially beneficial effects of the negative chronotropic effect on the other.

Surgical Treatment

There is a small place for surgical treatment. There are two operations available; septal resection or mitral valve replacement. Septal resection is indicated in patients whose symptoms do not respond to adequate medical treatment and who have appreciable septal hypertrophy with a consistent systolic pressure gradient of 50 mmHg or more in the left ventricle. Results are excellent for the relief of symptoms, and improvement in hemodynamics occurs (Reitz et al., 1975; Kuhn et al., 1978; Maron et al., 1978). There is no evidence that septal resection improves prognosis and the operation is essentially for the relief of symptoms. The mortality is not low (8% operative and 9% on late follow-up, making a total of 17%) (Maron et al., 1978). Therefore, septal resection should not be undertaken lightly.

Mitral valve replacement is indicated in patients in whom mitral regurgitation is severe and intractable and requires surgical relief in its own right. Such mitral regurgitation is usually the result of secondary damage to the mitral valve as a result of calcification, turbulance or infection. In addition to relieving mitral regurgitation, mitral valve replacement makes room in the crowded ventricle by removal of the large papillary muscles. A low profile disc type of mitral prosthesis must be used, as there is usually insufficient room in the hypertrophied left ventricle for a caged ball type of valve.

The presence of associated fixed outflow tract obstruction is an important additional reason for operation.

Relief of Ventricular Hypertrophy and Modification of the Progress of the Disease

It has been postulated that beta adrenergic blockade by relieving outflow tract gradients may lessen the stimulus to progressive hypertrophy but proof is lacking. Verapamil on the other hand has been considered to reduce hypertrophy in one series (Kaltenbach et al., 1979).

The possibility of retarding the progress of the disorder is the most compelling reason for treating asymptomatic patients with a good prognosis; that is, those with no family history of sudden death and whose signs are of mild or moderate disease on clinical and hemodynamic grounds, but the possible side effects of long term treatment and the uncertainty of benefit tend to outweigh these

considerations. Regular ambulatory monitoring is advisable and if arrhythmias are detected then treatment is needed, Amiodarone being the drug of choice at the present time, quinidine being the next best alternative.

Pacemaking Treatment

A pacemaker may be required occasionally to treat hypertrophic cardiomyopathy. Patients who develop an unacceptable degree of bradycardia on beta adrenergic blockade may require a pacemaker, while those who have evidence of sinus node dysfunction with alternating tachycardia and bradycardia require a pacemaker in addition to anti-arrhythmic drugs.

DILATED CONGESTIVE CARDIOMYOPATHY

The prognosis is usually poor, approximately 50% of patients dying in the first two years after diagnosis and around 70% within eight years. Just over 20% remain alive and have a reasonable prognosis (Fuster et al., 1982). In our own experience of 146 patients followed for up to 20 years, prognosis was related mainly to left ventricular function (MacArthur et al., 1982).

Treatment is essentially that for congestive cardiac failure and no specific therapy is known. General measures include advice against cigarette smoking and alcohol and prolonged reduction of activity after the heart failure has been treated. Prolonged inactivity has become a hallowed principle of treatment but there is, in fact, little evidence or hard data to support this view. Digitalis is indicated if there is atrial fibrillation but its use in sinus rhythm has been questioned on the grounds that it may not exert any effective action after the first few months of treatment. Nevertheless if there is substantial cardiomegaly due to cardiac dilatation, and heart failure in sinus rhythm, digitalis should be given provided that there are no contra-indications. Careful attention must be paid to avoid digitalis toxicity.

Recently treatment by new inotropic and vaso-dilator drugs have been successful in the short term. Salbutamol given as an intravenous infusion in severe heart failure improves cardiac function, mainly as a result of systemic vaso-dilatation but also because of some positive inotropic effect (Sharma and Goodwin, 1978). However, there are disadvantages, notably tachycardia, tachyphylaxis, increase in blood sugar and fall in serum potassium. Vaso-dilator therapy with Hydrallazine has been successful in reducing left ventricular end diastolic pressure and pulmonary artery pressure, and increasing cardiac output without a significant fall in blood pressure or increase in heart rate. These effects are seen not

only when the drug is given intravenously, but also over a period of weeks and months by mouth, in doses of up to 300 mg a day (Fitchett et al., 1979). Patients who are slow acetylators may experience side effects; headaches, flushing or lupus syndrome with high doses. If possible acetylator status should be determined before treatment is started as fast acetylators can probably be given the larger doses with impunity.

As an alternative to Hydrallazine, Prazosyn may be used but tachyphylaxis is common. In desperate situations, if the systolic blood pressure is not below 90 mmHg, Nitroprusside may be cautiously given intravenously and titrated against the blood pressure and clinical response. Also, in desperate cases the positive inotropic agents Dobutamine or Dopamine may be used, and in combination with Nitroprusside may be life saving. Unfortunately, improvement after such severe congestive cardiac failure is usually only temporary, although a few patients make dramatic and unexpected recoveries, so hope should not be abandoned lightly.

The angiotensin concerting enzyme inhibitor Captopril may prove to be an effective agent for reducing afterload in chronic heart failure (Sharp et al., 1980) and could be useful in congestive cardiomyopathy.

Beta adrenergic Blockade in Congestive Dilated Cardiomyopathy

The use of beta adrenergic blocking agents in dilated congestive cardiomyopathy remains controversial and the indications uncertain. The work of Hjalmarsson and his colleagues (Waagstein et al., 1975) indicated improvement in cardiac function, on clinical and hemodynamic evidence, after the use of graded oral doses of Metoprolol, a cardioselective agent. Improvement in prognosis was claimed also. The reason for the results are not understood. The patients who are most suitable for treatment may be those with disproportionate tachycardia who may have an excess of catecholamines in the heart (Waagstein et al., 1979). Certainly judicious slowing of the heart rate might be beneficial by allowing more time for ventricular filling. Treatment with beta adrenergic blocking agents should not be undertaken at random but should be part of a carefully controlled clinical trial.

The treatment of complications or threatened complications of dilated congestive cardiomyopathy is important. When there is severe congestive heart failure, with low cardiac output and peripheral edema and the patient is immobilized, anti-coagulants are advisable to protect against deep venous thrombosis and pulmonary embolism. In patients with atrial fibrillation anti-coagulants are indicated to avert systemic embolism.

The Implications of the Virus/Immune Theory of Causation on Treatment

The possibility that virus myocarditis or some infective agent might lead to dilated congestive cardiomyopathy either directly or by an auto immune mechanism was suggested a number of years ago (Kawii, 1971; Braimbridge et al., 1967). High titres to Coxsackie B viruses in the blood of patients with dilated congestive cardiomyopathy as compared to controls was reported by our group (Cambridge et al., 1979). In a number of patients endomyocardial biopsy did not show any evidence of myocarditis, and the possibility of a disturbance of cellular immunity resulting from the infection and causing progressive myocardial damage has been suggested. Work from Stanford University by Fowles et al. (1979) suggested a defect of suppressor cell function in dilated congestive cardiomyopathy, which might be explained in this way. However, the occasional finding of dilated congestive cardiomyopathy on a familial basis suggests that perhaps the opposite could sometimes be true; that is, that inherited defects in cellular immunity might predispose to virus infection. The matter is unsettled and the bearing on current treatment unclear. However, another recent report from Stanford by Mason et al. (1980) has therapeutic implications. These workers reported a small series of patients with a short history of congestive heart failure of unknown cause. Endomyocardial biopsy revealed evidence of myocarditis and treatment with immunosuppressive and steroid drugs caused improvement (as judged by clinical, hemodynamic and endomyocardial biopsy evidence) in the majority.

It is not clear whether these patients were suffering from myocarditis, or dilated congestive cardiomyopathy or that myocarditis led to the latter, but the implications for treatment are clear. Patients who apparently have dilated congestive cardiomyopathy of recent onset should be investigated by endomyocardial biopsy if possible, and if evidence of myocarditis is found, treatment with immunosuppressive drugs and steroids should be considered.

In the terminal patient, for whom all methods of treatment have proved unavailing, cardiac transplantation may be considered provided that there are no contra-indications.

In patients with arrhythmic cardiomyopathies (Goodwin, 1979) Amiodarone may be of great value for it is effective in controlling arrhythmias without depressing myocardial function. Most antiarrhythmic drugs have some negative effect on cardiac function, so Amiodarone is likely to be the drug of choice.

Restrictive/Obliterative Cardiomyopathy

Endomyocardial fibrosis is the usual cause of this type of cardiomyopathy. Since the hemodynamics and pathology are closely

similar in the tropical variety and in the temperate zone type known as Löffler's endomyocardial disease, treatment is basically the same and is in general unsatisfactory. The usual measures for congestive heart failure are indicated and anti-coagulants should be used because of the high risk of thrombosis in the ventricle. In Löffler's disease the eosinophilia, which may affect other organs, can be treated by cytotoxic drugs or steroids, but the result is usually unsatisfactory except in relieving exacerbations. The introduction of surgical means has improved prognosis. Resection of endocardial fibrous plaques and replacement of atrioventricular valves when severely involved in the disease has had considerable success in the short term, but long term results are not yet available (Dubost et al., 1976; Davies et al., 1981; Spry and Tai, 1976).

Table 1. Suggested Plan of Management for Hypertrophic Cardiomyopathy

1.	No family history, no symptoms, mild disease, no arrhythmia. Observe regularly. Holter monitoring.
2.	Ventricular arrhythmia: Amiodarone.
3.	Symptoms - no arrhythmia: beta blockade.
4.	Symptoms - but unsuitable for beta blockade: Verapamil if no AV conduction defect and LA pressure not significantly elevated. Watch carefully.
5.	Symptoms and arrhythmia: Propranolol, Amiodarone.
6.	Symptoms - gradient at or greater than 50 mmHg, no response to medical treatment. Septal resection.
7.	Severe complicated mitral regurgitation: mitral valve replacement with low profile mechanical prosthesis and excision of papillary muscle.

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HEART IN OUTER SPACE

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During the 25 years that have elapsed since the first sputnik in space, space research has scored spectacular achievements. This follows from the simple fact that 109 men of the Earth have already made space flights and that men and lunar vehicles have left their traces on the Moon. Space probes have informed us about the environmental parameters on the planets of the solar system and in outer space.

Many investigations, particularly of recent years, give evidence that within the solar system the Earth appears to be the only place where life exists. This inference is fraught with very important consequences: 1) it points to the unique significance of the Earth biosphere in our solar system, 2) it attracts our attention to the very specific place man occupies in the "Earth biological envelope", to his historical responsibility for the preservation of the biosphere and the healthy development of the human society, and 3) it strictly confines hypotheses on the origin and evolution of planetary systems and the conditions required for the emergence, existence and evolution of life in the Universe.

A space environment as such is alien to life and, therefore, it can be penetrated if living creatures, man included, are protected against its harmful effects. This is why space cabin atmospheres are normally very close to those on the Earth. Space flights are characterized by the so-called free fall which gives rise to the state of dynamic weightlessness. Weightlessness or zero-g is of particular interest for biologists and medical people because the entire biological evolution, at least that which began with the emergence of the cell and ended with the appearance of man, developed under conditions of a constant gravity field.

Space flights greatly stimulated interest in the biological role of gravity. Today we have the right to speak about a new branch of biology, gravitational biology, the purpose of which is to study in great detail the effect of weightlessness on the structure and function of living organisms, including cardiovascular physiology. This is the subject of the present paper.

MAN IN SPACE

Man, as the prominent astronomer Shapley remarked, was born by stars and, therefore, it is not surprising that he tries to go back to them. But the wise men of ancient times who used to say that the road to the stars is hard were also correct. The problems man may encounter in space flight are of great diversity. Among them mention should be made of those that may influence the health state of man: weightlessness, cosmic radiation, abnormal environmental parameters, and factors causing nervous-emotional stress.

It is known that the sources of cosmic radiation are: galactic cosmic radiation from the depth of outer space, radiation of solar flares, and Earth radiation belts. Careful dosimetric measurements carried out in every space flight demonstrated that the absorbed radiation dose during lunar flights (whose trajectory crossed the Earth radiation belts) was about 0.5 rem and never exceeded 1.0 rem. During long-term orbital flights of about 6 months in duration the dose increased and reached 5.0 rem. However, this is still lower th the permissible dose which is taken of 15 rem per flight (in the US allowable dose is 25 rem).

Thus, the radiation factor is not very dramatic at least today and cannot be viewed as one influencing the cardiovascular function in space flight.

In flight crewmembers live and work in a specific environment which reflects tendencies of our technically advanced society. It is, first of all, characterized by the man-made environment. It is a classical example of a compromise between human requirements and technical feasibility. Then, a confined enclosure - a small living space which amounts to several cubic meters per man. This diminishes motor activity, enhances hypokinesia, stimulates cardiovascular deconditioning.

At first sight, the physical and chemical parameters of the space cabin environment are stable. However, this is not quite the case, because the pressure, temperature, humidity and the concentration of the major constituents (O_2, N_2, CO_2) vary continuously; the content of trace contaminants, including anthropogenic, increases and the microbial composition changes. This gives rise to problems associated with the maintenance of the environmental parameters within normal limits because their marked

deviations may affect the physiological functions of crewmembers.

Of greater importance is a large group of various circumstances and factors that may induce and maintain nervous-emotional stress. As a matter of fact, flight starts still on the Earth and the first critical state is the launch. Space crewmembers typically show increased heart rate and a more or less increased blood pressure as the launch time approaches. Immediately after launch heart rate begins to go down, in spite of growing acceleration, thus reflecting a typical "prelaunch" reaction of sympatho-adrenal origin. The shock of novelty, great diversity of unexpected and memorable impressions, heavy duties combined with expected and actual complications and hazardous situations - these are specific features of space missions which may bring about more or less expressed reactions of the heart and the cardiovascular system on the whole. Before considering the role of zero-g, it is appropriate to discuss general problems of gravitational biology.

BIOLOGICAL ROLE OF GRAVITY

Gravity is a great and universal designer of the Universe - the force of gravity has determined the emergence, structure and environment of planets. Gravity is a constant and ubiquitous phenomenon which generates a very weak field. Therefore, living systems that have a different mass and spatial configuration show different sensitivity to gravity effects. Large living organisms, man included, distinctly react to gravity variations. Smaller animals, e.g. insects, respond much more markedly to surface tension. Microorganisms are indifferent to gravity and obey the laws of viscosity and Brownian movement.

In the course of evolution Nature has staged two important experiments associated with gravity effect. The first took place when living being moved from the ocean to the land, and the second when our ancestors developed the ability of uprightness. The success was due to the improvement of the structure and function of the cardiovascular and musculoskeletal systems. The development of two-leggedness, enlargement of the brain, ability for conceptual thinking and the upright body position in the Earth gravity field look like functionally ingenious achievements of Nature. They have armed man with great advantages, including social ones, and given rise to certain biological problems.

Since man spends a considerable and most active part of his life in the erect position, he has to maintain his body mass center in unstable equilibrium, use his muscles constantly, especially leg muscles in order to retain the posture. It is not at all surprising that the leg mass is close to half of the total body mass and the parameters characterizing motor activity are important in the control of homeostatic reactions. In contrast to man, very few mammals are capable to acquire the upright posture

and, if so, only for a short period of time. The fact that men do that without any problems is a considerable evolutionary achievement, because in this position the cardiovascular system actively counteracts gravity-induced blood redistribution. This ability is termed orthostatic tolerance and realized via a concordant interaction of the cardiovascular structure and regulatory mechanisms.

Concluding this Section, it should be emphasized again that the force of gravity, at least in the limits faced on the Earth or in space does not produce a direct effect at the molecular, subcellular or cellular levels. Its direct or indirect effect can be seen in organisms weighing several grams and very clearly in us, humans, who enjoy the privilege to view the world when standing.

The adaptation we have evolved is a philogenetic inherent property. In our individual life we can improve it or we can partially lose it. This normally occurs in aged or diseased people. But it may also develop in young healthy men who are travelling into outer space where weightlessness reigns.

WEIGHTLESSNESS

In the weightless state the body has no support, it is weightless and floats freely in the space cabin. On the Earth man can face nothing of the kind. In the course of his evolutionary development he was unable to acquire any specific mechanisms for compensating the effects associated with the weightless state. On the contrary, he developed the ability to counteract the gravitational stress. What are his responses to weightlessness?

From the intellectual and emotional point of view - these are more or less distinct disorders of spatial orientation, a wide spectrum of emotional reactions of positive or negative nature. With respect to autonomic functions these are equivalent physiological reactions which may arise on the ground when man drastically reduces his motor activity (hypokinesia, hypodynamics), assumes a horizontal position (bed rest) or goes down into the water (immersion).

Thus, weightlessness in space flight is a factor responsible for changes in the self-regulation of an intact organism which are aimed at establishing adequate relations with the lowered requirements of the environment. The exposure to zero-g is accompanied by the phenomena of disuse or unloading and then atrophy from disuse and deconditioning, which can be viewed as adaptive.

The functional changes that occur in weightlessness are primarily produced by the following principal factors: changes in the afferent compartment of the nervous system, withdrawal of the blood hydro-

static pressure, and the lack of weight load on the musculo-skeletal system. The responses to these changes develop at different time intervals. Besides, the physiological reactions have a characteristic time constant and may be modified by a simultaneous effect of other stress-factors of space flight. All this makes a very complicated picture of functional changes in response to the effects of weightlessness.

INVESTIGATION OF CARDIOVASCULAR FUNCTION IN SPACE FLIGHT

Methods of Research

After the historical flight of Yu. Gagarin in which ECG was recorded in one lead, a spectacular step forward was made in cardiovascular research techniques. The spacecraft and especially space stations are well equipped with sophisticated biomedical equipment furnishing diagnostic and scientific information. Several generations of the biomedical equipment were used in succession and now the Salyut-7 station is supplied with devices to record electrocardiograms in 12 leads, to perform dynamic electrocardiography in one lead continuously during 24 hours, to perform rheography of the head, body and limbs, to measure blood pressure by means of Korotkoff sounds, to record sphygmo - and tachooscilograms, seismograms, kinetocardiograms, ballistocardiograms and, finally, echocardiograms using the ultrasound Doppler method. In addition, many other parameters necessary for interpreting cardiovascular data, e.g. variations in body mass, leg volume, hematocrit, can be measured.

At first sight it looks as if space medicine has overcome the barrier of methodical problems. But this is not the case. The available methods enable one to get indirect and sometimes relatively inaccurate values of the main hemodynamic parameters, thus making difficult reliable evaluation of regulation mechanisms.

Another characteristic feature of cardiovascular investigations is their discrete pattern. The crewmembers are to perform many duties associated with flight control, scientific and engineering experiments, etc. In short-term flights a comparatively small amount of biomedical information is transmitted to the Earth almost every day. In long-term space flights instrumented investigations are carried out approximately once every 10 days which appears adequate for the medical control but less than it is necessary to study the functional variations and the adaptation process.

Without taking into account those who are aloft today and those who are on the verge of going into orbit there are 106 persons that have made space flights. Out of these 106 courageous space

travellers one (astronaut John Young) has made five flights, three have made four flights, twelve have made three flights, twenty six have made two flights and sixty four have made one flight, which is in total 169 man-flights and about 8 years 105 days of life in space.

When considering the scientific analysis of the accumulated data, it should be borne in mind that in the 169 space studies 106 crewmembers participated, each having specific features of his own. The scope of measurements in these flights varied substantially and each flight differed from another. All this makes it difficult to compare and analyze the findings, to evaluate their adequacy and to draw reliable conclusions concerning the effects to which man is exposed.

However, this problem can be somehow resolved with the aid of ground-based experiments simulating the physiological effects of weightlessness which include: bed rest (hypokinesia) and its modification - head-down tilt (antiorthostatic hypokinesia), and water immersion (at neutral temperature).

A large number of studies of this kind have been conducted. The results of ground-based studies and mathematical modeling widely used at present are employed to elucidate physiological reactions to weightlessness. This approach is undoubtedly well justified because it allows the conduct of an unlimited number of physiological experiments which can hardly be carried out on-board the spacecraft.

Primary and Delayed Cardiovascular Reactions

In weightlessness cosmonauts and astronauts experienced blood run to the head and nasal congestion, then head heaviness and pulsation. The level of these sensations varied. Sometimes they reported that the sensations persisted for 10-15 days and then disappeared. Sometimes they persisted throughout the flight although were less distinct. When looking at his partner, the crewmembers noted face puffiness, eyelid swelling and sclera hyperemia. Some of the symptoms augmented by the end of the working day or weakened during exercise or LBNP (lower body negative pressure) tests.

This is what happens after insertion into orbit when the hydrostatic pressure disappears and blood as well as the interstitial fluid move headwards. Their major portions go to the intrathoracic vessels, causing their enlargement. This also occurs in the cardiac cavities. The remaining portion of the fluids accumulates in the skin and in the subcutaneous tissue of the head and neck.

On the ground a similar situation occurs during the transition from the vertical to the horizontal position. In this case fluid redistribution is moderate; it is a normal physiological reaction which does not manifest in the healthy man. Many experiments carried out with the aid of different techniques yielded similar results. The volume of this, so to say, mobile fluid is about 500-700 ml on the ground.

Today we have reliable data on the dynamics of blood filling of different body compartments under various conditions and at a different level of activity. The results of many ground-based studies indicate that with respect to the effect of blood redistribution "laboratory models of weightlessness" can be ranked in a certain order with the head-down tilt at -6^{o} being the most accurate simulation of what happens in real weightlessness. This is now recognized by most researchers in the area.

Comparing a conservative number of indirect measurements in space flights with solid data of ground-based studies, we can evaluate the mobile fluid volume displaced from the lower body in the cranial direction. It is estimated to be twice as much as during the head-down tilt on the ground, i.e. about ±500ml. Figuratively speaking, weightlessness turns the man upside-down.

The measurements during the Salyut and Skylab flights demonstrated that the leg volume decreased by 12-18%, lowering most rapidly during the first days in weightlessness. An opposite situation occurred upon return to Earth. In flight the body mass center also shifted towards the head, being another indication of the cephalad fluid displacement. Orthostatic tests have shown that this reaction consists of two components: a rapid one associated with blood movement along the vascular bed, and a slow component induced by interstitial fluid accumulation.

It is obvious that the discussion of these primary, physically meaningful processes developing in the vascular system should take into consideration changes in the transcapillary fluid displacement. The final volume of the displaced fluid may largely depend on the contribution of the hydrostatic and osmo-oncotic pressure, as well as on tissue distensibility. These problems have been poorly studied. There are no data in the literature concerning the lymph flow. Therefore, fluid regulation requires further study both on Earth and in space.

It is believed that an increased blood inflow to the heart causes reflex and humoral reactions which result in a decrease of the plasma volume and, consequently, of the circulating blood volume.

The scope and rate of the plasma volume decrease in weightlessness can be understood with the aid of immersion studies which produce an effect that exceeds that of weightlessness. Following an early increase, the blood and plasma volume declines, the reduction being pronounced by the 2nd-3rd hour of the exposure. The greatest changes are seen by the 6-8th hour, when the plasma volume decreases by 8-15% (in some cases by 20%). Having reached this level, the plasma volume remains practically unchanged till the end of the exposure even if it lasts several days. This is easy to understand because this is an adaptive reaction aimed at establishing equilibrium between the volume of the intravascular fluid and that of the vascular bed.

This scheme gives a satisfactory explanation to the fluid volume regulation at acute stages of different experiments and space flights. However, as any other scheme, it disregards other possibilities and factors. It has been, for instance, shown that blood redistribution during ortho- and antiorthostatic reactions are noticeably affected by afferent somatic impulses that modify blood redistribution between the cranial and caudal body parts.

Similar reactions can be elicited from the chemoreceptor zones of the medulla oblongata ventral surface; this is of particular importance in view of increased blood filling of the head. The above hypothesis also ignores the role of hypothalamic osmoreceptors which are assumed to regulate ADH secretion. It should also be emphasized that in real life pathways of homeostatic equalibrium are at least doubled, as in the case with redundant spacecraft control systems. The actual reactions consist of combinations of different elements which may be of greater or lesser importance in different situations.

At the stage of acute adaptation when the circulating blood volume and plasma volume diminish, crewmembers exhibit, as a rule, losses of thirst and salt appetite, reduced water consumption and sometimes they are requested to drink water or juice. Due to the losses of plasma, the hematocrit level increases. These findings were documented in flight and confirmed by ground-based investigations. So far it is still unclear in what way the normal relation between the plasma volume and blood formed elements recovers, although it is known that the production of red blood cells declines and they have a tendency for microcytosis.

This essentially completes the early stage of adaptation to weightlessness. A new homeostatic level of the cardiovascular function is reached; it helps crewmembers maintain good health condition. In flights ECG was recorded many times. No unusual or abnormal changes in heart rate, rhythm (except for single cases of ectopic beats and one case of higeminy), pattern or parameters of ECG were observed.

Being very exacting, we can say that there was a difference between heart rates at night and in the daytime. In very few cases there were subjective complaints for discomfortable cardiac sensations which were never supported by ECG or other objective studies; there were no significant changes in blood pressure.

In a word, all the parameters measured and recorded at rest demonstrate complete well-being of crewmembers. But as soon as they increase their activity or start doing something difficult, they show unstable or hypertrophic cardiovascular responses as compared to those seen on the Earth. This was clearly seen during bicycle ergometry and LBNP tests. Preflight data make it possible to compare variations in their tolerance to exercises and orthostatic (LBNP) tests. As a rule, the tolerance declined, the reduction increasing as they diminished their exercises. However, the time-course of these variations was not ascertained because in all space flights of more than 2 weeks duration exercises were a mandatory prophylactic measure.

On the whole, we can see a very curious picture. Following the primary, acute phase of cardiovascular adaptation which completes within the first hours or days in orbit, a new homeostatic level is reached. This proceeds gradually and continuously for about a month or sometimes a month and a half. This level may be termed a relatively stable adaptation to weightlessness. Crewmembers adjust to the new environment, feel well, develop habits and skills necessary for their life and work in weightlessness, and face no serious problems. It can be said that a man of the Earth has becom e a man of space.

However, everyone who has made a space flight encounters certain, sometimes significant problems upon return to Earth. This uneasy and frequently difficult process of readaptation is the biological price of the privilege to fly into space.

Unlike flight studies, pre- and postflight examinations are wide, profound and less limited. However, the problem is that postflight the investigator deals with a person who is not in the weightless state but who was in zero-g but at the time of examination is at 1 g. This, obviously, makes data analysis and interpretation more difficult and speculative. Nevertheless, the data accumulated are extensive and can be used not only to evaluate readaptation reactions but also to take a glance into weightlessness per se.

After touch-down physicians try to perform examinations as soon as possible but sometimes fail to do that to a full extent. Nevertheless, during the first hours after recovery the following changes were demonstrated: losses of body mass, leg volume, and

circulating blood, increased heart rate, pulse variations, and trend for hypotension. X-ray examination showed a decrease of the heart shadow. Echocardiographic investigations suggested that the decrease of the heart size may be induced partially or totally by a reduction of the cardiac cavities rather than of the cardiac muscle mass.

The application of quantitative electrocardiography (e.g. according to Frank) and noninvasive methods to determine the phasic structure of the cardiac cycle did not reveal any abnormalities but indicated manifest individual variations. Biochemical and hormonal parameters of blood varied substantially and often in different directions. A consistent finding was a decrease of orthostatic tolerance and exercise tolerance, as shown by bicycle ergometry.

The level of the variations was not always correlated with the flight duration. It should be emphasized that a more active and regular use of different countermeasures - bicycle ergometer, treadmill, bungee cords, LBNP box - combined with other measures (adequate work-rest cycle, food and water consumption, water-salt supplementation) helped diminish the postflight reactions. In this context, mention should be made of the long-duration space flights onboard Salyut-6.

Nevertheless, we have to admit that our knowledge of cardiovascular reactions to weightlessness is still insufficient. In the near future we have to clarify the effect of cephalad fluid shifts and concomitant increased ventricular pressure on the morphofunctional characteristics of the heart; we have to ascertain whether a prolonged exposure to weightlessness may cause myocardial dysfunction; and we have to determine the role and importance of general and local mechanisms of cardiovascular regulation in weightlessness. This can be done provided that the existing methods are refined and the great achievements of modern cardiology are used in the best way possible.

It is hoped that the progress achieved within a short period of our space era will become more impressive. Many problems with which we are concerned today will be resolved but they will be replaced with new challenges which we will attempt to resolve with similar enthusiasm and dedication.

PROGRESS IN RADIONUCLIDE METHODS

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Radioisotopes are tools for observing the body at the molecular level of organisation practically without discomfort and risk to the patient. In this context, the method of imaging, for example, with a fast gamma-camera, is not an end in itself, but a first step for functional studies particularly at the molecular and cellular level in small body volumes. Thus, radioisotopes have generally proved useful for establishing diagnosis, for description and quantification of findings involving metabolic reactions in the evolution of disease and for control of therapy. Nuclear medicine has also become accepted in cardiology.

Radioisotopes are employed in cardiology mainly to study 1) cardiac function, 2) myocardial perfusion, 3) myocardial metabolism. This report emphasizes progress and covers these three topics.

1) CARDIAC FUNCTION

Cardiac function is essayed by measuring volumes, flow and wall motion at rest, and exercise for defining cardiac reserve.

Multiple Blood Volume Analysis

A rather novel technique uses the simultaneous imaging of blood pools of the heart, lung and liver.[1,2,3] Following equilibration of the tracer such as 99m-Tc labelled erythrocytes within the circulation counting rates relate to blood volume and are simultaneously measured over heart, lung and liver at frequent time intervals during stepwise increase of exercise load. In normal individuals the cardiac

blood volume shows an initial adjustment at the lowest work load level; then with increasing load it tends to decrease slowly. Over the lung, there is initially little change, then with more than about 60 watt the blood volume increases. There is little initial change in the liver blood volume and then a constant drop is seen. This thus indicates in normals an exercise induced diminution of total cardiac volume, an increase in pulmonary blood volume and a delayed mobilization of liver blood pool.

The responses are quite different in patients with latent cardiac insufficiency. There is an exercise induced increase in total cardiac volume, an immediate rise in pulmonary blood volume and an immediately induced mobilization of hepatic blood volume. The simplicity and potential applicability of this multiple blood volume analysis is obvious.

Cardiac Blood Pool Analysis

Cardiac function is most widely investigated by continuously observing the left ventricular blood pool, as initiated by Hoffmann and Kleine in 1965[4] and later adapted to the gamma-camera with ECG triggering.[5,6] Following tracer equilibration in the blood, the left ventricular blood pool is well recognized in the left anterior oblique projection.

a) Beat by beat observation. Beat by beat analysis of the total counting rate of the left ventricle is relatively simple by the nuclear stethoscope[7] proposed by Wagner, or more recently by a mini-probe with a sensitive solid state detector that may even be included in a garment to observe the patient throughout his daily activities.[8] These techniques permit continuous beat to beat analysis of left ventricular function and thus are a radiocardiographic equivalent of the continuous ECG monitoring.

b) Gated blood pool imaging. A commonly used mode permits the collection of the total counting rates from the left ventricle from at least 20 divisions of the cardiac cycle by ECG triggering, which are then added up over about 400-500 cycles so that good counting statistics for each cycle division are obtained. Thus, the counting rates during a representative cardiac cycle present a volume curve that lets one calculate left ventricular ejection fraction, filling-time and -rate, and ejection-time and -rate, as shown in Figure 1. The ejection fraction is given by the difference between enddiastolic and endsystolic counting rates divided by the diastolic counting rate after background substraction. Many investigators have shown the ejection fraction to have a high prognostic value, especially when the stress induced response is taken as major parameter.[9] A stress induced reduction of the ejection fraction by more than 5% indicates an about 60% incidence of a critical coronary artery lesion.[10]

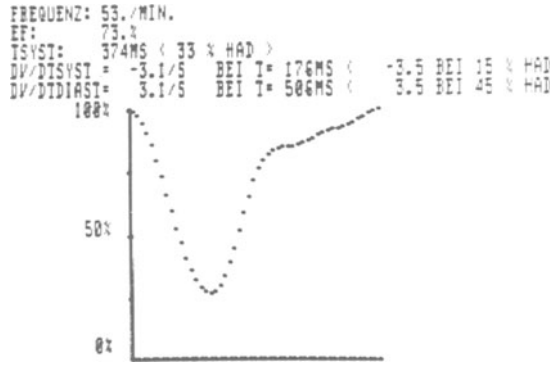


Fig. 1. Left ventricular volume curve obtained from gated blood pool images. The curve permits the calculation of the ejection fraction, systolic time and diastolic time and the corresponding ejection and filling rates.

The left ventricular volume curve may also be obtained from each individual image pixel and may be treated by Fourrier analysis, as suggested by Adam.[11] Each curve from a pixel is adjusted to a sinusoidal wave having a certain amplitude and phase. Whereas the amplitude relates to the extent of wall motion, the phase emphasizes the time at which contraction occurs. The values for phase and amplitude from each pixel may be displayed in colour for each individual pixel as shown in Figure 2. On the left there is a normal display of pixel amplitudes with the maximum in the mid ventricular region. On the right, the corresponding phase distribution image is seen; the dark color throughout indicates a synchronous beginning of ventricular contraction.

Figure 3 illustrates corresponding data from a patient with a left ventricular aneurysm. The upper 2 images are the enddiastolic and endsystolic unprocessed blood pool displays. After Fourrier analysis for each pixel curve, the lower left gives the amplitude image with a zero signal given in black in the mid ventricular region where the aneurysm is located; on the right, the phase distribution image shows an asynchronous contraction of the left ventricle with initiation of contraction at the cardiac base in dark color and late contraction in light color at the apical region.

Wall motion abnormalities are well correlated to the ejection fraction. Stress induced abnormalities were found in 67% of cases with 1 vessel disease, in 88% with 2 vessel disease and in about 75% of the cases with 3 vessel disease.[12]

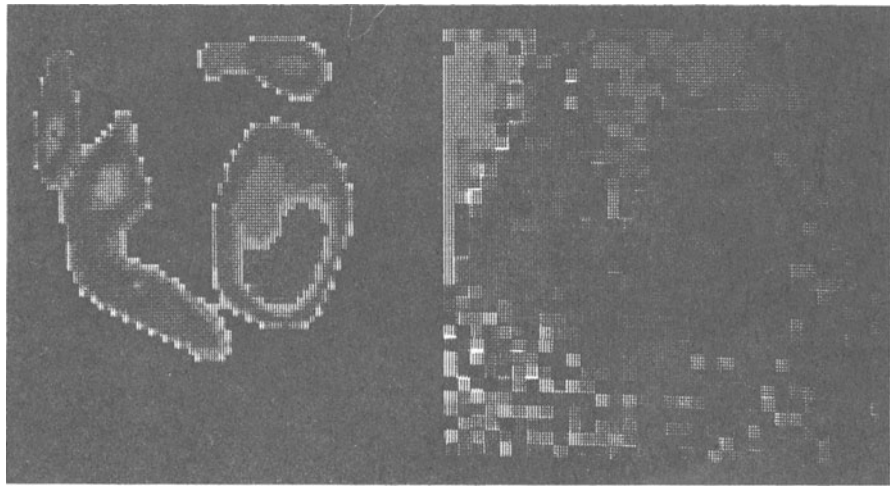


Fig. 2. Parametric images of a normal left ventricle with respect to amplitude (left) and phase (right) of ventricular volume curves obtained for individual image pixels.

What is now easily achievable with the left ventricle, is in most instances quite difficult with the right ventricle because of overlapping of atrial and ventricular volumes.

Still, the right ventricular blood pool may be analysed, even with difficulties, by the so-called first pass technique[13] and here preferably with short lived radionuclides, such as 81m-Kr.[14]

The assessment of both ventricular volumes from the first pass of an indicator bolus through the heart principally has the advantage of speed, and short lived radionuclides are generally preferred, for example 195-Gold.[15] Comparing right and left ventricular ejection fractions may help to analyse valvular regurgitation.[16]

It is difficult to determine absolute volumes from the first pass or from the gated blood pool technique.[17] A recent report by Maurer et al.,[18] describes an easy measurement of the attenuation of radiation coming from the ventricles. A gelatine capsule filled with 99m-Tc is measured before the camera head and again on its passage through the esophagus. The resulting attenuation coefficient

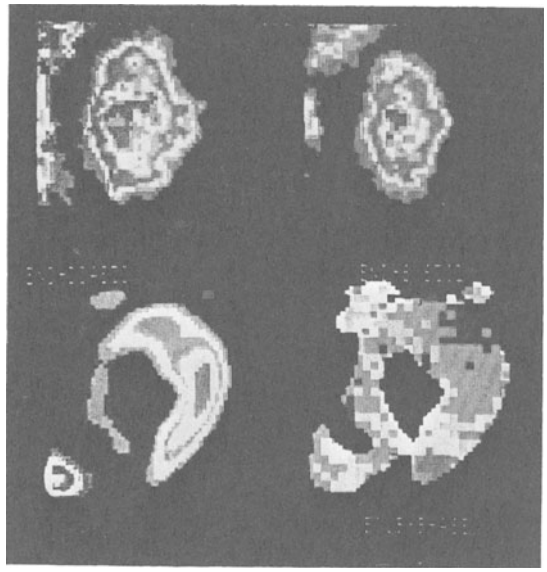


Fig. 3. Left ventricular aneurysm. The upper images show enddiastolic and endsystolic volume images. Below are the corresponding parametric images for amplitude and phase.

is then applied to a blood sample of the patient at the time of ventricular counting; this quickly permits the conversion of left ventricular counting rate into volume.

Flow Time, Minimal Transit Time

Quantification of the entire first pass of a radioactive bolus yields information on global function of all segments of the central circulation. For this purpose the minimal transit times have proved very useful, which my group began to measure in 1969.[19,20]

Following injection of a radioactive bolus, for example of 99m-Tc-pertechnetate, into an antecubital vein, the first pass is registered with the gamma-camera at a rate of at least 10 frames per second. Subsequently, frames are summed up for image display of the right and left side of the heart, as seen in Figure 4. Into these images regions of interest are placed for covering the superior caval vein, the right atrium, right ventricle, the pulmonary artery,

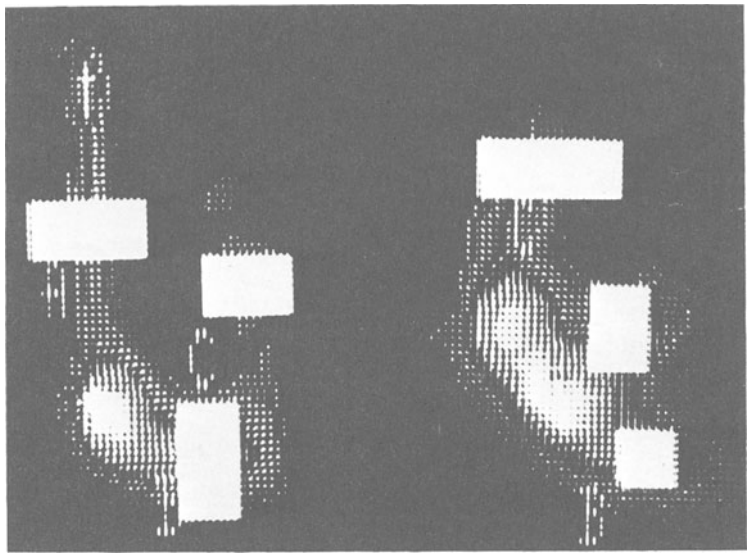


Fig. 4. The first pass of the indicator bolus through right and left heart may be quantified by measuring indicator arrival times at the indicated regions of interest for superior caval vein, right ventricle, pulmonary artery, left atrium, left ventricle and aortic arch.

the left atrium, left ventricle and the aortic root. It is not necessary that these regions fully cover the anatomical site, yet one must prevent the regions from extending into the intersegmental boundaries. Time activity curves are then generated from each region, as shown in Figure 5.

The time activity curves are illustrated in Figure 5, from the right atrium, right ventricle, pulmonary artery, left atrium, left ventricle and aorta; they are smoothed in such a way that the arrival of the tracer can be automatically detected.[21] The arrival of the indicator in the various regions is here defined by vertical bars on each corresponding curve. The time differences between consecutive arrivals are the fastest transport times or minimal transit times. They are a function of the ratio of volume to flow and thus are, in the absence of flow vortices, inversely proportional to the ejection fraction and heart rate. At a heart rate of 80 per minute, the total minimal transit time from the right atrium to the aorta in normal individuals, supine position, is 6 seconds ± 5% standard

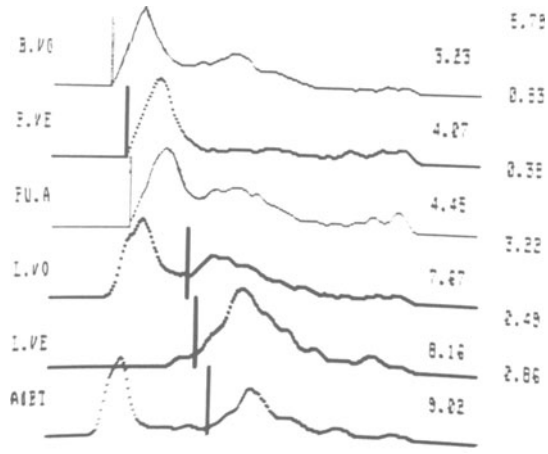


Fig. 5. Time activity curves from the regions of interest shown in Figure 4. The activity arrival is automatically recognized and is displayed here by vertical bars. The differences between consecutive arrival times are the fastest flow times or minimal transit times through the central circulation. These transit times are an expression of the ratio volume/flow.

deviation. The pulmonary minimal transit time is 3.2 seconds ± 8% standard deviation, the individual segmental minimal transit times through atria and ventricles vary between 0.5 and 1.0 second ± 15-20% standard deviation.[19] The values are slightly different in patients measured in the upright position.[22] It is thus relatively easy to describe the global function in terms of volume to flow ratio for each cardiac segment and the entire central circulation. Also the right ventricle is easier accessible than with the gated blood pool technique.

I propose to combine quantification of the first pass by the minimal transit times with the gated blood pool technique.[23]

Whereas the gated blood pool technique gives detailed information on left ventricular ejection fraction and wall motion, the minimal transit times give global information on all cardiopulmonary segments in terms of volume to flow ratios. The combined procedure was tested in 65 patients with coronary artery disease, in supine position. Thirty seven of the patients had a history of myocardial infarction, of which 17 had developed left ventricular aneurysm.

Figure 6 shows the relationship between left ventricular ejection fraction and cardiac minimal transit time, i.e. total transit time minus pulmonary transit time; the patients with aneurysm are shown by full dots. In patients without aneurysms, pathological minimal transit times are about similarly frequent as are diminished left ventricular ejection fractions. Patients with aneurysms have more frequently a diminished left ventricular ejection fraction than a pathological cardiac minimal transit time. It was interesting to note that patients with a normal minimal transit time usually had a better exercise tolerance than those with prolonged minimal transit times. It is justified to speculate whether minimal transit time readings may have prognostic value. A compact radiocardiograph was designed for easily and specifically measuring simultaneously total minimal transit time through the central circulation[24] and is now also being used for simultaneously assaying left ventricular ejection fraction, filling-time and -rate, and ejection-time and -rate.[24]

2) MYOCARDIAL PERFUSION

Myocardial scintigraphy with 201-thallium has proven to be of great diagnostic value in recognizing coronary artery disease with a sensitivity ranging from about 75-90%.[25,26] This technique has become the most widely used nuclear medical application in cardiology.

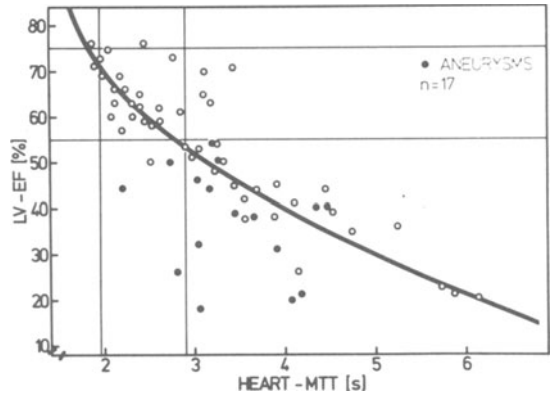


Fig. 6. Left ventricular ejection fraction, as measured by the gated blood pool technique, is correlated to the cardiac minimal transit time. Each individual sign relates to one patient. The solid dots relate to patients with left ventricular aneurysms.

It was tempting to check the potential diagnostic advantage of combining 201-thallium myocardial scintigraphy with the minimal transit time measurement and the gated blood pool analysis.

In a first study 33 patients with coronary artery disease including 9 with a history of myocardial infarction were investigated at rest and at peak exercise.

Figure 7 gives columns listing the number of patients with pathological findings either on the thallium-scan, upon gated blood pool analysis of ejection fraction and wall motion, or upon minimal transit time measurements, at rest and at exercise. A pathological score for minimal transit times was a significant prolongation; for the gated blood pool study it was a wall motion abnormality and/or a left ventricular ejection fraction below 55%; for the thallium-scan it was an accumulation defect. It is quite obvious that stress testing, of course, increases the sensitivity of all procedures. The highest sensitivity was obtained with the thallium-scan.

For evaluating the results of the 3 tests per individual at rest and exercise, the group of 33 patients with coronary artery disease was combined with additional 5 patients with the history of myocardial infarction but without obvious ischemia, 5 patients with cardiomyopathy and 10 normals, who, however, displayed some unspecific chest complaints. The 201-thallium-scan gave the highest sensitivity of 85% with a specificity of 60%. For the gated blood pool analysis

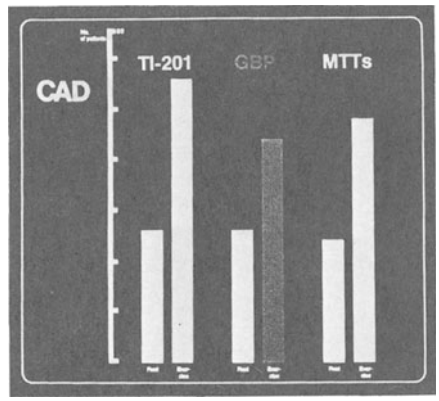


Fig. 7. Thirty three patients with coronary artery disease underwent examination for myocardial perfusion with 201-thallium, for cardiac function by the gated blood pool technique or by the minimal transit time measurement. The bars indicate the incidence of pathological findings at rest and exercise.

and the minimal transit time measurements about similar sensitivities of about 70% were obtained with a highest specificity of 87% for the gated blood pool study. When the values per patient were combined, sensitivity and specificity was highest with 91 and 93% respectively.

Even if the thallium-scan gives the highest score, the low gamma-energy, the relatively long half life of 75 hours, and the price per examination is somewhat disadvantageous. Efforts are being made to find 99m-Tc-labelled compounds that accumulate in the myocardium as efficiently as does thallium. This search has apparently been successful in studies on primates, as it was reported by Deutsch et al.,[27] for di-methyl-phosphino-ethane labelled with 99m-Tc. This compound yields in the baboon high quality scans similar to 201-thallium.

3) MYOCARDIAL METABOLISM

In order to study metabolic reactions in vivo, it is necessary to be aware of the principal metabolic pathways one wants to observe. Moreover, attention must be paid to the proper tracer, the proper substrate and the placement of the tracer on to the substrate so that the reaction of interest may become observable. Also, one should, whenever it is required, use counting techniques that permit the distinction between the labelled substrate and its labelled catabolite. Needless to say, the final choice for clinical use depends on the diagnostic information obtained by the procedure.

Positron emission tomography has been elegantly used for analysing uptake and turnover of various 11-C-labelled metabolites, especially fatty acids which are the main energy source of the myocardium in the fasting state.[28,29] Yet positron emission tomographs are expensive and rare and should be close to a cyclotron that produces the short lived positron emitters. But planar, i.e. simple gamma-camera imaging or single photon emission tomography is also applicable to metabolic measurements and is therefore of wide interest because of availability of the conventional imaging equipment.[30]

Prior to explaining the single photon imaging with labelled fatty acids a short review of the principal pathways of fatty acids in the myocardium is in order, as illustrated in Figure 8.

Free fatty acids are transported from the circulating blood through the interstial space into the cell, obviously without need of an active carrier system. Inside the muscle cell, there is a pool of free fatty acids partially attached to myoglobin. The free fatty acids are eventually bound to coenzyme-A on cytoplasmic membranes, and the coupling is a prerequisite for all further biochemical steps. There is evidence that some of the activated fatty acids rapidly enter mitochondria by the carnitine shuttle. A large frac-

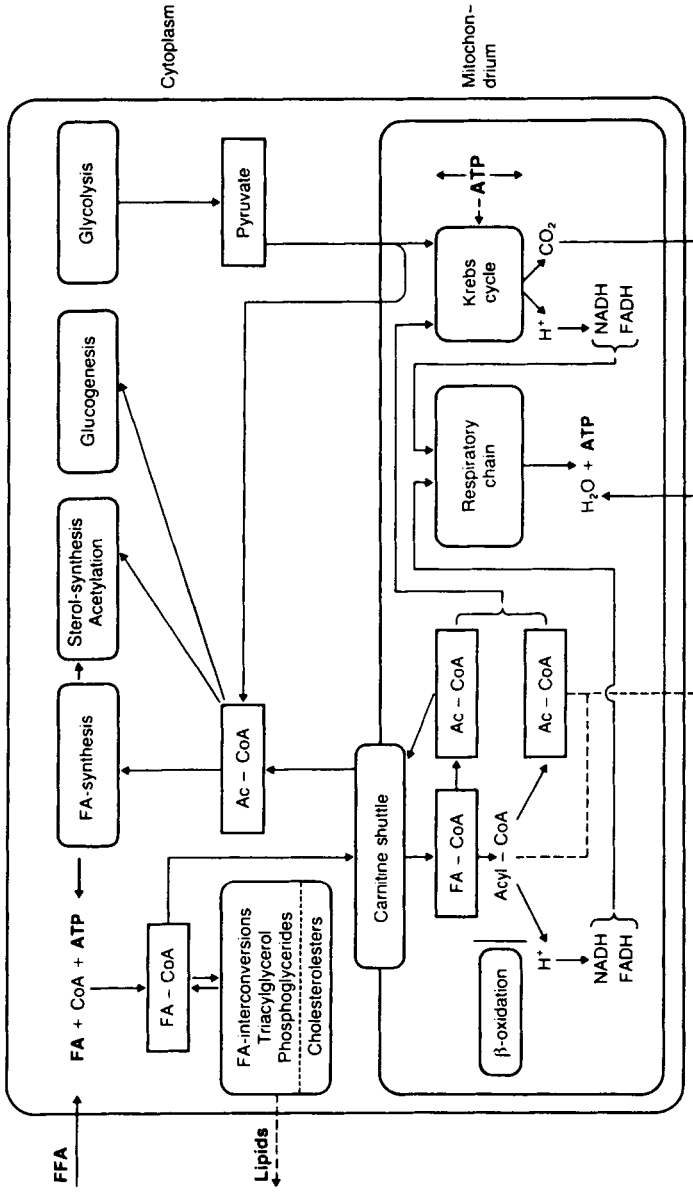


Fig. 8. Simplified scheme of fatty acid metabolism in the myocardium

tion of the fatty acid coenzyme-A is used for fatty acid interconversions and for the synthesis of triglycerides and phospholipids, also for cholesterol-esters. Mainly triglycerides and possibly phospholipids may serve as fatty acid reservoirs from which varying energy demands may be answered.

Having entered the mitochondria by the carnitine shuttle, fatty acids are metabolized by beta-oxidation yielding acetyl-coenzyme-A and protons. The latter are fed into the respiratory chain and the acetyl-coenzyme-A either returns to the cytosol or is fed into the Krebs cycle which produces CO_2 and protons which are again fed into the respiratory chain for oxidative phosphorylation. It is to be understood that acetyl-coenzyme-A in the cytosol is the central source for fatty acid de-novo synthesis, various acetylation reactions and sterol synthesis and also for glucogenesis. Thus, carbon fragments of fatty acids may be partially reutilized. Acetyl-coenzyme-A is also the result of catabolism of glucose and aminoacids and enters the Krebs cycle for generating protons for oxidative phosphorylation. When glucose supply is large, there is a reduced demand for fatty acid degradation.

Whereas fatty acid delivery to the cell largely depends on coronary flow and perfusion pressure, intracellular fatty acid degradation generally is governed by the energy demand and has been related to ventricular work load, oxygen tension, pulse rate and action of hormones. Metabolites, for example glucose, may partially substitute for fatty acids as major source of energy. Answering energy demands from any source depends on the integrity of the intracellular net work of the metabolic reaction chains, especially on the integrity of the mitochondrial apparatus.

It is clear that it is exceedingly difficult to measure by in vivo nuclear medical techniques a specific reaction in the fatty acid degradative system and relate this to a specific disorder. What has thus far been shown possible is the distinction between the accumulation of fatty acid inside the cell and the rate of release of activity. The latter will be shown below to signal fatty acid degradation.

Thus far, the fatty acids shown in Figure 9 have been employed for myocardial scintigraphy. 11-C-labelled palmitic acid obviously is a physiological substrate and has been successfully used with positron emission tomography for measuring fatty acid metabolism.[28]

123-I-heptadecanoic acid is an analogue of stearic acid; the final methyl-group is replaced by iodine.[31] Iodine labelled long chain fatty acids were first used by Evans[32] and by Poe[33] and shown to accumulate in the myocardium. 123-I-heptadecanoic acid was shown to behave kinetically similar to palmitic acid and was employed for measuring fatty acid utilization and turnover in the human myocardium.[34]

1 – ^{11}C	– PALMITIC ACID
17 – ^{123}I	– HEPTADECANOIC ACID
p – ^{123}I	– PHENYLPENTADECANOIC ACID
22 – ^{123}I	– ERUCIC ACID
17 – ^{131}I	– IODO-9-TELLURA-HEPTADECANOIC ACID
9 – [^{123m}Te]	– TELLURA-HEPTADECANOIC ACID
3 – [^{11}C]	– METHYL-HEPTADECANOIC ACID

Fig. 9. Some fatty acids presently employed in order to measure myocardial metabolism.

123-I-phenyl-pentadecanoic acid is artifical.[35,36,37] It does not follow the pathway of natural fatty acid but tends to become trapped mainly in the triglyceride pool with a turnover in the cell slower than palmitic acid. A similar behavior was shown for the long chain erucic acid.[38]

Other fatty acids have been especially synthesized to prevent beta-oxidation, thus permitting the observation of substrate accumulation in the cell. There is the long chain fatty acid which carries tellurium in the carbon chain and may be labelled by radioiodine or by a suitable tellurium radioisotope.[39] Another fatty acid in this category is the 3-methyl-heptadecanoic acid that was shown to be metabolically trapped in beta-oxidation.[40]

Thus there are 3 groups of labelled fatty acids: 1) natural fatty acids and their metabolic analogues, 2) the "artificial" fatty acids that differ from natural fatty acids in that they get specifically and temporarily trapped in pools, 3) fatty acids that are engineered in order to accumulate in mitochondria by virtue of being trapped in beta-oxidation.

For the development of a diagnostically useful assay of myocardial metabolism by planar imaging mainly 123-I-heptadecanoic acid was chosen. It is also easily applied to single photon tomography.

Single Photon Imaging of Fatty Acid Metabolism

High quality images and turnover measurements with 123-I-heptadecanoic acid required the development of a correction procedure that eliminates signal contribution from fatty acids in circulation or from catabolites such as free 123-iodine.[34] Frequent imaging over the first 40 minutes after tracer injection and using this correction technique thus creates time activity curves showing tracer accumulation and wash out from the myocardium.

The half times of the first component of tracer release from the total left ventricular myocardium and for the individual myocardial segments varied around 24 ± 5 minutes at rest, and were somewhat yet statistically not significantly shorter at exercise.[34]

Examination of patients with coronary artery disease led to the conclusion: 1) heptadecanoic acid accumulation defects correlate with coronary artery stenosis or occlusion; 2) heptadecanoic acid elimination rate for the entire myocardium may be normal; 3) heptadecanoic acid uptake and elimination is normal and areas with normal perfusion; regions with accumulation defects have diminished, normal or at times even increased elimination rates.[41]

As shown by Dudczak et al.,[42] the first and second components of the elimination rate of heptadecanoic acid in normal individuals as in patients with coronary artery disease promptly respond to an infusion of insulin and glucose, in fact indistinguishable from the response reported for 11-C-palmitic acid.[29] This further supports the elimination rates to be an expression of myocardial metabolism.

Both measurements, that of accumulation and of turnover, were applied to investigate the effect of rehabilitation training in 8 patients after myocardial infarction. The elimination half times for various myocardial regions after 1 year of rehabilitation training following myocardial infarction improved in 5 of the 8 patients, and 3 patients showed no change or even a slight deterioration.[41]

Of special interest is the application of 123-I-heptadecanoic acid for noninvasively recognizing cardiomyopathies. The accumulation images of patients with early or advanced congestive cardiomyopathy show gross, spotty heterogeneous distribution of the tracer in the left ventricular wall, similar to images at times obtained with 201-thallium and often not clearly distinguishable from those of patients with coronary artery disease (Figure 10). We have assayed the relationship between accumulation and turnover of the tracer in groups of patients with advanced and early congestive cardiomyopathy.[43,44] In contrast to results in coronary artery diseases, the quality of regional tracer accumulation does not correlate with the elimination half time in that region. Thus, accumulation and turnover of fatty acid in the various myocardial regions were unrelated and discordant. Discordance between distribution of accumulation defects and of altered elimination rates was also seen in patients with early stage of congestive cardiomyopathy as illustrated in Table 1. This data lets one speculate whether a systematic analysis of fatty acid accumulation and elimination may considerably aid the diagnosis of cardiomyopathy, in distinction from coronary artery disease.

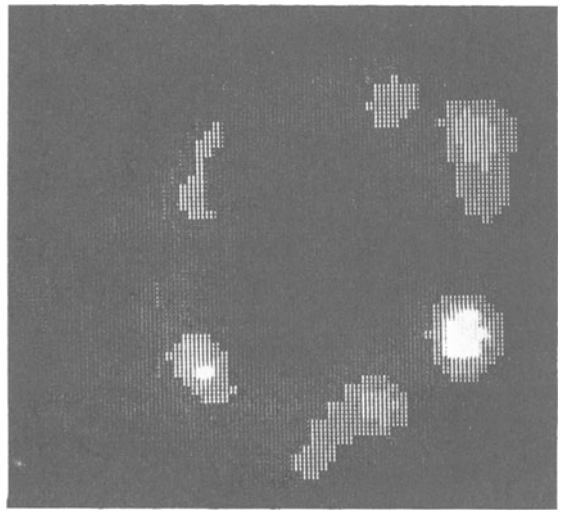


Fig. 10. Myocardial scintigram following the injection of 123-I-heptadecanoic acid in a patient with early stage of congestive cardiomyopathy.

Positron Emission Tomography

Metabolic measurements are the primary domain of positron emission tomography. This technique is presently limited to few institutions only.

A variety of 11-C-labelled substrates has thus far been employed for measuring myocardial metabolism; just to list a few:

(a) 11-C-palmitic acid[28,29,31] served to measure myocardial fatty acid metabolism and 18-F-deoxyglucose[45,46] has been used for measuring local glucose utilization and was shown to increase in areas of myocardial ischemia, where fatty acid utilization was depressed.

(b) 11-c-acetate,[47] 13-N-glutamat[48] and 13-N-ammonia[49] were tried and data correlated with cardiac disease.
Local perfusion may be measured with 15-O labelled CO_2.[50]

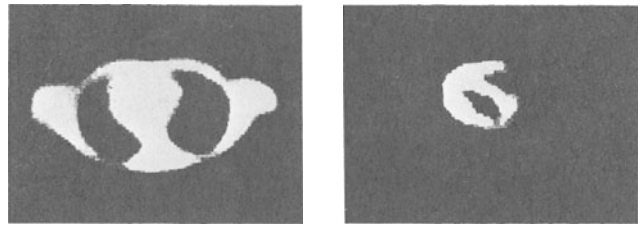
Table 1. Regional Myocardial Elimination Half Times (Min) of 123-I-Heptadecanoic Acid in Patients with Congestive Cardiomyopathy in Early Stage

PAT.	TOTAL HEART t 1/2	SEPTUM t 1/2 Acc	INFERIOR t 1/2 Acc	POSTERO-LATERAL t 1/2 Acc
M.W.	13.0	11.0 + 11.0 ++	9.4 –	12.8 + 12.4 +
K.H.	21.0	16.8 ++ 15.4 ++	16.5 +	31.4 + 19.6 ++
K.R.	46.5	47.6 + 35.0 +	38.8 ++	48.5 ++ 35.6 ++
B.M.	20.5	20.8 + 16.3 +	17.7 +	21.1 + 18.3 ++
F.B.	57.8	167.7 – 35.4 +	32.8 +	49.0 + 63.2 ++
R.A.	41.5	41.0 – 32.0 +	33.4 +	43.4 – 23.5 ++
D.R.	80.7	53.0 – 74.0 –	39.0 ++	344.0 – 277.0 ++
D.H.	30.4	28.8 + 26.6 ++	32.0 +	31.2 + 35.6 ++
R.A.	34.7	33.5 + 34.8 +	21.8 –	41.2 + 30.9 ++
M.G.	41.5	40.9 – 32.0 +	32.5 +	43.4 + 23.5 +

++ = Acc. normal, + = Acc. diminished, – = Acc. bad
a = basal part, b = distal part

I would like here to draw attention to the potential use of 75-Br amongst the newer positron emitting radionuclides.[51] This isotope is certainly easily handled chemically, and has the advantage of a long enough half life of 97 minutes for permitting transport from the cyclotron to the site of medical application. Figure 11 shows a dog heart on the left in a transmission image, and on the right is the ECG gated diastolic emission tomographic image of the myocardium taken at the level on the AV valves, after intravenous injection of 75-Br labelled phenyl-pentadecanoic acid. This approach appears practical and eventually useful for clinical research.

A potentially useful compound is 11-C labelled methylglucose.[52,53] This glucose analogue is transported like glucose across the cellular membrane, but it is not metabolised. It is transported back from the primary intracellular glucose pool into the circulation. After a single injection of the tracer into a suitable vein, activity



Transmission image Emission image

Fig. 11. ECG-gated diastolic image of the myocardium registered by positron emission transaxial tomography at the level of A-V valves, after intravenous injection of 15(p-75-Br-phenyl)-pentadecanoic acid in a normal dog. The transmission image is given for comparison on the left.

over tissue increases to a steady state between the concentration in tissue and that in blood. The 2 values finally give a fixed ratio for each tissue volume.

- By taking the blood concentration as internal standard, the transport of methylglucose into tissue to equilibrium may be selectively and quantitatively measured and relates to the rate of activity of the sugar transporting enzyme.[54] At steady state, tissue accumulation is readily seen when correction is made for the blood pool, as shown in Figure 12 giving the ECG gated diastolic image of the human myocardium registered by emission tomography at mid ventricular level, after i.v. injection of about 5 mCi 11-C-methylglucose.

- The dual analysis of blood and tissue concentration not only permits measuring glucose transport into the cell but also promises to yield local perfusion.[55] In normal myocardium local perfusion was 0.68 ml/min/g average; in the subendocardium local perfusion was 0.74 ml/min/g; and in the subepicardial region it was 0.67 ml/min/g.

- After transient ischemia in subendocardial areas the local glucose transport rate was significantly reduced whereas the local perfusion rate returned to normal values.

Conclusion

There has been an enormous advance over the past 10 years in using radioisotopes in cardiology because of improvement of counting and imaging devices, because of discovery of new useful radiopharmaceuticals, because of advancement of data processing techniques and of new concepts of evaluation of data. Most important is the inter-

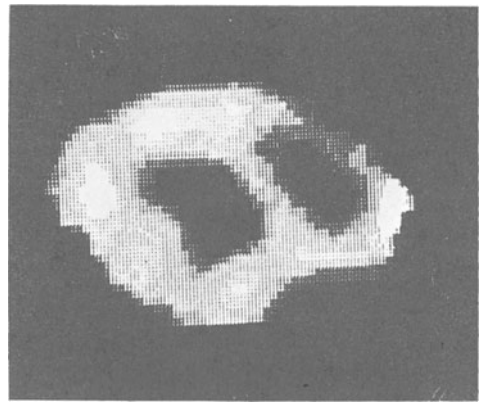


Fig. 12. ECG-gated diastolic image of a human myocardium obtained by positron emission transaxial tomography at mid ventricular level, after intravenous injection of 3-O-11-C-methyl-D-glucose in a normal individual.

disciplinary awareness amongst cardiologists, nuclear medical specialists, physicists, nuclear chemists, engineers and mathematicians. It is foreseeable that this broad approach continues to be rewarding particularly emphasizing the elucidation of defined metabolic reaction steps, i.e. biochemistry in vivo eventually with regard to many different enzyme reactions.

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CLINICAL TRIALS WITH ANTIAGGREGATING AGENTS IN THROMBOSIS

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Many agents capable of interfering with normal platelet function in vitro have been described in recent years; most are nonsteroidal anti-inflammatory agents. Not all these compounds have been shown to affect platelet aggregation and adhesiveness in experimental animals, but since animals do not develop spontaneous thrombosis, the relevance of such experiments to clinical medicine is rather tenuous. The only drugs suppressing platelet function that have been evaluated in clinical trials are aspirin, sulfinpyrazone, dipyridamole, hydroxychoroquine, clofibrate, flurbiprofen, and lidocaine, all of which were originally used for other therapeutic purposes. Even today one still has a poor understanding of the relationship between some of their effects on platelets and a potential antithrombotic effect. It is even uncertain whether the latter property is due to their impact on platelets or to some other unknown mode of action.

Aspirin does not inhibit the adherence of platelets to collagen and subendothelium but is a clear inhibitor of platelet aggregation and release induced by mild stimuli in vitro and in ex vivo experiments. Aspirin has however unequivocal effects in experimental venous, arterial and foreign shunt thrombosis in animals and was even shown to enhance thrombosis at high doses. Aspirin is unable to normalize shortened platelet survival in man.

Dipyridamole is a weak inhibitor of aggregation in vitro and ex vivo but was shown to prevent experimental thrombosis in most animal models. Moreover, it normalizes shortened platelet survival in animals and man.

Sulfinpyrazone is a poor inhibitor of platelet function in vivo and ex vivo although it has a protective effect against experimental thrombosis in animals. It also normalizes enhanced platelet survival in man. There is meagre evidence that sulfinpyrazone may be effective against the vessel wall.

PRIMARY PREVENTION OF MYOCARDIAL INFARCTION IN APPARENTLY HEALTHY AND IN AGED INDIVIDUALS

In a prospective American trial in over 2,000 hyperlipemic men (average age 42 years) free of coronary heart disease, treated with clofibrate (2 g daily), a reduction in the incidence of angina pectoris and non-fatal myocardial infarction but no statistically significant reduction in overall mortality was obtained (Krasno et al., 1972). However, major deficiencies in trial design and execution of the study prevent an unequivocal interpretation of the data.

The World Health Organization (WHO) sponsored in Edinburgh, Prague and Budapest a primary prevention, placebo-controlled trial with clofibrate on the incidence of ischemic heart disease in healthy men with a known cholesterol level (Report of the Committee of Principal Investigators, 1980). In total 15,000 healthy blood donors between 30 and 59 years were studied for an average of 5.3 years. The treatment group was given 1.6 g clofibrate per day. The overall lowering of non-fatal ischemic heart disease was 20% in the clofibrate group. However, the number of deaths and the crude mortality rates from all causes in the clofibrate group were significantly higher than those in the control group, the excess being particularly due to a group of noncardiovascular diseases including malignant neoplasms.

Evidence is also uncertain for aspirin. While the Boston Collaborative Drug Surveillance Group (1974) and Jick et al. (1976) had hinted a protective effect of aspirin in the prevention of myocardial infarction, subsequent follow-up studies, some of which involved very large numbers, showed no relationship between the regular use of aspirin and fatal or non-fatal myocardial infarction (Hammond et al., 1975; Hennekens et al., 1978).

Also in a prospective placebo-controlled trial carried out in a municipal old age home involving 430 elderly people (82% were females), there was no evidence of any prophylactic effect of aspirin (1 g/day) on the incidence of fatal or non-fatal myocardial infarction after 1 year follow-up observation (Heikinheimo, 1971).

A double-blind between-patient study was therefore carried out over 4 years in 291 institutionalized elderly males allocated at random to receive either sulfinpyrazone (0.6 g/day) or a placebo

(Blakely et al., 1975). At entry to the trial more than half of these elderly men had clinical signs of atherosclerosis (coronary, cerebral or peripheral arteries). No statistically significant reduction in mortality could be demonstrated in the sulfinpyrazone-treated patients.

The evidence available at present therefore does not allow the recommendation for use of clofibrate, aspirin or sulfinpyrazone for the primary prevention of myocardial infarction in apparently healthy men or in elderly individuals.

ANTIAGGREGATING AGENTS IN PREVENTION OF A FIRST MYOCARDIAL INFARCTION IN PATIENTS WITH ANGINA

In two Scottish trials 1.5 to 2 g clofibrate was given daily for 5 years to patients who had only angina when admitted to the trial (Research Committee of the Scottish Society of Physicians, 1971; Newcastle-upon-Tyne Physicians, 1971). There was a significant reduction in all cardiac deaths, sudden death and non-fatal death in the clofibrate-treated patients who had only angina when admitted to the trial.

While clofibrate looked promising in the prevention of a first myocardial infarction in patients with angina, this agent cannot - because of major side effects - be recommended for long term use (Committee of Principal Investigators, 1978, 1980).

PREVENTION OF RECURRENT MYOCARDIAL INFARCTION

The two Scottish trials discussed above and the Coronary Drug Project Research Group (1975) failed to show a significantly beneficial effect of clofibrate (1.8 g daily for 5 years) on cause-specific mortality or total mortality in patients who survived a previous myocardial infarct.

The first major double-blind trial of aspirin for the prevention of death after recovery from myocardial infarction was reported by Elwood et al. (1974). On the 1,239 males studied within 6 months after discharge from hospital, half were given a low dose of aspirin (0.3 g daily for 24 months) and half were given a placebo. After 12 months, the follow-up revealed that the treated group had a cumulative mortality of 12.2% and the control group of 18.5%; at no point during the study did this 24% reduction in mortality reach a level of statistical significance. One defect of this trial was that the 9% men who withdrew from treatment were not followed to ascertain death or survival.

Similar findings emerged from a second double-blind trial, also set up by Elwood et al. (1979) to evaluate the prophylactic effect of 300 g aspirin 3 times daily, started within 2 weeks after infarction. After 1 year the all-cause mortality was 12.3% in the aspirin-treated group and 14.8% in the control group, a not statistically significant difference of 17%. The major weakness of this trial is the high withdrawal rate from treatment (26%).

In the study of the Aspirin Myocardial Infarction Research Study Group (1980) 4,524 patients were randomly allocated to either aspirin (1 g/day) or placebo for three years. Endpoints determined before the beginning of the trial included total mortality, coronary incidence (that is the combination of coronary deaths plus proved non-fatal myocardial infarction) and fatal or non-fatal strokes. Entry was eight weeks to five years after myocardial infarction and analysis was by life tables and final outcome. As regards the three endpoints total mortality, coronary deaths combined with non-fatal myocardial infarction and fatal or non-fatal strokes, the results for aspirin and placebo were almost identical.

In a German-Austrian multicenter trial 946 survivors of a myocardial infarction were randomly allocated to aspirin, placebo or phenprocoumon and followed during 2 years (Breddin et al., 1980). There was little difference in total mortality among the three groups, but sudden death and non-fatal recurrent myocardial infarction combined were significantly lower in the aspirin group than in the placebo or phenprocoumon group.

In the "PARIS" Study (Persantine Aspirin Reinfarction Study Research Group, 1980) 2,026 patients were randomly allocated to three treatment groups; one group received 75 mg dipyridamole and 324 mg aspirin three times a day. The second group received the same dose of aspirin with a persantin placebo, and the third group (half the size of the two previous groups) placebo only. After 3 years of treatment, there was a trend in favor of aspirin and dipyridamole, and of aspirin as compared with placebo, but statistical significance was not achieved with regard to total mortality (respectively 10.7%, 10.5% and 12.8%), coronary deaths and coronary incidence. Life table analysis however showed that dipyridamole and aspirin were significantly better than placebo at 4, 8, 12, 16, 20 and 24 months, and aspirin was significantly better than placebo at 8 and 24 months. Subgroup analysis showed that the effects on the three primary endpoints were much more marked in patients who entered the study within six months of myocardial infarction.

Two major trials of sulfinpyrazone have been reported - the "ART" Study (carried out by the Anturane Reinfarction Trial Research Group (1978, 1980)) and the Anturane Reinfarction Italian Trial (1982). In the ART trial 1,629 patients who had suffered a myocardial infarction, were allocated at random to sulfinpyrazone

200 mg four times daily, or to placebo, for 12 to 24 months. Entry to the trial was 25 to 35 days after myocardial infarction. 43 deaths were regarded as "non-analyzible" according to criteria defined in the protocol. Deaths from all causes and cardiac deaths were reduced at up to 24 months, although conventional levels of significance were not achieved. Although deaths from myocardial infarction were almost identical in the treatment and placebo groups, there was a marked and significant reduction in sudden deaths (43%) which took place almost entirely during the first six months of treatment. An independent committee re-analyzed all deaths and when the principles of a so-called clinical efficacy trial are applied, their results are in general agreement with the original report, although the levels of statistical significance are less striking (The Anturane Reinfarction Trial Policy Committee, 1982). If the data from the "ART" study are analyzed on an "intention-to-treat" basis, including all patients and all deaths in the 24 months follow-up, there were 74 deaths in the 813 sulfinpyrazone-treated patients and 89 deaths in 816 placebo patients ($p > 0.05$) (McNicol, 1980).

The results of a similar trial with the same daily dose of sulfinpyrazone were recently published (Anturane Reinfarction Italian Trial, 1982). There are some important differences between the American and Italian sulfinpyrazone trials, eg., all patients who had a myocardial infarction, a stroke or a transient ischemic attack, withdrew from the Italian trial. The death rate during the first 6 months in Italian placebo-patients (cardiac death rate 4.4% and sudden death rate 2.5%) was much lower than in the American placebo-patients (cardiac death rate 10.3% and sudden death rate 7.0%), which may be due to less patients with recurrent infarction and heart failure in the Italian trial. Therefore it is not surprising that in contrast to the American trial, the Italian study did not demonstrate a reduction in either total mortality or sudden death rate. However, it did reduce the incidence of reinfarction during the entire 24 months observation period (75 in the sulfinpyrazone group, 34 in the placebo group).

PREVENTION OF GRAFT OCCLUSION AFTER AORTOCORONARY BYPASS SURGERY

Early occlusion of a coronary bypass is most often related to the level of blood flow through the bypass which depends mainly upon the size of the anastomosis and distal run-off. Late occlusion is more related to the progression of the basic disease in which platelet adherence to the graft wall could be an important factor.

A preliminary nonrandomized trial suggested that all groups of patients who received dipyridamole, aspirin or oral coumarin anti-coagulant drugs experienced higher survival rates over a 4 year observation period than control groups who did not receive

one of these drugs (Hall et al., 1974). These findings were not confirmed in a strictly randomized trial in which the protective effect of aspirin (1 g daily), dipyridamole (0.225 g daily) and oral anti-coagulants were compared to oral anti-coagulants. Six months after coronary bypass surgery, the occlusion rate was similar in the 4 treatment groups (Oblath et al., 1978). The treatment however was started 3 days after surgery which may be too late to prevent myo-intimal proliferation (Goldberg et al., 1979).

PREVENTION OF THROMBOEMBOLISM IN PATIENTS WITH PROSTHETIC HEART VALVES

Newer materials employed in modern prosthetic valves are less thrombogenic than the first artificial valves. Also the location of the artificial valve is important, the mitral location being associated with the highest thromboembolic risk. Oral anti-coagulants provide a satisfactory protection provided the level of anti-coagulation is effective, which requires frequent and accurate monitoring of the prothrombin time. The risk of bleeding complications still prevails and the potentially dysmorphogenic properties of oral anti-coagulants in the first trimester of pregnancy constitutes a problem in young women with cardiac valve replacement who wish to become pregnant. These drawbacks explain the interest in the possible use of antiaggregating agents, combined or not with oral anti-coagulants in patients with synthetic heart valves.

The first double-blind trial demonstrated that dipyridamole (0.4 g daily) given in addition to oral anti-coagulants resulted in a greater protection (1.3% arterial emboli) than oral anti-coagulants used alone (14.3% arterial emboli) (Sullivan et al., 1971). Several other trials confirmed these findings although they all had a less than satisfactory design (Arrants et al., 1970; Meyer et al., 1971; Pell, 1975; Groupe de Recherche Pacte, 1978).

Two trials in which aspirin (0.5 g or 1 g daily) was given in addition to oral anti-coagulants also had fewer thromboembolic complications than patients receiving anti-coagulants alone (Altman et al., 1976; Dale et al., 1977). Dipyridamole or aspirin prevention alone does not result in a satisfactory protection (Isom et al., 1973; Dale et al., 1977; Sutton et al., 1978). While a combination of oral anti-coagulants and either dipyridamole or aspirin results in a greater protection, one should not disregard the incidence of bleeding induced by aspirin, which may be 7 to 15% (Altman et al., 1976; Dale et al., 1977).

ANTIAGGREGATING AGENTS IN PATIENTS WITH OBLITERATIVE DISEASE IN LIMB ARTERIES

The most common cause of obliterative arterial disease in the legs is slowly progressive atherosclerosis which is eventually superimposed by thrombosis. The danger of the condition is the underlying disease, as most patients die from a cardiac or cerebral complication of atherosclerosis. The increased mortality in claudicating patients corresponds approximately to the mortality rate one would expect in an overall population aged 10 years older. The message is therefore very clear. A patient with claudication is to be treated first for the underlying illness and medical risk factors, and not for the presenting symptoms. Abstinence from smoking, appropriate diet and physical exercise are therefore the first treatment guidelines while percutaneous transluminal dilatation and reconstructive surgery often relieve the most severe cases.

There is a widespread interest in the use of antiaggregating agents in patients with atherosclerotic limb arteries as thrombosis of a stenotic lesion is a rather frequent event. Damaged endothelium and other atherosclerotic lesions may activate platelets which in turn accelerate the atherogenic obliterative process. Inhibition of platelet deposition, aggregation and release would then in the long run be beneficial in these patients.

There are very few well controlled trials demonstrating that antiaggregating agents are clinically effective in patients with intermittent claudication. A recent double-blind trial suggests that ticlopidine (0.5 g daily) decreases the extent of cutaneous lesions (Katsumara et al., 1980). This study has so far not been confirmed.

Aspirin (1.5 g daily) has been used after arterial thromboendarterectomy and found to increase the patency rate after 1 year from 11.2% in the control group to 22% in the aspirin-treated patients (Ehresmann et al., 1977). Similar findings were obtained after a 2 year follow-up with either aspirin alone (1 g daily) or in combination with dipyridamole (0.225 g daily) (Bollinger et al., 1978, 1981). This beneficial effect of aspirin was obtained in patients subjected to endarterectomy, not after venous bypass operation. It should be noted that arterial endarterectomy is at present an almost abandoned operation.

None of the published trials with sulfinpyrazone after peripheral arterial surgery resulted in a beneficial effect (Blakely et al., 1977; Rodvian et al., 1978).

Several reports have appeared on the beneficial effects of aspirin (0.3 to 1 g daily) in patients with thrombocytosis and intermittent arterial ischemia. The painful "blue toe" syndrome is

the consequence of a blocked microcirculation due to platelet emboli or spontaneously aggregating platelets. Aspirin immediately relieves the pain; treatment of the underlying disorder will prevent further ischemic attacks in toes or fingers (Vreeken et al., 1971; Bierme et al., 1972; Preston et al., 1974). Similar symptoms can also result from platelet emboli originating on ulcerated atherosclerotic plaques.

PREVENTION OF TRANSIENT CEREBRAL ISCHEMIA

The most important warning symptoms of impending stroke are Transient Ischemic Attacks (TIA's), which are focal cerebral dysfunctions, rapid in onset, variable in duration, lasting 2 to 15 minutes but no longer than 24 hours. Each attack leaves no persistent neurological deficit. In general, TIA's have a bad prognosis; 25 to 40% of patients with TIA's will eventually develop cerebral infarction within 5 years. The risk of stroke in the first year after TIA may be over 15%.

The origin of TIA's and stroke is multicausal and deals with a variety of heart diseases, blood vessel disorders, hypertension and blood disorders. Considering the multicausal origin of TIA's and stroke and the uncertainty of the role of platelets in many of them, it may be questioned whether administration of an antiaggregating agent may ever result in therapeutic large scale benefit in such a mixed population. The effectiveness of antiaggregating agents would certainly increase, could one predict which patients have an increased risk for fibrin-platelet emboli as the specific cause of their TIA.

A controlled clinical trial in America compared aspirin (1.3 g daily) in patients with a history of TIA's. The endpoints were the recurrence of TIA's, stroke and death. Aspirin did not reduce significantly these endpoints at 24 months of follow-up. Subgroup-analysis revealed that patients who had multiple TIA's before entering the trial and those with angiographically demonstrated lesions of the carotid artery had after 24 months a significant reduction of the absolute endpoints death and stroke combined (Fields et al., 1977, 1978).

The Canadian Cooperative Trial (1978) concerns patients who had experienced at least 1 TIA in the 3 months before entry in the trial. They were randomized for an average of 26 months to aspirin (1.2 g daily), aspirin (1.2 g) plus sulfinpyrazone (800 mg), sulfinpyrazone alone (800 mg) or placebo. Aspirin reduced the risk of continuing TIA's, stroke or death by 19% ($p < 0.05$) and also the risk for the harder, more important events of stroke or death by 31% ($p < 0.005$). For sulfinpyrazone, no risk reduction of TIA was observed and only a 10% ($p > 0.05$) reduction of stroke or death.

No overall synergism was observed between the 2 drugs. The aspirin effect was not observed in women and was less marked in patients with diabetes, hypertension or previous myocardial infarction.

In a more recent Swedish trial the combined effect of aspirin (1 g daily) and dipyridamole (0.15 g daily) appeared during the first year of follow-up to be in TIA patients as effective as oral anti-coagulants against stroke (Olsson et al., 1980).

One can therefore conclude that aspirin reduces significantly in the first 2 years the recurrence of TIA's, stroke or death in male patients with a recent transient ischemic attack. Many neurologists consider that patients with TIA's in whom arteriographic examination reveals a surgically accessible lesion, should be operated first and subsequently take aspirin. Considering the prominent role of hemodynamic processes which apparently are considerably more frequent than solely occlusive factors in the pathogenesis of TIA's in a general population (Herman et al., 1981), a complete clinical and technical examination of the patients is mandatory before any treatment is decided.

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ECHOCARDIOGRAPHY: PRESENT STATE OF THE ART

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Among the new diagnostic methods in cardiology, echocardiography is certainly the most informative. The field has grown so rapidly in recent years that it is almost impossible to keep up with the new technical developments and their application in patient care. The principle of the method is based upon the detection of echoes produced by a beam of short ultrasound pulses transmitted into the heart. Ultrasound is harmless at the energy levels used. The examination can thus readily and repeatedly be used without untoward effects to the patient making it the ideal method for serial analysis and follow-up studies.

It is important to appreciate some distinct differences between echocardiography and X-ray imaging for a better understanding of its specific applications. With ultrasound local changes in acoustic impedance along the sound beam pathway are registered. X-ray techniques register cumulated attenuation of energy along the pathway so that cardiac structures are superimposed in depth and seen as shadows. As a result, the specific details of intracardiac anatomy and pathology such as the attachment and morphology of the atrioventricular valves, the interventricular septum, mass lesions, etc. are better documented with ultrasound that with X-ray techniques.

Since its introduction in 1954 by Edler and Hertz[1] to the mid 1970's, M-mode echocardiography has been exclusively used and its clinical value and limitations are well established.[2,3]

The ultrasound beam is aimed manually at selected cardiac structures and a "diagram" showing how the position of these structures change during the cardiac cycle is obtained (time-motion

display of B-mode or intensity-modulated echoes). The high sampling rate (1,000 transmit-receive cycles/sec) permits recording of rapidly occurring events (e.g. valve opening, closure and fluttering) and facilitates measurement of cardiac dimensions and the analysis of time relationships with other physiological parameters (e.g. simultaneously recorded pulse and pressure tracings). The method however, does not provide information on the spatial relationships of different cardiac structures to each other. This can be accomplished by rapidly and automatically moving the ultrasound beam through a section of the heart to create a "tomographic image" yielding instantaneous structure information and thus cardiac anatomy in motion (Fig. 1).

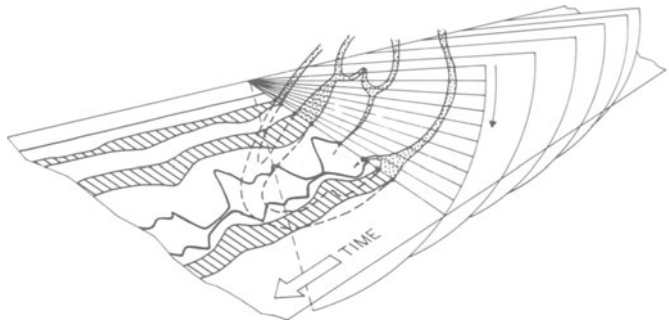

Fig. 1. Diagram illustrating the relationship between the two-dimensional and the M-mode echocardiogram. The motion pattern of the small part of the mitral valve hit by the sound beam is accurately tracted (1,000 transmit-receive cycles/sec). However, no information on its anatomical relationships is obtained. This is available from the two dimensional images.

The spatially oriented display of two-dimensional echocardiography allows information to be appreciated and utilized which is meaningless in the absence of such a spatial reference. This allows a multitude of cardiac cross-sections to be imaged from several chest wall transducer positions (parasternal, apical, subcostal and suprasternal) providing a wealth of diagnostic information.[4] Recently, the American Society of Echocardiography (ASE) has published recommendations for nomenclature and image orientation standards.[5] Such standards obviously are needed to make studies from different laboratories comparable. Building up a two-dimensional image requires time limiting the frame rate which is

25 or 30 frames/sec. As a consequence, two-dimensional echocardiography is less suitable for analysis of functional abnormalities. The relative advantages of M-mode and two dimensional echocardiography are listed in Table 1.

Table 1. Relative Advantages of M-Mode and Two-Dimensional Echocardiography

M-mode echocardiography
- Excellent time resolution
- Accurate dimensional measurements
- Timing of events against other parameters
- Easy storage and retrieval of data
Two-dimensional echocardiography
- Anatomical relationships
- Shape information
- Multiple tomographic views
- Complex motion patterns
- Easier to understand

It is obvious that both methods are complementary and best used in combination for a comprehensive analysis of cardiac conditions where anatomic and functional abnormalities overlap. Because the "ultrasonic windows" to the heart are limited by the ribs and lungs, all presently used two-dimensional imaging systems are of the sector type, either mechanical or electronic. Comparative advantages and limitations of each tupe of instrument are continually changing and their sophistication is increasing which makes differences between them less pronounced.[6]

Mechanical scanners use an oscillating or a rotating scan head. The principle of image formation in the oscillating systems relies on the rapid pivoting motion about a fixed axis of a single transducer by means of a magnetic deflection mechanism. In the rotating systems, several transducers are mounted on a spinning wheel inside a fluid-filled scanning head and are active when they pass over the heart. Signals from the scanning heads are used to steer the oscilloscope beam in the same manner as the ultrasound beam to create a tomographic image. Advantages of the mechanical scanners are their relative simplicity, the possibility to adapt them to existing M-mode units, and the retention of the straightforward resolution characteristics of a single crystal.

Phased-array scanners make use of a small stationary transducer with multiple small elements. They are all utilized in

producing each of the individual ultrasound beams which comprise the sector image. The ultrasound beam is steered electronically through the scan plane by firing them in an extremely rapid and precisely controlled sequence. These systems are very complicated and, hence, the most costly. Despite the necessary complexity, they offer the possibility for reducing the effective beam width by dynamic focusing, a feature not possible with mechanical scanners. Their major advantage is that they can simultaneously display the two-dimensional image while repeatedly sampling areas of the selected scan plane for M-mode recording. An alectronic cursor superimposed on the display is adjusted to the desired position and the appropriate B-mode lines are printed on a M-mode strip-chart recorder.

In order to document two-dimensional images we presently make use of digital scan converters which allow to change the image into a standard TV format. They comprise a matrix of memory cells (typically 512 x 512 elements, each called a "pixel"). When fully loaded, the memory matrix can be "read" in any desired sequence, e.g. as a series of horizontal lines to form a TV image for a video recorder, or as a series of vertical lines to enable it to be printed along with the M-mode on a strip-chart recorder. Digital stored images offer additional advantages since they can be manipulated in a number of ways. Alpha-numeric data (e.g. patient identification) can easily be added, various techniques can be employed to enhance the image quality, and measurements can be made directly from the displayed image using a light-pen or a joystick-controlled cursor and the results encoded in the image.

These capabilities allow accurate and reproducible measurements of the mitral valve orifice area from parasternal short axis views in patients with mitral valve stenosis.[7,8] They further enhance and simplify segmental wall motion analysis in patients with coronary artery disease.[9] Most of their attractiveness, however, lies in the measurement of left ventricular volume and quantitation of left ventricular function. The extreme accuracy reported in vitro and in animal studies proves that the models and formulas applied are satisfactory and suggest that quantitation is an obtainable goal.[10,11] The reported results in patients, however, are less convincing.[12,15] It seems that accurate measurement of left ventricular volumes in humans using two-dimensional echocardiography must await further instrument improvements and better display techniques.

Pulsed Doppler echocardiography is gaining increasing interest and currently undergoing intensive evaluation. The method uses the frequency shift of an ultrasound beam backscattered from the red blood cells and allows to detect the nature, direction and velocity of blood flow at one point (sample volume) within the heart and great vessels. Doppler echocardiography provides data

on the integrity of valve function and detection of septal defects but this information is qualitative at its best.[16] Doppler evaluation should nevertheless be considered as part of a comprehensive cardiac evaluation and is optimally used in conjunction with M-mode or preferably two-dimensional echocardiography. Some newer units offer these integrated capabilities, all information being obtained from one and the same transducer (Fig. 2). Such a combination with multigate, digital Doppler technique promises to obtain two-dimensional images similar to cineangiocardiograms, in which valvular regurgitation or shunting blood flow is translated directly into an anatomic form of display.

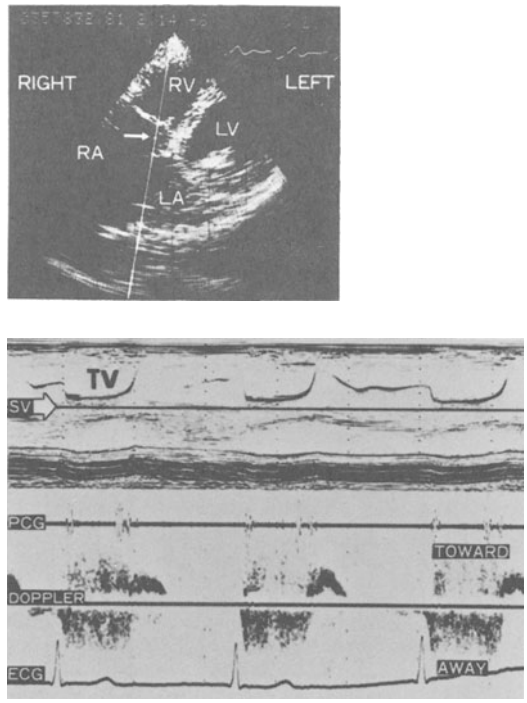

Fig. 2. Phased array sector image of a patient with tricuspid regurgitation. The imaging plane is a foreshortened four chamber view. The cursor indicates the B-mode line from which the M-mode echocardiogram is obtained and passes through the tricuspid valve. The position on the B-mode line of the range gate from which the Doppler is sampled is indicated by the arrow. The M-mode echocardiogram is shown below and the position of the sample

volume (SV) is here indicated by the straight line on the tracing. The Doppler shift indicating flow velocity and its direction and the M-mode cardiogram are recorded simultaneously. Laminar flow is represented by a narrow band of dots and turbulence by a broad band of dots. By convention, deflection of dots above the baseline indicates an increase in flow velocity toward the transducer and flow away from the transducer is indicated by the dots below the baseline. Note the systolic flow away from the transducer indicating tricuspid regurgitation. ECG: electrocardiogram, PCG: phonocardiogram, TV: tricuspid valve.

With presently available single sample volume instruments, accurate quantitation of volume flow is not possible and this possibility should probably be awaited with cautious optimism.

Contrast echocardiography is the technique of injecting an echoproducing biologically compatible solution into the bloodstream and using M-mode and two-dimensional techniques, observing the bloodflow patterns as revealed by the resulting cloud of echoes. The method is increasingly used and makes it possible to derive information that was heretofore available only from cardiac catheterization and angiocardiography. The source of ultrasound contrast is microbubbles of air present in the injectate.[17] Dextrose 5% in water, saline, and indocyanine green dye are the most commonly used contrast agents. Right sided structures are delineated by peripheral venous injections, and left sided structures are opacified by injections performed directly into the left heart chambers during cardiac catheterization (Fig. 3).[18] The clinical applications of contrast echocardiography are indicated in Table 2. Since echocardiographic contrast is entirely removed from the circulation by the pulmonary capillary bed, the appearance of contrast in the left heart after a peripheral venous injection is diagnostic for a right-to-left shunt. Shunts as small as 5% can be detected.[19] The method thus provides a sensitive means to diagnose uncomplicated atrial septal defects as a small right-to-left shunt is always present in this condition.

The technique is extremely useful in complex congenital heart disease helping to diagnose the various intracardiac connections and communications and to visualize the shunting blood flow (Fig. 4). The method is further helpful to diagnose tricuspid regurgitation.[20] Normally contrast injected in an upper extremity flows from the superior vena cava into the right atrium and right ventricle without retrograde flow into the inferior vena cava. In patients with tricuspid regurgitation, however, it can be detected in the inferior vena cava and hepatic veins during the "v" wave on the right atrial pressure tracing. Timing of its appearance is much easier from M-mode than from two-dimensional studies. Kerber et al. were using the technique during cardiac catheterization for the identification

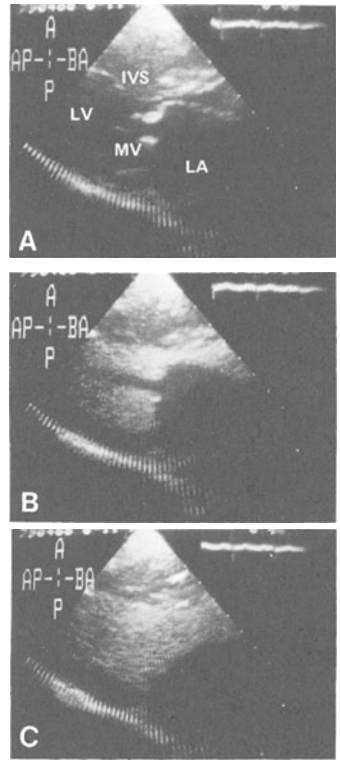

Fig. 3. Stop-frame photographs of parasternal long axis views of a patient with mitral valve stenosis before (panel A) and after injection of echo contrast via a catheter in the left ventricle. Panel B shows a frame recorded during diastole. The negative shadow caused by the noncontrast blood flowing from the left atrium into the ventricles visualizes the transmitral blood flow pattern. During systole (panel C), the echo contrast does not pass into the left atrium, excluding mitral incompetence. A: anterior; AP: apical; BA: basal; IVS: interventricular septum; LA: left atrium; LV: left ventricle; MV: mitral valve; P: posterior. (From Roelandt J et al: Contrast echocardiography of the left ventricle. In Rijsterborgh H (ed),"Echocardiology", M. Nijhoff, The Hague, 1981, p 219-232).

of valvular regurgitation and regurgitant volumes as small as 10% of the forward stroke volume can be demonstrated.[21] Recent studies have shown that injections in the pulmonary wedge position can cause contrast to appear on the left side of the heart and may enable us

Table 2. Clinical Applications of Contrast Echocardiography

- Structure identification (M, 2-D)
- Diagnosis (or exclusion) of shunts
Localization (2-D)
Direction shunting blood flow (M, 2-D)
Timing (M)
- Complex congenital heart disease (2-D)
- Valvular insufficiency
Venous injections: tricuspid and pulmonic (M)
Catheter injections: aortic and mitral (M, 2-D)
- Intracavitary and transvalvular flow patterns (2-D)
- Videodensitometric analysis of echo contrast
- Improved quantitation of left ventricle (2-D)
M, 2-D: indication for which points M-mode or two-dimensional echocardiography offer relative advantages.

to visualize left-to-right shunting.[22] There are several interesting additional future prospects for contrast echocardiography. Recent studies have suggested that quantitative videodensitometric techniques may allow to measure ejection fraction and cardiac output,[23] and to quantify intracardiac shunts[24] from two- dimensional contrast echocardiograms.

Exercise echocardiography is a recent experimental technique. Its important limitations at present are that successful exercise studies are technically difficult to perform and that there is a large proportion of patients in whom adequate study for analysis are unobtainable. Nevertheless there are some important advantages to this method. It allows to study cardiac physiology during exercise[25] and to detect left ventricular dysfunction in selected patients with coronary artery disease.[26] Because of the methodological difficulties, the method will probably not become a routine clinical screening method but may gain wider application in the investigation of normal and abnormal physiology in the future.

Summary

The combined use of two-dimensional and M-mode echocardiography complemented with contrast and Doppler techniques makes echocardiography an extremely accurate noninvasive method for the diagnosis of various cardiovascular diseases. It should be realized, however, that despite its apparent simplicity and safety, echocardiography is a difficult procedure to perform and that the interpretation of the results are subject to many pitfalls. It

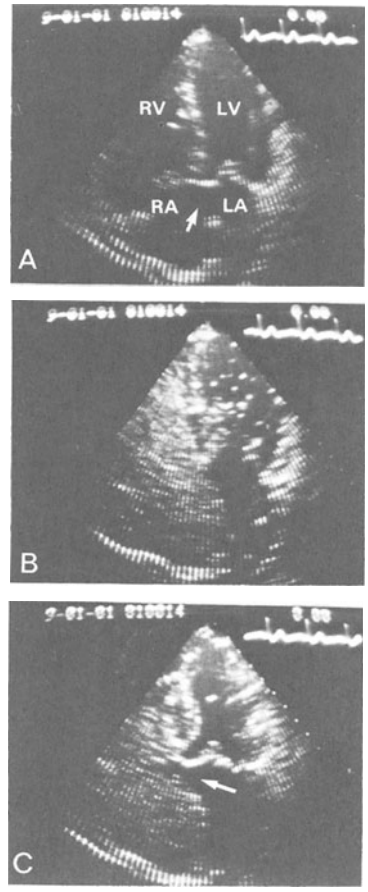

Fig. 4 Apical four chamber views obtained from a patient with an atrial septal defect of the primum type. The defect is indicated by the arrow on panel A. Panel B shows opacification of the right-sided heart after peripheral venous injection of 5 ml of dextrose 5% in water. Echo contrast is seen in the left ventricle indicating right-to-left shunting. The negative contrast effect proving the atrial septal defect is seen on panel C (arrow) when noncontrast blood flows from the left atrium into the right atrium. A bidirectional shunt is demonstrated. AP: apex; BA: basal; L: left; R: right; LA and RA: right and left atrium; LV and RV: left and right ventricle. (From Roelandt J et al: Contrast echocardiography: clinical applications. Verh Dtsch Ges Herz u Kreislaufforschg 47: 185-192, 1981)

appears that routine cardiac catheterization is presently being displaced from its dominant role for the preoperative assessment of patients with congenital and valvular heart disease.[27] Refinements of the technique will further increase its role in the diagnosis and management of cardiac patients.

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HIGH BLOOD CHOLESTEROL AND HIGH BLOOD PRESSURE: STATE OF KNOWLEDGE TODAY AND IMPLICATIONS FOR PREVENTION AND CONTROL OF EPIDEMIC ADULT CARDIOVASCULAR DISEASE

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INTRODUCTION

This presentation aims to review the key data sets on the role of elevated serum cholesterol and elevated blood pressure in the production of mass premature coronary heart disease (CHD). Its purpose is to set down the information needed by the health professions and the general public to act effectively in the growing effort to control the contemporary CHD epidemic in the industrialized countries and to prevent its emergence in the developing countries. To aid in accomplishing this objective, a most useful document is the just published report of the World Health Organization Expert Committee on the Prevention of Coronary Heart Disease[1]. This document summarizes current knowledge on these matters and on that basis presents an effective strategy for the health services and the general public for the effort to control this epidemic. (For an extensive review of the research literature in this field - i.e., the scientific basis of the preventive effort - see also the recently published Volume 2 of the Report of the Working Group on Arteriosclerosis of the U.S. National Heart, Lung, and Blood Institute[2].

THE STRATEGIC IMPORTANCE OF PRIMARY PREVENTION

First, a set of data dealing with the very important question of why a strategy emphasizing primary prevention - the prevention of the first CHD episode - is crucial for control of this disease (Figure 1)[3,4]. These are data from five major long-term prospective studies of populations in the United States, the Pooling Project study, involving 7,545 men originally aged 30 to 59, free at baseline of evidence of definite CHD and followed for almost 10 years.

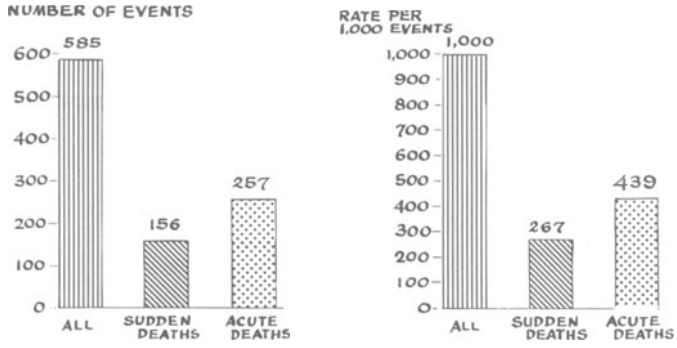

Fig. 1. U.S. national cooperative Pooling Project; sudden death and acute mortality with first major coronary events, men age 30-59 at entry, pooled data from five studies, experience over the first 10 years of follow-up; first major coronary events were definite non-fatal myocardial infarction, sudden (≤3 hours) coronary death, non-sudden coronary death (>3 hours-1 month)[3,4].

There were 585 first major coronary events during that time, of which more than a quarter (27%) were sudden deaths, and another 17% were non-sudden deaths with this first episode. (Sudden death was defined as observed death within 3 hours of onset of sickness in people previously well and without a history of definite CHD). Altogether, 44% of these first coronary episodes ended in death, in these men originally age 30 to 59, a typical experience with this disease all over the world[5]. About 70% of the deaths occur so rapidly that no medical care can be brought to bear and there is not time for hospitalization. For the 56% surviving the first attack, risk of dying in the next five years is increased about five times compared to similar men without a history of such an event, with most of the excess risk due to CHD, and with almost half the deaths being sudden death.

These data not only emphasize that this is a grim and difficult disease. More relevant practically, they also make the point that if we are to make major progress against a disease with this natural history, the fundamental strategic emphasis must be on primary prevention. Note that this is also fully in keeping with the lessons from past control of mass disease. The triumphs against infectious disease and against under-nutritional disease were achieved first and foremost by primary prevention, prevention of the disease from ever developing, based on approaches from infancy on, often long before people reached the stage of life when the mass occurrences of the diseases take place.

The second point I want to make on this matter is an elaboration of the remarks made by Finn Monahan on the roots of epidemic disease. Over a hundred years ago, one of the great founders of modern medicine - Rudolph Virchow - pointed out that when disease occurs at very high epidemic rates, this is due to, "... disturbances of human culture"[6]. In the case of CHD and the other atherosclerotic diseases afflicting both the middle-aged and the elderly in the industrialized countries at epidemic rates, the disturbances of human culture are mainly the "rich" diets, the cigarette smoking habit, the sedentary life-style all so widely prevalent in these countries in the second half of the 20th century, for the first time in human history. The consequence is that the major atherosclerotic disease risk factors, including high blood cholesterol and high blood pressure, are present at very high levels throughout the population, assuring the mass occurrence of these diseases.

SERUM CHOLESTEROL

Extensive data on the relationship of serum cholesterol to CHD risk are available both from studies comparing different populations (inter- or cross-population studies) and within-population studies. Figure 2 is from the invaluable Seven Countries Study of 16 cohorts in northern Europe (Finland, Netherlands), southern Europe (Italy, Greece, Yugoslavia), Japan, and the U.S.A.[7]. Each cohort is a large group of men age 40-59 at baseline and free of evidence of definite CHD. Each circle in this Figure represents the population average for serum cholesterol, i.e., a group value, one for each of the 16 cohorts. As is evident, the northern European groups had high mean values of serum cholesterol (especially the East Finns - about 270 mg/dl), as did the Americans, and high CHD rates. In contrast, the southern European and especially the Japanese cohorts had much lower mean cholesterol values and lower CHD rates. The relationship between population mean serum cholesterol and risk of fatal CHD is strong and significant statistically.

Table 1 takes us from comparisons of populations, in which the only statistic is the average for a whole population, to the situation within populations. In this case, data are again from five studies of the Pooling Project in the U.S.A.[8,9]. Each of 8,247 middle-aged men is classified - based on his entry serum cholesterol - into one of five quintiles, the lowest 20%, the next 20%, etc. From the third quintile on, the higher the serum cholesterol level, the greater the risk of a first major coronary event before age 65. Compared to men in quintiles 1 and 2, those in quintiles 3, 4, and 5 had a progressive increase in risk, of 15%, 64%, and 99% respectively. Thus, 60% of the population was at greater risk compared to the two lowest quintiles, with serum cholesterol values under 220 mg/dl. This clearcut relationship was registered with only one measurement, with all its limitations because of fluctuations of

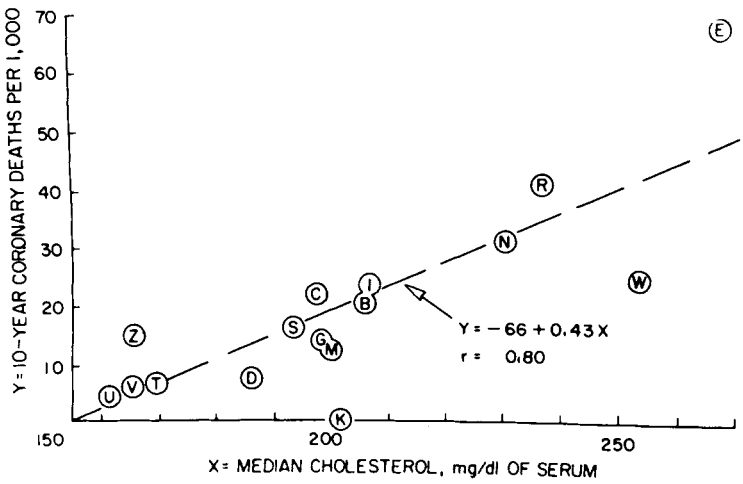

Fig. 2. Seven Countries Study; relationship between population cohort median serum cholesterol at entry and 10-year rate of coronary death; B is Belgrade faculty, Serbia, Yugoslavia; C is Crevalcore, Bologna Province, Italy; D is Dalmatia, Croatia, Yugoslavia; E is east Finland; G is Corfu, Greece; I is Rome-based Italian railroad workers; K is Crete, Greece; M is Montegiogio, Marche Region (near Ancona), Italy; N is Zutphen, The Netherlands; R is northwest U.S. railroad workers; S is Slavonia, Croatia, Yugoslavia; T is Tanushimaru (farming village), Kyushu, Japan; U is Ushibuka (fishing village), Kyushu, Japan; V is Velika Krsna, Serbia, Yugoslavia; W is west Finland; Z is Zrenjanin, Serbia, Yugoslavia[7].

individual levels and because of laboratory errors (even in research laboratories!).

Note the overall high risk of these American men in the 1950s and 1960s - 221.4 chances per 1,000 of a first major coronary event by age 65, almost 1 chance in 4, the epidemic onslaught of premature CHD.

Based on these data, it was possible to estimate the proportion of all these coronary attacks attributable to hypercholesterolemia, levels of about 220 mg/dl and above - about 25% of all attacks.

What are key environmental factors that influence the serum cholesterol level of populations and individuals? They are first and foremost the components of the habitual diet, especially its fat components. The higher the saturated fat and cholesterol in the diet, the higher the serum cholesterol level (all else being equal),

Table 1. Serum Cholesterol and Risk of a First Major Coronary Event Between Ages 40-64 (8,274 White Men, Pool 5, Pooling Project, Final Report)

Quintile of Level and Level	(mg/dl)	Number of Events	Risk of An Event per 1,000	Relative Risk	Absolute Excess Risk per 1,000	Per Cent of all Excess
I+II	≤218	166	162.7	1.00	---	---
I	≤194	86	172.4	---	---	---
II	194-218	80	153.0	---	---	---
III	218-240	104	186.8	1.15	24.1	8.3%
IV	240-268	167	266.3	1.64	103.6	35.8%
V	>268	210	324.1	1.99	161.4	55.8%
ALL		647	222.4	---	---	---

$$\frac{\text{QIII-QV Excess Events, 3,000 Men}}{\text{All Events, 5,000 Men}} = \frac{289.1}{1,112.0} = 25.6\%,$$

of all events are excess events, attributable to hypercholesterolemia.

as well illustrated in the data from the Seven Countries Study* (Figure 3, upper)[7]. (In contrast, dietary polyunsaturated fat has a modest serum cholesterol lowering effect). And the higher the saturated fat and cholesterol in the diet of populations, the greater their coronary risk (Figure 3, lower)[7]. This is true also for individuals within a population[10]. In addition, diets low in fiber, as well as high in cholesterol and saturated fat, tend to be associated with high serum cholesterol levels.

Correspondingly, changes in diet pattern - with sizeable decrease in saturated fat and cholesterol intake, moderate increase in intake of polyunsaturated fat and fiber (particularly of the pectin and gum types, from fruits and from legumes) - produces marked reduction in serum total cholesterol and its most atherogenic fraction (LDL-cholesterol) and in serum triglycerides, with little change in

*Dietary cholesterol was not measured in the Seven Countries Study. However, extensive data from other epidemiologic studies, from controlled nutritional experiments in man, and from animal research, indicate its important influence on serum cholesterol and risk of atherosclerotic disease[2].

(a)

(b)

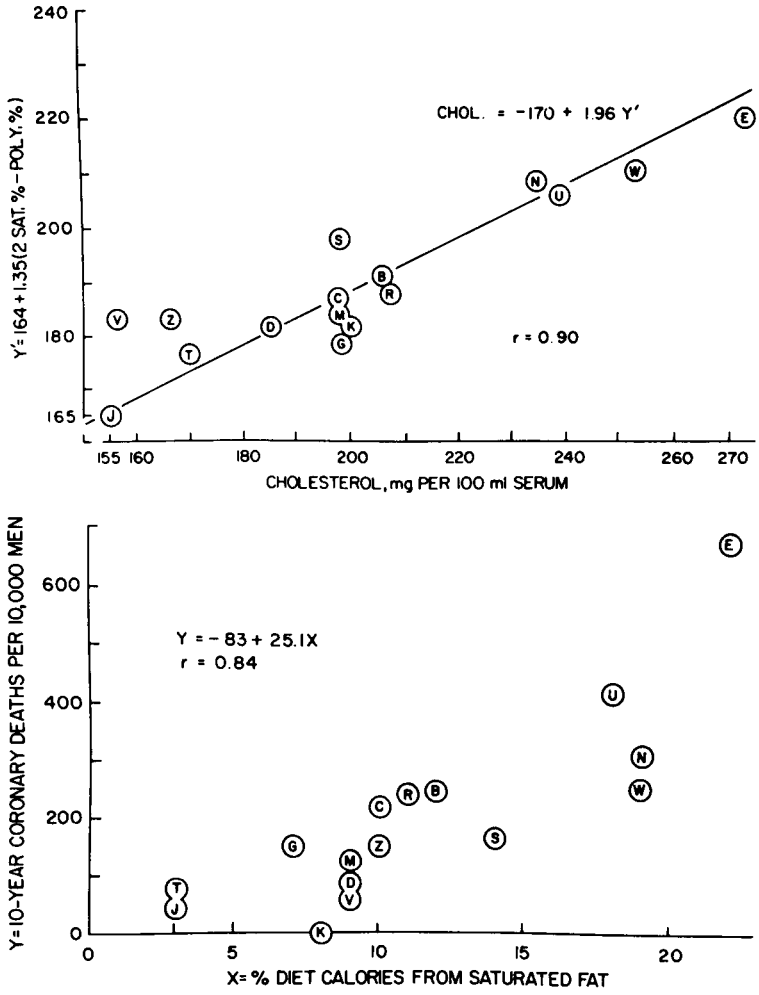

Fig. 3. Seven Countries Study; upper graph: relationship between population cohort mean intake of dietary saturated fat and polyunsaturated fat as per cent of total calories, and median serum cholesterol level at entry; lower graph: relationship between population cohort mean per cent of total calories from saturated fat at entry and 10-year rate of coronary death; for identification of the cohorts[7], see legend for Figure 2.

the putatively protective fractions (HDL and HDL_2). These changes are well illustrated by the findings with Diet C of the recent study on Dutch monks (Table 2)[11]. Note that the fall in serum total

Table 2. Diet Composition and Change in Serum Lipids-Lipoproteins Controlled Experiment, 12 Healthy Dutch Monks

Nutrient	Reference Diet A	Fat-Modified Diet B	High-Fiber Fat-Modified Diet C
Protein-% Cal.	14	14	14
Vegetable Protein-% Protein	34	34	52
Fat-% Cal.	40	27	27
Polyunsaturates-% Cal.	5.2	8.5	8.7
Saturates-% Cal.	19.3	8.4	8.7
Cholesterol-mg/2,500 Cal.$^{\Delta}$	617	245	252
Available Carbohydrate-% Cal.$^{+}$	46	59	59
Fiber-g/2,500 Cal.$^{\Delta}$	19	20	55
Pectin-g/2,500 Cal.$^{\Delta}$	1.2	1.8	6.3
Serum Lipid-Lipoprotein-Per Cent Change	**Reference Diet A**	**Fat-Modified Diet B**	**High-Fiber Fat-Modified Diet C**
Total Cholesterol	---	-21.6%	-29.2%
LDL Cholesterol	---	-26.5%	-34.5%
HDL Cholesterol	---	-12.0%	-10.6%
HDL_2 Cholesterol	---	-34.6%	-11.5%
Total Triglycerides	---	0.0%	-20.8%
VLDL Triglycerides	---	+ 4.8%	-19.0%
Ratios			
Total Chol./HDL Chol.	4.49	4.02	3.56
LDL Chol./HDL_2 Chol.	17.7	19.9	13.1

$^{\Delta}$Energy intake varied from 1,550 to 4,250 Cal/day.
$^{+}$Mono- and di-saccharides contributed 18-20% of calories.

cholesterol was a substantial 29%; in LDL-cholesterol, 34%. Corresponding results were obtained in a Boston study of a population group eating a predominantly vegetarian diet low in animal products (Table 3)[12].

Figure 4 illustrates - from the experience of the Framingham population - the effect of caloric balance on serum cholesterol (and glucose, uric acid, and blood pressure as well)[13]. Caloric

Table 3. Serum Cholesterol and Lipoprotein Levels in "Macrobiotic" Vegetarians and Controls in Boston

	Serum Cholesterol-mg/dl				
Group	Total	LDL	VLDL	HDL	Weight kg.
115 Controls	184±37	118±34	17.2±11.0	49±12	73±15
115 Vegetarians	126±30	73±24	11.8± 7.0	43±11	58± 9
Mean Difference	58±48***	45±44***	5.4±13.3***	6±15***	15±16***
% Difference	-31.5%	-38.1%	-31.4%	-12.2%	-20.5%
Mean Difference$^{\Delta}$	55±53***	39±46***	4.6±14.4*	9±17**	0± 9

$^{\Delta}$Weight matched pairs - N = 42.
* $p \leq .05$
** $p \leq .01$
*** $p \leq .001$

balance is, of course, a resultant of both diet and physical activity. With weight gain, especially on a diet high in saturated fat and cholesterol, serum cholesterol rises; with weight loss on an ordinary or a fat-reduced diet, it falls. The changes can be sizeable, even with modest changes in weight. The Multiple Risk Factor Intervention Trial demonstrated this in extenso in its large group of free-living men[14]. For obese men, weight loss of 4 to 5+ kg. on its diet - reduced in saturated fat and cholesterol, with a moderate increase in polyunsaturated fat - was associated with favorable changes in all plasma lipid-lipoprotein components. Plasma total cholesterol, LDL-cholesterol, VLDL-cholesterol, total triglycerides, VLDL-triglycerides all fell sizeably, and HDL rose. The MRFIT progressive eating pattern emphasized delectable fare based upon Mediterranean and Far Eastern cuisine, with concomitant attention to dietary fat composition and level, fiber intake from fruits and vegetables, grains and legumes, and moderation in sodium intake.

Is it possible by dietary means to influence the occurrence of atherosclerotic disease, i.e., severe atherosclerosis of the coronary, cerebral, trunk, and peripheral arteries? In the first decade of this century, Anitschkow working as a young man with colleagues in Petrograd demonstrated in rabbits the relationship among dietary lipid (dietary cholesterol in particular), serum cholesterol, and the atherogenic process[15]. This was a major breakthrough in experimental medicine, the basis for the great acquisition of knowledge by animal experimentation in this field over the last 70 years. Anitschkow was probably the first scientist to demonstrate that with

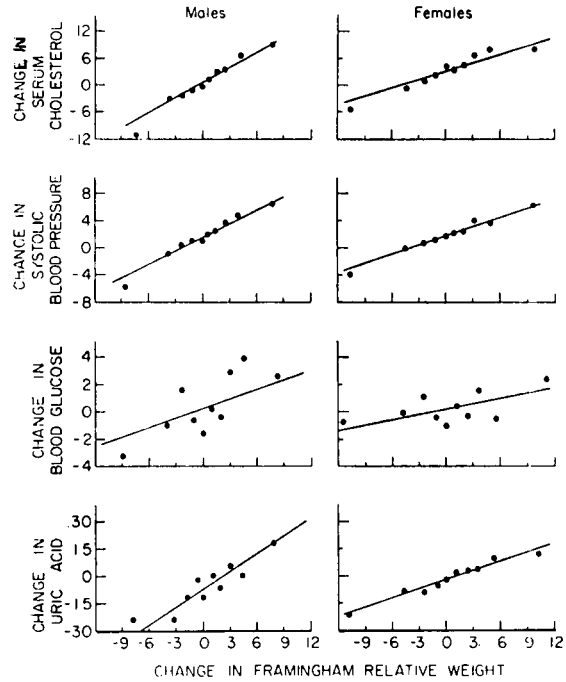

Fig. 4. Framingham Heart Study; relationship between change over time in relative weight and change in serum cholesterol, systolic blood pressure, blood glucose, serum uric acid[13].

cessation of cholesterol-fat feeding, and consequent restoration of low normal serum cholesterol in animals, atherosclerotic lesions regressed. Figure 5 illustrates this phenomenon, from a modern classic experiment in monkeys[16]. For 18 months a diet high in egg yolk (hence high in dietary cholesterol) was fed, and sustained hypercholesterolemia was thereby induced. One-third of the animals were sacrificed to study the resultant atherosclerosis and narrowing of the coronary arteries (upper 4 frames, Figure 5). The two-thirds of the animals remaining alive were then taken off this atherogenic diet and were maintained for 40 months on a diet low in saturated fat and free of cholesterol, so that the serum cholesterol was quickly restored to low normal levels. As is evident from the lower six frames of Figure 5, after these 40 months marked reversal of lesions took place and the coronary arteries opened up, with only minimal residual thickening of the arterial walls. Another study in monkeys, recently completed, showed that such regression of lesions can be achieved if the serum cholesterol is brought down to a level of about 200 mg/dl, but regression does not take place at 300 md/dl[17]. (Figure 6 and Table 4). Thus, the disease is not only preventable by nutritional means but is also within limits reversible.

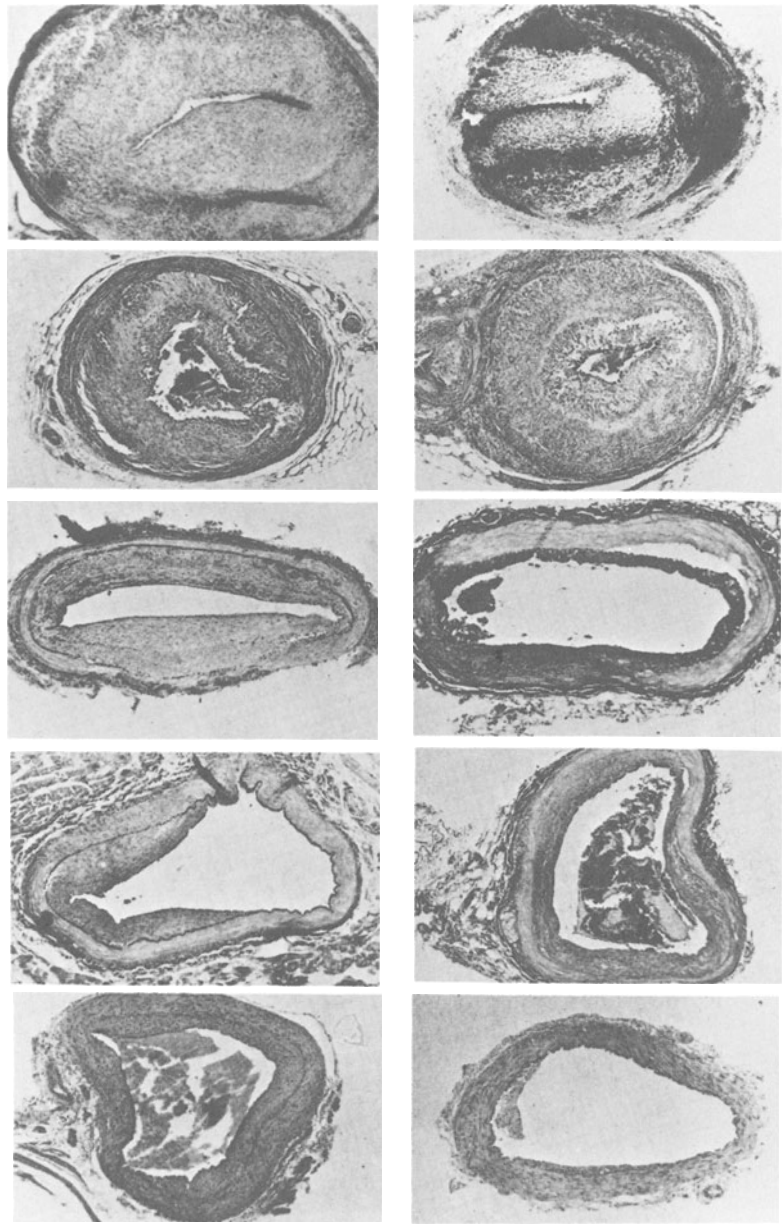

Fig. 5. Experiment on regression of coronary atherosclerosis in male rhesus monkeys; upper 2 rows (4 figures) - coronary atherosclerosis after 18 months on a high-cholesterol diet from egg yolk; lower 3 rows (6 figures) - regression of atherosclerosis 40 months after cessation of feeding of the high-cholesterol diet[16].

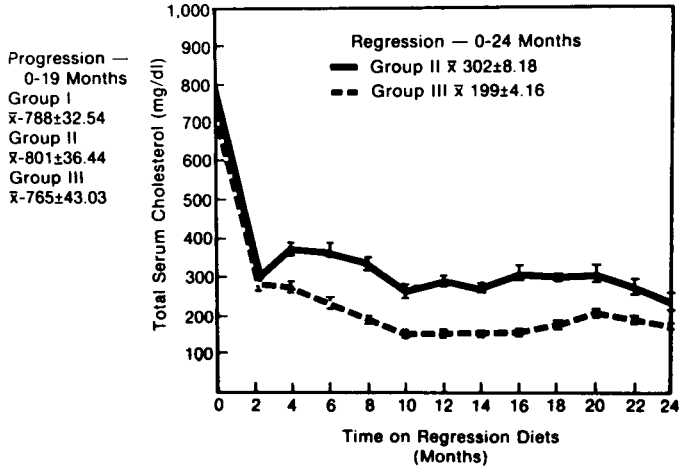

Fig. 6. Experiment on regression of atherosclerosis in macaca mulatta monkeys; mean serum cholesterol concentration after 19 months of feeding an atherogenic diet high in cholesterol and fat, and over the next 24 months, for Groups II and III, fed diets to yield mean serum cholesterol levels of approximately 300 mg/dl and 200 mg/dl respectively[17].

Table 4. Primate Regression Study Coronary Atherosclerosis

Group	Stenosis %	Fat %	Medial Damage %	IEL Damage %
I	25±5.2	41±3.9	40±8.0	25±5.8
II (Chol 300)	35±4.7	20±2.9	37±5.3	22±3.5
III (Chol 200)	17±4.2	6±1	20±5.4	17±4.1

Data are available on the human species supporting this thesis, although such data are much more difficult to obtain. Figure 7 illustrates such findings from an important study of 846 American men in late middle-age, residing in a home for veterans in Los Angeles[18]. The data are on the incidence in 8.5 years of major atherosclerotic

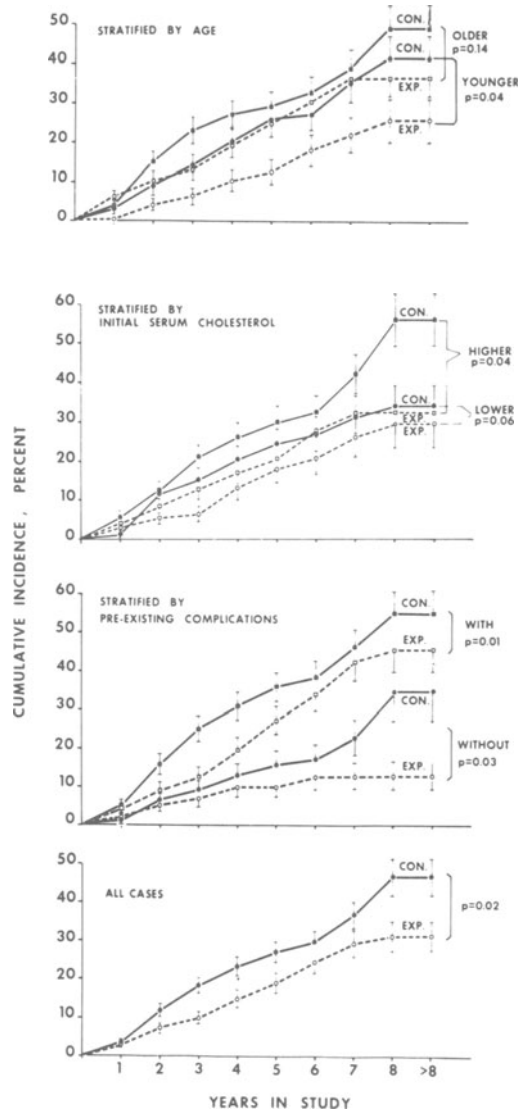

Fig. 7. Los Angeles Veterans Administration Domiciliary Facility double blind trial on dietary fat modification to reduce serum cholesterol and atherosclerotic disease; cumulative incidence rates over 8.5 years in experimental and control groups overall (bottom graph), and stratified by entry age (top graph), entry serum cholesterol level (graph 2nd from top), and entry findings with respect to clinical evidence of atherosclerotic disease; the incidence end point was a combination of "hard" atherosclerotic disease events, including coronary death, definite non-fatal myocardial infarction, definite cerebral infarction[18].

disease events, fatal and non-fatal, involving chiefly the heart and brain, in two groups - one eating a usual American diet, the other a fat-modified diet, reduced in cholesterol and saturated fat, increased in polyunsaturated fat, with a resultant reduction in serum cholesterol of about 10 to 15% over the years. Overall there was a statistically significant 34% reduction in the incidence of the major atherosclerotic diseases (lowest frame, Figure 7), even though these men were already rather elderly - median age 65.5 years - when the study began. Benefit was more clearcut and greater for the 50% of men under 65.5 years at entry (top frame, Figure 7) and for men hypercholesterolemic at entry (2nd frame, Figure 7). Both those without and with clinical evidence of atherosclerotic disease at entry benefitted, i.e., there was significant achievement in both primary and secondary prevention (although not in regard to all causes of mortality).

Table 5 presents similar positive findings from the primary prevention study in Oslo (Norway), for men age 40-49 at entry. This study involved both diet intervention for these markedly hypercholesterolemic (normotensive) men and an effort to achieve cessation of cigarette smoking[19]. Over the five years of the trial, the intervention group had a serum cholesterol level 42.6 mg/dl (13.0%) lower than the controls, and a greater degree of weight reduction by 3 kg. Of the intervention group, 31% quit smoking cigarettes; of the controls, 19%. No differences in blood pressure were recorded. Incidence of CHD and all cardiovascular diseases was markedly lower in the intervention group compared to the controls, by over 40%, and striking positive results were also obtained in regard to death rates[19]. (Table 5). Thus, as difficult as it is to acquire accurate information on ability to prevent atherosclerotic disease in man (properly designed and executed large-scale randomized controlled trials are very complex undertakings), meaningful results are indeed available, and they support all the other sets of data - from observational epidemiology, clinical medicine, autopsy studies, and animal experimentation - on the relationship among diet, serum cholesterol, and atherosclerotic disease.

BLOOD PRESSURE

Figure 8, again from the Seven Countries Study, shows the independent relationship of blood pressure, in addition to serum cholesterol, in contributing to differences across populations in risk of premature CHD[7]. Again, each population is treated as a single unit. The higher levels of both blood pressure and serum cholesterol at baseline for the northern European and U.S. population samples play a key role, in an additive way, in accounting for their much higher 10-year CHD death rates, compared to the Greek, Italian, Yugoslav, and Japanese cohorts. Measured only once at the outset of this prospective epidemiologic study, these two factors by themselves explain a good deal of the differences in CHD risk among the populations.

Table 5. 5-Year Intervention Results, Oslo Trial

End Point	Intervention Group-604 Men		Control Group 628 Men		% Difference*	p Value
Coronary heart disease incidence	19[Δ]	31.5[°]	36	57.3	-45.0%	.028
Cardiovascular disease incidence	22	36.4	39	62.1	-41.4%	.038
Sudden CHD mortality	3	5.0	11	17.5	-71.4%	.024
Coronary heart disease mortality	6	9.9	14	22.3	-55.6%	.086
All cardiovascular disease mortality	8	13.2	15	23.9	-44.8%	.168
Cancer mortality	5	8.3	8	12.7	-34.6%	---
All mortality	16	26.5	24	38.2	-30.6%	.246

*$\frac{\text{Intervention group rate minus control group rate}}{\text{Control group rate}} \times 100$

As to the effect of blood pressure on the risk of individuals within a population, Table 6 gives representative data, again from the national cooperative Pooling Project in the U.S.A., again with combined findings from five long-term studies, with the population divided into quintiles based on the entry diastolic pressure reading[8,9]. Again, there are the limitations of only one measurement. Nevertheless, diastolic pressure is highly related to long-term CHD risk (as is also systolic pressure). Even for men with diastolic readings in the 80 to 88 mm Hg range (quintile 3), usually regarded as normal for middle-aged men, there is already a 42% increase in risk of a first major coronary event by age 65. Given such findings, it seems wise to begin designating such levels as not simply normal, but at the very least, as what we are now calling high-normal. As one goes further up the scale of diastolic pressure, to 90 and over, risk increases progressively. Again, a simple calculation leads to the estimate that over 30% of all cases of heart attack in this middle-aged U.S. population of men in the 1950s and 1960s were attributable to levels of blood pressure above the optimal. Note that such elevated levels were recorded at baseline for 60% of the population (quintiles 3, 4, and 5). This is not a minority phenomenon. Almost 40% of these men had frankly elevated or hypertensive readings, based on the currently accepted medical standard of 90+ mm Hg diastolic.

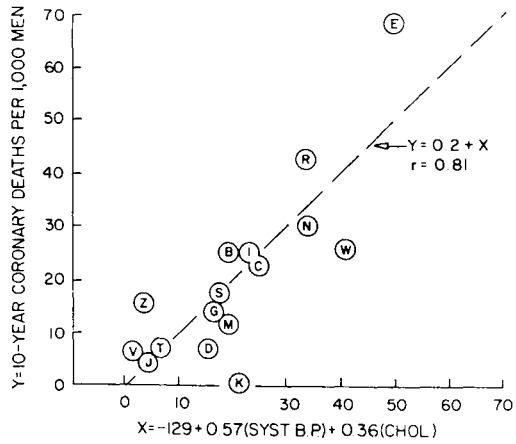

Fig. 8. Seven Countries Study; relationship between population cohort average values for systolic blood pressure and serum cholesterol at entry, and 10-year rate of coronary death; for identification of the cohorts[7], see legend for Figure 2.

Table 6. Diastolic Pressure and Risk of a First Major Coronary Event between Ages 40-64 (8,381 White Men, Pool 5, Pooling Project, Final Report)

Quintile of Level and Level	(mmHg)	Number of Events	Risk of An Event Per 1,000	Relative Risk	Absolute Excess Risk Per 1,000	Per Cent of all Excess
I+II	≤80	182	149.7	1.00	---	---
I	<76	91	162.6	---	---	---
II	76-80	91	136.7	---	---	---
III	80-88	126	212.4	1.42	62.7	18.1%
IV	88-94	142	239.7	1.60	90.0	26.0%
V	≥94	208	343.5	2.29	193.8	55.9%
ALL		658	223.0	---	---	---

$$\frac{\text{QIII-QV Excess Events, 3,000 Men}}{\text{All Events, 5,000 Men}} = \frac{346.5}{1,094.9} = 31.6\%$$

of all events are excess events, attributable to elevated blood pressure.

What makes high blood pressure so common in our populations? Table 7 gives typical data on one important trait that has repeatedly been shown to be related to occurrence of elevated pressure, namely overweight[20,21]. This is the case for older and younger, men and women, all ethnic groups, persons with and without a positive family history of high blood pressure. Prospective studies - e.g., as illustrated in Figure 4 above[13] - confirm that overweight and weight gain are indeed associated with increased risk of developing high blood pressure, for persons originally normotensive. Thus, a major risk factor for high blood pressure is overweight, and one key strategic aspect of improving blood pressure levels of populations is the prevention and control of overweight.

Figure 9 illustrates the role of another trait influencing prevalence of high blood pressure in the population, i.e., level of sodium intake[22]. Again, this is a modifiable aspect of modern human eating habits.

Another trait that is a predictor of high blood pressure is a rapid heart rate[23]. This too is amenable to influence by regular frequent rhythmic (isotonic) exercise to improve cardiopulmonary fitness.

Another important trait related to prevalence and incidence of high blood pressure is heavy consumption of alcohol - 50 or 60 or more grams of alcohol per day (4, 5, or more drinks per day), irrespective of type (beer, wine, whiskey, vodka, etc.)[24]. (Table 8).

Table 7. Family History of Hypertension, Weight and Prevalence of Hypertension by Age, Chec Screening, One Million Americans, 1973-5

Weight	Family History	Age-Averaged Prevalence of Hypertension$^{\Delta}$ per 1,000 Age 20-39	Age 40-64
Normal	Positive	82.8	309.4
	Negative	46.7	181.7
	P/N†	1.84	2.02
Overweight	Positive	182.4	443.5
	Negative	118.2	293.2
	P/N†	1.66	1.92

$^{\Delta}$DBP≥95 mmHg or reported current use of antihypertensive medication.

$^{\dagger}P_1\ (1{,}000.0 - P_2)/P_2(1{,}000.0 - P_1)$

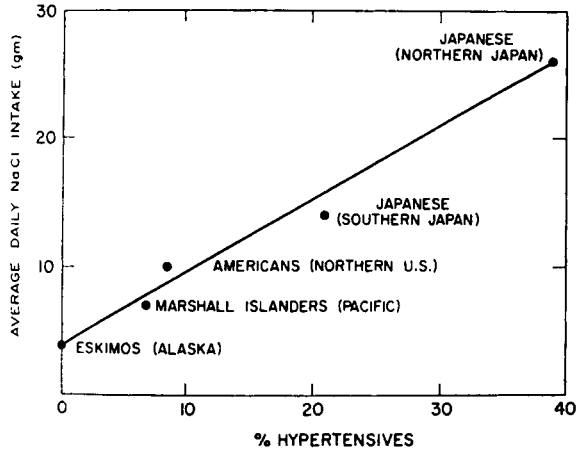

Fig. 9. Relationship between population average daily sodium chloride intake and prevalence of hypertension, based on data from 5 populations[22].

Finally, as illustrated in regard to serum cholesterol, so in regard to high blood pressure, it is possible to control or prevent the development of high blood pressure[25] (Table 9). This can be achieved by safe nutritional-hygienic means - without drugs - in many people with high-normal or already frankly elevated blood pressure levels, by reduction of weight, moderation of sodium intake, improvement of physical fitness by regular rhythmic exercise, and control of alcohol intake. For many of those who are frankly hypertensive, of course, it may also be necessary to use modern pharmacologic treatment for the long-term control of high blood pressure. And recent trials have shown that it is indeed possible to improve prognosis in regard to death from all causes and from the major cardiovascular diseases by such treatment, both for those with so-called "mild" and for those with more marked hypertension[26-28].

In summary, there is a great deal of information available on the contribution of hypercholesterolemia and hypertension to risk of coronary heart disease and the other atherosclerotic diseases. There is also much information available on the main aspects of life style of populations that influence these two traits. There is also now considerable information on the ability to apply all that knowledge to prevent these traits from developing in the first place, or to control them if already present - and the utility of these approaches for control of epidemic atherosclerotic disease. In a number of countries this knowledge is being progressively applied. There is good reason to believe that the favorable changes in life

Table 8. Per Cent with High Blood Pressure Among Problem Drinkers and Non-Problem Drinkers

Variable	Problem Drinkers N = 38	Non-Problem Drinkers N = 1195	t	Adjusted* t
Chicago Peoples Gas Company 1233 White Males Age 40-59 in 1958				
SBP ≥ 140	55.3	33.1	2.85	2.92
SBP ≥ 160	18.4	10.7	1.50	1.68
DBP ≥ 90	34.2	20.4	2.06	2.66
DBP ≥ 95	15.8	9.1	1.39	1.95
Chicago Western Electric Company 1899 White Males Age 40-55 in 1957				
	Heavy Drinkers N = 117	Non-Heavy Drinkers N = 1782		
SBP ≥ 140	59.0	36.9	4.75	4.30
SBP ≥ 160	28.2	11.7	5.18	4.66
DBP ≥ 90	64.1	42.1	4.65	4.27
DBP ≥ 95	40.2	20.4	5.02	4.75

*Adjusted by analysis of covariance for age, serum cholesterol, pulse, relative weight, and cigarettes/day.

style and medical care that have resulted are related to recent declines in CHD and stroke mortality, and all causes mortality as well. That is, the trail has been blazed indicating that prevention of these diseases is possible in the population as a whole.

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Table 9. Sequential Mean Changes in Weight, Relative Weight, Pulse Rate, Blood Pressure and Serum Cholesterol, Nondropouts, Not on Antihypertensive Drugs, Chicago Coronary Preventive Evaluation Program

Years in CPEP	No.of Men	Δ Weight lbs.°	Δ Relative Weight	Δ Pulse Rate beats/min.	Δ Systolic Pressure mm Hg	Δ Diastolic Pressure mm Hg	Δ Serum Cholesterol mg/dl
Men with Last Baseline Diastolic Pressure ≥ 90 mm Hg							
1	115	-11.9***	-7.9***	-3.1***	-11.5***	-8.1***	-28.1***
3	99	-10.1***	-6.7***	-2.5*	-12.5***	-8.6***	-22.8***
5	96	- 8.4***	-5.5***	-5.1***	-10.8***	-8.0***	-21.8***
7	65	- 8.1***	-5.4***	-3.6*	-13.4***	-9.7***	-23.2***
9	35	- 6.3***	-4.4***	-3.9	-11.8***	-12.6***	-25.3*
Baseline Value 115 Men	--	197.3	129.6	81.2	147.7	96.3	248.5
Men with Last Baseline Diastolic Pressure 80-89 mm Hg							
1	101	-10.6***	-7.1***	-2.3**	- 7.9***	- 4.4***	-22.4***
3	97	- 9.8***	-6.5***	-3.5***	- 7.3***	- 4.0***	-14.7***
5	89	- 7.7***	-5.1***	-3.2**	- 7.0***	- 3.3***	-12.8***
7	72	- 6.7***	-4.4***	-2.7**	- 2.8*	- 3.1***	-13.7***
9	46	- 8.4***	-5.5***	-3.5*	+ 0.1	- 3.0*	- 7.2
Baseline Value 101 Men	--	189.9	126.2	78.1	131.5	82.9	259.5

*p≤.05 **p≤.01 ***p≤.001 Δ is "Change in". °Relative Weight is the ratio of observed weight to desirable weight for height and sex, from the 1959 actuarial data[6,18].

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DETECTION AND TREATMENT OF VENTRICULAR ARRHYTHMIAS

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INTRODUCTION

During the past 15 years the techniques available to detect ventricular arrhythmias in hospitalized and ambulatory patients have been developed to permit characterization and quantitation of the ventricular arrhythmias.[1] These advances have permitted the assessment of ventricular arrhythmia in many cardiac disorders and determination of their importance in the natural history of each syndrome.[2] Recently, a number of new antiarrhythmic drugs have been developed to provide the cardiologist a number of highly effective therapeutic agents for almost all ventricular arrhythmias.[3] However, each advance has raised new problems such as stratifying patients requiring treatment, documenting the effects of treatment and assessing the impact of treatment, on the natural history of the cardiac disorder. The report focuses on the latest available information about the detection and treatment of ventricular arrhythmias.

MAGNITUDE OF THE PROBLEM

In the USA there are more than 300,000 arrhythmic sudden deaths annually. Almost 90% of these patients have coronary artery disease (CAD), but other cardiac disorders are associated with ventricular arrhythmias and sudden death (Table 1).

Since CAD is the largest single cause of ventricular arrhythmias and is the area where sufficient studies have been performed to permit conclusions, we focus our presentation on the early and late occurrences of ventricular arrhythmias in the CAD patients.

Table 1. CV Disease Associated with Arrhythmic Death

1. Coronary Artery Disease
   a) Post-myocardial infarction
   b) Unstable angina
   c) Stable angina
   d) "Sudden Cardiac Death Syndrome"
   e) Coronary spasm
2. Cardiomyopathy
   a) Dilated forms (advanced)
   b) Hypertrophic - obstructive and non-obstructive
3. Aortic Valve Disease
4. Prolapsed Mitral Valve
5. Congenital Disease
   a) Pre-excitation syndrome
   b) Repaired shunt lesions
   c) Conduction disease
6. Idiopathic

Early Post-Myocardial Infarction Ventricular Arrhythmias

Electrical inhomogeneities occur within seconds of coronary occlusion and ventricular arrhythmias are almost universal. The ventricular arrhythmias and sudden death rate decreases thereafter almost logarithmically with time. While the concept of "warning ventricular arrhythmias" occurring before ventricular fibrillation in the CCU was developed in the 1960's, it is now well documented that many patients have ventricular fibrillation with no or very short warning times. In addition, CCU's without computer monitoring do not detect many serious ventricular arrhythmias and there is marked delay or failure to commence therapy. For these reasons, I and others have advocated the prophylactic administration of lidocaine (or other agents) for all patients following acute infarction.[4,5,6] With new understanding of the pharmacokinetics of lidocaine, the ability to monitor plasma concentration and understanding the influence of disease on the disposition of the drug, it can be administered safely and effectively in most patients in the CCU for 36 to 48 hours.[7]

Late Post-Myocardial Ventricular Arrhythmias

After myocardial infarction late occurring ventricular arrhythmias are seen in from 52 to 88% of all patients depending on the duration of monitoring.[9,10] Many studies have documented a higher sudden cardiac death rate for 6 to 12 months after myocardial

infarction, but stratification of risk depends on quantitation of ventricular arrhythmia and the association of decreases in ventricular function due to myocardial loss.

In patients with the "sudden death syndrome" almost 90% have underlying severe CAD, but only approximately 16-25% experience myocardial infarction at the time of ventricular fibrillation. Untreated, these patients have 46-60% recurrence of ventricular fibrillation in one year.

MONITORING AND INTERPRETATION OF VENTRICULAR ARRHYTHMIAS

A number of technologies permit monitoring of ambulatory patients for ventricular arrhythmias. These include: telemetry units in hospitals, analog (Holter) tape units, ambulatory event recorders which activate when ventricular arrhythmias are detected and record "snapshots" and telephone coupling transmission devices. The greater the period of monitoring, the higher yield of detected events.[2] However, the costs, complexity of analysis, the need for a computer system for quantitation, and the inability of the patient to keep electrodes functional for long periods limits recording times.[2] Indications for ambulatory monitoring are outlined in Table 2.

Table 2. Indications for Ambulatory Monitoring in CAD

1.	Prognostic stratification of post-MI patients
2.	Symptoms
	a) Palpitations
	b) Syncope or presyncope
	c) Stroke or cerebral ischemic attacks
3.	To assess results of therapy
4.	Post-resuscitation of sustained VT/VF

Interpretation of ventricular ectopic activity to determine its prognostic value and when it should be treated remains controversial although careful clinical studies permit tentative conclusions.[8,10] Frequent and complex ventricular ectopic activities (VT, couplets, R on T, and multiformed ventricular ectopic activities) are common: greater than 10-20/hour in 7-26% of studies; complex forms in 27-43% and VT in 1-14%, of reported studies in post-myocardial infarction groups. Spontaneous variability is considerable.[11,12] And all types of ventricular ectopic activities are more common with CHF,[10] reduced ejection fractions and ventricular aneurysm. However,

I conclude that frequent and complex ventricular ectopic activities have independent prognostic value.

TREATMENT OF VENTRICULAR ARRHYTHMIAS POST-MYOCARDIAL INFARCTION

A number of studies have documented that beta-blocking drugs reduce overall mortality and sudden deaths when given after myocardial infarction. They should be used when not contraindicated.

Studies do not definitively document that treatment of ventricular arrhythmias with Class 1 antiarrhythmic agents reduces mortality. However, existing studies have problems demonstrating that effective treatment has been given, that suppression of ventricular ectopic activities is required and that the observed reduction of ventricular ectopic activities is not just spontaneous variation.[11,12] For these reasons I recommend Class 1 antiarrhythmic agents when frequent and complex ventricular ectopic activities are detected (Table 3).

Table 3. Ventricular Ectopic Activity to Treat

1.	When sustained VT occurs (greater than 20 beats
2.	Greater than 20 VEA's/hour
	a) Nonsustained VT (less than 20 beats)
	b) Couplets
	c) R on T
	d) Multiformed VEA's
3.	When 1 or 2 (above) occur with heart failure exceptional effort

Several methods have been developed to determine effectiveness of treatment. First, use plasma levels of antiarrhythmic drugs to be certain each patient is in therapeutic range and do not attempt ventricular ectopic activity monitoring.[7] Secondly, do two ambulatory monitorings before instituting treatment and 4 to 7 afterwards with probability statistical analysis.[13] Thirdly, two pretreatment quantitations with analysis of variance and confidence intervals established for judging one or more post-treatment monitoring results.[13] Fourthly, to require 90% suppression of all ventricular ectopic activities. Further studies are clearly needed.

Schema for Post Myocardial Infarction Management

Based on clinical studies, the new technologies available, and the potential for salvaging large numbers of patients, I offer a schema for post-myocardial management (Table 4).

Table 4. Schema for Post-MI Management

1.	Beta-blocker to all with no CHF, exercise ECG ischemia or frequent or complex VEA's.
2.	For angina, exercise ECG ischemia, and complex and frequent VEA's - coronary angiography and stratification for surgery or medical treatment.
3.	For suitable patients - CABG based on symptoms and anatomy.
4.	For suitable CABG patients - Class 1 antiarrhythmic treatment a) Vessel anatomy b) Ventricular function poor
5.	If CABG not required - antiarrhythmic drugs based on electrophysiologic induction study.

While such an approach is experimental at this time, background suggestive supporting data are available for each new technology, for each new quantitative approach and for a larger group of safe and effective drugs.[14] The potential salvage of young patients with CAD makes it imperative to launch such approaches in the near future.

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CLINICAL EFFICIENCY AND SIDE-EFFECTS OF OLD AND NEW ANTIARRHYTHMIC DRUGS

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The number of antiarrhythmic drugs has increased greatly during the last three decades. From the triad of digitalis, quinidine and procainamide in 1951 to now a multitude of drugs. The purpose of the present paper is to assess the therapeutical potentials of this large armory of drugs. Focus will be on long-term prophylaxis of symptomatic paroxysmal tachycardia and fibrillation with a few comments on the treatment of ventricular extrasystole.

Drug Action Through Suppression of Arrhythmia Generating/Triggering Event (Calcium Channel Blockers and Beta Blockers).

In the more infrequent case, the mechanism of drug action is causal in that it suppresses the pathophysiological event underlying the arrhythmia, or protects a functionally unstable myocardium against the arrhythmia triggering influence of an increased adrenergic tone.

Calcium channel blockers, both those with effect on the AV nodal cells (verapamil and diltiazem) and the one without (nifedipine), act against arrhythmias related to Prinzmetal's variant angina.[1] By their vasodilatatory effect coronary artery spasm is prevented and with it attacks of chest pain and arrhythmia. While chest pain may be slight and not reported by the patient at the history taking, arrhythmia symptoms like fainting may prevail in the clinical picture. The ST elevation preceding the arrhythmia and signaling transmural myocardial injury may not be visible in the monitoring lead, or the ECG registration may first have been started after the onset of the arrhythmia. Thus, occasionally all signs of myocardial

ischemia may be missing and with them the clues for the choice of the proper antiarrhythmic treatment.

By protecting the myocardium against adrenergic influence, beta-blocking agents are capable of:

- suppressing attacks of torsade de pointes ventricular tachycardia in a substantial part of the patients with congenital long QT syndrome with or without associated hearing defect[2], and
- alleviating exercise/emotion induced attacks of ventricular tachycardia/fibrillation with fainting in the child or adolescent with normal ECG[3], or in the individual with mitral valve prolapse and T wave anomalies.[4] Exercise induced ventricular tachycardia/fibrillation related to coronary artery disease is best managed by coronary artery surgery.[42]

The therapeutical response is usually all to none with complete alleviation of the attacks of tachycardia/fibrillation. The efficacy of an initiated antiarrhythmic treatment can often be challenged by provocation test: by hyperventilation or ergonovine test in Prinzmetal's variant angina[1], by abrupt awakening in the early morning in a few of the patients with long QT syndrome[3,41], and by work load on a bicycle ergometer or treadmill in the patient with exercise induced fainting.[3] Particularly in the younger age groups, failure to comply with regular medicine intake can result in recurrence of the attacks of tachycardia and sometimes in sudden death.[3]

Drug Action Through Modification of Cardiac Cell Electrophysiological Properties (Quinidine, Mexiletine, Propranolol, Amiodarone and Verapamil

In the great majority of patients, drugs exert their effect directly on the cardiac cell by modifying its electrophysiological properties. The mechanism of action can be that of a suppression of enhanced ectopic activity; or the evening out of differences in myocardial refractoriness within a potential reentry circuit; or the conversion of an unidirectional block in such a circuit to a bidirectional one blocking for further impulse circling.

To create some form of order in the electrophysiological potentials of the multitude of antiarrhythmic drugs, they are usually divided into four classes according to their dominant effect on the action potential (Table 1). The digitalis glycosides form an additional fifth class of their own, being the only drugs which increase vagal heart tone on a chronic basis.[6] A clinically more valid classification is a differentiation between "wide spectrum" antiarrhythmic drugs with action potential against both supraventricular and ventricular arrhythmia and more "narrow spectrum" ones with effect primarily on either supraventricular or ventricular arrhythmia (Table 2).

Table 1. Classification of anti-arrhythmic drugs from their dominant effect on the cellular action potential (AP). Class 1 drugs are divided in class 1 A drugs (quinidine-like drugs) which slightly prolong the duration of AP, and class 1 B drugs (lidocaine-like drugs) which shorten or do not affect the duration of AP. Modified after Vaughan Williams.[39]

Class 1		Class 11	Class 111	Class 1V
Depressors of max. rate of rise of AP (Fast response inhibitors)		Beta-blocking agents (Adrenergic tone inhibitors)	AP-prolongators (Refractoriness prolongator)	Calcium channel blockers (Slow response inhibitor)
A	B			
Quinidine	Lidocaine	Propranolol	Amiodarone	Verapamil
Ajmaline	Aprindine	Acebutolol	Sotalol	Diltiazem
Disopyramide	Encainide	Atenolol		
Procainamide	Ethmozin	Metoprolol		
Propafenone	Flecainide	Pindolol		
	Lorainide	Sotalol		
	Mexiletine	Timolol		
	Tocainide			

Table 2. Classification of antiarrhythmic drugs from their effect on supraventricular arrhythmia (SVA) and ventricular arrhythmia (VA)

Wide Spectrum Drugs for SVA and VA	Narrow Spectrum Drugs for SVA	Narrow Spectrum Drugs for VA
Class 1 A drugs	Digitalis[a]	Class 1 B drugs
Beta-blocking agents	Verapamil	(Verapamil)[b]
Amiodarone		

[a] Digitalis should not be given to the adult patient with WPW.

[b] Verapamil is usually not effective in ventricular tachycardia but may be so in Gallavardin's repetitive ventricular tachycardia[40]

Independent of arrhythmia type and mechanism sensitivity and tolerance to various drugs varies greatly from one individual to another. The optimal result of drug prophylaxis is usually restricted to a relative bettering of the arrhythmic status with a reduction in rate, duration and/or severity of the arrhythmic events. Complete elimination of the arrhythmias is the exception.

Selection of the Optimal Drug

Within the framework of the statements in Table 2, selection of the optimal drug is based on the cumbersome trial and error method. An alternative approach in advanced cardiological centers is invasive electrophysiological investigation with programmed electrical stimulation (PES).[7] PES is performed with the intention of deliberately provoking arrhythmia before and after drug administration to challenge the antiarrhythmic efficacy of drug treatment. Prospects of the method seem good for selecting the optimal drug and, particularly for excluding the dangerous one which may worsen the arrhythmia and endanger the life of the patient.[8,9] However, long-term validity of these optimistic preliminary results awaits corraboration by large scale, well planned investigations. The resource demand of the method will also have to be dramatically reduced before it can significantly influence management of the average arrhythmia patient. The method seems not equally applicable for the assessment of all antiarrhythmic drugs. Thus, a poor correlation has been reported between PES inducibility of the tachycardia and the often very impressive reduction in symptomatic events observed in the amiodarone treated patient.[10]

Drug Prophylaxis in Chronic Ventricular Extrasystole

Individual patients with chronic ventricular extrasystole (VE) often show large degrees of spontaneous variability. Repeated 24-hour Holter monitoring was performed in an own series of 28 clinically stable patients - 11 with chronic coronary artery disease, 5 with cardiomyopathy and 12 without organic heart disease. The selection criterion was an average VE count of 200/hour or more at a previous 24-hour Holter monitoring. The spontaneous variations from the first to the second investigation with 95% and 99% confidence limits are shown in Fig. 1. Through a further statistical evaluation (regression analysis) it was found that in a clinical trial a reduction of at least 80% in average number of VE/hour is required to confirm drug efficacy. Figures of 65 - 83% have been reported by former investigators.[11,12]

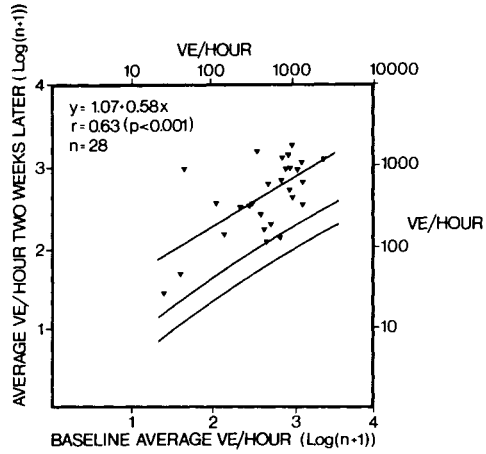

Fig. 1. Spontaneous variability in ventricular extrasystole (VE) frequency of 28 patients investigated with 24-hour monitoring at 14-day intervals. VE/hours at the first investigation is indicated along the abscissa, from the second investigation along the ordinate (log. scale). Linear regression analysis was used to describe the relationship between the two recordings and to establish 95% and 99% one-tailed confidence intervals for this relationship. To distinguish a true drug response from spontaneous variability at the 0.05 and 0.01 level of significance, the coordinate corresponding to no-treatment/post-drug response must fall below the 95% or 99% confidence limits, respectively.

Judged by these criteria, several corss-over studies[14,15] convey the impression that most antiarrhythmic drugs of class I A and B possess a reasonable rate of success in the suppression of ventricular extrasystole. The individual sensitivity/tolerance varies from patient to patient. No single agent seems to be out-standing in efficacy over the other, but in the individual case the one may work where the other has failed. Beta-blocking agents seem somewhat less efficient in suppressing ventricular extrasystole than class I drugs. When tried in an open series, the class III drug, amiodarone, proved efficient in a substantial number of cases where other drugs had failed.[16] Verapamil is usually non-effective.

Drug Prophylaxis in Symptomatic Paroxysmal Supraventricular Tachycardia

The episodic nature of the paroxysms and lengthiness of the required study period preclude the use of expensive Holter monitoring in the evaluation of the patient with paroxysmal supraventricular tachycardia. With all of its inbuilt shortcomings we have tried to assess the problem by persuading a series of patients to keep a diary of their attacks over periods extending from several months to several years. At intervals, the accuracy of the diary keeping has been checked by telemetric ECG monitoring during hospital admission. Irrespective of the type of supraventricular arrhythmia and underlying heart disease, if any, arrhythmic activity was usually erratic and highly unpredictable in appearance (Fig. 2). The wide spontaneous variability in attack rate, duration and severity makes it obvious that any proper assessment of drug efficacy in paroxysmal supraventricular tachycardia ought to build on the outcome of controlled trials or preferably double-blind cross-over studies extending over a period of at least several months. The extent of such trials is extremely limited.[17,18] What we "know" today about antiarrhythmic drug efficacy in paroxysmal supraventricular tachycardia is based primarily on the outcome of non-controlled trials supported by evidence from electrophysiological investigations in the cardiac laboratory with programmed electrical stimulation (PES). The drugs presumed to be efficient are the ones listed in the first two columns of Table 1. Each of the two class I A drugs, quinidine and disopyramide, are often administered in combination with digitalis to counteract their vagolytic effect, which in the event of atrial fibrillation/flutter may otherwise result in high ventricular rates. In infants with paroxysmal tachycardia digitalis is the drug of first choice. The main object of digitalis in this context is to counteract heart failure, and it is often given as the only drug.[19] As a general rule digitalis should not be given to adult patients with Wolff-Parkinson-White syndrome since it may decrease refractoriness in the accessory pathway causing a very high ventricular rate in the event of atrial fibrillation with accompanying risk for development of ventricular fibril-

ation.[20] Due to possible side-effects, amiodarone, at least in the hands of the authors, is used as the drug of second or third choice although it will often prove effective in a substantial number of cases where the other drugs will fail (see special section on amiodarone).

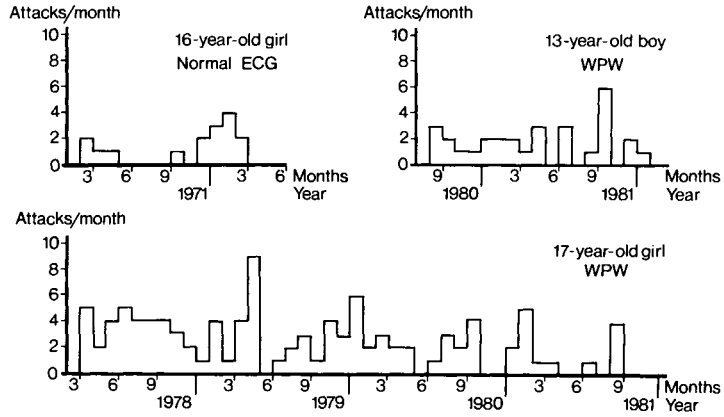

Fig. 2. Spontaneous variability in paroxysmal supraventricular tachycardia. Three adolescents without organic heart disease, two with, one without WPW. Diagrams based on patient diaries.

With these limitations drugs are usually tested on a trial and error basis with no universally accepted rank of order for the testing. It is obvious from a view of Fig. 2 that spontaneous variations in the attack rate of paroxysmal supraventricular tachycardia can mimic drug-induced bettering or worsening. To avoid this fallacy it is recommendable to start out with a no-treatment or placebo period establishing the individual "attack profile" as baseline for drug efficacy in the subsequent periods of treatment (fig. 3). After selection of the better drug for continued treatment it might be worthwhile to check the accuracy of the baseline and the assumed antiarrhythmic efficacy of the selected drugs by renewed account of attack rate/duration during another no-treatment or placebo period. For various reasons it may be necessary to omit the initial no-treatment or placebo period completely and instead establish an attack profile from the case history in the knowledge that this frequently will be inadequate.

Drug Prophylaxis in Symptomatic Paroxysmal Ventricular Tachycardia

A great spontaneous variability is also seen in symptomatic paroxysmal ventricular tachycardia (Fig. 4). Judged from non-controlled

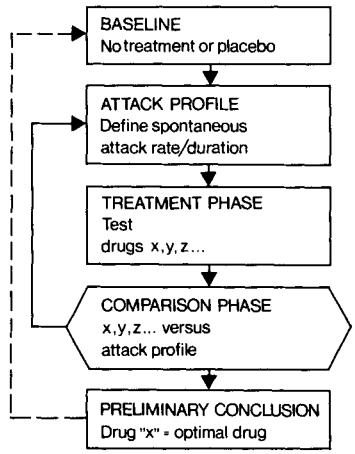

Fig. 3. A "trial and error" model for selecting the optimal drug treatment in paroxysmal supraventricular tachycardia.

series, class I antiarrhythmic drugs or beta-blocking agents given alone or in combination will often reduce the attack rate. Again, when tried, amiodarone often proves effective where other drugs have failed (see special section on amiodarone).

Drug Prophylaxis in the Prevention of Sudden Death in Coronary Artery Disease.

It has been widely assumed that reducing ventricular extrasystole in the patient with coronary artery disease is synonymous with reducing the risk of sudden death due to paroxysms of ventricular tachycardia/fibrillation. An incredible number of Holter monitoring hours have been spent in first finding the patient with a high incidence of ventricular extrasystole and next following the effect of various antiarrhythmic drugs in reducing the frequency of ventricular extrasystole. However, for evaluating drug efficacy in preventing sudden arrhythmic death, reduction in ventricular extrasystole appears to be a faulty substitution parameter. Thus, in four large long-term controlled trials with various class I antiarrhythmic drugs in acute myocardial infarction survivors, all drugs proved efficient in decreasing the frequency of ventricular extrasystole. However, in no case was the reduction in ventricular extrasystole followed by a significant lowering of sudden death mortality.[21] In contrast, drug prophylaxis with beta-blocking agents exerting less suppressive effect on ventricular extrasystole have proved efficient in lowering both total mortality and mortality

related to sudden death, probably because they are able to protect the myocardium against ischemic events and hereby intervene in the disease process.[21]

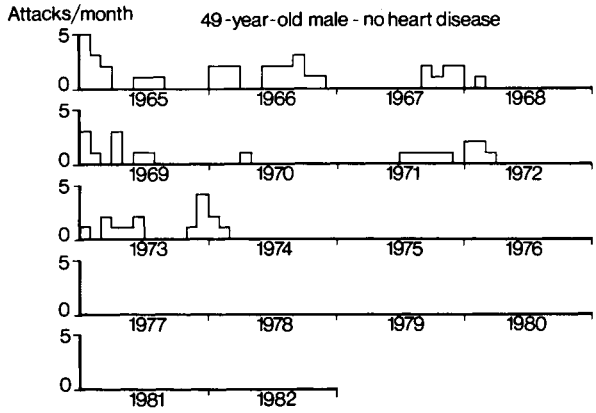

Fig. 4. Spontaneous cessation of paroxysmal ventricular tachycardia after repeated attacks over a period of 9 years. The patient is a 49-year-old male with normal ECG and no sign of heart disease (normal coronary arteriography and ventriculography). From 1965 to 1974 repeated attacks of sustained ventricular tachycardia lasting for hours to days which often had to be stopped by DC conversion. During the same period of time treated with many various drugs without definite benefit. Since 1974 and up to the last follow-up in 1982 - no treatment and no attacks. It appears that the start of a drug treatment in the last month of 1973 might have conveyed the impression of high efficacy with the recommendation that the treatment should be continued on a lifetime basis.

Side-Effects of Antiarrhythmic Drugs

Side-effects, cardiac and non-cardiac side-effects, are common in antiarrhythmic drug treatment. They can be due to high plasma concentrations of the drug, but in a substantial number of cases are not dose dependent and can occur at drug concentrations definitely within the therapeutical level. Interactions can occur. Thus, quinidine[22], amiodarone[23], and to some extent verapamil[24], when given combined with digoxin, will increase the plasma concentration of digoxin increasing the risk of digoxin intoxication. Cardiac arrhthmias are potential complications of any antiarrhythmic drug regimen. They can take the form of

Worsening of the original arrhythmia which can occur in up to 10% of the patients deduced from electrophysiological studies using programmed electrical stimulation (PES)[8,9] and a single clinical series.[25]

Torsade de pointes ventricular tachycardia preceded by QT prolongation observed particularly during treatment with quinidine[26,27], disopyramide[28], and aprindine[29], but occurring also with the beta-blocking agent sotalol[30], and with amiodarone.[31]

Increase in ventricular rate in atrial fibrillation or flutter related to the vagolytic effect of disopyramide and quinidine or in WPW syndrome due to digitalis or more rarely verapamil, lowering the refractory period in the accessory pathway.

SA block, AV block, bradycardia and/or asystole complications developing primarily in patients with latent or manifest dysfunction of the SA and AV conduction system.

In general, antiarrhythmic agents (except digitalis) have a depressing effect on cardiac contractility. The depressant effect on cardiac contraction is regarded as primarily true of disopyramide[32], and is considered to be rare with amiodarone.[33] Among the many non-cardiac side-effects attention will be called only to those of amiodarone as listed below.

Amiodarone, the Better Drug and the Drug with the Unusual Side-Effects

Amiodarone is a drug of several virtues, thus-

- evidence from several open series states that it is effective in a substantial part of patients both with supraventricular and ventricular tachycardia where other drugs have failed, and that it more often than other drugs leads to total suppression of the attacks of tachycardia/fibrillation[16,33,34], results from own series of 50 patients support this view (Table 3);
- it is particularly effective in the prevention of tachycardia in WPW syndrome and is the drug of choice for slowing down the ventricular rate in WPW related attacks of atrial flutter/fibrillation.[35]
- its long halflife (1 month) ensures continued drug effectivity by a single daily oral dose, not endangered even by a few days' lapse in medicine intake.

Meanwhile, more general use of amiodarone is restricted due to a number of unusual, worrisome side-effects including: thyrotoxicosis, myxoedema, goitre, photosensitivity, pneumonitis and pulmonary

fibrosis.[10,34,36,37,38] Incidence of thyrotoxicosis in own series (Table 4) was as high as 10%; much smaller percentages have been reported by other investigators.[37] Corneal micro-deposits are observed in almost all patients receiving amiodarone, but these rarely interfere with vision, are dose related and disappear after some time when treatment is discontinued. Because of the thyroid complications, monitoring of thyroid function is essential in all patients on long-term therapy.

Table 3. Amiodarone in 50 patients with symptomatic paroxysmal tachycardia resistant to treatment with a variety of other antiarrhythmic drugs. 27 patients were without signs of organic heart disease (4 with WPW); 11 had ischemic heart disease; 3 congenital heart disease, hereof 2 with WPW; 3 congestive cardiomyopathy; and 6 miscellaneous forms of heart disease.

Effect of Treatment	Paroxysmal Tachycardia	
	Supraventricular	Ventricular
No symptomatic events	8	7
Fewer attacks	15	-
Unchanged	12	7
Worsened	-	1[a]
Total no. of patients	35	15

[a] Repeated attacks of torsade de pointes ventricular tachycardia.

CONCLUSIONS

The state of the art of antiarrhythmic therapy continues to be a difficult balancing between therapeutic and toxic effect of the applied drugs. The expansion in recent years with the introduction of a vast number of class I drugs has brought with it only limited progress. It can be hoped for the future that:

- new drugs of the amiodarone type will be in the offing, but drugs without its unusual and worrisome side-effects and with simpler pharmacocinetics and easily measureable plasma concentration.

Table. 4. Side effects of amiodarone in 50 patients (same series as in Table 3). Treatment duration 1 month to 5 years, average 2½ years. Development of thyrotoxicosis was observed in 5 patients treated for periods varying from 16-29 months in the two patients during treatment and in the three patients 6-9 months after cessation of treatment.[36]

Side-effect	No. of patients
Corneal micro-deposits	50
Thyrotoxicosis	5
Goitre, eumetabolic	3
Photosensitization	3
Aggravation of SA block	2
Torsade de pointes VT	1
Vasculitis	1

- electrophysiological testing will live up to its promise and evolve into a less resource demanding method.
- pacing and surgery may father better treatment alternatives for the many patients with arrhythmias who are presently poorly helped by medicine.

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GENERAL AND LOCAL FACTORS IN ARTERIAL THROMBOSIS:

PLATELETS, PROSTAGLANDINS AND POSTULATES

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The discovery by Moncada et al., (1976) that vessel fragments could convert prostaglandin endoperoxide G_2 in an unstable metabolite with a potent antiaggregating as well as vasodilating property appeared to be a major step in the understanding of arterial thrombosis. The fact that this substance was mainly produced in the endothelial cells of the vessel wall (Moncada et al., 1977) seemed to be an additional explanation to the non thrombogenicity of the endothelium. These discoveries were followed by the emergence of the concept of antagonistic substances, namely platelet thromboxane (TX) A_2 and vascular prostaglandin (PG) I_2 playing a key role in the thrombocyte-vessel wall interactions. According to this concept, a balance might exist between TXA_2 and PGI_2 for the homeostatic regulation of platelet-vessel wall interactions. This very tempting hypothesis has stimulated a tremendous effort in the cardiovascular field, aimed at the verification of this theory as a likely explanation of most thrombotic disorders. However, at the present time, it appears that the TXA_2 - PGI_2 axis may only be one part of the mediators that play a role in vascular disorders. Plasma factors, membrane components, blood cells other than platelets... may be involved in those processes. In this chapter, we will try to review some of these aspects.

DYSFUNCTION OF CYCLOOXYGENASE REGULATIONS IS SUPPOSED TO BE RESPONSIBLE FOR THROMBOTIC EVENTS

Let us consider some pathophysiological situations for which an alteration of the TXA_2/PGI_2 balance has been claimed to be the main explanation.

Non Steroidal Antiinflammatory Drugs as Antithrombotic Molecules

The finding that aspirin irreversibly acetylated platelet cyclooxygenase (Roth and Majerus, 1975) and therefore inhibited PGH_2 as well as TXA_2 production was a good justification to its use as an antithrombotic drug. However, although a lot of wide scale trials have been conducted in cardiovascular diseases, the best estimate of reduction in total mortality was just less than 10% (Sweetnam and Elwood, 1982). At the present time, it is not clear whether aspirin should be more active in some defined categories of patients nor why it is less effective in women in the prevention of strokes. Further, although the administration of aspirin to patients was intended to act as an antiplatelet drug it is known that platelet aggregation undergoes via several metabolic pathways. Actually, no one knows exactly which pathway of aggregation or which mediator predominates in many situations where aggregation seems to play an important role in the disease. Among the cyclooxygenase-independent pathways, PAF-acether has been claimed to aggregate platelets independently of TX formation. We have shown in in vitro studies that platelet activation (i.e. release reaction involving the liberation of vasoactive substances such as 5-Hydroxy-tryptamine from the dense bodies or of products facilitating cell-cell interactions such as fibrinogen or fibronectin stored in the α granules) did not depend on TX synthesis but rather of the triggering of internal Ca^{2+} fluxes (Levy-Toledano et al., 1982). These facts emphasize the need for designing new drugs more adapted to antithrombotic functions than anti-inflammatory drugs.

Key Role of PGI_3 in the Diet

In 1978, Dyerberg et al., who were doing epidemiological studies on the Greenland eskimos mainly fed on fish diet observed a very prolonged bleeding time that they related to the rarity of atherosclerosis. They hypothesized that the replacement of arachidonic acid by fish eicosapentaenoic acid in the membrane phospholipids of those individuals caused a synthesis of vascular PGI_3 (whose biological activity was comparable to PGI_2) contrasting with an inactive platelet TXA_3. However biochemical regulations turned out to be more complex since Hornstra and collaborators (1981) showed that experimental animal diet with fish oil resulted in an equal decrease of TXA_2 and PGI_2 suggesting a competition of eicosapentaenoic acid with arachidonic acid at the cyclooxygenase level rather than conversion. Further it was also shown (Berlin et al., 1980) that dietary fat modification also altered the fluidity of platelet membranes leading rather to physical perturbations of the cell membrane reactivity rather than a modification of the TXA_2/PGI_2 balance.

Circulating PGI_2 as a Hormone

Since a number of years it was admitted that PG were autacoids rather than hormones. However in 1978, the report of the existence of circulating PGI_2 (Gryglevski et al., Moncada et al.,) in the peripheral blood brought the concept of prostacyclin playing a beneficial antiaggregating and vasodilator role as a circulating hormone which may be responsible for the resting state of platelets under normal conditions. Since then several groups could neither detect circulating PGI_2 nor the stable hydrolysis product, 6-Keto-$PGF_{1\alpha}$ (Siess and Dray, 1982), by direct measurement and it is well accepted that there is no circulating prostacyclin.

PGI_2 as the major Cyclooxygenase product of Endothelial Cells

Isolated human endothelial cells from the umbilical cord have been shown to produce high amounts of PGI_2 contrasting with the small concentrations of $PGF_{2\alpha}$ or PGE_2 (Weksler et al., 1977). However, although this situation seems the most frequent it has then been hypothesized that regional vascular differences may exist and that PGI_2 may not be the unique regulatory PG of vessel function. The findings that bovine endothelial cells in culture (Ingerman - Wojenski et al., 1981) or rabbit intrapulmonary arteries (Salzman et al., 1980) synthesize PGI_2 and TXA_2 may raise pathophysiological arguments concerning a possible heterogeneity of the vasculature.

PROSTANOID DERIVATIVES AS MODULATORS OF THE THROMBOTIC EVENTS

Modification of Platelet Adhesion to Collagen by PGI_2, PGD_2, and PGE_1

After a vascular lesion, adhesion to subendothelium is the primary step of platelets with the different macromolecules exposed to the blood. Several in vitro techniques have been designed to investigate platelet adhesion. The Baumgartner and Muggli technique (1976) using deendothelized rabbit aorta placed in a perfusion chamber through which the blood is passed at different shear rates allows a quantitative estimation of both adhering and subsequently aggregating platelets. Using this technique, we were able to show, in a joint work with the Wellcome group, that PGI_2 could inhibit platelet adhesion to subendothelium although this inhibition was much less potent than that observed on the subsequent formation of thrombi (Higgs et al., 1978). However the interference of plasmatic factors or biorheologic parameter may modify dramatically this primary step. Therefore a quantitative method was developed in our laboratory to estimate specifically the platelet adhesion to collagen without other interference (Legrand et al., 1979). In this situation we could show that inhibition of adhesion to collagent, by PGI_2, was

correlated to the enhancement of the intracellular cAMP level by adenylate cyclase (Karniguian et al., 1982). These effects were also found using PGD_2 and PGE_1 and they were potentiated by the phosphodiesterase inhibitor, theophylline.

On another hand, structural and amino acid sequences have allowed the finding of a collagen-derived peptide which represents the smallest part of the molecule that can interact with platelets, thus providing an inhibitor of collagen-platelet reactions, independent of PG modulation (Legrand et al., 1980). Such approach may be a basis for a development of a novel strategy of antithrombotic drugs.

Platelets-Von Willebrand Factor Interactions in the Presence of PGI_2

It appears that the fixation and attachment of platelets to damaged endothelial cells depends mainly on a complex molecule present in the circulating plasma and platelets as well as in the endothelium or in subendothelial regions, namely the von Willebrand factor. Factor VIII/von Willebrand factor (F VIII/WF) is composed of a pro-coagulant portion and of a ristocetin cofactor activity necessary for the aggregation of normal platelets in the presence of the antibiotic. It has been shown (Moake et al., 1981) that PGI_2 could reverse ristocetin-induced platelet agglutination although it did not suppress the ristocetin induced binding of FVIII/WF to platelets. From these data it was concluded that PGI_2 could suppress platelet-platelet cohesion sites without suppressing the binding of platelets to subendothelial FVIII/WF necessary for hemostasis.

IMPORTANCE OF OTHER BLOOD FACTORS IN THE INTERACTION WITH VASCULAR CELLS

Red Blood Cells

Diabetes mellitus is associated with a high prevalence of microvascular and atherosclerotic disease; it has also been observed that whole-blood viscosity was increased and that the erythrocyte deformability was reduced. Recently, the adhesion of erythrocytes from diabetic patients to endothelial cells was studied in our laboratory (Wautier et al., 1981). After labeling of washed erythrocytes with ^{51}Cr, the cells were incubated with confluent endothelial cells cultures from umbilical veins. After sequential washings, it was found that the percentage of adhering red cells from diabetic patients was higher than when they were from controls ($P<0.005$). These results could be correlated to a vascular score which suggested that in diabetes an intrinsic erythrocyte abnormality could be related to vascular diseases.

Von Willebrand Factor

Recently there have been increasing indications that after an endothelial injury, platelets and lipid entry in the arterial wall may be involved in the early stages of atherosclerosis. A recent important finding (by Ross and Vogel, 1978) is that a substance derived from platelets would be responsible for the smooth muscle cell proliferation of the atherosclerotic lesion. Fuster et al., (1978) using homozygotes von Willebrand pigs lacking this factor, have shown that such animals have a marked impairment in platelet attachment to the vascular wall. These pigs are resistant to the initiation and progression or atherosclerosis either spontaneous or induced by a mildly high-cholesterol diet. This contrasted with the heterozygous animals with milder impaired platelet function which were less resistant to atherosclerosis.

Leukocytes

In contrast to our increasing understanding of the mechanisms of platelet adherence, the specific factors that favor leukocytes adherence to the injured vascular wall are poorly understood although they may be quite important. We studied the effect of thrombocytopenia in rats, using an antiplatelet serum (Kovacs and Caen, 1979): examination of the micro-circulation exhibited a striking increase in the number of marginating, i.e. rolling, leukocytes. Such an increased adhesion of leukocytes to endothelial cells could have some relevance in the development of the ensuing microvascular lesions. Since this work was done, the discovery of leukotrienes (Samuelsson et al., 1979) and their potent biological activities (increased adhesion of leukocytes to endothelial cells, changes in vascular permeability) may bring a new insight in leukocyte - endothelial cells - interactions. In a recent work with Borgeat (Maclouf et al., 1982) we were able to show that platelets and leukocytes may cooperate for leukotrienes biosynthesis. We could show that platelet-derived lipoxygenase product, 12-HPETE, could initiate or amplify the leukotriene biosynthesis thus providing a new example of blood cells cooperativity.

CONCLUSIONS

In summary, it is quite clear that we have only surveyed part of the complex factors that play a role in arterial thrombosis. The brilliant, though so far incomplete PGI_2-TXA_2 balance hypothesis as governing platelet-vessel wall interactions has had the advantage of stimulating efforts in the cardiovascular research in biochemical, pathophysiological or pharmacological approaches. The cyclooxygenase metabolites are undoubtedly involved in the modulation of the normal homeostatic function of platelet-vessel wall interaction either by

cAMP mediation or via the modulation of receptor sites of plasmatic macro-molecules such as fibrinogen or the Von Willebrand factor. Other cells such as red blood cells or leukocytes may also be of the upmost importance in the genesis of atherosclerotic disease. Blood cells cooperation such as platelet leukocyte interactions mediated by the non cyclooxygenase pathway, i.e. lipoxygenases, may also contribute to a more complex although more realistic picture of multifactorial causes of arterial thrombosis.

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SYMPOSIA

INFLUENCE OF DIFFERENT FACTORS ON MILD HYPERTENSION

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Mild hypertension (MH) should be considered according to two equally important aspects of arterial hypertension. One of them is directly associated with treatment of patients. Another aspect relates to the pathophysiological mechanisms of the disease and factors responsible for the process of subsequent evolution (progressing and regression of the disease). These two important aspects are determined by heterogeneity of cases which is reflected in clinical forms of the course of the disease with their own distinctive features, extent of involvement into the pathological process of vitally important organs, risk of complications, differences in prognosis.

The significance of the MH problem is especially well demonstrated in studies based on long-term observations (Veteran Administration Cooperative Study, 1972; US Public Health Service Hospitals, 1972; Kannel et al., 1980; Helgeland, 1981; Morgan, Meyers, 1981; etc.).

The main findings arising from these studies are the following.

It has been ascertained that moderately elevated level of blood pressure (BP) in adults is the most common medical problem in industrially developed countries (Epstein, 1979). Mild hypertension is heterogeneous by its course and prognosis. The latter depends not only on the degree of BP elevation, but rather on the range of BP fluctuations, age and previous injury of some vitally important organs (heart, brain, kidneys), on different risk factors (Veteran Administration Cooperative Study, 1972; Kannel et al., 1980). A very important conclusion has been drawn that some types of hypotensive therapy can differently influence the course and prognosis of the disease.

The results of these studies demonstrate the necessity of selection of MH patients for treatment and more careful choice of therapy. For example, myocardial infarction and sudden coronary death occurred more frequently in elderly patients (older than 65) with mild hypertension who were treated by diuretics (Morgan, Meyers, 1981). And it was indicated that during treatment by diuretics the development of hypertriglyceridemia and hyperuricemia was possible which could increase the risk of coronary complications. Taking into consideration that these patients were elderly it cannot be excluded that moderate BP elevation characteristic of patients with mild hypertension reflects a certain degree of adaptation of circulation system to the initial (or preexistent) organ disorders of atherosclerotic genesis. Due to this BP decrease could increase the vulnerability of patients to cardiovascular complications.

On the basis of the analysis of the data obtained in various studies on evaluation of the effectiveness of the long-term treatment of patients with mild hypertension, Alderman (1980) defines three groups of patients proceeding not from BP indices but rather from clinical characteristics, response to treatment and prognosis: group 1 - with no risk of sudden death and other complications, group 2 - with risk of complications and no results of treatment, group 3 - with risk of complications which can be modified and eliminated by active treatment. It is not excluded that these groups of patients have differences in the nature of the disease.

It is known that differentiation of patients on the basis of renin-sodium profile suggested by Laragh actually correlates with the extent of risk of complications (myocardial infarction, brain stroke) which has been shown in studies by Brunner et al. (1972).

From the presented data it is obvious that on the basis of the analysis of the results of long-term observations for MH patients it is possible to evaluate the natural course of the disease, to determine what external and internal factors can influence the disease including action of different drugs, to understand better the nature of the disease. In this connection it is reasonable to present the results of the studies performed at the A. L. Myasnikov Institute of Cardiology which demonstrate the effect of disorders of the heart, vascular wall, kidneys and excessive salt content in food on the course of mild hypertension.

We will consider consecutively the available data.

1. We carried out 3-5 year observations for 163 patients aged 25-54 with labile (LH) and stable (SH) hypertension without clinical signs of IHD and heart failure. In these patients we re-studied central hemodynamics and anatomo-functional state of the heart by echocardiography. It was found out that without regular treatment in 10% of patients with LH BP level came to normal, in 36% of patients progressing of the disease occurred, and in 54% the course of the

disease did not change. At the same time in 25% of patients with LH reverse development of left ventricular hypertrophy (LVH) occurred, i.e. left-ventricular myocardial mass (LVMM) decreased > 10 g. In 42% of patients we observed increase in LVH extent, mainly at the expense of increase of interventricular septum thickness (T_{ivs}), in 33% of patients LVMM did not change significantly.

In 60% of SH patients we observed LVH aggravation, in 33% - no changes and only in rare cases - LVH decrease. As for the course of the disease in these groups of patients, progressing signs were noted in 49%, the course did not change in 44% and improvement was observed in some patients.

The presented observations indicate that there was no direct parallel link between the dynamics of the disease course and changes in direction and manifestation of LVH. Thus, the known conclusion has been confirmed according to which LVH development in hypertension is not limited to the direct association with elevated BP, but has a more complicated mechanism (Tarazi and others). And changes in LVH extent occur more often and are found earlier than changes in the course of the disease. Nevertheless, the performed studies revealed the direct correlation between LVMM value and increase of systolic BP ($r = +0.58$, $p < 0.001$ - in LH; $r = +0.71$, $p < 0.001$ - SH). In cases of reverse LVH development in LH patients there were found its distinct correlations with decrease of the total peripheral vascular resistance ($r = +0.57$, $p < 0.05$). These findings are in agreement with literature data indicating relationship of LVH development with increase of systolic BP and LVH regression with initially lower diastolic BP (Inrahim et al., 1981). Besides the mentioned above data, the results on changes of some indices of intracardiac hemodynamics are also very important. Increase of end diastolic volume (EDV) and end diastolic size (EDS) of the left ventricle were observed initially, i.e. at the first examination, in cases of reverse development and also in progressing of LVH in patients with hyperdynamic type of circulation. LVH regression was characterized by subsequent increase of volume indices of the left ventricle in diastole. As known, EDV increase testifies to increased venous flow which in hypertension, especially at early stage in elevated cardiac output, was reported by many authors and associated with increase of central cardiopulmonary blood volume as a result of re-distribution of blood from periphery to the center due to venoconstriction. There are reasons to believe that at early stages of the disease the indicated hemodynamic shifts have a functional basis which determines predisposition of these patients to either progressing or reverse development of hypertension. It should be emphasized that subsequent development of the disease is not always combined with increase of LVH extent (as also, by the way, normalization of BP with LVH regression), but further LVH increase (according to LVMM, T_{ivs}) usually indicates to the progressing of the disease and this index should be taken into consideration in evaluation of the dynamics of the disease course.

It is a matter of principle to have objective criteria which could be used for prognosis of possible LVH evolution in arterial hypertension, especially in application of different hypotensive drugs which, as known from literature, are effective differently in respect of reverse LVH development (Tarazi et al., 1979). In this connection there should be mentioned the data obtained by our researchers. These findings testify to the direct and close correlation between LVMM changes and the values of intramyocardial stress of left ventricular wall (6). This has been well demonstrated in cases of reverse LVH development in patients treated by diuretics, beta-adrenal blocking agents (E. G. Diyakova, A. P. Yurenev), when decrease of 6_{max} value by more than 60% of the initial index preceded LVMM decrease. These observations are in an agreement with the data obtained by Ibrahim et al. (1981) on a higher dependence of LVMM on changes in the value of LV intramyocardial stress compared to other factors.

Taking into account the results of the effectiveness of long-term treatment with some hypotensive drugs (diuretics, in particular), a special attention should be paid to functional consequences of LVMM decrease and other shifts in the organic and systemic levels in decrease and normalization of previously elevated blood pressure. From this point of view the presented above data on the significance of studies of the value and dynamics of 6_{max} changes of left ventricle in treatment with hypotensive drugs are very important.

Other practically important data have been obtained by our researchers (A. A. Klembovsky, A. P. Yurenev). It was found that BP decrease by more than 25% of the initial level in patients with marked arterial hypertension and LVH under the effect of i/v injection of sodium diazoxide leads to decrease of myocardial contractile function.

2. Morphofunctional changes of vascular wall should be considered among other factors which are associated with manifestation and duration of arterial hypertension, on the one hand, and degree and rate of LVH development and other vascular and organic lesions, on the other hand.

Vascular system and heart in anatomo-functional relation represent a common system together with the mechanisms of their regulation, therefore it can be assumed that structural, in particular hypertrophic changes developing in hypertension are not limited to the heart but also spread to vascular wall. In fact, in studies of SH patients our researchers (V. V. Paniflov, N. N. Usubaliev) found close correlation between LVH (according to LVMM determined by echocardiography) and value of minimal resistance (R_{min}) reflecting ability of vessels to dilation in reactive hyperemia in response to limb ischemia performed by the method of venous occlusive plethysmography with tensio-transmitter. Thus, for the first time it was shown that LVH development and structural changes of vascular wall could occur simultaneously.

This means that LVMM determination to a certain extent can be used as an index of manifestation of hypertrophic changes of vascular wall in SH patients. At the same time indices characterizing anatomo-functional state of peripheral vessels restudied in dynamics can be also used in evaluation of the degree of progressing or possibility of reverse development of hypertension.

As for patients with mild hypertension, the obtained data indicate that in normokinetic type of hemodynamics there is statistically significant increase of vessel resistance in maximal vasodilation, i.e., signs of structurally adaptive changes of resistive vessels. In contrast, such changes were not observed in hyperkinetic type. It is not excluded that the revealed regularities can be in a certain extent connected with the nature of subsequent course of the disease. It is likely that patients with marked hypertrophic changes of the heart and vessels are prone to progressing of hypertrophy. However, long-term observations for the circulation dynamics are necessary to confirm this assumption.

We have gained some experience of dynamic studies in treatment with diuretics (Usubaliev N. N.) We have studied patients with mild (BP_d < 105 mm Hg) and stage II B (BP_d > 105 mm HG) hypertension aged 28-50. In these patients we determined peripheral blood flow and regional resistance of crus vessels by venous occlusive plethysmography.

According to the results of the study improvement of peripheral hemodynamics was observed only in SH (BP_d > 105 mm Hg) and in good hypotensive effect of short-term therapy (2 weeks) by furosemide and hypothiazide. This was expressed in decrease of vascular tension, significant R_{min} decrease indirectly indicating increase of vessel lumen resistance. Nevertheless, in spite of R_{min} decrease, i.e. increased ability to vasodilation, its values were higher than in the control.

It is true that decrease of vessel resistance at rest and maximal dilation under the effect of diuretics reflects increase of arteriole lumen due to normalization of water-electrolytic content of vascular wall and decrease of its swelling (Tobian, 1952). However, maintenance of higher R_{min} as compared to the control in patients treated with diuretics indicates the presence of structural changes of vascular wall of other nature not connected with water-saline disturbances.

3. Renal mechanisms play a key role in the development of a number of forms of arterial hypertension and in pathogenesis of hypertension, in particular. According to numerous literature data the kidneys and pathophysiological mechanisms connected with them widely participate in the onset, development and progressing hypertension.

We will discuss here only a number of questions directly related to pathogenesis and clinical aspects of mild hypertension.

we present data obtained in clinico-morphological study of kidneys in hypertensive patients. 42 patients (26 women and 16 men, aged from 17 to 44) with mild hypertension (stage II A according to A. L. Myasnikov classification) were studied. All these patients underwent percutenaous renal biopsy. The bioscopic material was studied by light-optical and electron microscopic analysis. The patients were followed-up for 3-5 years. Patients whose history data included diseases which could cause renal changes were excluded from the study. In all patients hypertension was diagnosed. The duration of the disease varied from 2 to 6 years and was not accompanied by urinary syndrome and any disturbances in renal function.

In all 42 patients bioscopic material was heterogeneous. In 9 cases it revealed a healthy kidney; in 19 patients we found insignificant changes such as roughness of mesangium in some glomeruli, swelling of epithelium of proximal tubules in unchanged JGA, arteriole hyalinosis with stenosis of their lumen at some points. In 1/3 of cases (14 patients) we detected marked changes: some glomeruli had initial elements of sclerosing, other glomeruli had partial hyalinosis with replacement of one or several loops by hyaline, dystrophic changes of tubule apparatus, and in some cases small foci of interstitial sclerosis. In patients of this group we revealed sclerosing of renal vessels with thickening of intima of arteries of medium and small sizes, hyperelastosis, some lumen stenosis and initial arteriole hyalinosis. 6 out of 14 patients had morphological signs of JGA hyperfunction which was judged by hypertrophy and hyperplasia of JGA, hypergranulation of epithelioid cells.

Out of 42 cases 27 patients were followed-up for 5 years in an out-patient clinic and were not treated regularly.

It can be seen that out of 5 patients with initially normal kidneys in 4 cases AH course did not change, and 1 patient had further development of hypertension.

Out of 11 patients with initially insignificant renal changes in 5 cases the course of the disease did not change, and 6 patients had stabilization and progressing of AH. During all the period of observation the patients did not have changes in urine, renal function was within normal levels, there were no complications in the brain and heart. Progressing of the disease was determined only according to BP indices and nature of AH course.

Fundamental differences in the course of the disease were found in patients of group 3 with initially more marked morphological changes: distant results were obtained in 11 patients (7 patients did not have regular treatment, and 4 patients had regular treatment). It is essential that out of all 11 patients in 10 cases we observed AH stabilization at a higher level (BP_d > 110 mm Hg) and signs of progressing (increase of resistance to hypotensive therapy in the absence of urinary syndrome). Especially distinct picture of progressing was observed in cases of interstitial sclerosis.

Thus, it can be concluded that in patients with mild hypertension real danger of further development, and in some cases progressing of the disease, can occur in the presence of structural changes in glomeruli, tubules, renal vessels of medium and small sizes. And these patients do not have changes in urine, and irregular treatment by hypotensive drugs is of low effectiveness. The obtained data testify to the possibility of inclusion of renal factor in the process of formation of patients not only as functional shifts of renal blood flow, but also as structural changes of renal vessels. It cannot be excluded that in 1/3 of patients with morphologically marked JGA activation renin-angiotensin mechanism plays a certain role in progressing of the disease.

Not repeating well-known reports (Tobian, Laragh and others) on the important etiological and pathogenetic role of excessive salt content in food in arterial hypertension, we present summarized clinical data obtained in joint studies by researchers of A. L. Myasnikov Institute of Cardiology (A. A. Nekrasova, Yu. I. Suvorov, I. F. Patrusheva, S. E. Ustinova, N. A. Chernova) and Uzhgorod University (M. I. Fatula). We studied population of one of the rural regions of the Transcarpathians where NaCl content in water is from 2 to 5 times higher than in normal drinking water. Due to reduction of sensitivity of gustatory receptors to salt, people of this region usually add higher amounts of salt in food, consuming 400 mEq Na daily.

Among these people we observed elevation of BP > 160/95 mm Hg in 13.8%, in the boundary zone it was found in 13.2%, while these values were 3.4 and 7.4%, respectively, in the control group (normal salt consumption).

The population of this region was followed-up for 20 years. Studied subjects had excessive salt consumption and the first examination showed labile arterial hypertension > 160/95 mm Hg. These studies demonstrated that normalization of BP occurred in 7%, improvement - in 5.4%, progressing of the disease with stabilization and further elevation of arterial hypertension - in 10%. In 77% cases the course of the disease remained the same. Patients of the control group with labile elevation of BP > 160/95 mm Hg, but with normal salt consumption (< 180 mEq per day) had different dynamics of the disease: BP normalization - in 19.7%, improvement - 13.4%. progressing of the disease - in 2.6%, and the course did not change in 64%.

Thus, it was demonstrated that excessive salt consumption had unfavourable effect on hypertension course.

In the process of this study we revealed some peculiarities of systemic dynamics in excessive salt consumption, i.e. higher indices of total peripheral resistance as compared to the controls, decrease of cardiac output and increase of the volume of circulating blood.

Besides hemodynamic changes, prolonged excessive salt consumption causes changes in the functional state of renin-angiotensin-aldosterone system (RAAS). Such changes can even occur in healthy subjects subjected to chronic salt overloading and are aggravated in hypertensive patients with excessive salt consumption. This is expressed in the absence of decrease of plasma renin activity (PRA) and aldosterone concentration in blood plasma (ACBP) in high salt overloading or tendency to their increase in patients with labile (mild) hypertension, significant increase of PRA (instead of its decrease) by more than twice of the initial value in stable and high hypertension (BP_d > 110 mm Hg). Complete dissociation in PRA and ACBP reactions is observed at different loadings in stable hypertension.

The indicated above changes of hemodynamics and RAAS in hypertensive patients in prolonged excessive salt consumption can, evidently, determine some clinical peculiarities of hypertension course in these cases, in particular predisposition to sodium retention, high total peripheral resistance and possibility for further progressing of the disease.

Studies based on the evaluation of water and sodium transport by the kidneys in hypertensive patients are very important taking into account the effect of salt consumption on the course of mild hypertension. According to the studies performed by our researchers, patients with labile hypertension have three types of reactions on salt loading. Two types of them (II and III) reflect disorders in regulation of sodium and water transport processes and are characterized in one subgroup of patients (II) by excessive natriuresis and diuresis (phenomenon of exaggerated natriuresis) with reduction of renal ability to concentrate urine on the 1st and 2nd days after salt loading, and in contrast, in another subgroup (III) they are characterized by significant decrease of sodium excretion in small urine volume of high concentration (osmomolarity).

It is assumed that in the first case the observed effect is associated not with structural changes of tubules, but with inadequate reaction of hypothalamo-hypophysial system on salt loading as insufficient ADH release in blood. In the second case such effect is, probably, connected with primary disorders of renal function regulation leading to increase of sodium reabsorption in proximal tubules or ascending part of Henle's loop. It is likely that these patients are especially sensitive to salt overloading which promotes not only further development but also progressing of the disease. Similar to reactions observed in experiments in rats with spontaneous hypertension (Yu. V. Postnov et al.), patients with this type of reaction on salt loading can be considered as subjects with hereditary predisposition to increased sensitivity to sodium. Thus, we assume that on the basis of response to salt loading in mild hypertensive patients it is possible to define two subgroups (2/3 of all patients with labile hypertension) of patients with functional disorders of sodium and water transport by the kidneys.

The presented data let us make some conclusions which may serve as a basis in discussion of questions related to nature, course, therapy and prognosis of mild hypertension.

1. Mild hypertension is not homogeneous by biochemical (hormonal) and functional (hemodynamic) indices, by extent of involvement of organs (heart, kidneys) into the process, response to treatment and by rates and direction of disease evolution.

2. Early involvement of the heart and kidneys in pathological process is prognostically significant for further development and possible progressing of the disease. This includes LVH and IVS combined with changes of cardiac volume indices per diastole, and different degrees of structural changes of glomerular and tubular apparatus of the kidneys, primary hyalinosis of renal arterioles, hyperplasia and hypertrophy of JGA, elements of interstitial sclerosis (by renal biopsy data).

3. Significance of chronic and prolonged salt overloadings is associated with existence of sensitive and insensitive groups of patients to sodium chloride which can be identified by nature and degree of diuresis and natriuresis in response to salt loadings.

4. In order to find methods for higher effectiveness of treatment and prevention of possible complications it is necessary to conduct long-term observations for patients with mild hypertension and define different clinical groups according to the course of the disease.

EPIDEMIOLOGY AND NATURAL PROCESS

OF MILD HYPERTENSION

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INTRODUCTION

Most studies on the pathophysiology of essential hypertension have suffered from being done in selected subjects referred to hospitals or hypertension clinics. In 1963/64 a screening of the blood pressure (BP) in the Bergen population gave us the opportunity to start a long-term study of the natural process of mild hypertension in a small group of patients recruited from an epidemiological survey.

The BP was screened in 77% of the total adult population (92.000) and in 98.1% of a 10% random sample[1,2]. All subjects with diastolic BP above 120 mmHg and a randomly selected group with moderate and mild hypertension (diastolic 90-120 mmHg) were called for a diagnostic follow-up. Male subjects who at this follow-up and at one additional examination had mild or moderate previously untreated essential hypertension and no other diseases (obese subjects were excluded) were candidates for a study of the circulatory disturbances.

CENTRAL HEMODYNAMICS - CROSS SECTIONAL STUDIES

Central hemodynamics were studied at rest and during exercise in 77 hypertensives 17-66 years old and in 48 age matched normotensive controls[3]. In the youngest age group, 17-29 years, with a mean value of BP at rest sitting of 150/92 mmHg, the cardiac index (CI) was 13% higher ($p<0.05$) than in the age matched controls. The heart rate (HR) was 16% higher ($p<0.01$), and the stroke index (SI) was not significantly different. The total peripheral resistance index (TPRI) was not significantly different. In the hypertensives 30-39 years with BP 160/99 mmHg, the SI was subnormal, but since HR was

increased, the CI was normal. The TPRI was significantly increased. In hypertensives in their 40-ties and 50-ties the cardiac index and stroke index were subnormal and the TPRI increased. Similar results have been found in studies from different parts of the world (reviews in 4, 5).

The high CI and HR in subjects supposed to be in the early phase of essential hypertension formed the concept of a hyperkinetic circulation with a "luxury" perfusion of the tissues at this stage of the disease. To protect the tissues from overirrigation the arteriolar resistance was then expected to increase and thus reduce the blood flow. The hypertension would then be maintained by the increase in TPRI (the "whole body autoregulation" theory). However, an argument against this theory is that our study as well as three others demonstrated an increased oxygen consumption (VO_2) (about 12%) in the young mildly hypertensive subjects, and the CI was not increased when related to the metabolic demands[5]. In other words, no overirrigation in the tissues seemed to take place and the whole body autoregulation theory can not explain the increased TPRI in established essential hypertension.

The cause of the high CI, HR and VO_2 in early essential hypertension is not known, but an overactivity in the sympathetic nervous system is usually postulated but difficult to demonstrate[6].

Studies in the spontaneously hypertensive rats (SHR), the animal model with many similarities to human essential hypertension, have shown that even in young, mildly hypertensive animals structural changes appear very soon in the heart and in the resistance vessels[7]. Based on our studies of hemodynamics during rest only, it might seem that the heart pump function and the arterioles are not affected in early human hypertension. However, studies performed during physical exercise change this picture. Even during mild exercise (50 Watt) the young hypertensives are definitely no longer hyperkinetic. The CI related to VO_2 is then subnormal. The HR is still increased but the SI is then subnormal. Furthermore TPRI does not fall to the same low levels as in normotensive controls. The exercise studies seem to indicate that structural changes are present in mild hypertension already in the third decade and recently a reduced compliance in the left ventricle has been demonstrated at an early stage of essential hypertension[8].

CENTRAL HEMODYNAMICS - PROSPECTIVE STUDIES

The results from cross-sectional studies should indicate that the circulatory system in essential hypertension would change from a "high-flow low-resistance" system in the early phase to a "low-flow high-resistance" system in the later. Our group of patients from 1964/65 who had been studied hemodynamically have now been

followed for 17 years. After 10 years clinical data were available in 75 of the 77 subjects and 33 subjects who had been untreated, were restudied hemodynamically by the same methods as 10 years before. After 17 years clinical data were available in 75 and a third hemodynamic study in those still untreated have started. All living subjects have remained hypertensive over these 17 years.

10-year follow-up. In the two youngest age groups treatment had been started in four subjects while 28 were untreated, apparently healthy, and they were restudied hemodynamically. (One subject died from heart and lung insufficiency). In age group III (40-49 years) only 5 patients were untreated. All patients in the oldest group (50-66 years) started treatment and 12 of 16 had died after 10 years.

In age group I (17-29 years) the VO_2 during rest had decreased 8% ($p<0.05$) but was almost unchanged in group II. The BP at rest showed remarkably few changes. Only during 150 W exercise was there a significant increase in MAP from 133.1 to 141.0 mmHg (6%) in age group I, and from 140.7 to 147.3 mmHg (5%) in age group II. In spite of the small changes in BP the CI at rest had fallen in both age groups, 15% and 23%, respectively. During 150 W load the mean values had decreased about 15% in each age group. In both age groups the reduction in CI was associated with a significant decrease in SI of about 8-12% at rest and during exercise. The HR during rest and exercise showed only small changes. TPRI had increased significantly during rest sitting, the increase was about 25-30% in both age groups. Figure 1 shows the changes in age group 17-29 years. The small group of patients 40-49 years demonstrated the same changes as the two younger age groups, but the increase in TPRI and BP was much greater.

Thus the long-term study demonstrated that the expected increase in TPRI had indeed occurred, although the increase in BP was relatively modest.

17-year follow-up. For ethical reasons treatment has been started when diastolic BP has increased to > 100 mmHg. After the 10-year restudy and over the next 7 years the blood pressure has increased to such levels in most subjects and after 17 years only 6 out of 36 subjects in age group I and II are still untreated. Preliminary results from the third hemodynamic restudy in untreated subjects seem to indicate further decline in cardiac index and increase in total peripheral resistance.

CONCLUSION

The present and a few other long-term studies in untreated subjects with essential hypertension have demonstrated that mild hypertension is usually a progressive disorder with slowly developing pathological changes in the heart pump and in the resistance vessels,

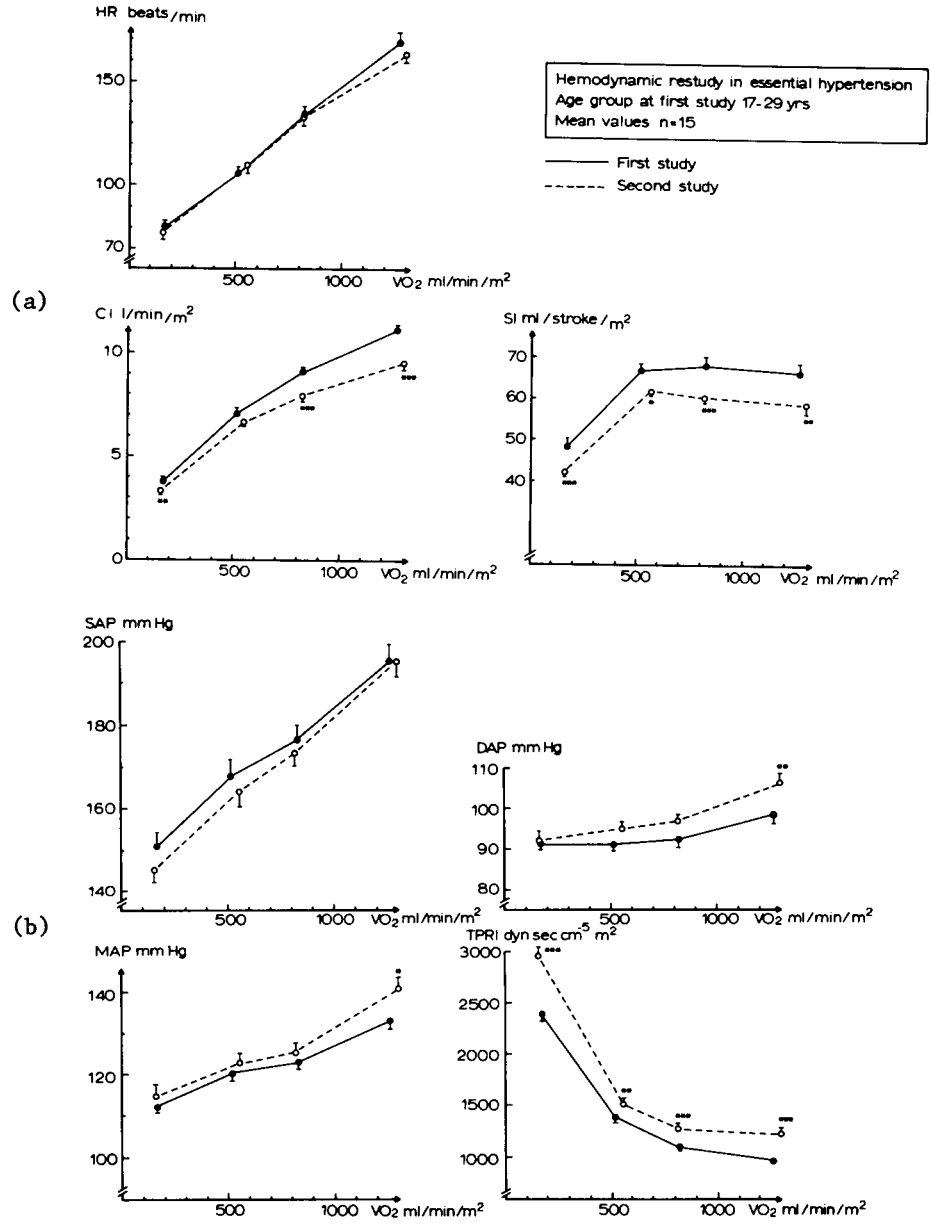

Fig. 1(a). Heart rate (HR), cardiac index (CI) and stroke index (SI) in age group 17-29 years. Mean values and SEM. *p<0.05, **p<0.01, ***p<0.001.

Fig. 1(b). Systolic (SAP), diastolic (DAP) and mean arterial pressure (MAP) and total peripheral resistance index (TPRI) in age group 17-29 years. Mean values and SEM. *p<0.05, **p<0.01, ***p<0.001.

although the increase in BP is usually not impressive in subjects in their 20-ties and 30-ties during the first 10 years. From then on increase in BP seems to be more common. Thus mild hypertension (defined as BP during rest 140/90 - 170/105 mmHg on several occasions, weeks apart) is perhaps a more important threat to the cardiovascular system than what is commonly believed. Our presently available antihypertensive agents reduce blood pressure through quite different mechanisms resulting in widely different hemodynamic patterns[9]. From a hemodynamic point of view, an antihypertensive agent, devoid of side-effects, correcting the hemodynamic disturbance would seem to be the logic approach to treatment of mild hypertension.

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MILD HYPERTENSION : TREATMENT AND PROGNOSIS

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Two major studies on the effects of antihypertensive drug treatment in mild hypertension have recently been published. Both have demonstrated that patients with mild hypertension treated with antihypertensive drugs have a lower death rate and fewer vascular complications than patients either untreated or less vigorously treated.

The High Blood Pressure Detection and Follow-up Program[1] conducted a large study in several centers in the United States of America. Patients with diastolic blood pressures above 90 mmHg were admitted to the study. No patients were excluded because of pre-existing vascular or associated disease. Patients were randomly allocated to one of two groups. One of these, the "stepped care" group were seen regularly in special treatment clinics and given free aggressive anti hypertensive therapy. The other group, the "referred care" group were referred back to their own doctors, who may or may not have treated their hypertension. The mortability in the stepped care group both from cardiovascular and non-cardiovascular causes was lower than in the referred care. The reduction in all cause mortality was larger in blacks than whites. The reduction in mortality in the mild group, with diastolic pressures 90-105 mmHg was 13 per 1,000 persons over five years. In the HDFP reduction in stroke incidence was more obvious than reduction in ischaemic heart disease.

The Australian Therapeutic Trial in Mild Hypertension[2] differed in several important respects from the HDFP. Entry diastolic pressures were 95-109, using the average of six readings. The control group received matching placebos and both groups attended the same special clinics. Persons with evidence of previous stroke, myocardial

infarction, renal failure, diabetes or angina were excluded from the study. In spite of the differences in design the Australian Study and the HDFP came to broadly similar conclusions. Mortality from cardiovascular disease was reduced in the Australian Study by 2 per 1,000 persons per annum, and morbidity, mainly from stroke, by 5 per 1,000 persons per annum. The major end points in both groups were due to ischaemic heart disease, and in this category there were 33 myocardial infarctions in each group. The incidence of other manifestations of ischaemic heart disease was similar in both groups.

The total incidence of trial end points in the placebo treated group was low. This seems to have been due to three factors. Firstly, patients with pre-existing evidence of vascular complications were excluded. Secondly in the placebo group 198 patients (12%) whose diatolic pressures consistently rose above 110 mmHg were given active treatment, but remained in their original randomization to the placebo group. Finally, in all the patients receiving placebos, diastolic pressures fell to levels below 95 mmHg.

Analysis of the relationship between the average levels of blood pressure achieved during the period of observation showed that in both the actively treated and placebo groups increasing levels of diastolic blood pressure was associated with a higher rate of trial end points. At the very lowest diastolic pressures the rates in the active and placebo groups was almost identical. However, 87% of the actively treated group had average diastolic pressures below 94 mmHg, whereas 55% of the placebo group were in this category. At higher average levels of diastolic pressure, the rate of trial end point occurrence was higher in the actively treated than in the placebo treated group, suggesting that if blood pressure is not well controlled, the administration of antihypertensive drugs may be associated with an additional risk, presumably related to the drugs themselves. An alternative explanation is that the small group of mild hypertensives apparently resistant to the antihypertensive effects of drugs may constitute a small sub group with a worse prognosis than the average patient.

In the placebo group as a whole there was a fall of diastolic pressure over a period of three years. Over half the placebo group had diastolic pressures below 99 mmHg at three years, and 12% of the group developed pressures persistently above 110 mmHg. On the average those whose pressures fell below 95 mmHg had lower than average pressures originally, while those whose pressures rose above mild limits had higher than average initial pressures. No characteristics emerged, however, which allowed a prediction to be made in an individual person as to whether the blood pressure would rise or fall.

SUMMARY AND CONCLUSIONS

1. Mild hypertension can be defined as the presence of diastolic blood pressures consistently between 95-109 mmHg.
2. In persons with mild hypertension prognosis is considerably better in those with no evidence of cardiovascular disease; in this group mortality is approximately 2 per 1,000 per annum. In persons with mild hypertension and pre-existing cardiovascular complications, mortality is much higher.
3. Ischaemic heart disease, which is the commonest complication of mild hypertension, is not significantly influenced by anti-hypertensive drug treatment.
4. The incidence of stroke is significantly reduced in treated mild hypertension.
5. A prolonged observation period is helpful in deciding which patients to treat. In over half the patients initially found to be mildly hypertensive, blood pressure falls to levels at which treatment provides no benefit.

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ANTIHYPERTENSIVE THERAPY IN PATIENTS ABOVE AGE 60: A report of the European Working Party on High Blood Pressure in the Elderly (EWPHE)

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The following centers are collaborating in the EWPHE:

1. University Hospital St. Raphael, Leuven, Belgium.
2. Zuiderziekenhuis, Rotterdam, The Netherlands.
3. University Hospital, Gent, Belgium.
4. Geriatric Hospital Le Valdor, Liège, Belgium.
5. Hammersmith Hospital, London, United Kingdom.
6. University Hospital St. Luc, Brussels, Belgium.
7. Medisch Centrum voor Huisartsen, Leuven, Belgium.
8. Hôpital Charles Foix, Ivry, France.
9. University Hospital Santa Maria, Lisboa, Portugal.
10. St. John's Hospital, London, United Kingdom.
11. North Karelia Project, Kuopio, Finland.
12. Istituto di Ricerche Cardiovascolari, Milano, Italy.
13. University Hospital Haukeland, Bergen, Norway.
14. Victoria Geriatric Unit, Glasgow, Scotland.
15. University Hospital Köln, West Germany.
16. St. Charles Hospital, London, United Kingdom.
17. Royal College of Surgeons, Dublin, Ireland.
18. Aberdeen Royal Infirmary, Aberdeen, Scotland.

Seven hundred and ninety two hypertensive patients above the age of 60 have entered the double blind multicenter trial of the European Working Party on High blood pressure in the Elderly (EWPHE). Half were treated with one capsule daily containing 25 mg

hydrochlorothiazide and 50 mg triamterene and half were given placebo. If blood pressure control was not adequate in those receiving active treatment, a second capsule was given and if necessary up to 2 g of methyldopa/day.

No significant differences between the groups were present prior to randomization. A significant blood pressure difference of 25/10 mmHg was obtained between the groups and maintained during five years of follow-up. No major disturbances in serum potassium or serum sodium were noted.

On the other hand, during the initial phase an increase in serum creatinine and serum uric acid was noted in the actively treated group, which was maintained during the later years. This increase in serum creatinine was related to the decrease in sitting systolic blood pressure. Also, changes in serum uric acid correlated with changes in serum creatinine both in the placebo and in the actively treated group, but were independent of the change in creatinine; the serum uric acid was on average 1 mg higher in the actively treated than in the placebo group.

Fasting blood glucose did not change significantly in the placebo treated group, but it did so in the active treatment group.

A favorable influence on prognosis by active treatment can be expected on the basis of the blood pressure reduction and in the absence of major electrolytes disturbances. However, the balance between this decreased risk and the increase produced by the rise in blood glucose and the other treatment effects remains to be determined. The trial continues and more patients are being admitted.

THE CLINICAL RELEVANCE OF PARTIAL AGONIST ACTIVITY OF BETA-ADRENOCEPTOR BLOCKING DRUGS

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Introduction

Beta-adrenoceptor blocking drugs are often considered first choice therapy in patients with mild hypertension who require medical therapy. This is due not only to their effectiveness but also to the low incidence of side-effects during long-term administration.

There is a general consensus that it is β-adrenoceptor blockade which is responsible for their therapeutic effectiveness in the treatment of hypertension as well as angina pectoris (Simpson and Waal-Manning, 1970; Prichard, 1974). Of the different ancillary properties, membrane stabilizing action seems to be of no practical relevance, whereas clinical advantages have been attributed to partial agonism - also called intrinsic sympathomimetic activity or ISA - and to β_1-adrenoceptor selectivity.

Partial Agonist Activity

The potential advantages of partial agonist activity long remained a matter of debate and only in recent years have the advantages theoretically to be expected been confirmed in the clinical situation.

A β-blocker with partial agonist activity occupies β-adrenoceptors in the same way as a drug without this property and effectively prevents the access of stimulatory agonists in a competitive manner. In addition it provides some stimulation to the receptors. The clinical pharmacological peculiarities of a β-adrenoceptor blocking drug with partial agonist activity have recently been reviewed (Aellig, 1982a).

Beta-adrenoceptor blocking drugs with partial agonist activity

Acebutolol
Penbutolol
Alprenolol
Oxprenolol
Practolol
Bopindolol
Pindolol

Fig. 1.

The stimulant action of the β-adrenoceptor blocking drugs listed in Figure 1 is either smaller than that provided by normal resting sympathetic activity (this applies to acebutolol, penbutolol, alprenolol, oxprenolol and practolol) or just about as high as resting sympathetic activity (bopindolol and pindolol). The stimulant action therefore totally or partly compensates for the loss of resting sympathetic drive resulting from β-adrenoceptor blockade. The effects of increased sympathetic stimulation during physical or mental stress, however, are reduced to the same extent as by drugs devoid of stimulant activity. One of the most obvious differences is therefore that drugs without ISA nearly always reduce resting heart rate - even if initial values are very low - whereas the effect of drugs with ISA depends on pre-existing sympathetic activity.

Heart Rate at Rest

An example for the correlation between the effect of a β-adrenoceptor blocking drug with partial agonist activity and resting

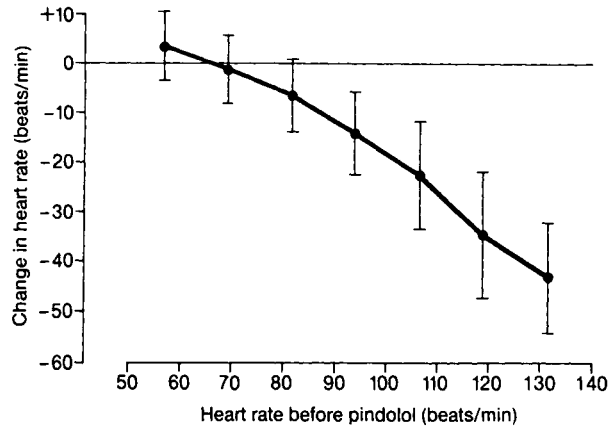

Fig. 2. Changes in resting heart rate, related to pre-treatment values in patients with essential hypertension during oral therapy (mean ± s.e.). Reproduced from Rosenthal et al., 1979, with kind permission of British Journal of Clinical Practitioner.

heart rate is shown in Figure 2. In a multicenter study Rosenthal et al. (1979) studied over 7,000 hypertensive patients treated with pindolol. In patients with pre-treatment resting heart rates of about 70 beats/min pindolol produced no net changes. The ISA of the drug therefore just about replaced the effect of resting sympathetic activity. The higher the pre-treatment heart rate, however, the greater was its reduction. When initial heart rate was low, the ISA of pindolol led to a slight increase.

The maximum stimulant activity of the partial agonists currently used is already reached with low therapeutic doses and is independent of the dose over a range wider than that generally used in clinical practice. Carruthers and Twum-Barima (1981) studied oral doses of pindolol from 2.5 to 57.5 mg, and found no difference in the effects on resting heart rate.

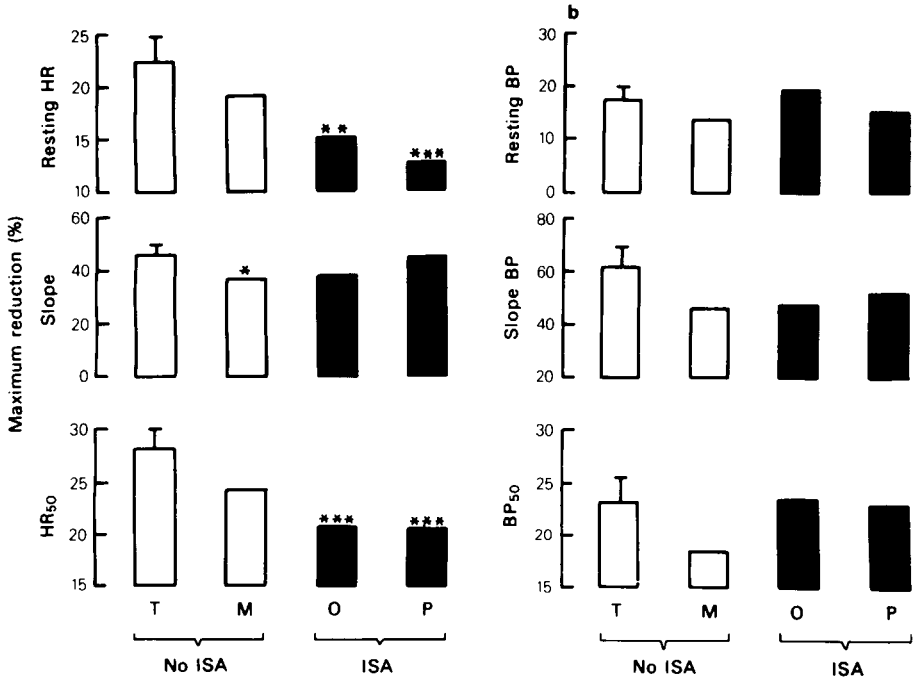

Fig. 3. Maximum reduction in resting heart rate and blood pressure, in the slope of the heart rate and blood pressure rise during exercise and in the heart rate and blood pressure observed during 50% of working capacity after oral administration of timolol (T), metoprolol (M), oxprenolol (O) and pindolol (P). Error bars represent the standard error of the difference between any two columns (analysis of variance). **P < 0.01, ***P < 0.001 for difference from timolol. Reproduced from Jennings et al., 1981, with kind permission of the British Journal of Clinical Pharmacology.

Exercise-induced Tachycardia

ß-adrenoceptor blocking drugs with partial agonist activity inhibit the effect of beta-adrenoceptor stimulation on the heart to the same extent as drugs devoid of this property. Tachycardia due to exercise or to infused isoprenaline is therefore reduced in a dose-dependent manner (Aellig, 1982b). Four drugs with and without ISA were compared by Jennings et al. (1981). Several doses were administered and the results in Figure 3 show the maximum effects reached with each drug. As expected resting heart rate was more reduced by timolol and metoprolol - drugs without ISA - than by oxprenolol and pindolol - drugs with ISA, whereas resting blood pressure fell to the same extent after all drugs. The reduction of the blood pressure and heart rate rise during exercise was identical after all compounds, whether they possessed ISA or not.

Erikssen et al. (1982) exercised 10 healthy subjects with increasing work loads before - and 24h after one week of oral treatment

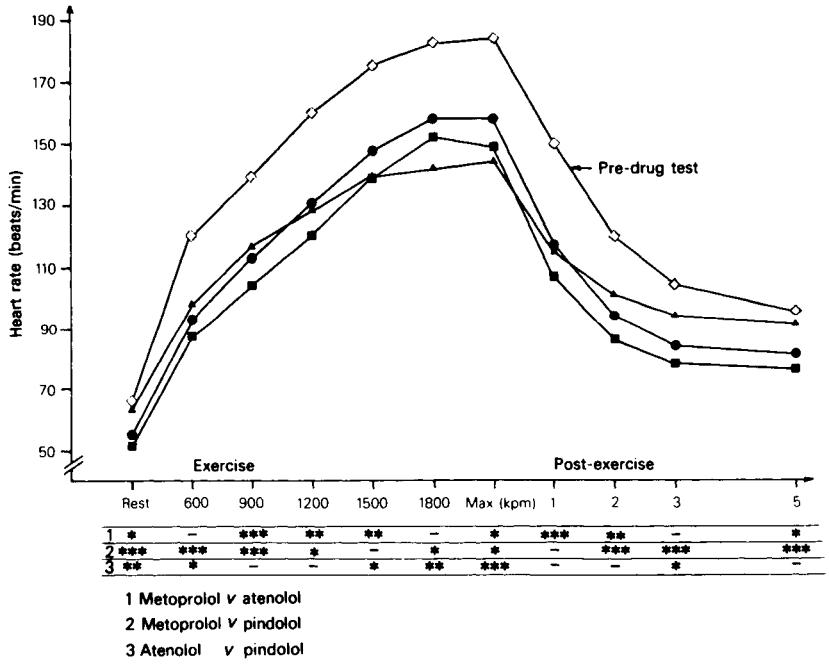

Fig. 4. Effects of metoprolol 300 mg/d (■), atenolol 100 mg/d (●) and pindolol 15 mg/d (▲) on heart rate before, during, and after exercise measured 24h after 6 days of therapy. *P < 0.05, **P < 0.01, ***P < 0.001. Reproduced from Erikssen et al., 1982, with kind permission of the British Journal of Clinical Pharmacology.

(Figure 4). Metoprolol (300 mg/d) and atenolol (100 mg/d) reduced resting heart rate from about 65 to about 55 beats/min whereas pindolol (15 mg/d) was practically without effect. During exercise, however, the increase in heart rate was reduced after all three β-adrenoceptor blocking drugs and, in the doses used in this experiment which represent frequently used therapeutic doses, heart rate during maximum exercise was even lower after pindolol than after metoprolol and atenolol.

Thus, β-adrenoceptor blocking drugs with ISA reduce the effects of increased sympathetic activity as effectively as drugs lacking this property, but they do not interfere with cardiac function at rest.

Peripheral Resistance in Acute Studies

If resting heart rate is unchanged or only slightly reduced then cardiac output will be unchanged or only slightly lowered. As a result one would expect drugs with a clinically relevant ISA to produce little change in peripheral resistance.

In an experiment in 6 healthy volunteers blood flow in the lower extremities was measured using venous occlusion plethysmograph (Aellig, 1979). The results are summarized in Figure 5. After placebo blood flow was somewhat reduced during the 4 hours of observation. Blood flow values after 10 mg pindolol were practically identical to those after placebo. After 160 mg propranolol,

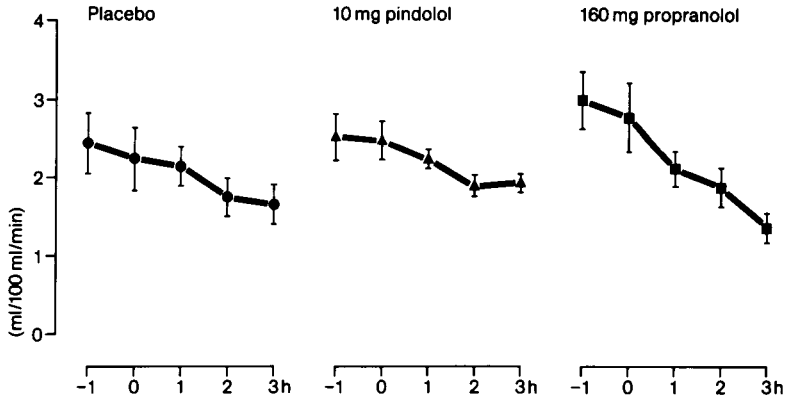

Fig. 5. Blood flow in the calf before and after oral administration of placebo, 10 mg pindolol or 160 mg propranolol to 6 healthy volunteers (mean ± s.e.). Data from Aellig, W. H., 3. Basel Hypertoni Symposiet, Stockholm, 1979, ed. L. Hansson and O. Thulesius.

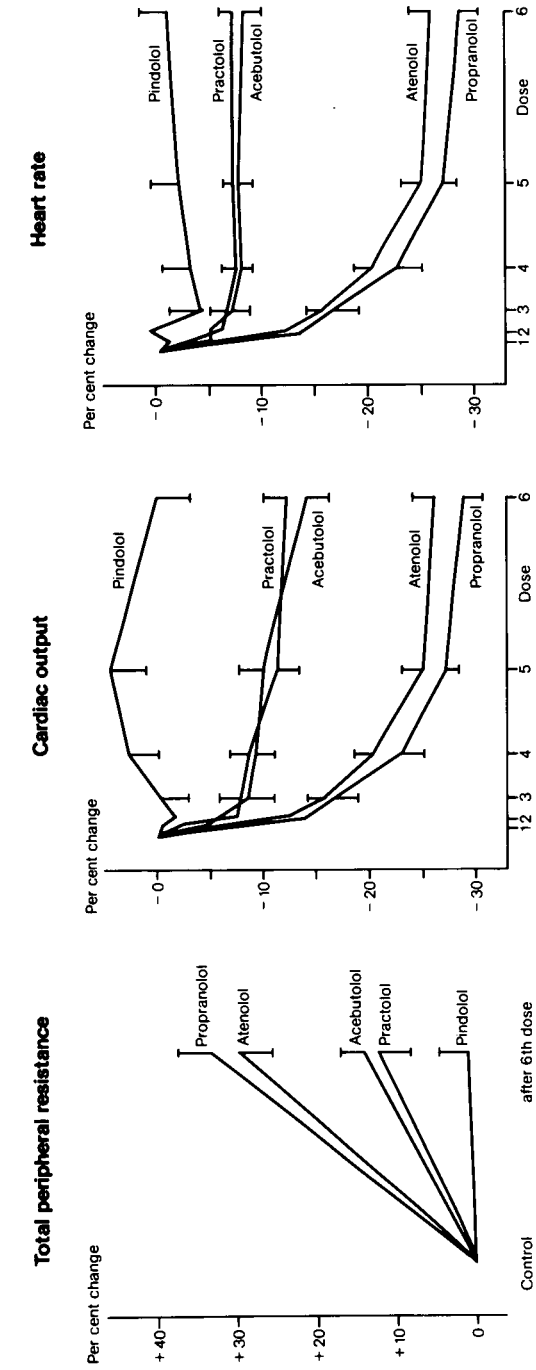

Fig. 6. Effects of different β-adrenoceptor blocking drugs on total peripheral resistance, cardiac output, and heart rate after intravenous administration to patients with angina pectoris.
After Svendsen et al., 1981, with kind permission of Clinical Pharmacology and Therapeutics.

however, the reduction in peripheral blood flow was significantly greater than after either placebo or pindolol.

Svendsen et al. compared the acute haemodynamic effects of β-adrenoceptor blocking drugs with and without ISA or with β_1-selectivity in healthy volunteers (Svendsen et al., 1979) as well as in patients with angina pectoris (Svendsen et al., 1981). The results of the latter study are summarized in Figure 6. As expected, resting heart rate is only slightly influenced after pindolol but markedly reduced with propranolol and atenolol, drugs without ISA; practolol and acebutolol lie in between. Cardiac output is not reduced with pindolol and falls most markedly with propranolol and atenolol. Peripheral resistance, however, increased most markedly after propranolol and atenolol. The results with atenolol show that β_1-adrenoceptor selectivity does not protect against the increase in total peripheral resistance which is secondary to the reduction in cardiac output. Peripheral resistance was less influenced by practolol and acebutolol - which possess ISA - and remained unaltered after pindolol. Haemodynamic effects similar to those obtained in this study in man were reported by Clark et al. (1982) in dogs.

Peripheral Resistance During Treatment of Essential Hypertension

In the treatment of hypertension it is of course important to study the haemodynamic effects of a drug also during chronic oral therapy. Man in't Veld and Schalekamp (1982) analysed the literature on the acute and long-term haemodynamic effects of β-adrenoceptor blocking drugs in patients with essential hypertension. After acute administration blood pressure remained practically unaltered and drugs without ISA (timolol, atenolol, propranolol and metoprolol) markedly reduced cardiac output and increased peripheral resistance. Pindolol, with a marked ISA, had practically no influence on both parameters. Penbutolol, acebutolol, oxprenolol, practolol and alprenolol, with less ISA, showed an intermediate effect (Figure 7).

During chronic therapy, when blood pressure is reduced, at first sight the figure looks very similar (Figure 8). All peripheral resistance values, however, are now lower than in the acute situation. Timolol, propranolol, metoprolol and atenolol, which reduce cardiac output, still elevated peripheral resistance but less so than after acute administration. Pindolol and practolol, which did not reduce cardiac output, now exhibited a clear reduction in peripheral resistance below pre-treatment values. The drugs with a smaller degree of ISA showed intermediate effects.

During chronic therapy therefore β-adrenoceptor blocking drugs with ISA lower resistance to pre-treatment values or in the case of practolol and pindolol even to values clearly below pre-treatment

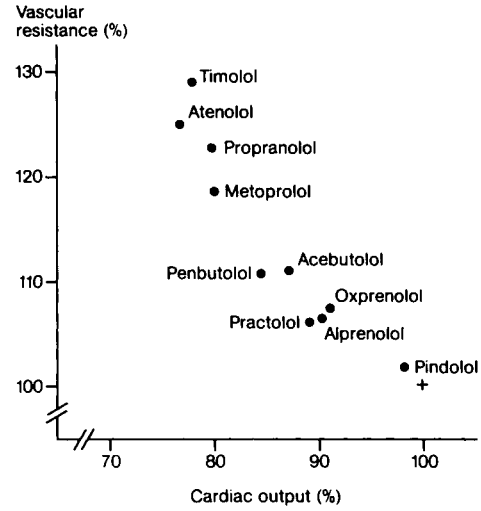

Fig. 7. Acute haemodynamic effects of β-adrenoceptor blocking drugs in relation to pre-treatment values (indicated by the black cross). Reproduced from Man in't Veld and Schalekamp, 1982, with kind permission of the British Journal of Clinical Pharmacology.

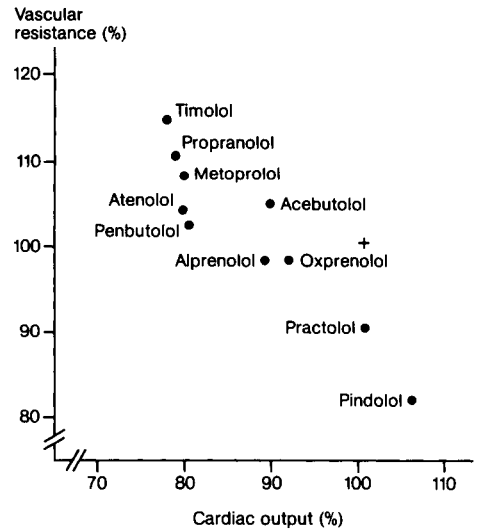

Fig. 8. Long-term haemodynamic effects of β-adrenoceptor blocking drugs in relation to pre-treatment values (indicated by the black cross). Reproduced from Man in't Veld and Schalekamp, 1982, with kind permission of the British Journal of Clinical Pharmacology.

values. These differences are of potential clinical importance and drugs with a clinically relevant ISA appear to offer haemodynamic advantages in the treatment of hypertension.

Other Aspects

Other advantages have been attributed to ISA, for example that drugs with ISA do not unfavourably alter the ratio between HDL and LDL cholesterol (Pasotti et al., 1982; Leren et al., 1982; Lehtonen et al., 1982), that they are less likely to cause bronchoconstriction in susceptible patients (Louis and McNeil, 1982) and that they are less likely to give rise to a rebound phenomenon on sudden withdrawal (Walden et al., 1982; Rangno et al., 1982, Szecsi et al., 1982, Prichard, 1982).

An impairement of glucose metabolism in some insulin-dependent diabetics was reported during treatment with a β-adrenoceptor blocking drug (Waal-Manning, 1979). This effect was smaller with β_1-selective drugs. In order to establish whether the presence or

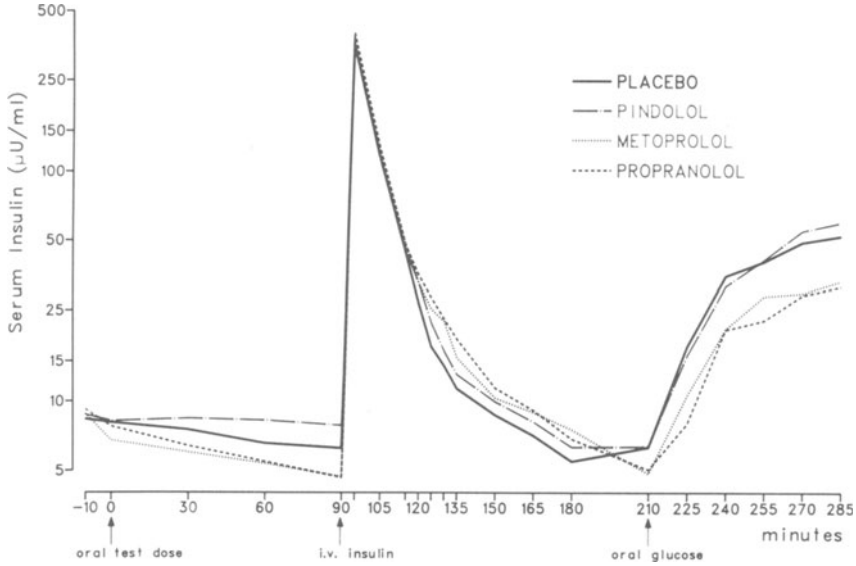

Fig. 9. Effects of an oral dose of placebo, pindolol (15 mg), metoprolol (200 mg) or propranolol (160 mg), during hypoglycaemia induced by insulin (0.1 U/kg body weight) and after an oral glucose load (100g) on serum insulin concentrations in volunteers (n = 8). Mean ± s.e. mean results. Reproduced from Schlüter et al., 1982, with kind permission of the British Journal of Clinical Pharmacology.

absence of ISA is relevant in this respect, Schlüter et al. (1982) administered oral doses of placebo, 15 mg pindolol, 200 mg metoprolol and 160 mg propranolol to 8 healthy volunteers. 90 min later insulin (0.1 U/kg body weight) was given intravenously. After identical serum insulin peaks, insulin levels underwent a slower decline after β-adrenoceptor blocking drugs (Figure 9). The effect was greatest after propranolol, smaller but significant after metoprolol (the relatively β_1-selective drug) and only slight and not significant after pindolol (the drug with ISA). When an oral glucose load was given two hours after insulin the rise of serum insulin was significantly delayed and reduced after propranolol and metoprolol, but not after pindolol.

The adrenaline peak during hypoglycaemia (Figure 10) was more than doubled after propranolol, less increased after metoprolol but not influenced after pindolol. These experimental studies in healthy subjects require confirmation in diabetic patients.

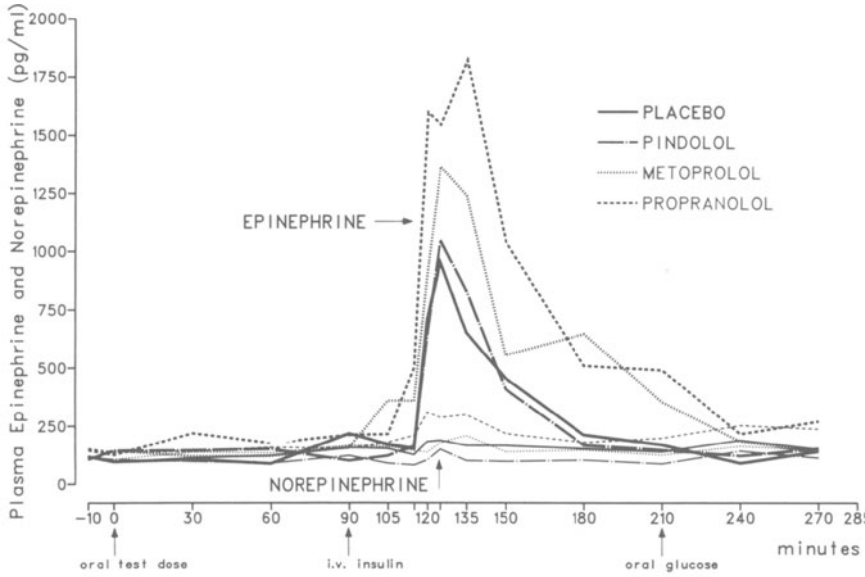

Fig. 10. Effects of an oral dose of placebo, pindolol (15 mg), metoprolol (200 mg), or propranolol (160 mg) during hypoglycaemia induced by insulin (0.1 U/kg body weight) and after an oral glucose load (100g) on plasma adrenaline (epinephrine) and noradrenaline (norepinephrine) concentrations in volunteers (n = 8). Mean ± s.e. mean results. Reproduced from Schlüter et al., 1982, with kind permission of the British Journal of Clinical Pharmacology.

Conclusions

The results of the different clinical and clinical pharmacological studies reported here indicate that a β-adrenoceptor blocking drug with a clinically relevant ISA may have a series of therapeutically important advantages in the treatment of hypertension.

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THE WITHDRAWAL OF BETA ADRENERGIC BLOCKING DRUGS

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INTRODUCTION

It was found fairly soon after the introduction of beta adrenergic blocking drugs for the treatment of angina pectoris that when they were stopped abruptly an exacerbation of ischaemic symptoms sometimes occurred to above pre-beta blockade levels. The first report was in a trial of oxprenolol in angina where seven cases of status anginosus occurred out of nineteen patient in the trial when placebo was substituted for active treatment (Wilson et. al. 1969). Two cases of severe ischaemia were reported in a trial of propranol when placebo was substituted in twenty patients with angina. One patient required hospital admission for acute coronary insufficiency and one patient died from coronary occlusion the day after the propranolol (40 mg qds) was stopped (Zoster and Beanlands, 1969). In a multidose study of propranolol in angina pectoris the patients experienced a higher incidence of chest pains during the first week of placebo compared with the second week (Prichard and Gillam, 1971). Slome (1973) reported two cases of myocardial infarction that occurred when propranolol was stopped in patients with angina pectoris. Alderman et. al. (1974) described six cases of severe ischaemia in angina patients when propranolol (80 or 160 mg/day) was stopped. All of them experienced unstable angina, three had infarcts one of whom died; there was one case of sudden death, and one other developed multiple ventricular ectopics.

CLINICAL STUDIES WITH PROPRANOLOL

Miller et al., (1975a) reported a series of twenty patients with stable angina where there were two ischaemic deaths, one sudden death, one acute anterior infarct with shock, and four other serious reactions, three cases of intermediate coronary syndrome and one ventricular tachycardia when propranolol was stopped. These patients had received propranolol for a total period of twelve weeks; 160 mg daily for six weeks and 320 mg a day for the final period. There were four additional patients who experienced increased angina attacks on placebo compared to prior to propranolol administration. These ten patients had significantly more severe angina (as suggested by more angina attacks per week and trinitrin consumption) than the remaining ten who had no excessive exacerbation of the manifestation of ischaemia after propranolol was stopped. On the other hand they had not observed any episodes in a "large number" of "symptomatic coronary patients" who had propranolol stopped abruptly prior to cardiac catheterization. This might be taken to suggest that it was the limitation of physical activity that provided protection against serious exacerbations of the disease on drug withdrawal. In a retrospective study in a series of patients with angina, propranolol, average 149.2 mg (range 40-320 mg) was abruptly stopped for coronary arteriography on fifty three occasions in fifty one patients (Myers and Wisenberg, 1977). These patients included fourteen with single vessel disease but with greater than 70% stenosis, the rest had significant narrowing, ie. over 50% of two or three vessels and all but four at least 70% stenosis. There were two patients who had experienced unstable angina on propranolol, one had a recurrence off propranolol which responded to restarting propranolol, the other patient, however, experienced a fatal myocardial infarction ten days after stopping propranolol. Again, as was the case in the report of Miller et al., (1975a), these patients with more severe disease appeared to be at greater risk. Mizgala and Counsell (1976) reported fifteen acute coronary events in fourteen patients with severe angina when propranolol was stopped, six transmural infections, three intramural infarctions with one ventricular fibrillation and six episodes of acute coronary insufficiency.

Myers et al., (1979) reported a series of one hundred patients with angina admitted to hospital for coronary arteriography. Propranolol was abruptly stopped after an average duration of treatment of 8.2 months and at an average dose of 216 mg. Three patients had minor increases in chest pain and two patients had non-transmural infarctions prior to the time propranolol was stopped; and the same number of minor and major episodes occurred after the cessation of the drug. The occurrence of ischaemic episodes was not related to stopping propranolol but to the severity of the disease; all four patients who developed non-transmural infarction had Class IV New York Heart Association symptoms. In another survey, fifty five patients who had propranolol, average dose 127 mg (range 20-320 mg) stopped prior to cardiac catheterization revealed only one patient who

experienced any increase in pain. Another forty seven patients continued on propranolol, average 143 mg (range 80-320 mg). The overall incidence of chest pain in the two groups while they were in hospital was 27% in those who stopped propranolol, and 28% in those continuing propranolol. However, there was one patient in the group which stopped propranolol who had a ventricular dysrhythmia and myocardial infarction, but this was following selective coronary artery dye injection. The authors concluded that propranolol withdrawal syndrome is infrequent in hospital patients (Shiroff et al., 1978). In a study of blood pressure and incidence of atrial arrhythmias in patients undergoing coronary bypass surgery (Kadish et al., 1979), it was found that stopping propranolol 48 hours or 10 hours preoperatively was associated with a greater rise in blood pressure during intubation and a higher incidence of post-operative atrial arrhythmias than in patients who had half of their usual dose of propranolol on the morning of operation and 1 mg iv qds in the intensive care unit.

WITHDRAWAL OF OTHER BETA BLOCKING DRUGS

The withdrawal of propranolol and metoprolol in twenty patients with angina pectoris resulted in one patient experiencing a fatal infarct when placebo was substituted for metoprolol and in the same circumstances another patient experienced severe angina (Frich and Luurila, 1976). Meinertz et al., (1979) also reported a case of myocardial infarction in an anginal patient when metoprolol was stopped; however, this coincided with the commencement of nifedipine treatment which may have contributed as its administration can be associated with a tachycardia (Beeley and Talbot, 1979).

THE EFFECT OF GRADUAL WITHDRAWAL OF BETA ADRENERGIC BLOCKING DRUGS

The withdrawal of propranolol which had been given for over three months, over six days in one patient and nine days in two patients was still associated with increase in sensitivity to isoprenaline (Nattel and Rangno, 1978). However when fifteen hypertensive patients given propranolol 80 mg qds for over a month were studied, the six who had the dose reduced gradually to 10 mg qds for fourteen days had no evidence of increase in sensitivity to isoprenaline or increase in blood pressure or plasma catecholamines compared to the ultimate stable levels in contrast to the nine patients in whom propranolol was abruptly stopped (Rangno and Nattel, 1980). Similarly when metoprolol 300 mg/day was first reduced to 25 mg bd for ten days before the drug was stopped only minimal sensitivity to isoprenaline was seen (Rangno et al., 1982).

TIMING OF THE WITHDRAWAL PHENOMENON

There is some difficulty in being certain of the incidence of delayed reactions after beta blockade withdrawal as ischaemic events may occur due to reasons other than the recent administration of beta blocking drugs. The timing however, seems to vary considerably from one to fourteen (Miller et al., 1975; Mizgala and Counsell, 1976) or possibly twenty one days (Alderman et al., 1974), after the cessation of beta blocking treatment.

WITHDRAWAL OF BETA BLOCKING DRUGS IN NON-ISCHAEMIC PATIENTS

Goldberg et al., (1977) performed continuous intra-arterial monitoring studies that suggested that withdrawal of beta blocking drugs (oxprenolol in seven patients, propranolol in one) in hypertensive patients was not associated with an overshoot of blood pressure in contrast to clonidine. Other investigators have also found no overshoot of blood pressure when propranolol was stopped (Vlachakis and Aledort, 1980). However, hypertensive patients may complain of palpitations, tremor, sweating and similar symptoms after the stopping of beta blocking drugs metoprolol and propranolol (Pedersen et al., 1979). Williams et al., (1979) have also described a hypertensive patient who experienced palpitations associated with a tachycardia and marked ST segment depression when metoprolol was stopped.

MECHANISM OF BETA ADRENERGIC WITHDRAWAL PHENOMENON

There have been several suggested mechanisms to explain the withdrawal phenomenon.

EFFECT OF BETA ADRENERGIC RESPONSES

Boudoulas et al., (1977) observed an increase in sensitivity of heart rate responses to isoprenaline infusions 24-48 hours following the end of two days of administration of propranolol at a dosage of 40 mg qds to six normal volunteers aged 20-24. It was suggested that the propranolol withdrawal phenomenon was the consequence of the formation of additional beta receptors during propranolol therapy which persisted on propranolol withdrawal for a short period leading to augmented sympathetic responsiveness. Similarly the administration of propranolol average 240 mg/day (range 160-320 mg) was stopped in nine hypertensive patients who had been on treatment for at least three months, an increased responsiveness to isoprenaline began 2-6 days later lasting for 3-13 days after propranolol was withdrawn (Nattel et al., 1979). The average bolus dose of isoprenaline required to produce an increase in heart rate of 25 beats/minute six days after was 1.2 μg while it was an average of 2.3 μg after responses were

stabilized by day 14. In another investigation with the cardioselective agent metoprolol (300 mg/day) in eight hypertensives for over six weeks a 52% average increase in cardiac sensitivity to isoprenaline was seen 2-8 days after withdrawal (Rangno et al., 1982). In seven other patients when metoprolol dosage was first reduced to 25 mg bd only minimal hypersensitivity was seen.

In our studies in a between subject study in normal volunteers, we titrated doses of propranolol (final average dose 614 mg/day) atenolol (634 mg/day) and pindolol (18 mg/day), with drug administration for a total of about three weeks so that maximum inhibition of exercise tachycardia was obtained. Isoprenaline hypersensitivity was seen after atenolol (n=6) at some time in the withdrawal phase, with measurements at 1 day and then alternate days until Day 13 post drug. Hypersensitivity was observed with two of the six subjects after propranolol but not after pindolol in any of the volunteers (Walden et. al., 1982). In another study Rangno and Langlois (1982) found a steady restoration of isoprenaline sensitivity in ten hypertensive subjects with no overshoot after pindolol was stopped. Likewise after oxprenolol (160-320 mg given for 8-18 months) was withdrawn in six hypertensive patients the sensitivity to isoprenaline gradually increased with observations at 1, 2, 3, 6 and 13 days post drug, there was no overshoot with hypersensitivity of heart rate responses (Bolli et al., 1981).

Goldstein et al., (1981) studied normal volunteers (n=14) that were given propranolol for 24 days in a dose (80-240 mg) sufficient to reduce the exercising heart rate at 160 watts by 20 beats/minute. Studies were performed 1, 2 and between 6-9 days after stopping propranolol, no increase in isoprenaline sensitivity was seen. These investigators gave infusions of isoprenaline, 4 minutes at each dose level, sufficient to increase heart rate by 20 beats/minute over preinfusion levels or to give a rate of 120 beats/minute. Lindenfeld et al., (1980) administered infusions of adrenaline to ten normal subjects and ten patients with angina; in neither group was there an increase in the heart rate and systolic blood pressure responses 4 days after two weeks of administration of propranolol 160 mg a day compared to control, or at least 6 days in the angina group that was studied for a longer period.

Nattel et al., (1979) did not find any overshoot of heart rate measured at rest. Likewise Lindenfeld et al., (1980) and Goldstein et al., (1981) found no overshoot of supine heart rate after propranolol was stopped. Ross et al., (1981) also observed no overshoot of heart rate at rest supine or standing when propranolol 160 mg one day (80 mg bd) was given for one week (n=6) or six weeks (n=12) to normal volunteers. On the other hand, Pedersen et al., (1979) found a significant overshoot of standing heart rate with levels of 95 at 48 hours after the last dose falling to 89 on the seventh day after metoprolol (150-300 mg a day, n=5) or propranolol (160 mg daily, n=1).

Goldstein et al., (1981) observed an overshoot at higher levels of sympathetic activity. After tilt following stopping propranolol, an increase of heart rate of 6 beats over levels prior to propranolol was observed ($p<0.05$), following tilt plus the administration of atropine (0.8 mg iv), an increase of 8 beats/minute compared to before propranolol ($p<0.02$). There is some difficulty with these results as the subjects were in-patients (see below). When increased sympathetic activity was induced by vasodilation from 1 mg sublingual glyceryl trinitrate an enhanced tachycardia was seen in the immediate withdrawal phase before heart rates stabilized. There also appeared to be an enhancement of the tachycardia of Valsalva's manoeuvre in the withdrawal phase after six weeks propranolol (Ross et al., 1981). Other experiments suggested four days propranolol administration was not long enough for the withdrawal overshoot of post vasodilation tachycardia (n=6). However, although there was no peak followed by a decline after propranolol was stopped, in contrast to the other groups, the post vasodilation heart rate remained high (120/min) compared to the post six week propranolol group (100/min); control heart rates before beta blocking drug administration were not reported. Similar overshoot of the post vasodilation heart rate was seen in normal volunteers after atenolol 200 mg/day for six weeks (n=6), oxprenolol 160 mg/day for one week (n=6) and acebutalol 400 mg/day for one week (n=6). Overshoot of the post vasodilation heart rate was also seen in hyperthyroid patients with propranolol 160 mg/day for six weeks (n=6), and oxprenolol 160 mg/day for six weeks (n=6) (Ross et al., 1981).

Jackson et al., (1979) found a significant overshoot of exercise systolic blood pressure after propranolol (80 mg qds for one week) at 42 and 54 hours post drug, and after atenolol (100 mg daily for 1 week) at 66 hours, but no overshoot of heart rate was found. Goldstein et al., (1981) observed that the heart rate at 160 watts of exercise averaged 165 beats/minute, a rise of 12 over control ($p<0.02$) 48 hours after withdrawal of propranolol 80-240 mg daily. These results are difficult to interpret as the subjects were not trained, one baseline exercise being performed, one at 4 days or more on drug, one 2-7 days before propranolol was stopped, besides 1, 2 and 7 days after drug. Propranolol was given for 24-79 days, average 36 days. Throughout the subjects were in-patient, and although it is stated they maintained physical activity, being an in-patient represents the possibility of change of activity level, thus a 'negative' training effect seems possible. A higher exercising heart rate than control might be expected therefore once beta receptor occupation from propranolol had completely declined, ie. at Day 2, when overshoot was reported. Rangno and Langlois (1982) observed a tachycardia of exercise of 158 the second day after stopping pindolol (10 mg bd for one month in ten hypertensives), whereas the rate at Day 20 was 144. However, the probability of a training effect makes these results difficult to interpret as no readings were obtained until the day pindolol was stopped. However others have not

found an overshoot of the exercise tachycardia. It was not seen after propranolol (160 mg/day) in ten normals with observations made at 2 and 4 days post drug, or in ten patients with angina at 2, 4 and 6 days post drug (Lindenfeld et al., 1980). Walden et al., (1982) studied trained normals and overshoot was not seen with daily observations for 13 days in the withdrawal phase after propranolol (average 614 mg/day) atenolol (average 634 mg/day) or pindolol (average 18 mg/day).

Experiments in animals have also revealed evidence of increased beta responses after beta blockade withdrawal. Hypersensitivity was seen after withdrawal of propranolol (120 mg qds) in dogs, to the cardiac responses to isoprenaline (Webb et al., 1981). However no evidence of post drug increased beta receptor sensitivity was seen in dogs treated with four weeks oral propranolol at a dosage of 40 mg, 8 hourly (Myers and Horwitz, 1978). Experiments in rats also showed a hypersensitivity to isoprenaline after propranolol withdrawal (Botting and Gibson, 1979). Also in rats hypersensitivity to isoprenaline was seen after the administration of oxprenolol or metoprenolol. This response appeared to be oscillatory; on some days hypersensitivity was seen but not on others (Manning et al., 1981). While Le Roy et al., (1978) did not demonstrate any increase in the amount of left ventricular cyclic AMP levels following the administration of isoprenaline in the mouse from 8-40 hours post propranolol compared to saline, Manning et al., (1980) found an increase in cyclic AMP and dP/dt max in rat hearts after the withdrawal of three weeks oxprenolol or metoprolol. Hypersensitivity in rats to heart rate responses to isoprenaline was confirmed with propranolol but was not found after the administration of atenolol or LL 21,945, a long acting non-selective beta blocking drug (Botting and Crook, 1981). Three to six days following the termination of three weeks oral propranolol to guinea pigs, Dennis et al., (1980) found an increase in incidence of arrhythmias during pre-ischaemic aerobic perfusion; during re-perfusion the incidence of arrhythmias was increased and there was a large rise in the occurrence of ventricular fibrillation.

BETA RECEPTOR POPULATION

A reduction in the number of frog erythrocyte beta receptors has been found after the chronic administration of isoprenaline, but propranolol administration failed to alter their number. However, experiments in rats revealed an increase in beta adrenergic receptor binding sites in ventricular membranes after two weeks administration of propranolol, the receptor assay being made 8 hours after the last dose of propranolol (Glaubier and Lefkowitz, 1977a). The reduction of beta receptor stimulation from the administration of guanethidine or 6-hydroxydopamine was also associated with an increase in beta receptor population (Glaubiger and Lefkowitz, 1977b). Studies with

homogenized whole rat hearts however failed to reveal any increase in receptor population after propranolol administration (Baker and Potter, 1980). Aarons et al., (1980) demonstrated with radioligand studies in man a 43% average increase in beta receptor density in human lymphocytes that reached a maximum after 5 days of propranolol 160 mg daily administration. This increase in receptor population declined to pre-propranolol levels between 4 and 7 days after propranolol was withdrawn. However, while others found an increase in beta receptor population in human leucocytes during the administration of propranolol 240 mg/day for 4 weeks (Fraser and Wood, 1979), because levels fell below control within 2 days of stopping propranolol it was felt by these investigators that it was improbable that a continuing raised beta receptor population was the explanation of the withdrawal phenomenon. Finally, Goldstein et al., (1981) in nine normal subjects found no increase in white cell beta receptor sites or in cyclic adenosine monophosphate produced in response to isoprenaline after propranolol withdrawal. These subjects were in-patients during propranolol administration and possibly relatively inactive; this may therefore have influenced these results (see above).

CATECHOLAMINE LEVELS

Catecholamines do not increase after beta blocker withdrawal. Plasma levels of adrenaline and noradrenaline were not found to increase after withdrawal of metoprolol (five patients), propranolol (one patient) in a group of hypertensives (Pedersen et al., 1979). Maling and Dollery (1979) in fact observed a fall in plasma noradrenaline in five hypertensive patients after propranolol average dose 344 mg (range 240-640 mg) was stopped. Likewise Lindenfield et al., (1980) found a fall in serum adrenaline levels back to control after propranolol 160 mg daily for 14 days was stopped in ten normal volunteers and ten patients with angina pectoris. Propranolol blocked the adrenaline induced increase in free fatty acid levels, which then returned to baseline with no overshoot when propranolol was stopped. Similarly, measurements of both adrenaline and noradrenaline at 48 hours post propranolol (up to 240 mg daily for 6-8 weeks) fell back to control (Vlachakis and Aledort, 1980). No evidence ot overshoot of plasma noradrenaline when propranolol was stopped was seen in seven normals studied by Goldstein et al. (1981), in eight hypertensives after metoprolol (Rangno et al., 1982) or in six hypertensives after oxprenolol Slow Release 160 of 320 mg. (Bolli et al., 1981). Our investigations in normals with propranolol (average 614 mg/day) atenolol (average 634 mg/day) or pindolol (18 mg/day) revealed a fall in serum noradrenaline with drug administration, and no overshoot at 5 or 7 days post drug (Walden et al., 1982). Likewise work in animals has indicated that an overshoot of noradrenaline does not occur when propranolol is stopped (Webb et al., 1981). Vanillylmandelic acid (VMA) excretion was not found to be increased after the administration of propranolol in normal subjects (Pantano and Lee, 1976).

On the other hand, Nadeau et al., (1980) found a 55% ($p<0.02$) increase in urinary adrenaline levels 24 hours after the withdrawal of 4 weeks of propranolol 240 mg/day which returned to normal after 48 hours. The urinary noradrenaline levels showed no overshoot but were increased by 26% during propranolol treatment. Rangno and Nattel (1980) found an overshoot of plasma catecholamines when propranolol (80 mg qds for over one month) was withdrawn.

RENIN LEVELS

Plasma renin levels are reduced by beta adrenergic blocking drugs. No overshoot of renin levels was seen when oxprenolol was withdrawn in six hypertensives by Bolli et al., (1981), or in normal volunteers after atenolol, pindolol or propranolol (Walden et al., 1982).

THYROID HORMONES

Three cases, two hypertensives and one ischaemic patient were reported who developed symptoms after propranolol withdrawal confirmed by laboratory findings of thyrotoxicosis that apparently developed during propranolol treatment (Shenkman et al., 1977). There have been suggestions that increased levels of thyroid hormones may be at least partly responsible for increased beta receptor responsiveness. Free tri-iodothyronine levels were found to be raised in four out of five hypertensive patients who developed tachycardia, sweating, and tremor 2 to 6 days after propranolol (160-480 mg daily for 2 to 18 months) withdrawal, no change in thyroxine or total thyroid hormone levels were found (Kristensen et al., 1978). The increase in free tri-iodothyronine correlated with the serum propranolol concentration on its last day of administration. However, in this study no pre-propranolol or follow up levels of thyroid hormones were reported.

Ross et al (1980) did not observe any change in thyroid hormones in euthyroid subjects after propranolol (160 mg/day for 4 to 8 weeks) was stopped, but an increase in tri-iodothyronine and free tri-iodothyronine though not throxine or free thyroxine levels in hyperthyroid subjects so treated; again hormone levels before propranolol were not quoted. Lindenfeld et al., (1980) did not find any increase in tri-iodothyronine levels after 2 weeks of propranolol 160 mg daily was stopped, free tri-iodothyronine levels were not reported. We observed a fall in serum free T_3 in normals with the administration of atenolol, pindolol and propranolol, with a return to baseline after the drugs were stopped with measurements made 1,3,5,7,9 and 11 days post drug (Walden et al., 1982). Rangno et al., (1982) found no evidence of increased levels of serum T_3 and T_4 after withdrawal of metoprolol (300 mg/day) in eight hypertensive patients; likewise there

was no effect on these hormones after oxprenolol (160-320 mg daily) was withdrawn in six hypertensives (Bolli et al., 1981).

EFFECT OF BETA ADRENERGIC BLOCKING DRUGS ON PLATELET AGGREGATION

Observations in patients with angina pectoris indicate that they have an abnormally low threshold to aggregation to adrenaline or adenosine diphosphate (ADP) compared to age and sex matched normal controls. Frishman et al., (1978) for instance found the average concentration for the biphasic response and maximal platelet aggregation in ten normals at rest was 3.72 μM for ADP and 6.46 for adrenaline. The corresponding figures for twenty patients with angina at rest was 1.55 μM for the ADP and 1.26 μM for adrenaline. When propranolol 160 mg daily for 16 weeks was given to ten of the ischaemi patients, 3.43 μM ADP was then required for aggregation, ie. similar to normal controls, compared to a control level of 1.32 μM ($p<0.01$) in those patients; the results after 50 weeks were similar to those at 16 weeks. After propranolol was stopped only 1.0 μM was required for aggregation 48 hours later, but this was not significantly lower than prior to propranolol. The responses to adrenaline showed a similar pattern, after propranolol 12.9 μM of adrenaline was now required for aggregation in contrast to 1.02 μM before propranolol ($p<0.01$), then two days after propranolol was stopped an average of 0.57 μM adrenaline was required. These results were not significantly different overall from pre-propranolol, but six patients were hyper-aggregable compared to pre-propranolol. The ten ischaemic patients who received placebo showed no change in their abnormally high degree of sensitivity of platelet aggregation to ADP and adrenaline during the period of observation. Studies in hypertensive patients revealed an increase in the threshold to maximum platelet aggregation at rest by adenosine diphosphate from propranolol (6-8 weeks) which declined t control values two days after propranolol was stopped. Values returned to control levels although some patients showed an increased sensitivity compared to control (Vlachakis and Aledort, 1980). Exercise resulted in a decrease in the amount of adenosine diphosphate required for platelet aggregation both before (0.55 μM) and after propranolol (0.25 μM), the differences were not significant. The administration of propranolol 80-240 mg/day for 24 to 79 days in normal subjects has indicated that platelet survival is reduced (7.8 days) during the withdrawal phase compared to control (10 days, $p<0.05$) (Goldstein et al., 1981). Aggregation of platelets liberate the prostaglandin thromboxane A_2 which is a coronary vasoconstrictor (Ellis et al., 1976) this may be a possible basis for a platelet dependent withdrawal phenomenon.

PROGRESSION OF THE DISEASE PROCESS

In some cases the withdrawal of beta blocking drugs after prolonged treatment could lead to the unmasking of underlying progression of the disease process, at least as a contributing factor. In the absence of β-blockade, oxygen supply might not be sufficient to meet the requirements of the heart even at rest (Diaz et al., 1974).

EFFECT OF INTRINSIC SYMPATHOMIMETIC ACTIVITY

The beta adrenergic blocking drugs vary in their pharmacodynamic properties, presence or absence of a number of associated properties, intrinsic sympathomimetic effect, membrane stabilising activity and cardio-selectivity, and this has been used as a basis of classification (Prichard, 1978). It seems possible that post beta blockade sensitivity may be due to the generation of additional beta receptors, thus a beta blocking drug with some partial agonist effect might provide sufficient beta stimulation to prevent the generation of additional beta receptors. There is evidence, as discussed above, with the partial agonist beta blocking drugs pindolol and also oxprenolol, of the absence of post beta blockade hypersensitivity to isoprenaline. However, in the case of oxprenolol, which possesses less partial agonist effect, there have been reports of the withdrawal syndrome in ischaemic patients (Wilson et al., 1969).

CONCLUSION

There is little doubt that the beta blocker withdrawal syndrome is a real phenomenon although the incidence may be low. In some experimental studies this may be explained by the relatively short period of observation. In addition to stopping the beta blocker, exertion may usually be a prerequisite for the development of significant clinical consequences where serious reactions appear to have occurred in patients with more severe ischaemic disease. When it is necessary to stop a beta blocking drug, patients should have the dose reduced gradually and be advised to minimise exertion as the dose is reduced and for two to three weeks after the drug is stopped. It is advisable to maintain the final low dose for at least two weeks.

The mechanism of the withdrawal phenomenon may be the generation of additional beta receptors during the period of beta blockade, thus when the beta blocker is withdrawn the increased beta receptor population readily results in excessive beta stimulation. This will be important when the delivery and use of oxygen is finely balanced, as frequently occurs in ischaemic disease. There is some evidence that the possession of intrinsic sympathetic stimulating properties by a beta blocking drug may no longer mean that additional receptors are formed during the administration of the beta blocking drug.

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THE MANAGEMENT OF HYPERTENSION

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In his stirring remarks at the opening of the IXth World Congress of Cardiology in Moscow Dr Mahler, Director General of the World Health Organization, made some critical comments about high technology medicine. He also made some favorable references to the results of treating hypertension. Many will agree with Dr Mahler, but I see a danger in his criticisms of modern technology, for it confuses ends and means. The end is better prevention and/or treatment of disease by simple, effective and, preferably, cheap methods. Evolution of those methods may require application of the most sophisticated technology. The treatment of hypertension is a case in point. The medicinal chemistry, pharmacology, and clinical research that have led to therapeutic use of antihypertensive drugs involved a great deal of very sophisticated science. The output of that science may have been in medicine that a nurse or medical assistant can use, but if the resources of medical care had all been devoted to training medical auxiliaries there would have been no medicines for them to use.

In the case of hypertension the discovery of effective antihypertensive drugs has redefined the nature of the problem.

Over the past 30 years hypertension has evolved from a medical emergency, patients presenting with left ventricular failure, renal failure or hypertensive encephalopathy, to an exercise in preventive medicine that tests the effectiveness of the medical care delivery system as much as the clinical skill of the doctor. I can illustrate this change by referring to the results of treatment in 660 severely hypertensive patients who began treatment at the Hammersmith Hospital Hypertension Clinic in the 5 year period between 1962 and 1966 (Bulpitt and Dollery unpublished). This was a time when treatment

methods had stabilised after the era of ganglionic blockade. The patients had relatively severe hypertension with a mean pre-treatment diastolic pressure in excess of 120 mmHg. They have now been followed in the Hypertension Clinic for a minimum of 15 years. Those who died have been ascertained by the Office of Population Censuses and Surveys, the organization which maintains the mortality records in the United Kingdom.

The survival of these patients has been remarkably good. In the case of patients who were between 30 and 49 years old at diagnosis, over 75% were still alive after 15 years. Thus these patients fell into the pressure range of the first Veterans' Administration trial. The data show that the favorable effects of treatment can be maintained over very long periods of time. Those who died had on average a higher blood pressure, smoked more cigarettes, had a higher blood urea, were older and included more males than those who lived. There were two noteworthy exceptions to the profile of risk factors. The mean serum cholesterol and body weight were slightly higher in those who lived than in those who died. The survival of black patients was also slightly better than of whites because fewer of them died of myocardial infarction. In the white patients myocardial infarction was the most important cause of cardiovascular death.

This and other studies have enabled us to redefine the problem of hypertension. The acute bursting vascular pathology that leads to cerebral haemorrhage or renal failure is readily controlled or prevented by drug treatment. The strengthening of the heart and blood vessels by hypertrophy of muscle is halted, and to some extent reversed, by treatment. The same cannot be said for fibrous replacement of muscle in blood vessels and the heart. My own studies in the retina and those of Dr Tarazi in the heart suggest that infiltration by collagen is not remodelled during treatment. This had led to the rather fanciful terminology referring to the collagen infiltrated heart of the treated hypertensive as a "beating scar".

The principal problem remaining is that of occlusive, especially atheromatous, vascular disease. Much less progress has been made with this condition and there is a distinct shortage of ideas about how to tackle it. We may hope that earlier treatment of hypertension and more effective antismoking measures may in time make an impact on this problem. We shall need to pay attention to possible adverse effects of drugs upon atheroma as a disease (e.g. the hypoglycaemia and hyperuricaemia caused by thiazide diuretics, and the hyperlipidaemia caused by beta adrenergic blockade).

Finally, a word about the structure of this symposium. We have deliberately not scheduled a standard talk about management of the uncomplicated patient. Step care regimes which include a beta blocker, a diuretic and a vasodilator, have been generally adopted. Whether the beta blocker should be beta-1 selective or non selective,

the diuretic should be potassium conserving or not, and the precise type of vasodilator are a matter of personal preference. No clinical trials have been published which tell us which of these is best in terms of outcome. My own choice currently falls on atenolol as the beta blocker, moduretic as the diuretic, and hydrallazine or minoxidil as the vasodilator. I anticipate that the choice of vasodilator will move towards the calcium antagonists in the future. The role of angiotensin converting inhibitors remains to be defined. They appear to have fewer pharmacodynamic side effects than beta blocking drugs. However, the favorable pharmacodynamic effects of beta adrenergic blockade in patients who have suffered a myocardial infarction may not be shared by the angiotensin converting inhibitors.

For the future some of the most important arguments about the management of hypertension relate to the role of sodium, the use of angiotensin converting enzyme inhibitors, and beta adrenergic blocking drugs. The one aspect of atheromatous disease in which there have been favorable developments is the use of balloon dilatation of renal artery stenosis, and we are pleased to have Dr Heiss to address us on that topic.

SODIUM AND BLOOD PRESSURE IN HUMAN HYPERTENSION

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The role of sodium in the pathogenesis of hypertension is not agreed. It is likely that excess dietary sodium or excessive retention of sodium is capable of raising blood pressure in some circumstances in man. In Conn's syndrome, for example, the body content of sodium is abnormally increased, the increase is positively related to arterial pressure[1], and both abnormalities are corrected by surgical removal of the causative tumour[1]. It is also well recognized that increased dietary sodium can raise blood pressure in patients with chronic renal failure[2]. However, in essential hypertension the role of sodium is much less certain. Although blood pressure and dietary sodium are higher and essential hypertension is commoner in 'civilized' than in primitive societies, within a civilized society the essential hypertensives do not, as a rule, eat or excrete more sodium than normal individuals[3]. Nor do patients with essential hypertension have an excess of body sodium and yet amongst such patients arterial pressure and body sodium are positively related, while normal subjects show no correlation of arterial pressure and body sodium[4,5] (Figure 1). Some workers consider it unlikely that dietary sodium is important in the pathogenesis of essential hypertension[3,6], others have suggested that patients with essential hypertension differ from normal subjects in showing a greater change of arterial pressure for a given change of dietary sodium (Figure 2). It is the response to change of diet, not the magnitude of the change of diet which is abnormal.

DIETARY SODIUM

An increase of dietary sodium raises blood pressure in some patients with essential hypertension[8]; on decreasing dietary sodium

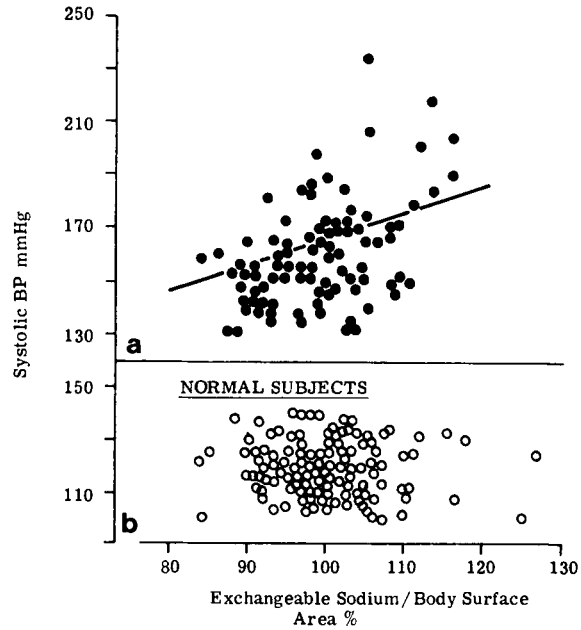

Fig. 1. (a) Essential Hypertension. (b) Normal Subjects. Relation of systolic blood pressure with exchangeable sodium in patients with untreated essential hypertension and in normal subjects. Details of the patients and controls are given in refs. 4 and 5. The significance of these and other correlations with arterial pressure are given in the table.

blood pressure falls[7,9 10]. Changes of arterial pressure are less marked in normal subjects during the same increase of dietary sodium[8]. This is compatible with the idea of an altered response but it does not establish the point as the dietary studies referred to are of a few days duration, while the hypertensive state presumably develops over years. Also, the response of blood pressure to a particular treatment is of limited value in identifying the cause of high blood pressure. Arterial pressure is regulated by more than one mechanism and essential hypertension can be corrected by drugs acting on different mechanisms. A patient with essential hypertension may respond with a reduction of blood pressure when given a low sodium diet or diuretic and when given inhibitors of the renin-angiotensin system or agents which interfere with transmission in the peripheral and central nervous systems. As the primary fault in essential hypertension is unlikely to lie in all these sites a good response to low sodium diet does not necessarily prove that a high sodium diet is the primary fault.

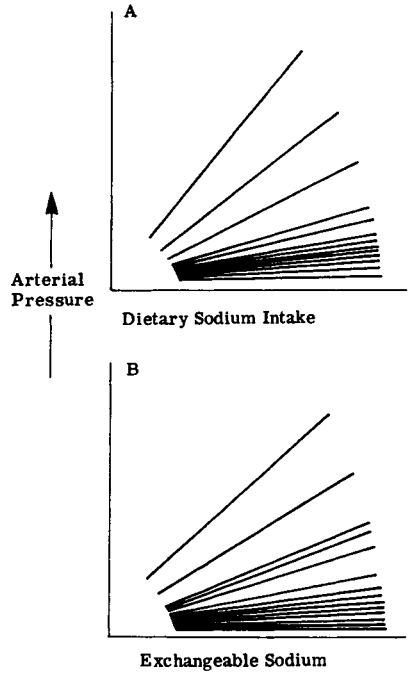

Fig. 2. A. Hypothetical relation of arterial pressure and dietary sodium intake in different people, each represented by a single regression line. Most people show little or no increase in pressure on increasing sodium intake, while a few show a distinct increase.
B. Hypothetical relation of arterial pressure with exchangeable or total body sodium. Again, most people show little change on changing body sodium while a few show a distinct change.
Reproduced from the Brit. Med.J. (ref.4) with permission.

A practical issue arises from the success of low sodium diets in essential hypertension. Most workers demonstrating a good response to such diet recommend their adoption in the treatment of hypertension as a more 'natural' alternative to treatment with hypotensive drugs. Superficially this is an attractive idea but with every hypotensive regime there are two points to be established: that the regime lowers blood pressure and that the lowering of blood pressure reduces the excess mortality associated with high blood pressure. The second is more difficult to prove, but ultimately more important. Before using earlier hypotensive regimes clinicians have insisted on proof of benefit. Furthermore, demonstration of benefit from one hypotensive agent has not been considered sufficient evidence to recommend all agents. Separate tests of benefit were considered

necessary for treatment in severe and moderate hypertension[11], for treatment in mild hypertension[12,13] for treatment in the elderly and for treatment with beta-adrenergic blocking agents and thiazide diuretics[12].

The attitude to testing benefit with low sodium diets in hypertension is different. As noted earlier, several studies have shown that blood pressure can be reduced by a low sodium diet, but we know of no study which has tested or aims to test the beneficial qualities of such diets. Meanwhile, there are numerous recommendations, some from political agencies, suggesting that dietary sodium should be reduced either in the whole population or in hypertensive individuals[7,9,14,15]. A trial testing the assumption of these recommendations is needed urgently, in our view.

BODY SODIUM AND ARTERIAL PRESSURE

The content of sodium in the body can be measured by isotope dilution as exchangeable sodium[16] and by activation analysis[17] which measures the total body content of sodium. Where both methods are used in the same individual the two estimates correlate well (Figure 3). Total body sodium is generally higher since some body sodium is not exchangeable[16]. It can be seen in Figure 1 and in the Table 1 that total body sodium and NaE correlate significantly and positively with arterial pressure in patients with essential hypertension, no significant correlation of NaE and blood pressure emerging in normal subjects. This is compatible with the idea that essential hypertensive individuals are those in whom a given change of body sodium leads to a greater than normal change of arterial pressure. If true, the hypothesis would explain both the rise of arterial pressure with increasing dietary sodium and the response of hypertensive patients to diuretic agents and low sodium diets. Data in Figure 4 are compatible with this since they show that changing body sodium leads to a greater change of blood pressure in hypertensive than in normal subjects, but it can be seen also that data derive from different studies. Comparison of normal and hypertensive subjects in one laboratory would be a better test.

There are other important reservations. When our data are analysed in subgroups no correlation of blood pressure and body sodium is found in women or in young hypertensives[4,5]. Thus, if essential hypertension is a progressive illness and if the younger patients in our study represent an earlier stage of the disease found in older patients, the absence of the correlation between body sodium and blood pressure in the young suggests that its presence is not the primary fault leading to essential hypertension. Instead, the disease may pass through several stages becoming progressively more dependent on sodium[5].

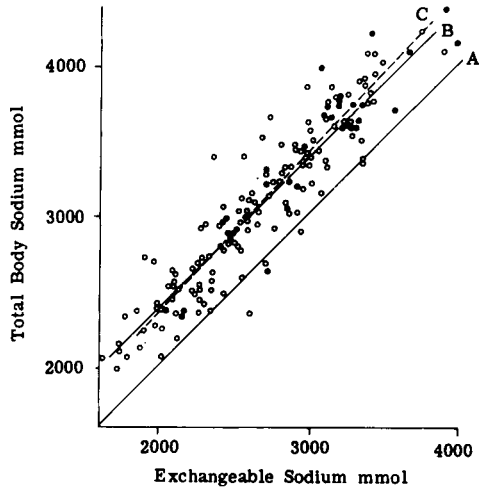

Fig. 3. Relation of exchangeable sodium and total body sodium measured by activation analysis. Solid points are from 38 essential hypertensive patients having both measurements (details of these patients are given in refs. 4 and 5). Open circles are for all other measurements made in our patients, most of whom have renal artery stenosis or Conn's syndrome before and after treatment. 'A' is the line of identity; 'B' the regression for the 38 essential hypertensives; 'C' the regression for other conditions.

Another point of interest is that in young patients with essential hypertension there is an abnormal relation of arterial pressure with plasma potassium concentration (Table 1). There are links between the potassium concentration of extracellular fluid and the activity of the autonomic nervous system and overactivity of the autonomic nervous system is suspected by some workers as a pathogenic factor in the early stages of essential hypertension[18,19].

IN SUMMARY

Excess dietary sodium and excess body sodium can lead to hypertension but patients with essential hypertension show no evidence of either excess. This does not rule out a role for sodium since hypertensive individuals may be those in whom arterial pressure rises when dietary sodium and body sodium are increased. This abnormality is more likely to be present in old than in young patients. An abnormal mechanism involving potassium and autonomic nervous overactivity may be more important in the earlier stages.

Table 1. Body Sodium and Potassium and their relation with Blood Pressure

	Whole group		Age <35 yr		Age >50 yr	
	Normal	Essential Ht.	Normal	Essential Ht.	Normal	Essential Ht.
n	121	91	63	30	37	29
A. Mean ± SD						
Blood pressure mmHg	120/76±11/9	160/102±20/15	118/73±11/10	154/98±14/3	126/80±11/8	165/104±21/13
Plasma Na $mmol.l^{-1}$	140.1±2.0	139.3±2.3*	140.7±1.5	138.3±2.4**	141.8±1.0	140.1±2.2*
Exchangeable Na						
% surface area	100.0±7.3	99.1±7.1	100.2±7.1	96.6±5.2*	99.1±8.0	102.6±7.7
% leanness index	100.0±7.3	97.8±6.6**	99.8±7.7	95.3±5.1**	99.4±7.9	101.3±7.0
B. Correlation with BP						
Exchangeable Na	Syst. Diast.	Syst. Diast.	Syst. Diast	Syst. Diast.	Syst. Diast.	Syst. Diast.
% surface area	0.04 -0.03	0.44** 0.31*	-0.07 -0.09	0.12 0.05	-0.15 0.12	0.64** 0.48**
% leanness index	0.02 0.01	0.41** 0.33**	-0.03 -0.07	0.22 0.15	-0.08 0.16	0.69** 0.59
Total body Na		0.55*** 0.44**				
Plasma potassium	0.17 0.09	-0.32** -0.41***	-0.08 -0.13	-0.41* -0.51**	0.13 0.11	-0.36 -0.16
KE	- -	-0.28* -0.28*	- -	-0.39 -0.52*	- -	-0.38 -0.27
Total body K	- -	-0.38** -0.39**	- -	-0.32 -0.52**	- -	-0.03 -0.14

Mean values ± SD are shown in the upper part of the table, *p <0.05; **p <0.01; ***p <0.001.
Total body sodium was measured in 38 patients.

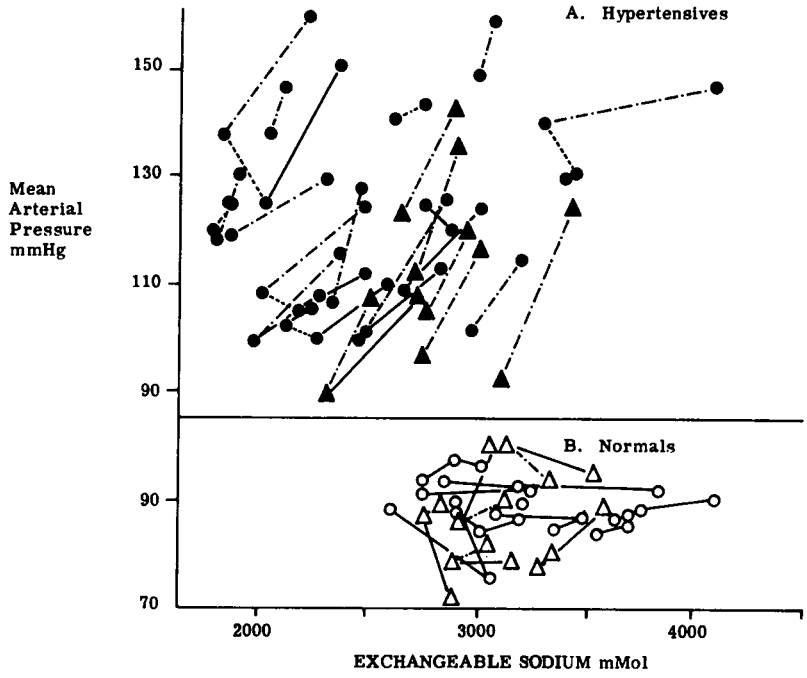

Fig. 4. Relation between exchangeable sodium and mean arterial pressure before and during changes of dietary sodium intake. Open symbols, normal subjects; solid symbols, patients with essential hypertension. Lines join measurements in the same individual. A dashed line indicates measurements made during sodium restriction, a solid line, measurements made during sodium loading and a dotted line, measurements made under identical dietary circumstance. Solid circles are from hypertensive patients studied by Dahl and his colleagues (refs. 19 and 20). Solid triangles are for essential hypertensives studied in the Glasgow Unit; open circles are from normal subjects studied by Kirkendall and his colleagues (ref. 21); open triangles are from normal subjects studied in the Glasgow Unit.

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INHIBITION OF THE RENIN ANGIOTENSIN SYSTEM

IN THE TREATMENT OF HYPERTENSION

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Renin is a proteolytic enzyme that is synthetized, stored and secreted mainly by the juxtaglomerular apparatus of the kidney. The release of renin is regulated a.o. by renal baroreceptors, by sodium-sensitive mechanisms at the level of the macula densa, by the sympathetic nervous system, prostaglandins and angiotensin II. Renin acts on its substrate, angiotensinogen, an alpha-2-globulin produced in the liver and present in the plasma to form the decapeptide angiotensin I, which is practically devoid of pressor activity. The converting-enzyme, a peptidyl-dipeptidase, converts angiotensin I to angiotensin II by splitting off the dipeptide histidyl-leucine at the C-terminal of the decapeptide. The pulmonary circulation is the main site of conversion, but converting-enzyme is also present in the plasma, in the splanchnic system, in the kidney and in several other tissues. Angiotensin II is the effector hormone of the system in man. Its most important actions are direct pressor effects on the arteriolar smooth muscle, stimulation of the aldosterone secretion by the adrenal gland, and effects on the central and peripheral nervous system; these actions result in an increase of blood pressure and in salt and water retention.

The renin-angiotensin system may be blocked more or less selectively at several levels. Pharmacological agents which depress sympathetic activity decrease renin release, as is the case with beta-adrenoceptor blocking agents. The action of renin on renin substrate has been antagonized by nonspecific proteases such as pepstatin, and antibodies to renin and to angiotensinogen as well as structural analogues of the latter have been developed. Peptide and nonpeptide converting-enzyme inhibitors interfere with the conversion of angiotensin I into angiotensin II, but also with the degradation of bradykinin. Angiotensin II analogues, acting as

antagonists by competing with angiotensin II at its receptor sites, have been synthetized, but are not completely devoid of agonist actions.

This paper will deal only with the angiotensin II analogues and with the converting-enzyme inhibitors.

ANGIOTENSIN II ANALOGUES

In the first approach to the development of angiotensin II antagonists a thorough study of structure activity relationships of angiotensin II was performed[1]. As a result of these studies, it was shown that the side group in position 8 on angiotensin II was responsible for the transmission of the information which caused smooth muscle contraction. The information led to the development of the 8-substituted analogues that are competitive inhibitors of angiotensin II. Moreover it was found that substitution of sarcosine in position 1 potentiates the biological activities already existing in the molecule.

The derivate of angiotensin II which has been used most in clinical and/or experimental studies is 1-sarcosine-8-alanine-angiotensin II (saralasin)[2]. This substance has antagonist properties, but also agonist angiotensin II-like activities which is not surprising for an agent that arose from modification of angiotensin II.

In a few studies[3-6] saralasin has been infused into supine sodium replete hypertensive patients, whose sodium intake was between 95 and 150 mmol/24 hours. A vasodepressor response, usually defined as a decrease of diastolic or mean arterial pressure of 7-8 mmHg, occurred in 0-40% of the patients. Differences between studies may be explained by the etiology of hypertension, prevailing plasma renin levels and possibly by differences in plasma volume which may have been decreased with a consequent rise in plasma renin in severe hypertensives[7]. Furthermore, stimulation of the renin-angiotensin system in the seated position[8] may increase the number of responders. In several studies saralasin was administered in both sodium replete and sodium depleted conditions[3-6]. Whereas in these studies 0-40% of the patients responded to saralasin with a significant vasodepressor response in sodium replete conditions, the percentage was 30-90% after sodium depletion. Differences between studies are mainly due to patient's selection, but also to the degree of sodium depletion. Indeed, the number of responders and the magnitude of the pressure fall rose with increasing degree of sodium depletion[6] and even low renin patients do respond to saralasin after severe sodium depletion[9]. When blood pressure decreases in response to saralasin this is usually related to a reduction of systemic vascular resistance[10].

The use of angiotensin II antagonists for the treatment of hypertension is up to now limited by the fact that they have to be administered parenterally. Therefore they can only be used for short periods of time. Brunner et al.,[11] reported that when given to seven patients with malignant or advanced hypertension, saralasin lowered blood pressure to close to normal levels in three patients, whose peripheral plasma renin activity was elevated, and reduced the blood pressure slightly or not at all in the remaining four with normal or low renin levels. A similar experience was reported by Streeten et al.,[12] but they found intravenous infusions of nitroprusside more reliable in those circumstances.

CONVERTING ENZYME INHIBITORS

The nonapeptide teprotide, which is found in snake venom, blocks the conversion of angiotensin I into angiotensin II but has to be administered intravenously. Recently orally active converting enzyme inhibitors have been synthetized, of which the most thoroughly investigated is captopril (2-D-methyl-mercaptopropanoyl-L-proline; SQ 14225).[13] It effectively inhibits the pressor effect of exogenous angiotensin I in man[14] and suppresses the levels of endogenous angiotensin II.[15] However the converting enzyme is also responsible for the degradation of bradykinin, and accumulation of kinins may contribute to the hypotensive response to captopril.[16] There are at present no convincing data that non specific mechanisms, independent of inhibition of converting enzyme are operational.[17]

Captopril lowers blood pressure to a variable extent in patients with hypertension.[18] When a group of subjects with a wide range of plasma renin or angiotensin II levels is studied, the hypotensive effect is correlated with these levels, but the relationship is better for the acute hypotensive effect than for prolonged treatment.[18,19] In sodium replete patients with uncomplicated essential hypertension blood pressure decrease averages 10 to 25%, and is comparable to the effects of thiazides or beta-blockers.[18] The response appears to be greater in patients with severe renovascular hypertension, in some with 'resistant' of 'malignant' hypertension, or after renin-stimulating procedures such as sodium depletion. In high-renin patients with contracted plasma volume dramatic blood pressure falls may however occur with the first dose of captopril.[20] The dose response curve of captopril in hypertensive man suggests that 3 x 50 mg per day guarantees effective biochemical expression of converting enzyme inhibition.[21] Smaller doses, even a few mg, are effective, but then the effect is only of short duration. Hemodynamically the fall in pressure is based on a reduction of systemic vascular resistance both at rest[19] and during exercise,[22] with little or no increase of heart rate and cardiac output; also pulmonary capillary wedge pressure falls with captopril.

Treatment with captopril has been associated with side effects such as rash and taste disturbance, hematological effects as neutropenia and agranulocytosis, and proteinuria.[18]

COMPARISON OF SARALASIN AND CAPTOPRIL

When saralasin and captopril are administered to slightly sodium-depleted hypertensive patients, the changes of pressure of both drugs are related to the prevailing plasma renin level. However, the hypotensive effect of captopril is about 10 mmHg greater than that of saralasin.[15] The difference may be due to the intrinsic pressor effect of saralasin, or to kinin accumulation during captopril.

These data indicate that there is still room for the development of more specific inhibitors of the renin angiotensin system.

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CATHETER TREATMENT OF RENOVASCULAR HYPERTENSION

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Renovascular hypertension can be treated effectively in nearly all patients with modern antihypertensive drugs. But medical therapy is accompanied by disadvantages such as lacking patient compliance, and side effects of the medication. Long-term medication - as necessary in younger patients - is expensive, and the reduced blood flow to the affected kidney is not improved. In the past vascular surgery was the treatment of choice in patients with renal artery stenosis with hyperreninemia and a short history of hypertension. Operative pain and risk, costs and an uncertain treatment result restricted vascular surgery to critically selected patients.

Four years ago percutaneous transluminal catheter dilatation (PTRD) was added to the therapeutic spectrum of renovascular hypertension.[2,3,5,7] In the meanwhile we treated 76 patients with this method and gained plenty of data to evaluate the benefits and limitations of the procedure.

INDICATIONS

PTRD is indicated in patients with a combination of hypertension, hyperreninemia, and renal artery stenosis regardless of its nature (Table 1). Age is no limiting factor for PTRD, but it may be difficult to dilate renal arteries in infants younger than 2 years because of the small vascular dimensions. In older children PTRD may be performed successfully.[8,9] In patients older than 60 years the blood pressure cannot be expected to return to normal values, but most of these patients are treated for renal insufficiency due to bilateral kidney disease.[4] Kidney grafts with anastomotic or postanastomotic renal artery stenosis caused by intimal proliferation

Table 1. Indications for PTRD in Renovascular Hypertension

Renal Artery Stenosis: 70% or more
Atherosclerosis
Fibromuscular Dysplasia
Neurofibromatosis
Intimal Proliferation in Kidney Graft
Postoperative Anastomotic Stenosis
Renal Artery Occlusion
Rarely in Atherosclerosis in
Connection with Renal Insufficiency
Renal Insufficiency
Caused by Stenosis or Occlusion

are suited for PTRD, but the success rate is not as high as in our other patients depending on the implantation technique and anatomical details.[6] Patients with post-traumatic or atherosclerotic renal artery occlusions are furtheron candidates for surgery.[1]

PERFORMANCE

PTRD is carried out in local anesthesia under light sedation. Long acting antihypertensive drugs are discarded before the intervention to avoid threatening pressure drops. The blood pressure should not exceed 200 mmHg during PTRD to prevent hemorrhage at the puncture site. In critical patients this is achieved by sodium nitroprusside infusion under continuous pressure surveillance.

The normal approach for PTRD is the femoral artery at the groin. A transaxillary route is chosen when the renal artery originates with an acute angle from the aorta or when the iliac arteries are obstructed.[10]

Two catheter systems are available for PTRD.

1. Single catheter technique: after probing of the renal artery with a diagnostic catheter a guide wire is placed with its tip beyond the stenosis, the diagnostic catheter is exchanged against a dilatation catheter (F-7 or F-5), and the stenosis is dilated.
2. Coaxial catheter technique: a guiding catheter (F-9 or F-8) is placed before the ostium of the renal artery. A small dilatation catheter (F-4.5) is passed through it and enters the narrowed renal artery. Thereafter, dilatation is performed.

The coaxial catheter set is advantageous when a high degree stenosis cannot be overcome by the thicker single catheter, but will produce a bigger puncture hole.

During PTRD the pressure is measured in the renal artery determining the pressure gradient before and after dilatation. At the end of the dilatation treatment a control angiogram is performed. On the first day the patients receive 20,000 U of heparine, and medication with platelet aggregation inhibitors is started, and should be continued for a least 6 months.

RESULTS

PTRD was attempted in 76 patients with 87 stenoses. The patients had a mean age of 45 years with a range from 5 to 75 years. The nature of the stenoses is shown in Table 2. PTRD was finished in 5 patients without success, in whom the obstruction could not be passed by the dilatation catheter (2 stenoses in fibromuscular dysplasia and 2 in kidney grafts, 1 renal artery occlusion). PTRD was technically successful in 82 of 87 stenoses (94%). In 19 patients hypertension persisted or recurred. In 14 of them a control angiogram was performed and revealed a normal renal artery in 9 and a recurrent stenosis in 5 patients. In 2 cases the recurrent stenosis was redilated improving the hypertension considerably. In all 3 patients with renal insufficiency the kidney function was improved by PTRD, but the blood pressure was not reduced significantly (Table 3).

Case 1: 22-year old woman with a blood pressure of 240/110 mmHg. Angiography revealed bilateral fibrodysplastic renal artery stenosis. The stenotic segment of the renal artery was dilated on both sides, and the blood pressure declined within 6 hours to 120/80 mmHg (Figure 1). The patient is normotensive for 8 months without medication.

Case 2: 58-year old man with a history of hypertension for 2 years and a renal artery stenosis on the left side. After PTRD a residual stenosis of 20% remained, but the blood pressure returned to normal values without any medication.

The patients came for follow-up studies every 3 months in the first year and every 6 months thereafter. The mean follow-up amounts to 23 months with a range from 1 to 58 months. The results were evaluated using the life table method. 24 of 76 patients were cured with blood pressure values below 150/95 mmHg without antihypertensive drugs. In 33 patients the blood pressure declined after PTRD, but the patients still need saluretics or β-blocking agents to keep the blood pressure in the normal range. Most of these patients belong to the group with an atherosclerotic stenosis and are older than 45 years. In 19 patients the blood pressure remained unchanged after PTRD. 7 of these patients had nearly normal renin values before

Table 2. Nature of Renal Artery Stenosis

Atherosclerosis	68	78%
Fibromuscular Dysplasia	13	15%
Kidney Graft	6	7%

Table 3. Results of PTRD

Patients	76	100%
Normotensive (<150/95 mmHg)	24	32%
Improved	33	43%
Hypertensive	19	25%
Dead	2	2.5%

PTRD and 3 patients suffered from terminal renal insufficiency. In 2 females with fibromuscular dysplasia hyperreninemia vanished, but hypertension persisted. In one patient with malignant hypertension cerebral hemorrhage occurred 2 months after PTRD, and had a fatal outcome.

Renin concentration was determined in renal vein blood samples before and after PTRD in 68 patients. Renin values were elevated in 51 of them (74%), 6ng/ml/h being the upper normal limit. The predictive implications were impressive: 90% of the patients with hyperreninemia had a blood pressure decrease after PTRD. But renin concentration and renin ratio are of little use for the indication of PTRD, because in 59% of the cases with normal to borderline renin values hypertension was improved after PTRD, too.

COMPLICATIONS

Complications were encountered in 3 patients (4%) and required surgical intervention in one case of renal artery dissection treated by an aortorenal bypass graft. Two patients developed a hematoma at the groin which was conservatively managed.

CONCLUSIONS

PTRD was performed in 76 patients with renovascular hypertension with a primary success rate of 94%, and a blood pressure decrease in 75% of the patients. 5 patients (6%) developed a recurrent stenosis during a mean follow-up of 23 months which was redilated in 2 of

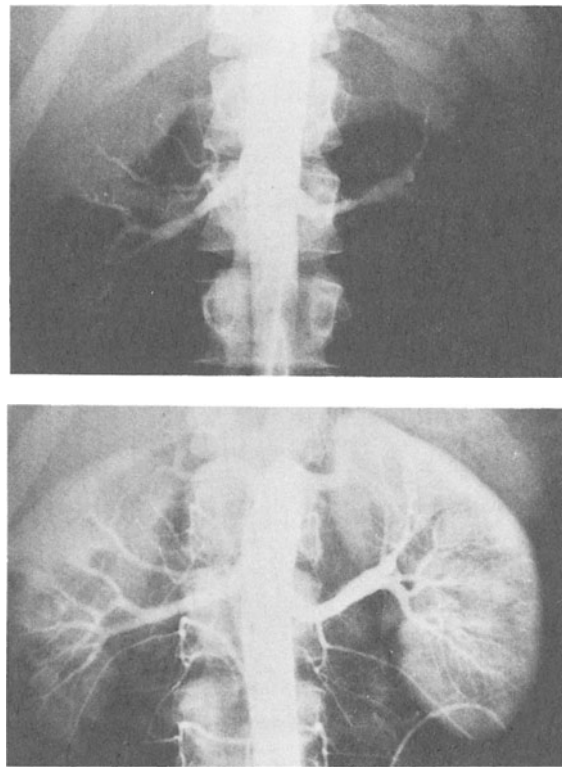

Fig. 1. 22-year-old female with bilateral fibrodysplastic renal artery stenosis which could be completely removed on both sides.

them (2.5%) with good result. These data, confirmed by other groups working with PTRD, underline the efficacy of the procedure in renovascular hypertension.[2,3,5,7]. Therefore, in our opinion there is enough evidence that PTRD should be attempted in all hypertensive patients with renal artery stenosis with a narrowing of more than 70%, regardless of renin values. Vascular surgery should be restricted to patients with additional vascular problems as aneurysm of the renal artery, aortic aneurysm or iliac artery occlusion, and to those with technical treatment failures of PTRD. Patients older than 60 years should be managed conservatively, and are only referred to PTRD when the blood pressure cannot be controlled sufficiently by antihypertensive medication or when renal insufficiency develops.

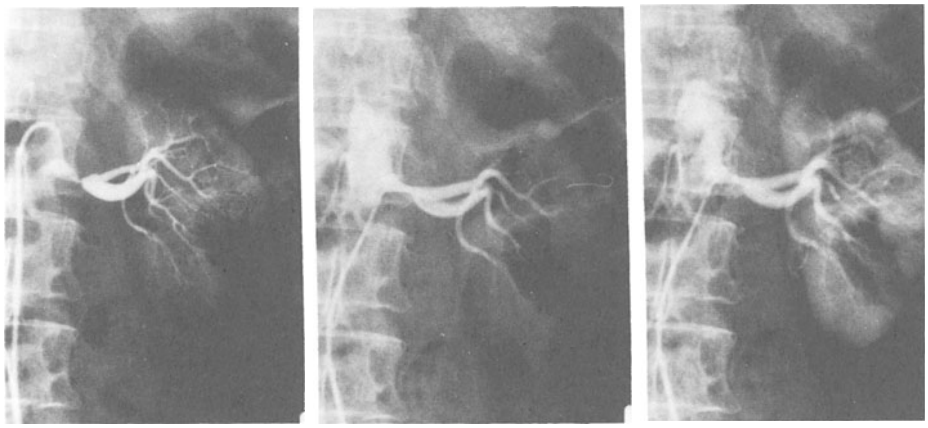

Fig. 2. 58-year-old man with left sided atherosclerotic renal artery stenosis, considerably improved by PTRD. Better filling of the intrarenal arterial braches after dilation.

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PATHOGENETIC MECHANISMS IN ESSENTIAL HYPERTENSION

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INTRODUCTION

Major parts of the etiology and pathogenesis of hypertension remain fragmentary and hypothetical. Over the past few years,[1-5] investigative attention has focused more and more on the hypothesis of Dahl[6] that a circulating natriuretic substance might be the culprit of the increase in arterial pressure. This concept proposes that a genetic defect of sodium metabolism causing a discrete positive sodium balance might stimulate secretion of a naturiuretic hormone. Thus, normal sodium balance in the prehypertensive patient could be achieved by continuous high levels of naturiuretic hormone. However, sodium transport inhibition would occur not only along the nephron but in other tissues as well.[4,7,8] In vascular smooth muscle it would increase the reactivity and tone of arteriolar and venous smooth muscle, thereby elevating arterial pressure and diminishing the venous compliance. Constriction of capacitance vessels would, in turn, shift the total blood volume from the periphery toward the cardiopulmonary area. This expansion of cardiopulmonary volume would further perpetuate the secretion of the sodium transport inhibitor from the hypothalamus (even in the presence of a normal or contracted total blood volume) and a vicious circle would ensue.

This hypothesis is supported by hemodynamic, fluid volume, and endocrine changes that have been documented over the natural history of arterial hypertension as it evolves from its prehypertensive state to borderline and established hypertension, and reaches its

final stage in congestive heart failure. The following is a review of this pathogenetic sequence that takes place as hypertensive cardiovascular disease progresses.

PREHYPERTENSION

Dysregulatory mechanisms leading to established essential hypertension may have their onset in early childhood. Normotensive children or adolescents of hypertensive parents are at an increased risk of developing high blood pressure at a later stage and thus provide a study population that is to some extent prehypertensive. In borderline or established hypertension, alterations in the activity of the sympathetic nervous system, in hemodynamics, in endocrine factors, or in vascular reactivity could represent the consequence rather than the cause of the disease. In contrast, in these prehypertensive adolescents secondary effects of an elevated arterial pressure are absent. Several subtle alterations have indeed been documented over the past few years: offspring of parents with essential hypertension have been found to have abnormalities of sodium transport in white and red blood cells which possibly reflect a generalized defect in cellular sodium transport. [8] Also, Falkner et al [9] demonstrated that normotensive adolescents who had a significant family history of hypertension showed a sustained diastolic pressure response to mental stress which was similar to patients with borderline hypertension and different from normotensive subjects without a positive family history. Moreover, these prehypertensive subjects had significantly elevated plasma catecholamines levels after mental stress.[9]

Similarily, McCrory et al[10] found abnormalities in blood pressure and plasma catecholamines response to orthostatic stress in normotensive siblings of hypertensive children. Thus, some normotensive adolescents who have a genetic risk of essential hypertension exhibit a hemodynamic and endocrine response to stress that is similar to the one observed in borderline hypertension. This clearly indicates that similar adrenergic dysfunction can be even encountered already in the normotensive prehypertensive stage and does not arise as a consequence of the blood pressure elevation.

BORDERLINE HYPERTENSION AND OBESITY HYPERTENSION

Non-obese young patients with borderline hypertension are characterized by an elevated cardiac output and cardiac index, an increase in heart rate, and an augmented renal blood flow.[11-20] Symptoms and signs of an increased sympathetic outflow such as palpitations, chest pain, and mitral prolapse are common.[21,22] Total blood volume is slightly contracted and redistributed to the cardiopulmonary circulation.[12,17,21,23] At the same time, higher

circulating norephinephrine levels can be observed.[21,24] Although total peripheral resistance has been found to be numerically normal in these patients, it is still inappropriately elevated with regard to their level of cardiac output.[12,25] These hemodynamic changes with cardiopulmonary translocation of the intravascular volume suggest vasoconstriction in the arteriolar as well as the venous vascular bed. An increase in tone in vascular smooth muscle together with elevated heart rate and cardiac contractility reflects enhanced participation of the sympathetic nervous system or decreased parasympathetic tone.[27-29] Thus, an adrenergic dysfunction seems to be the predominant underlying mechanism for the discrete elevation of arterial pressure in these young borderline hypertensive patients.

Obese normotensive persons are also at a higher risk of developing established hypertension than comparable lean subjects.[30,31] Obesity, therefore, represents another prehypertensive state. However, in contrast to the hyperadrenergic borderline hypertensive state, described above, the elevated cardiac output in obese patients is associated with an expanded intravascular volume but a normal distribution within the central and peripheral circulation.[21] Cardiac output in obesity parallels the expansion of total blood volume and both seem at a first glance to merely reflect the increase in metabolic requirements that occur because of the additional (adipose) body mass. However, we have recently shown that the augmented cardiac output observed in obese patients cannot be explained by the increased requirements due to adipose perfusion alone.[21] Thus, the elevation in cardiac output seems inappropriate with regard to the amount of fat tissue. These hemodynamic changes of obesity with an elevated cardiac output, expanded total blood volume, and a decreased peripheral resistance are remarkably similar to the experimental situation that occurs initially with salt-loading in a partially nephrectomized dog (Fig 1). Many experiments have shown (in this model) that an expanded blood volume associated with renal arterial constriction will result in arterial hypertension.[32-34]

Transition to Established Hypertension (Fig 2)

When hyperadrenergic borderline hypertensive patients grow older and their disease progresses to established hypertension, cardiac output falls toward normal values and total peripheral resistance continues to rise.[19,35-37] As a consequence, intravascular volume becomes progressively more contracted.[38] This shift in the hemodynamic profile from a high cardiac output hypertension to high vascular resistance hypertension could be mediated through changes in receptor responsiveness or receptor density and/or by progressively increasing circulating norephrinephrine levels.[19,24,39] Thus, the hemodynamic pattern in established essential hypertensive patients is characterized by normal cardiac output, contracted intravascular volume, and elevated total peripheral resistance.[40-42] Renal

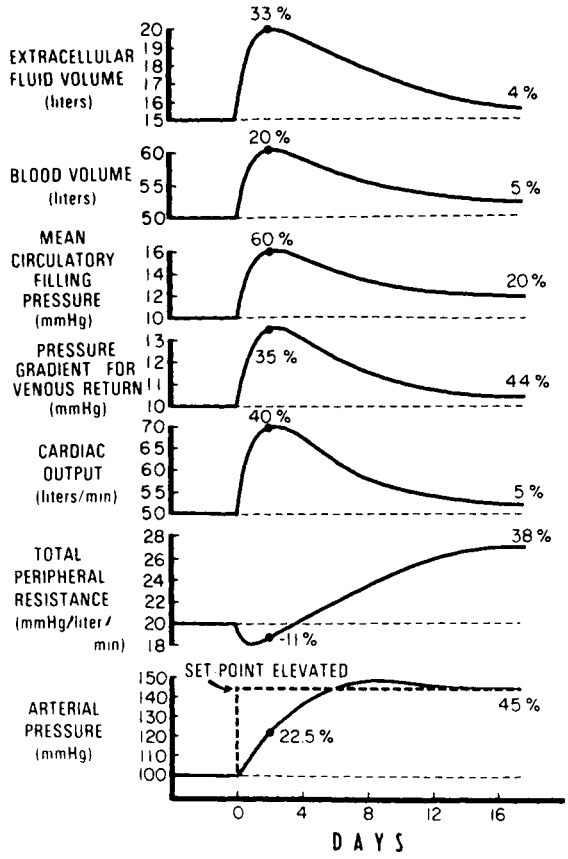

Fig. 1. Pathogenesis of hypertension with salt-loading in partially nephrectomized dogs (from Guyton AC, Circulation 64:1079, 1981, with permission)

blood flow seems to fall relatively early in the disease process, whereas glomerular filtration rate remains normal.[43,44] As a consequence, filtration fraction becomes elevated. Circulating norepinephrine levels, although progressively increasing with age, 19,24,39 are most often within normal range.

Obese patients progress from the normotensive to the hypertensive stage by a progressive vasoconstriction without change in cardiac output.[45] Vasoconstriction seems to be a response to the chronically inappropriately elevated systemic blood flow (i.e., cardiac output) in obesity. Consequently, the low total peripheral resistance progressively increases and the initially expanded intravascular volume becomes smaller.[45] Since the effects of obesity

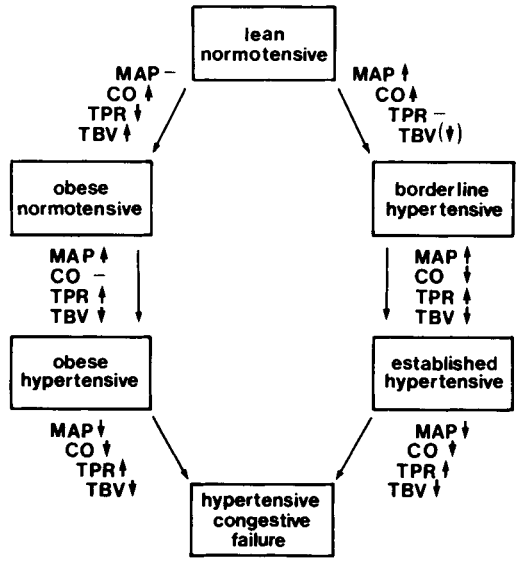

Fig. 2. Pathogenesis and evolution of arterial hypertension in obese and nonobese patients
MAP: Mean arterial pressure
CO: Cardiac output
TPR: Total peripheral resistance
TBV: Total blood volume

and arterial hypertension on intravascular volume and total peripheral resistance offset each other, an obese hypertensive patient may be characterized by a high cardiac output, normal total peripheral resistance and a normal total blood volume.[46] However, as hypertensive cardiovascular disease becomes more severe, intravascular volume becomes contracted and total peripheral resistance elevated. Cardiac output has a tendency to fall in patients with morbid obesity because of early left ventricular dysfunction.[47] Thus, the transition of hemodynamic profile from the obese normotensive to the obese hypertensive patient seems to be mediated predominantly by volume and autoregulatory mechanisms without much involvement of the autonomic nervous system. Perhaps then, the pathogenesis of obesity hypertension in man may represent the clinical counterpart of the famous Guytonian model[35] of experimental hypertension in the salt-loaded, partially nephrectomized dog.

HYPERTENSION IN THE ELDERLY AND CONGESTIVE HEART FAILURE

As the patient grows older, blood pressure increases progressively and target organ involvement progresses. The hemodynamic picture of the nonobese patient is characterized by a high vascular resistance and a normal to low cardiac output.[40] As a consequence of long-standing elevated vascular resistance, target organ damage such as nephrosclerosis, systemic vascular disease, and retinopathy may develop. In constrast, since obesity is associated with a lower vascular resistance, it can be expected, at least to some extent, to mitigate systemic vascular disease.[48] These pathophysiologic observations lend credence to the findings of Pererra et al of a higher prevalence of accelerated hypertension (as characterized by retinopathy and arteriolar necrosis on renal biopsies) in lean than in moderately obese women.[48]

However, although obesity may exert a beneficial effect on systemic vascular disease, it greatly enhances the risk of congestive heart failure. The increased stroke volume and the increase in end-diastolic pressure result in a high preload to the left ventricle that is already burdened by a high afterload of arterial hypertension.[46] The heart adapts to this double burden with eccentric hypertrophy. The two evils, hypertension and obesity, take a heavy toll of the heart and distinctly increase the longterm risk of congestive failure. Not surprisingly, obese hypertensive patients have been documented to be at a high risk of premature congestive failure regardless of their level of arterial pressure.[49,50]

Irrespective of body weight, a progressive increase in arterial pressure will ultimately lead to congestive failure. At that stage, cardiac output declines and total peripheral resistance becomes greatly elevated. For this prefinal stage the term "decapitated" hypertension has been used: the left ventricle is no longer able to maintain the arterial pressure at its high level and a fall in pressure ensues. Although intravascular volume still may be contracted, activation of the renin-angiotensin-aldosterone system occurs and thereby promotes progressive sodium and fluid retention. Consequently the classical clinical picture of congestive failure with low cardiac output, impaired organ perfusion and generalized edema ensues.

THE HYPERADRENERGIC VS THE AUTOREGULATORY PATHWAY

From a study of the two extremes, hypertension in the lean and hypertension in the obese, a dissection can be made of a predominantly hyperadrenergic mechanism from one that seems to involve more volume and autoregulation (Fig. 3.). Thus, in lean patients in a very early phase of the disease, evidence of increased adrenergic activity can be documented. Conceivably, the blood pressure may be maintained at a high level by an enhanced adrenergic outflow with relatively little or no involvement of volume factors. On the other

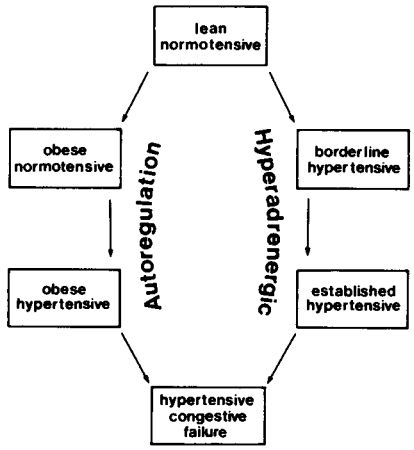

Fig. 3. The hyperadrenergic versus the volume/autoregulatory pathway in the pathogenesis of essential hypertension.

hand, in the distinctly obese patient, an evolution as originally proposed by Guyton may take place.[34] Volume expansion and an elevated cardiac output lead by an autoregulatory process to a progressively increasing peripheral resistance which thereby elevates arterial pressure. Both pathogenetic pathways are comparable with Dahl's hypothesis:[6] the rise in vascular resistance could be triggered and/or accelerated by increased circulating levels of sodium transport inhibitor.

However, pathogenesis of arterial hypertension in the very lean and in the very obese patient merely represent two extremes of a continuing spectrum. Since most patients with essential hypertension are somewhat overweight, it is fair to assume that generally a combination of both pathophysiologic mechanisms may be operating. Established essential hypertension most probably represents the end result of a pathogenesis that involves the hyperadrenergic as well as the autoregulatory pathway.

SUMMARY

Disparate pathogenetic mechanisms are involved in essential hypertension in lean and in obese patients. In lean subjects, evidence of early adrenergic dysfunction with elevated cardiac output, venous constriction and expanded cardiopulmonary volume as well

as increased circulating norepinephrine levels can be demonstrated. In contrast, obese subjects are characterized by volume expansion, lower vascular resistance although their cardiac output may be elevated to a similar level as in lean borderline hypertensive patients. It is proposed that blood pressure elevation is maintained by enhanced adrenergic outflow causing postcapillary venoconstriction without or less apparent involvement of volume factors in lean subjects. However, volume and autoregulatory mechanisms are more apparent in the mediation of the pathogenesis of hypertension in obese patients. Pathogenesis of hypertension in the very lean and the very obese are two extremes of a continuum. Since most patients with essential hypertension are somewhat overweight, generally a combination of both pathophysiologic mechanisms may be operating.

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NEURAL FACTORS IN ESSENTIAL HYPERTENSION

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Since the discovery that the autonomic nervous system represents a mechanism of primary importance in the regulation of circulation the idea that essential hypertension might depend on a derangement in neural cardiovascular control has been a leading one in the investigation related to this pathological condition.

This presentation will not touch all complex and controversial aspects of this topic but rather concentrate on a few points derived from studies on both experimental and human hypertension. It is hoped that these points will show some of the basic information that has been gained during these years, but also underline the limitation in our knowledge of this field.

Mention of results obtained in studies on experimental hypertension is important because this approach has answered two key questions that are still unsolved in human hypertension. The first question is whether neural factors may produce a condition of high blood pressure, i.e. whether they may have an initiating role in this disease. By showing that in several animal species a variety of neuropsychological manipulations can produce a permanent blood pressure elevation this question has received a positive answer. The following are a few examples:

(a) a prolonged blood pressure rise has been shown to occur in monkeys subjected to adverse conditioning, i.e. to a stressful procedure that requires the animals to act in a way that can avoid them the delivery of unpleasant stimuli[1,2];

(b) a hypertension has been shown to occur in male mice which behave as dominant within their colony, in contrast to the lower blood pressure values (and the lower incidence of cardiovascular complications) of other mice of the colony whose social behaviour appears to be subordinate to the dominant ones;[3]

(c) neural factors have also appeared to be responsible for the initiation of the hypertensive state in the Okamoto strain of rats. It has been shown[4] that these rats display exaggerated pressor responses to stressful stimuli even at an age that precedes the development of hypertension. Furthermore such development has been shown to be prevented or slowed either by early treatment with "antisympathetic" drugs or by a drastic reduction in the amount of natural stimuli that occur in rats' lives. Thus, this hypertension model has reproduced the condition that is believed to characterize essential hypertension, i.e. an interaction between environmental and inherited factors.[5] It has also shown that the latter may consist in a hyperactivity of the neural structures involved in the integration of the defence mechanisms, the permanent blood pressure elevation being probably the consequence of behaviourally-induced excessive phasic blood pressure rises.

The second question is whether neural influences, beside being capable of playing an initial role, are important as secondary factors, i.e. whether they represent a mechanism that maintains blood pressure high in non-neurally initiated hypertensions. Animal studies have provided a positive answer also to this question. For example, destruction of sympathetic nerve structures either at a peripheral or a central site has been shown to be followed by a large blood pressure reduction not only in neurally induced hypertensions but also in hypertension models of a non-neural origin (DOCA hypertension, renovascular hypertension, etc.).[6] Furthermore, the development of renovascular hypertension in rats has been slowed or prevented not only by diffuse sympathetic destruction, but also by restricted lesions placed centrally in the hypothalamus.[7] Finally, data of our group have shown that in secondary hypertension normalization of blood pressure values can be achieved also by spontaneous reduction of the existing sympathetic vasoconstrictor tone.[8] These findings are exemplified in Figure 1 which represents average blood pressure values recorded intra-arterially during a large number of wakefulness-sleep cycles in a normotensive and a renovascular hypertensive cat. Both animals had been subjected to sino-aortic denervation to allow an unbuffered depression of sympathetic vasoconstrictor tone to occur during sleep, particularly during its REM phase.[8] In the normotensive cat the REM sleep-induced sympathetic depression reduced blood pressure to low values. The same low values, however, were attained during REM sleep by the renovascular hypertensive cat, whose blood pressure elevation thus depended on activation of sympathetic tone. The mechanisms through which stenosis of renal arteries or DOCA injections lead a sympathetic acti-

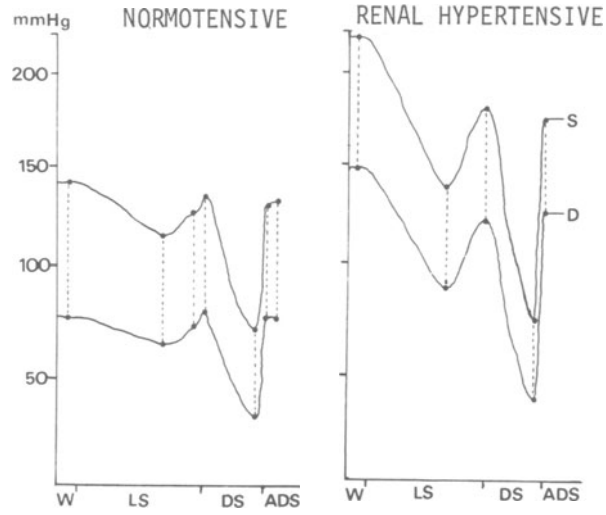

Fig. 1. Systolic (S) and diastolic (D) blood pressure during the wakefulness-sleep cycle in a normotensive and a renovascular hypertensive cat, both with sino-aortic denervation. W: wakefulness; LS: light or synchronized sleep; DS: deep or REM sleep; ADS: after deep sleep. Each point represents the average of 10 different episodes. (From Mancia and Zanchetti, "Physiology during Sleep", p.1, 1980, Academic Press, by permission).

vation that takes over the responsibility of the hypertensive state represent a third basic question. This question is still unanswered. Current hypotheses focused on the relationship between sodium and norepinephrine stores and release in the nerve terminals[6] and on the stimulating properties of angiotensin II at various central and peripheral sympathetic sites.[9,10]

Coming to neural factors in human hypertension it is important to emphasize that this field is encompassed with two methodological difficulties. The first difficulty is represented by the fact that in humans hypertension is commonly seen at a variably advance stage from its inception which prevents the separation between primary and secondary factors that has been so profitably applied in experimental animal models. The second and more important difficulty is that the approaches that have so far been employed to quantitate neural cardiovascular influences have major limitations. This applies to old approaches (personality patterns, assessment of haemodynamic responses to stress, assessment of depressor responses to "antisympathetic" drugs, basal haemodynamics, etc.) but also involves the new ones. For example measurement of plasma norepinephrine levels, though capable of signaling large and diffuse stimulation or

depression of sympathetic activity, may represent a relatively insensitive index of more moderate alterations in sympathetic tone, particularly when comparisons have to be made among subjects.[11,12] Direct assessment of this tone in skeletal muscle circulation, besides representing only a fraction of the overall sympathetic activity, may also be difficult to quantitate and compare among subjects. Due to these limitations it is not surprising that studies on the importance of neural factors in essential hypertension have produced many conflicting and inconclusive results.

There is a further approach to this problem, however, that avoids the elusive quantitation of sympathetic activity and addresses to clarification of the mechanisms that are involved in neural cardiovascular regulation of human beings and to their evaluation in normotensive and hypertensive individuals. A mechanism that has been extensively investigated is the reflex control of the cardiovascular system exerted by the arterial baroreceptors. Several studies that have examined the baroreceptor control of heart rate have shown that in hypertension its range is reset towards the elevated blood pressure values and its sensitivity is reduced.[14] These studies have raised the possibility that essential hypertension is due to a reduction in the reflex inhibition that physiologically modulates sympathetic noradrenergic activity.

However, data obtained recently by us and by others[15,16] do not support this possibility. If a reduction in baroreflex sensitivity were important in producing hypertension, baroreceptor denervation should be followed by a permanent blood pressure elevation. Table 1 shows that this may not be the case. In unanaesthetized unrestrained cats blood pressure was continuously recorded for several hours before and a week after section of the sino-aortic nerves. Computer analysis of the tracings showed that blood pressure was much more variable after than before the sino-aortic nerve section, a finding to be expected in consideration of the buffering role of the

Table 1. Effects of Sino-aortic Denervation Alone or Combined with Vagotomy on Blood Pressure Mean Values and Blood Pressure Variability*

	n = 8			n = 6		
	Intact	SAD	P<	SAD	SAD+vagot	P<
Mean arterial pressure (mmHg)	99±7	93±7	NS	103±8	101±6	NS
Variation coefficient (%)	6±1	12±2	.01	13±2	12±6	NS

*Data represent means (±SE) from recording performed in unanaesthetized, unrestrained cats.

arterial baroreceptor afferents on the blood pressure oscillations. However, mean blood pressure values did not differ significantly in the two conditions. This was the case even when, in another series of cats, blood pressure recordings were performed first after sinoaortic denervation and then 1-7 days after bilateral vagotomy, a procedure which removed the residual inhibitory influence reflexly exerted on sympathetic tone, namely that arising from cardiopulmonary receptors.[17] Thus even complete baroreceptor denervation (a condition that might be equated to "0 sensitivity" of the baroreflex) does not appear capable of producing hypertension.

Further evidence against involvement of the arterial baroreflex in essential hypertension was obtained in human studies through the use of the neck chamber technique, which allows carotid transmural pressure to be increased or reduced in a measurable and gradual amount, and to relate these alterations to the resulting mean arterial pressure responses, thereby obtaining the sensitivity of the most important baroreflex function, i.e. blood pressure control[18]. A study performed in normotensive subjects and in subjects with moderate and severe essential hypertension[19] showed that (a) in normotensive subjects the blood pressure changes caused by reducing carotid transmural pressure, i.e. by deactivating baroreceptors, were greater than the blood pressure changes caused by increasing carotid transmural pressure, i.e. by stimulating baroreceptors, (b) the blood pressure effects of baroreceptor deactivation progressively fell, and those of baroreceptor stimulation progressively increased in size on going from normotensive to moderate and severe hypertensive subjects, (c) the sums of the blood pressure effects of baroreceptor deactivation and stimulation, and the maximal slopes of these responses, were similar in the three groups (Figure 2). These findings indicate that, at variance with heart rate control, baroreceptor control of blood pressure is largely preserved in essential hypertension. They confirm that a resetting of the baroreflex occurs in this condition and suggest that this phenomenon may be so marked as to displace the set-point of the reflex in a direction opposite to that predictable, on the basis of the blood pressure elevation, i.e. towards threshold rather than towards saturation.[20] In a further study we were able to show that an unchanged sensitivity and a marked resetting of the baroreceptor-blood pressure control also characterize subjects with renovascular hypertension.[21] Taken together these findings do not support the possibility that the arterial baroreflex represent a causal factor in essential hypertension. They do show, however, that an important modification of the baroreflex, namely a marked resetting, can occur as a result of blood pressure elevations. Such resetting may represent a factor that defends the raised blood pressure and secondary contributes to this condition. It may also be viewed, however, as an adjustment that preserves the baroreflex ability to protect hypertensive subjects against further acute pressure rises and that by favouring maintainance of sympathetic vasoconstrictor tone allows the antihypertensive action of sympathetic agents to take place.

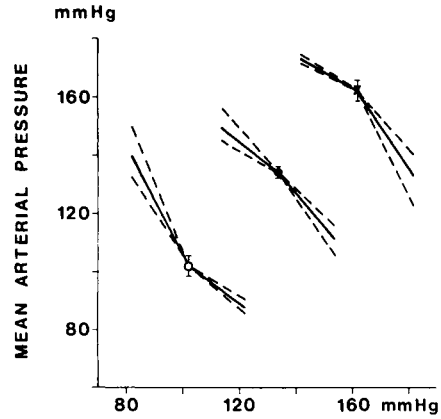

Fig. 2. Reductions and increases in mean arterial pressure (vertical line) induced by increases and reductions in carotid transmural pressure (horizontal line). Data represent average values from a group of normotensive subjects (n = 11), a group of subjects with moderate essential hypertension (n = 18) and a group of subjects with severe essential hypertension (n = 17). The open circle, closed circle and the cross represent average (±SE) baseline mean arterial pressure of the 3 groups respectively. The continuous lines departing from these symbols represent the slopes of the mean arterial pressure in relation to the baroreceptor stimuli, the dashed lines representing the standard errors. Note the reverse asymmetry of the pressor and depressor responses in the severe hypertensive as compared to the normotensive subjects (adapted from Mancia et al., Circ. Res. 43:170, 1977, by permission).

Another approach we have followed is analysis of spontaneous blood pressure variability due to the demonstration that this phenomenon may be largely neural in nature. Figure 3 shows the results obtained in an ambulant subject in which blood pressure was invasively monitored for 24 hours by the Oxford technique.[22] The blood pressure signal was analyzed by a computer which provided mean and standard deviation values of each of the 48 half hours of the 24 hour period. Average of the 48 standard deviations was then calculated and used as an index of the blood pressure variability within half hours, i.e. a rather short-term variability. The standard deviation observed by averaging the 48 mean values was also calculated and used as an index of the blood pressure variations among half hours, i.e. a longer-term variability. The same analysis was performed for heart rate. In 38 subjects blood pressure variabilities showed a weak and often non-significant correlation with the arterial baroreflex sensitivity, suggesting that the differences in the buffering action of this mechanism that may exist within the population may not be

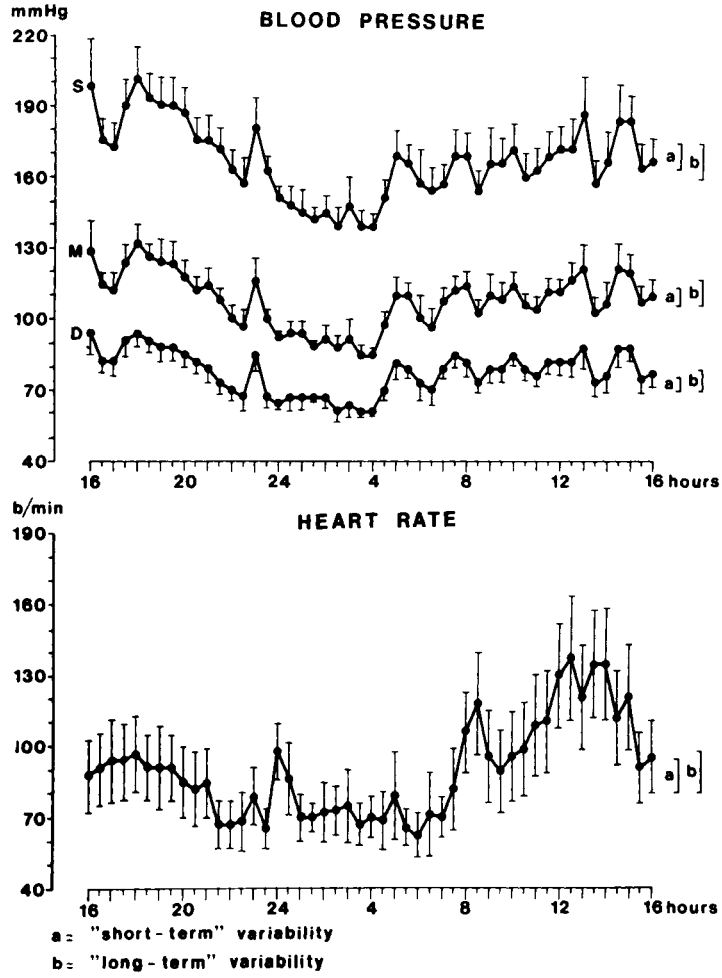

Fig. 3. Computer analysis of systolic blood pressure, diastolic blood pressure, mean arterial pressure and heart rate obtained during a 24 hour intraarterial blood pressure monitoring in an ambulant subject. The blood pressure signal was sampled by a computer every 60 msec. Data are expressed as mean values and standard deviations for each of the 48 half hours of the recording. a: standard deviation within half hour, i.e. short-term variability; b: standard deviation among half hour, i.e. long-term variability. For further explanations see text.

crucial in determining the extent of this phenomenon.[23] On the other hand the short-term blood pressure variability showed a striking reduction during sleep and its changes throughout the 24 hours were

closely paralleled by alterations in short-term heart rate variability. Blood pressure and heart rate mean values also showed a concomitant close parallelism. These findings can be explained by postulating the existence of central influences affecting consensually cardiac and vascular targets whose importance in daily cardiovascular modulation largely predominates over negative feed-back systems that would alter these targets in opposite directions.

If blood pressure and heart rate variabilities mainly reflect centrally originated influences, their comparisons at normal and high blood pressure may bear a special interest. Such comparison was made in subjects classified on the basis of their 24 hour mean arterial pressure values as being normotensive or as having an essential hypertension of moderate or severe degree.[23,24] Figure 4 shows that short-term absolute mean arterial pressure variability (i.e. standard deviation within half hours) showed a tendency to increase progress-

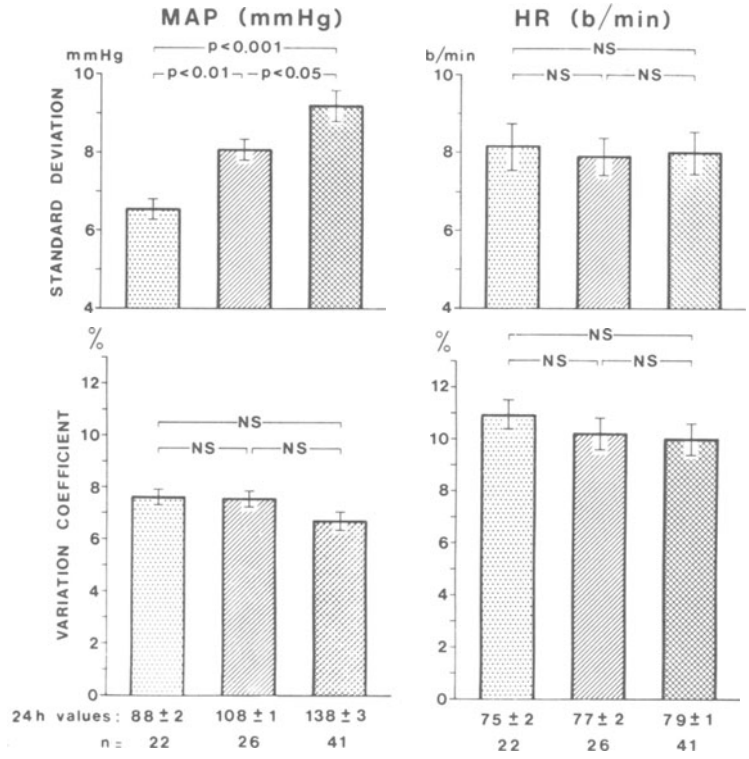

Fig. 4. Short-term mean arterial pressure and heart rate in 3 groups of subjects with normal blood pressure, moderate and severe essential hypertension. Figures at the bottom represent 24 hour average mean arterial pressure and heart rate values. Age was similar in the 3 groups.

ively to normotensive to severe hypertensive subjects. However, when relative variability was used instead, namely when variation coefficient was adopted to correct for differences in baseline blood pressure values, such tendency was lost and variability appeared to be similar in the various groups. This was the case for systolic and diastolic blood pressure, and also occurred when the analysis was performed on long-term blood pressure variability. Heart rate variabilities, either absolute or relative, also were similar regardless of the 24 hour mean blood pressure values. From the postulate outline above one might conclude that there is no alteration in central modulation of circulation in essential hypertension. This feature, and the reset but unimpaired baroreceptor-blood pressure control, might also allow to conclude for an overall normality of neual cardiovascular control in this condition. Whether this normality is the result of a secondary adjustment or truly reflects lack of participation of neural factors in the genesis of this disease remains to be clarified.

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RENOVASCULAR HYPERTENSION:

NEW ASPECTS OF PATHOGENESIS AND TREATMENT

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Although it is now nearly half a century since Goldblatt and his colleagues demonstrated that hypertension could be produced experimentally by the application of a constriction to a renal artery,[1] the pathogenesis of this condition remains imperfectly understood. The issue is clinically relevant, because in man hypertension is often associated with renal artery stenosis, and in a proportion of such patients, although by no means all, blood pressure can be lowered either by renal arterial reconstruction or by excision of the kidney distal to the stenosis. The importance of the renin-angiotensin system in initiating and maintaining renovascular hypertension remains particularly controversial.

In 1974 we proposed[2] a schema relating the evolution of renovascular hypertension to changes in the renin system. In the first phase, which appears within minutes of the application of a renal artery stenosis, plasma renin and angiotensin II, and arterial pressure, rise together, and fall, also in parallel, if the stenosis is relieved. Within days this is succeeded by a second phase in which, while blood pressure remains high, plasma renin and angiotensin II are proportionately less markedly elevated. This dissociation has cast doubt on the importance of the renin system in phase II, although blood pressure may still be lowered either by correction of the renal artery stenosis or removal of the affected kidney. Clinical renovascular hypertension is not often observed earlier than phase II. Much later a third phase supervenes in which blood pressure remains high while the renin system continues relatively suppressed; in phase III surgical measures are ineffective in lowering arterial pressure. Almost certainly the renin-angiotensin system is not pathogenically relevant in phase III. Clini-

cally, the distinction between phases II and III is of major importance in deciding on surgical measures.

Recent studies in this department[3] have attempted to define more precisely the evolution of renovascular hypertension in relation to the renin-angiotensin system. Rats with both kidneys remaining were studied, a unilateral renal artery clip being applied to one group, while control animals had a sham operation. In an attempt to minimise artifacts, blood samples for the assay of plasma renin and angiotensin II, and arterial pressure measurements, were obtained in conscious animals.

On the day after operation, when the first measurements were made, blood pressure, plasma renin and plasma angiotensin II concentrations were significantly elevated in the rats with unilateral renal artery stenosis. Two weeks after operation, however, plasma renin and angiotensin II had subsided in the clipped rats to values no different from those seen in the controls, although blood pressure remained significantly elevated. From the fourth week after operation onwards, plasma renin and angiotensin II again rose in the rats with unilateral renal artery stenosis, and to a very variable extent in different animals, while the hypertension became more severe. Blood pressure, renin and angiotensin II remained significantly elevated in the clipped rats up to 20 weeks from operation, when the study was terminated. A significant positive correlation was demonstrable ($r = 0.48$, $n = 21$, $p<0.05$) between plasma angiotensin II and arterial pressure in measurements made on the first and second days after applying the unilateral renal artery clip.

We have previously shown[4] similar related changes in blood pressure and plasma angiotensin II immediately after renal artery constriction in conscious dogs; in these dog studies an almost identical relationship between blood pressure and angiotensin II was obtained during acute intravenous infusions of exogenous angiotensin II. Thus it appears that in the first phase of renovascular hypertension, the blood pressure increase can be explained by the immediate rise in plasma renin, and hence the direct pressor effect of the resultant increase in plasma angiotensin II.

Between 8 and 20 weeks after clipping, in the rats,[3] endogenous plasma angiotensin II was also significantly correlated with arterial pressure ($r = 0.51$, $n = 47$, $p<0.001$), and the regression was no different in slope from that describing the similar relationship at 1 and 2 days after clipping. However, at 8-20 weeks, blood pressure was markedly higher for concurrent plasma angiotensin II than was the case 1 and 2 days after clipping.

These findings in the rat corroborate and extend our earlier observations in man.[2,5] In a series of untreated patients with hypertension associated with a renal or renal arterial lesion, a

significant positive correlation was found between endogenous plasma angiotensin II and arterial pressure; however, for any given value of plasma angiotensin II, blood pressure was distinctly higher than could be achieved by brief elevation of plasma angiotensin II during intravenous infusions of the peptide.

What are the possible factors involved in the upward shift of angiotensin II: blood pressure relationship during the evolution of renovascular hypertension? This changed relationship could well be independent of any alterations in the renin-angiotensin system. However, two observations raise the possibility that chronic but perhaps quite modest elevation of plasma angiotensin II might be responsible for resetting its own pressor dose-response curve.

First, in a patient with a renin-secreting tumour,[6] in whom there was chronic elevation of plasma angiotensin II, and in whom this increase was almost certainly the sole ultimate cause of the hypertension, a similarly enhanced angiotensin II : blood pressure relationship was seen. Removal of the tumour restored both plasma angiotensin II and blood pressure to normal. Second, infusion of angiotensin II into conscious dogs for 2 weeks, at a dose which elevated mean plasma angiotensin II only from around 25 to 50 pg/ml - well within the physiological range - caused a progressive rise in arterial pressure and advancing elevation of the angiotensin II: blood pressure dose-response curve.[7]

These considerations therefore sustain the possibility that the renin-angiotensin system could still be centrally involved in the second phase of renovascular hypertension. Raised plasma angiotensin II might both begin and maintain hypertension due to renal artery stenosis. Nevertheless, this remains far from certain and the resetting of the angiotensin II : pressor dose-response curve could have other causes. Several possible mechanisms of this resetting, some of which might be angiotensin II-dependent, have been considered.

First, Folkow[8] has emphasised the structural changes in arterial and arteriolar walls in hypertension, and has pointed out that an increased wall : lumen ratio can per se have a progressive pressor effect. Such structural alterations could be initiated and perpetuated by increased levels of angiotensin II.

Second, Cowley and DeClue[9] found that part of the pressure increase seemed to result from a rise in cardiac output, possibly as a consequence of decreased vascular compliance.

Third, angiotensin II has a variety of central and peripheral sympathetic nervous actions that might well potentiate its initial pressor effect.[10,11] These include an excitatory action on the area postrema; stimulation of the adrenal medulla and sympathetic ganglia; potentiation of postganglionic neurotransmitter biosynthesis

and release; and inhibition of neurotransmitter re-uptake.

Fourth, the prolonged infusion of angiotensin II at a low dose is accompanied by resetting of the baroreceptors.[9]

Fifth, chronic exposure of the adrenal cortex to increased levels of angiotensin II potentiates the aldosterone-stimulant effect of angiotensin II.[12] In renal hypertension in man there is evidence that the plasma aldosterone concentration is higher for a given plasma angiotensin II concentration than in normal subjects acutely infused with angiotensin II.[13] Such a phenomenon would require corresponding elevation of arterial pressure to balance the resultant tendency to retain sodium. Furthermore, the enhanced aldosterone level might lead to increased sodium accumulation in vascular walls that in turn could raise the wall : lumen ratio in resistance vessels, and also enhance the response to circulating vasoconstrictors. This effect on aldosterone might be relevant to renal hypertension in man, but it is unlikely to explain the progressive pressor effect of angiotensin II in the chronically infused dogs,[7] because in the latter plasma aldosterone was not significantly raised. However, in renal hypertension, an increase in plasma aldosterone concentration is not theoretically an obligatory requirement for a heightened tendency for sodium retention,[14] either in the entire body, or selectively in vascular walls, and hypertension can develop in adrenalectomised dogs maintained on constant replacement therapy.[15]

Sixth, Lucas and Floyer[16] have provided evidence of a hormone, of renal origin, that is responsible for altering tissue compliance, and whose release is inhibited in renal hypertension. It is possible that angiotensin II might modify the release of this hypothetical hormone.

In summary, it appears that circulating angiotensin II is entirely responsible, by acute vasoconstrictor effect, for the initial rise in pressure that follows renal artery constriction. Later, other mechanisms come into play. However, angiotensin II has undoubted pressor actions of slow onset, and these could, at least in part, be responsible for later phases of renal hypertension, both clinical and experimental; this remains unproved.

Sodium retention seems not to be a necessary accompaniment of evolving renovascular hypertension. In the one-clip two-kidney rat model, no differences in exchangeable sodium between hypertensive and sham-operated animals were seen up to 7 weeks after operation.[17] In man, renovascular hypertension shows indeed a tendency to sodium depletion, with a significant inverse relationship between arterial pressure and exchangeable sodium in a series studied by us.[18] With severe unilateral renal artery stenosis or occlusion, pronounced sodium depletion with secondary aldosterone excess may present as a striking hyponatraemic syndrome.[19]

If the renin-angiotensin system is involved in phase II of renovascular hypertension, antagonists and inhibitors of the system might be expected to correct the high blood pressure. The acute effects of agents such as saralasin - a competitive antagonist of angiotensin II - or captopril - an angiotensin I converting enzyme inhibitor - are consistent. With both types of drug an immediate fall in blood pressure is seen, in proportion to the pre-treatment plasma level of renin or angiotensin II.[20,21] In the case of captopril, the acute blood pressure fall is also in proportion to the acute fall in plasma angiotensin II.[21]

Although interesting, however, such immediate changes are not necessarily relevant to a slow pressor component of the action of angiotensin II, to unmask which more prolonged inhibition of the renin system might well be required.

Riegger et al.,[22] studying rats with one-clip two-kidney hypertension 28-60 days after operation, found that infusion of saralasin or of converting enzyme inhibitor for 11 hours slowly returned blood pressure to normal, while brief administration did not have this effect. This slow antihypertensive effect was not significantly related to pre-treatment plasma renin value.

The availability of orally-active converting enzyme inhibitors such as captopril and enalapril has permitted an evaluation of prolonged suppression of angiotensin II formation in renovascular hypertension in man.[23,24] Long-term administration of both of these agents has been shown to cause sustained reduction of plasma angiotensin II with converse increases in circulating renin and angiotensin I concentrations. The initial blood pressure reduction was proportional to the initial fall in angiotensin II but this early blood pressure change often related poorly with the long-term response.

Some severely hypertensive sodium-depleted patients had a very marked initial blood pressure fall,[19,23] while the long-term effect, after sodium balance was restored, was more modest. By contrast, other patients showed a gradual reduction in pressure over 1-3 weeks of continuous converting enzyme inhibition.[23] This latter type of response was observed in some patients whose pre-treatment plasma angiotensin II concentrations were within or just above the upper part of the normal range, and the effect might therefore have been due to reversal of the slow pressor component of angiotensin II. However, the converting enzyme inhibitors might lower blood pressure by mechanisms additional to suppression of angiotensin II formation, so that interpretation must, of necessity, be cautious.

Can long-term inhibition of the renin-angiotensin system aid in the selection of those patients with renal artery stenosis whose hypertension will respond to renal or renovascular surgery? In

particular, can such a measure help distinguish between phases II and III of renovascular hypertension?

The issue is an important and difficult one. As we have commented elsewhere[25] "..... patients whose blood pressure will fall most after surgery are likely to have hypertension of recent onset, to be young, to have fibromuscular hyperplasia, to have increased renin, angiotensin II and aldosterone in peripheral blood with high renal vein renin ratio, reduced blood flow in the affected kidney, and well-maintained flow in the untouched kidney. At the opposite end of the spectrum, surgery is likely to fail in elderly patients with long-standing hypertension caused by atheromatous renal artery stenosis, with other vascular disease, impaired renal function, reduced blood flow in the untouched kidney, a normal renin in peripheral blood, and a normal renal vein renin ratio. The decision on surgery is not difficult in extreme examples of this sort. In practice most patients fall between, with a mixture of favourable and unfavourable features." Is it possible that the long-term use of captopril or enalapril might summate the varied effects of all these factors?[23]

In a small series, we found the initial response to captopril a poor guide to eventual surgical outcome.[23] However the long-term captopril response related well with the later response to renal arterial reconstruction or nephrectomy, predicting, in absolute terms of systolic and diastolic pressure, successes and failures alike. Interpretation must necessarily be cautious with the few patients studied; if however, the early promise were fulfilled in larger series, it could have not only prognostic value but also important pathophysiological implications concerning the role of renin in pathogenesis.

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ALTERATION OF CELL MEMBRANES IN PRIMARY HYPERTENSION

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The past few years have witnessed a rapid accumulation of facts which give reason to believe that primary hypertension (essential hypertension in humans as well as spontaneous hypertension in Kyoto-Wistar rats) is causally associated to a wide-spread alteration of the cell membrane's regulation of intracellular free calcium and transport of univalent cations. In 1975-1978 we suggested that the hypertensive syndrome in primary hypertension is the manifestation of a certain type of membrane pathology[19-21]. Within a short period of time this hypothesis gradually began to gain ground. Below we present our data.

Erythrocytes

The first findings on the disturbance of the cell plasma membrane function of non-cardiovascular tissues in primary hypertension were obtained in studies of erythrocyte permeability. It was found that the rate constant of steady-state Na^+/Na^+ exchange through the erythrocyte membrane is increased both in spontaneous hypertension in rats (SHR, Kyoto-Wistar) and in essential hypertension[29,25,31]. These alterations are not connected with the Na-pump function[25,31], but are due to an increase of passive membrane permeability and/or to disturbances in the systems of counter-transport[3] and co-transport[7] of cations.

It is known that transmembrane transport of univalent cations is controlled by the concentration of free calcium in the cytoplasm[18,32]. This control is evidently achieved by changes in calcium content on the inner membrane surface[18]. Therefore, our further studies were directed to the study of intracellular calcium distribution. It was found that in both forms of primary hypertension the Ca-binding

ability of erythrocyte ghosts was reduced[26], which was due to a decrease in the number of calcium binding sites on the inner membrane surface[24]. Similar data for rats with spontaneous hypertension were also obtained by Devynck et al.[5] The technical difficulties in the determination of free calcium-concentration in the cytoplasm are well known. Quite recently Losse[10] laboratory developed microelectrodes which allowed to solve this problem for erythrocytes. It was found that free calcium concentration in the cytoplasm in erythrocytes of spontaneously hypertensive rats was 50-60 times higher than in normotensive control rats[10]. In erythrocytes this parameter is determined by the rate of calcium influx into cells along the electrochemical gradient and by the rate of Ca^{2+} efflux at the expense of Mg-Ca-ATPase activity[34]. The latter system is controlled by calmodulin[33].

In studies of the rate of ^{45}Ca influx into the erythrocytes of SHR no differences were found[5]. There were also no differences in the Mg-Ca-ATPase activity of erythrocyte membranes pre-treated with EGTA[30]. However, both in spontaneous hypertension[16] and in patients with essential hypertension (Table 1) the calmodulin effect on the rate of ^{45}Ca accumulation by the inside-out vesicles was significantly less than that of controls. In this connection it can be assumed that the alteration of calmodulin interaction with Mg-Ca-ATPase is the most probable cause of the Ca^{2+} concentration increase in the erythrocyte cytoplasm in primary hypertension.

Adipose Tissue

Compartmentation analysis of the kinetics of ^{45}Ca efflux was used in studies of calcium distribution in adipose tissue. In these experiments it was demonstrated that the content of intra-cellular exchangeable calcium is increased at the expense of a higher calcium content in mitochondria[23], both in spontaneous hypertension in rats[22] and in essential hypertension[27]. Calcium overload of adipose tissue in primary hypertension can probably be explained, as in the case of erythrocytes, by a decrease of the calmodulin-dependent component of Mg-Ca-ATPase activity of the adipocytes' plasma membrane. The presence of the defect itself in the plasma membrane of adipocytes is confirmed by the decrease of their Ca-binding ability[23] similar to that found in erythrocytes,

Vascular Smooth Muscles

The existence of alterations in the excitation-contraction coupling mechanism for vascular smooth muscle cells in hypertension was revealed from numerous experiments on the variation in their contractility, reactivity and sensitivity (see review[15]). Later on it was shown that these changes in SHR are due to a disturbance of

Table 1. Affinity of Ca-pump to Ca^{2+} (K_m) and its Maximal Activity (V_{max}) in Erythrocyte Membranes in Essential Hypertension

Groups	n	Calmodulin 1.2×10^{-7}	K_m (μM)	V_{max} nmol Ca (mg protein IOV)$^{-1}$ min^{-1}
1. Control (normotensive patients)	11	-	1.72 ± 0.09	5.32 ± 0.61
2. Control (normotensive patients)	11	+	0.33 ± 0.04	25.11 ± 1.17
3. Essential hypertension	10	-	1.68 ± 0.10	4.98 ± 0.50
4. Essential hypertension	10	+	0.52 ± 0.04	10.32 ± 0.71
$P_{1,3}$			non-significant	
$P_{2,4}$			0.001	0.001

Erythrocyte membranes were treated with EGTA at the stage of isolation. IOV - inside-out vesicles. n - number of examined patients.

univalent cation transport[8] and also, as in the case of erythrocytes, are not related to a disturbance in the Na-K-ATPase activity, but to an increase in the rate of cation passive transport[1]. Calculations by Jones[9] showed that these disturbances lead to a partial depolarization of the sarcolemma and to enhanced excitability of SHR vascular smooth muscle cells.

In addition to disturbances of univalent cation transport in cells of SHR vascular smooth muscles, changes in their Ca-binding and Ca-accumulating systems were also found. Decrease by 40-60% of the ^{45}Ca accumulation rate was observed for crude cell microsome fractions of SHR aorta smooth muscles[2]. In subsequent studies similar alterations were also described for the sarcolemma fraction, where the decrease of Ca-binding ability of SHR membranes was detected together with a reduction of the ATP-dependent accumulation of ^{45}Ca[17,35].

Synaptosomes

Already the alteration of the plasma membrane function in erythrocytes, in adipose tissue cells and in vascular smooth muscles indicates the wide-spread character of the membrane defect in both forms of primary hypertension. Moreover, the decrease of plasma membrane's Ca-binding was observed in rats with spontaneous hypertension in cardiomyocytes, hepatocytes and synaptosomes of the brain[6]. The latter observation is especially important since it indicates the possibility of changes in the mechanism of neurotransmitter secretion by nerve terminals, which largely accounts for increases of vascular tone in primary hypertension. To elucidate the state of plasma membranes in the cells of the central nervous system in spontaneously hypertensive rats, we studied transport systems of Ca influx and efflux in isolated synaptosomes and microsomes from the brain tissue. It is known that the activity of these systems determines the cytoplasm calcium balance and finally, neurotransmitters output and reuptake by nerve endings. These studies were carried out jointly with G. M. Kravtsov and N. I. Pokudin.

It was found that basal calcium uptake (i.e. calcium uptake in physiological concentrations of sodium and calcium in incubation medium) in SHR synaptosomes was 1.5-2 times higher than in normotensive Kyoto-Wistar rats (Figure 1). The differences remained during membrane depolarization obtained by lowering the extracellular K^+ concentration and disappeared in saturated concentrations of this cation. It can be assumed that the initial difference in Ca uptake is due to the existence of a constant partial depolarisation of the plasma membrane of nerve cells in SHR. The following data have been obtained to prove this assumption.

1. Calcium uptake by synaptosome of SHR and control rats does not differ if a specific inhibitor of potential-dependent Ca-channels (verapamil) is present in the incubation medium.

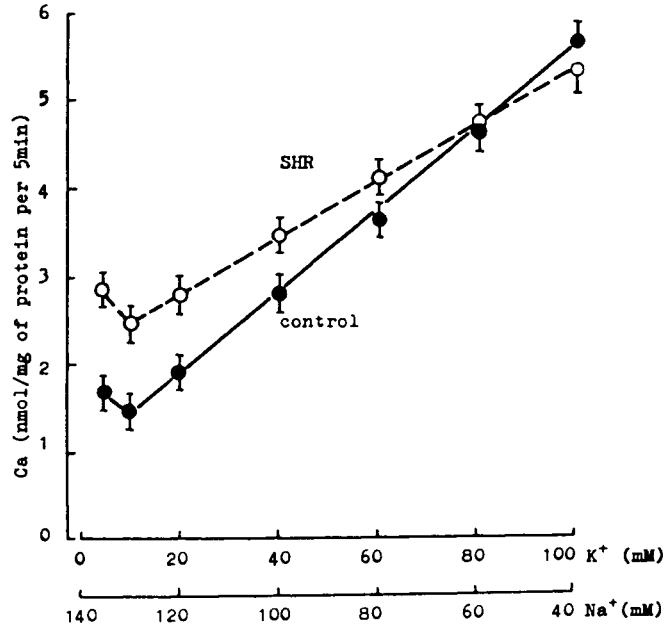

Fig. 1. Dependence of calcium uptake by synaptosomes in rats with spontaneous hypertension (SHR) and normotensive controls (NKWR) on the ratio of sodium and potassium concentration in the incubation media.

2. Synaptosome permeability for Na^+ (the most important factor for the formation of action potential in excitable tissues) is significantly increased in SHR as compared to normotensive controls (Figure 2).

In subcellular brain tissue fractioning, besides the mitochondria, one more membrane fraction with ATP-dependent calcium uptake ability was found. According to the distribution of marker enzymes (presence of Na-Ca exchange system, K^+-dependent Ca^{2+} uptake and calmodulin effect on ATP-dependent calcium uptake), this fraction was characterized as a fraction of plasma membrane fragments of nerve cells and their dendrites. In studies of the dependence of the ^{45}Ca uptake rate of this fraction on Ca^{2+} concentration in the incubation medium (Figure 3), it was found that the maximal rate of ^{45}Ca uptake in SHR was 40% lower than in the controls. It should be emphasized that, similarly to the Ca-transporting erythrocyte system, the calmodulin effect on Ca^{2+} transport in the plasma membranes in SHR's brain was significantly lower than that of normotensive animals (Figure 3).

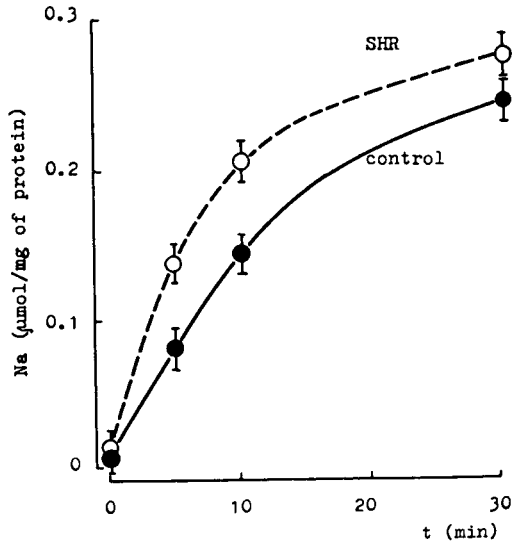

Fig. 2. The kinetics of sodium uptake by synaptosomes in rats with spontaneous hypertension (SHR) and normotensive control rats (NKWR).

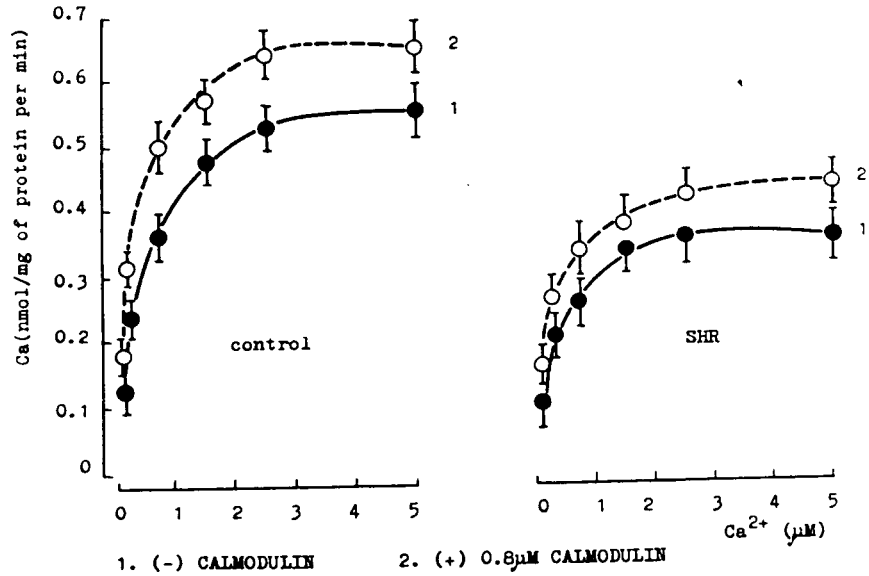

Fig. 3. Dependence of the rate of calcium accumulation by microsome from the brain of spontaneously hypertensive rats (SHR) and normotensive control rats (NKWR) on calcium concentration. 1) without calmodulin; 2) +0.8 uM calmodulin.

The presented data give reasons to suggest that free calcium concentration in the cytoplasm of nerve terminals in SHR is increased. This may serve as one of the main causes of the increased activity of peripheral part of the sympathetic nervous system in SHR and, evidently, in essential hypertension in humans as well.

Molecular Mechanisms of Membrane Defect

The first attempts to study this problem were started simultaneously in our laboratory and in the laboratory of Prof. Meyer[11] by the technique of fluorescence probes. Using diphenyl-hexatrien, whose fluorescence polarization is determined by the rate of rotational diffusion of molecules in the hydrophobic regions of the membrane, an increase of microviscosity of these parts of erythrocyte membrane was observed in rats with spontaneous hypertension[11,12]. A reduction in the rate of lateral diffusion of molecules (especially significant in the region of protein-lipid contact) was found by means of another probe - pyrene both in spontaneous hypertension in rats[12] and in essential hypertension in humans[14]. Now some data are available indicating that the defect of structure of plasma membrane is not limited to erythrocytes, but also manifests itself in the plasma membranes of hypatocytes, synaptosomes and cardiomyocytes[4]. It can be assumed that the defect in the structure of the plasma membrane is the cause of disturbances of its cation-binding and cation-transporting function.

Conclusion

The present paper contains a short review of data on the studies of cell membranes in primary hypertension. These data confirm a wide-spread, if not universal, alteration of cell plasma membranes in primary hypertension, both in rats and in man. In excitable and non-excitable cells alterations in plasma membrane structure, alteration of the membrane's permeability to monovalent cations, decrease of its Ca-binding ability in combination with a disorder of the Ca-transporting systems were found. This is the basis of the membrane defect, which is manifested in the insufficiency of membrane control over free calcium concentration in the cytoplasm.

The membrane defect was observed at stages previous to the development of the hypertensive syndrome[25]; it was not detected in secondary hypertension both in experimental models[13,26] and in patients with chronic renal hypertension[14]; there was no dependence of the membrane defect on the activity of the sympathetic nervous system[28] and the secretory function of the adrenal cortex[22,25].

It should be noted that the wide-spread (not limited to one type of cell) membrane alteration in primary hypertension and its

obvious genetic origin are indicative of the fact that the initial inherent defect of the genetic apparatus concerns the few basic genes unaffected by repression during tissue differentiation.

As for the pathogenesis of hypertension, the following consequences of the alteration of cell membranes should be taken into consideration:

1. the disturbance in the excitation - contracting coupling in contractile cells of the cardiovascular system;

2. the alteration of Ca transport in the neurolemma (presynaptic membrane), indicating the presence of an increased free calcium concentration in nerve endings, intensifying neuromediator quantation and changing synaptic transmission.

3. the change in cell calcium distribution in tissues which are the target for the sympathetic nervous system, serving as the basis for the alteration of its functional status (since both sensitivity a reactivity of such targets are changed).

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HIGH DENSITY LIPOPROTEINS AND ATHEROSCLEROSIS

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The change in the ratio between lipoprotein (LP) concentration with the density higher or lower than 1.063 g/ml was first observed in patients with ischemic heart disease as early as 1951[1]. Later Gofman et al.[2] also pointed to the increased blood concentration of low density lipoproteins (LDLP) and decreased content of high density LP (HDLP) in patients with clinical atherosclerosis. However, at that time attention was paid to the increasing level of LP in atherosclerosis leaving in shade the low levels of LP.

In 1970 T. N. Lovyagina and E. B. Ban'kovskaya[3] found that in man or animals (rabbit, guinea-pig, pig, monkey) with spontaneous or easily induced experimental atherosclerosis greater part of plasma cholesterol is contained by LDLP or very low density lipoproteins (VLDLP). In case of animals (suslik, rat, cat, dog) which never suffer from spontaneous atherosclerosis and are resistant or immune to experimentally induced atherosclerosis the major part of cholesterol is contained by HDLP. It is characteristic that such cholesterol distribution is preserved in the above mentioned animals (rat excluded) fed cholesterol-rich diets. It is interesting to note, that in case of cholesterol fed susliks the plasma cholesterol concentration level amounted to extremely high figures (more than 700 mg/dl), but the atherosclerosis did not develop in these animals since most of the cholesterol taken up by the organism was used to form HDLP. Soret et al.[4] reported on the lame attempt to induce atherosclerosis under experimental conditions in chinese hamsters, probably, also due to high HDLP blood level.

In 1975 G. Miller and N. Miller[5] reported that in patients with coronary atherosclerosis the level of HDLP cholesterol was evidently lower and that this might be important in pathogenesis.

Further clinical and population studies carried out in many countries of the world, due to the simplicity and availability of the CD-HDLP determination method, showed the lower cholesterol content of HDLP in ischemic heart disease (IHD) patients than in patients without any IHD signs (see review N 6). The often observed difference in the CS concentrations of HDLP of 3-6 mg/dl proved to be statistically significant. Thus, the population study of more than 7000 male sex residents of Moscow and Leningrad, aged 40 to 59, with signs of IHD (according to ECG at rest and Rose questionnaire) showed that the mean level of CS-HDLP amounted to 49.6 ± 17.2 mg/dl, while in persons without IHD it amounted to 53.5 ± 17.2 mg/dl ($p < 0.001$). So the difference is 3.9 mg/dl.

In line with the published data, HDLP cholesterol as an IHD predictor proved to be eightfold more sensitive than the total CS concentration and fourfold more sensitive than LDLP cholesterol[7]. We have suggested to calculate the so-called cholesterol coefficient of atherogeneity as the "predictor" which is represented by the following ratio[8,9]:

$$C = \frac{\text{LDLP cholesterol + VLDLP cholesterol}}{\text{HDLP cholesterol}}$$

In clinic it is more convenient to calculate this coefficient by determining the total and HDLP cholesterol:

$$C = \frac{\text{Total CS - HDLP CS}}{\text{HDLP CS}}$$

The higher the coefficient is (in healthy persons it does not exceed 3 units), the higher is the danger of IHD. In IHD patients the value of the coefficient often amounts to 5-6 and more units.

If at the beginning of the study of the physiologic role of HDLP they were termed "non-atherogenic" LP contrary to LDLP and VLDLP, there are all grounds now to consider them "anti-atherogenic". In this context, the mechanism of antiatherogenic HDLP action assumes special concern.

ON ANTIATHEROGENIC HDLP ACTION

At present, there is no unified theory to explain the antiatherogenic action of HDLP or their subspecies.

One of the first viewpoints on this question[10,11] assumed the competition between HDLP and LDLP for the receptors on the cell surface and the higher the blood HDLP concentration is the fewer LDLP (and consequently less CS) penetrate into the cell. However, HDLP affinity for cell receptors appeared to be on the whole 200-fold lower as compared to that of LDLP[12]. Yet this affinity increases with the increase of apoprotein E concentration in HDLP and it is represented to a greater extent in $HDLP_c$ (c means cholesterol induced)[13].

The following facts may be opposed to the attempt aimed at explaining HDLP antiatherogenic action by their competition with LDLP for cell receptors. It is known that the receptor-bound cellular uptake to LDLP helps the blood LP homeostasis[12]. Receptor function disorders result in the increase of CS and LDLP blood levels. Thus, the competitive LDLP prevention from the receptor-bound cellular uptake must lead to the development of atherosclerosis rather than to its inhibition as it occurs in the hereditary receptor insufficiency[12].

Moreover, as the studies by Miller[14] have shown the increase instead of the inhibition of LDLP uptake by cells through the receptor mechanism which followed the preincubation of human skin fibroblasts with HDLP. Cholesterol removal from the cells during their preincubation with HDLP stimulated the receptor synthesis and the uptake of additional amounts of LDLP.

Unfortunately, there are no data as to the HDLP influence on non-specific endocytosis LDLP uptake by the cells, particularly, by endothelial cells, in other words the effect on the process which is likely to promote the unregulated LDLP and CS accumulation in the endothelial and intimal cells.

In 1968, Glomset[15] presupposed that HDLP are involved in the removal of excess CS from the peripheral tissues and its transport to the liver for subsequent oxidation to bile acids. If we combine this hypothesis with receptor hypothesis of Goldstein and Brown[16], there appears the general pattern of CS transport into the cell by VLDLP-LDLP system and the reverse CS transport by HDLP (Figure 1). Evidently, at the insufficient blood level of HDLP they fail to remove CS from the cell.

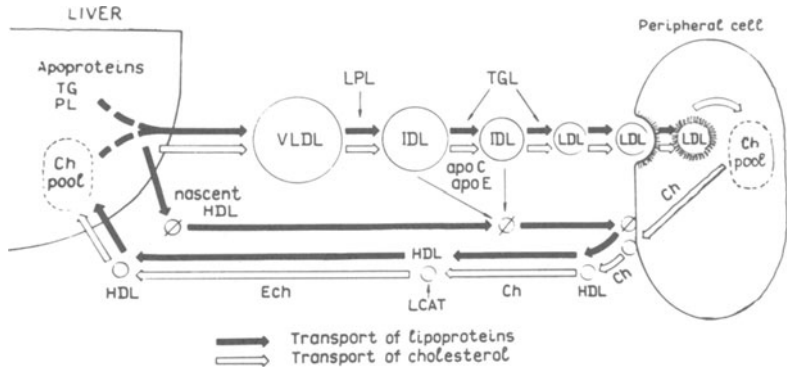


Fig. 1. Transport of cholesterol from the liver into the peripheral cell and the reverse cholesterol transport.

Analogously, the antiatherogenic HDLP action may be assumed to result from partial cellular uptake of CS by these LP due to their easy penetrability into the arterial wall, and CS removal through the paravasal system of adventitia[8]. This conception is supported by the studies showing that as a result of the interaction in vitro between HDLP and CS the former are capable of dissolving CS crystals[17], and due to the interaction with liposomes[18] and erythrocytes[19] they are capable of removing from them a part of CS. It is significant that HDLP fractions accepting a part of CS from the erythrocyte membrane, augmented in size[19]. It is likely, that in the studies mentioned above the main acceptors of CS were $HDLP_3$ and the formed large particles - $HDLP_2$. Further, the data obtained in our laboratory[20] showed that during incubation of HDLP with intima isolated from human aorta afflicted by atherosclerosis the enrichment of $HDLP_3$ and to a much lesser extent of $HDLP_2$ by non-esterified CS took place. So, one of the HDLP subspecies, namely $HDLP_3$, is capable of binding additional amounts of CS. Alongside this it is significant that being saturated with non-esterified CS, $HDLP_3$ fractions became lighter and were translated to $HDLP_2$-like particles. We cannot still define to what extent the capability of $HDLP_3$ to bind non-esterified CS is manifested in vivo, though it is worth mentioning the study of Sarma et al.[21] who showed that

in perfusion of pug coronary artery by homologous plasma with the addition of HDLP to it, inhibited the penetration of the labeled CS into the arterial wall.

Another possible mechanism of antiatherogenic HDLP action is connected with the fact established by Nikkila[22]: at high HDLP blood level the highest rate of the lipolysis of triglyceride (TG)-rich lipoprotein is observed. This is in accord with many studies showing the reciprocal correlation between the HDLP level (or CS HDLP) and TG VLDLP. A decrease in the content of the latter in blood caused by various means (the lowering of body mass, physical exercise, chlorphibrate intake) leads to the increase of HDLP level. All these data served as the ground for the assumption and then demonstration of direct VLDLP-HDLP interaction[23]:

$$\text{VLDLP} + \text{HDLP}_3 \xrightarrow{\text{lipoproteinlipase}} \text{HDLP}_2 + \text{LDLP}$$

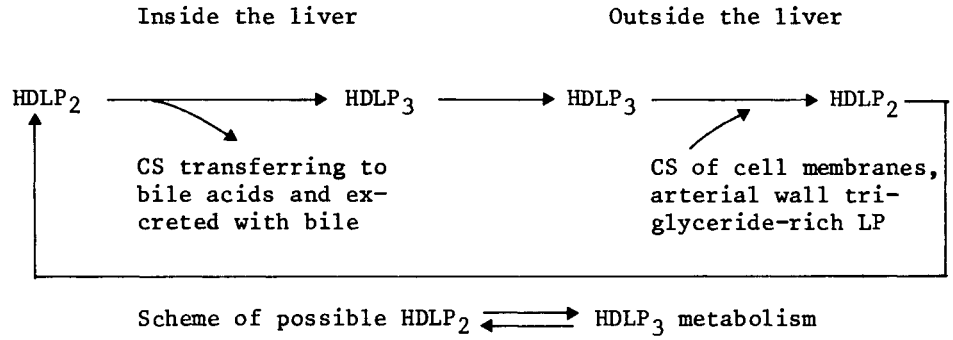
As a result of this reaction catalyzed by lipoproteinlipase the decrease of the VLDLP level is observed. Is it of any importance to the organism? VLDLP according to Zilversmit[24], penetrates into the arterial wall primarily as highly atherogenic remnant particles.

In line with our data[25] among all the LP species VLDLP have most pronounced autoantigeneity leading to the formation of the autoimmune complex VLDLP-IgG circulating in blood. Like all other autoimmune complexes it may afflict the endothelium initiating the development of atherosclerotic process. In line with the above mentioned, it is possible to assume that the decrease of the VLDLP level favors the prevention of atherosclerosis. However, the reaction mentioned above results in the formation of LDLP which according to generally accepted viewpoints are the most atherogenic. So it is difficult to determine whether or not the reaction plays a protective role in atherogenesis. Nevertheless, it is likely to assume that the formed $HDLP_2$ are more antiatherogenic than $HDLP_3$. In connection with such an assumption there are some population and clinical data. It is known, for instance, that the difference in HDLP blood levels in men and women concern mainly $HDLP_2$. Long-distance runners have an increased level of $HDLP_2$ instead of $HDLP_3$. Just the same increase of the $HDLP_2$ level is observed with the administration of drugs decreasing the TG blood level. Namely this LP subspecy, contrary to $HDLP_3$ increased in hyper-α-lipoproteinemia. On the other hand, the increase in the $HDLP_2$ level may be treated as a result of the antiatherogenic action either of nascent (disco-like) HDLP or other products on the way of formation of native HDLP or, finally, $HDLP_3$. Similar conception was put forward by Assman[26], for instance. It is also probable that $HDLP_2$ and $HDLP_3$ extracted by preparative ultracentrifuging, are not quite

equal to those HDLP circulating in blood. No doubt, additional studies are necessary to determine the most antiatherogenic subspecy of HDLP.

As was noted above, $HDLP_3$ are capable of non-esterified CS binding. It can be CS from cell membranes, CS settled extra-cellularl in the intima of arterial wall, or either CS released upon the hydrolysis of triglyceride-rich LP in plasma. Evidently, physico-chemical composition and structure of $HDLP_3$ particles lead to such an uptake of CS irrespective of its source. As $HDLP_3$ with taken CS transfer to $HDLP_2$ or the similar particles the process of $HDLP_3$ regeneration must occur in the organism. Nikkila et al.[27] assume, that the inverse process of $HDLP_2$ metabolism to $HDLP_3$ takes place in the liver involving the hepatic lipase in it.

The whole cycle of HDLP metabolism is in general represented by the following scheme:



Scheme of possible $HDLP_2 \rightleftharpoons HDLP_3$ metabolism

In conclusion we would like to mention our lame attempt to inhibit the development of experimentally-induced atherosclerosis in rabbits by multiple intravenous administrations of heparologic (horse) HDLP[28]. Despite the fact that on the whole within 10 weeks each animal received 58 intravenous injections with 3143 mg of HDLP (as protein), the experimentally-induced atherosclerosis was not inhibited, but on the contrary its development intensified. It is characteristic that in animals injected with HDLP the level of total CS and especially of TG was low. By the end of the experiment the plasma of the animals injected with HDLP was transparent (probably, due to the lower content of VLDLP), whereas plasma of the control animals was turbid. Probably in these experiments the effect of immunologic factors was clearly seen, conditioned by the manyfold administration of heterologic HDLP which was followed by the intensification of atherosclerosis. HDLP extracted from plasma were likely to have weak antiatherogenic action either because of the lack of nascent HDLP among

them or the loss of antiatherogenic properties during isolation (LCAT etc.).

Finally, it is worth noting that rabbits possess extremely low activity of hepatic lipase (triglyceride lipase) in blood both basal and postheparin[29]. If this enzyme is involved in the $HDLP_2 \longrightarrow HDLP_3$ metabolism in the liver[27], one may assume that the low rate of metabolism in the rabbit liver does not lead to enough regeneration of $HDLP_3$ and their uptake by the blood. Probably, this leads to the easy development of experimentally induced atherosclerosis in rabbits.

We failed to delay the development of the experimentally induced atherosclerosis in rabbits by administrating homologous HDLP (unpublished data). In these experiments within 10 months each animal was subjected to 60 intravenous injections of HDLP, isolated from plasma of a great amount of healthy rabbits with the density ranging from 1.063 to 1.210. Each rabbit received totally 290 mg HDLP (as protein). As upon the administration of heterologous LP, the evident decrease of the total CS and TG content was observed in plasma of animals.

Though the experiments on the delay of the development of experimental atherosclerosis were not a success the obtained results showed that artificially induced increase in the blood level of HDLP in rabbits by multiple intravenous administration of heterologous homologous HDLP leads to the decrease of the total content of CS and TG which is in accord both with the above scheme put forward by Patsch et al.[23] and with general ideas about the scavenger activity of HDLP - i.e. the ability to utilize and accelerate CS catabolism and lipids upon the lipolysis of triglyceride-rich LP in blood[30].

The antiatherogeneity of HDLP is accepted to depend on the concentration ratio of $HDLP_3$ and $HDLP_2$ in it, free and esterified CS, phospholipids, apoproteins as well as the activity of LCAT and lipoprotein lipase in plasma and lipase in the liver.

The following hypothesis attempting to explain antiatherogenic HDLP action should be noted: inhibitory effect of HDLP on proliferation of aortic smooth muscle cells induced by LDLP[31]; inhibiting of the synthesis of glycosaminoglycans in aortic smooth muscle cells[32]; solubilization of glycosaminoglycan-LDLP complexes in arterial wall by HDLP[33]; the decrease in the thrombogenic LDLP action[34]; stimulation of the oxidation of CS to bile acids in the liver[35] etc.

CONCLUSION

There is yet no unified theory to explain the role of HDLP in the pathogenesis of atherosclerosis. The theories and hypotheses put forward consider the atherogenic action of HDLP at different physio-

logic levels - individual cell, arterial wall and plasma. Special attention is paid to the possible involvement of HDLP in CS removal from tissues, interference of HDLP into cellular uptake of CS and interrelation between HDLP and triglyceride-rich lipoproteins in plasma.

No matter what kind the atherogenic action of HDLP, of their subspecies or precursors was, we may consider as clearly established the low blood level of HDLP which reflects the state promoting the development of atherosclerosis and ischemic heart disease while the high blood level inhibits them. Probably, dislipoproteinemia characterized by both high blood level of LDLP or VLDLP and low blood level of HDLP, is the most dangerous for the organism.

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REGULATION OF PLASMA HIGH DENSITY LIPOPROTEIN CONCENTRATION BY LIPOPROTEIN LIPASE AND HEPATIC ENDOTHELIAL LIPASE

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A number of epidemiologic and angiographic studies have established that the risk of developing ischemic heart disease (IHD) and the extent of coronary atherosclerosis bear a direct relationship to plasma LDL cholesterol concentration but are inversely correlated to HDL cholesterol.[1,2] The latter association seems to be mostly mediated by the HDL_2 subfraction.[3] The question on the possible additional role of elevated plasma triglyceride and VLDL levels in increasing the risk of IHD has remained controversial.[4] Many authors agree that hypertriglyceridemia is common among patients with IHD and also predicts the risk of developing new IHD[5,6] but multivariate analyses considering also plasma HDL have consistently eliminated triglycerides as an independent risk factor.[4] In other words, elevated VLDL indicates an increased risk of IHD only because of its close inverse association with HDL. Thus, high VLDL is not atherogenic if not accompanied by low HDL. It is also evident that the significance of HDL as a protective factor of IHD increases with increasing LDL cholesterol concentration. Accordingly, among populations with low LDL levels (usually due to diet) a simultaneous low HDL cholesterol is not atherogenic.

Many of the factors which are known to be positively or negatively associated with IHD are also related to plasma HDL cholesterol and influence it in such direction that HDL can be thought to be the biochemical link between the primary risk factor and the IHD. To mention a few examples, it is well documented that alcohol, estrogenic hormones and endurance exercise all increase the plasma HDL and decrease the risk of developing IHD whereas smoking, overweight, low physical activity and progestin-containing oral contraceptive pills tend to decrease HDL and increase the incidence of IHD.

The mechanism(s) by which the HDL or HDL_2 (or even a subfraction of the latter particle) inhibits the accumulation of lipids into arterial intima is not completely understood. Of plasma lipoproteins only HDL can promote efflux of cholesterol from cultured cells in vitro[7] and it is currently believed that the "reverse transport" of cholesterol from extrahepatic tissues to liver is the function of HDL which best accounts for its antiatherogenic effect.

Irrespective of the underlying mechanism the strong evidence relating low HDL cholesterol to atherosclerotic heart disease has raised major clinical interest for factors regulating plasma HDL (HDL_2) levels and for possibilities to increase low HDL by dietary or pharmacologic measures. It has been established that the metabolism of HDL particles is a complex process involving enzymes, transfer proteins and nonenzymatic exchange reactions most of the events occurring in circulating blood. This paper will review only the role of two endothelial lipolytic enzymes in the regulation of HDL levels. These are lipoprotein lipase and hepatic lipase (known also as hepatic endothelial lipase to avoid mixing with other lipases of the liver).

LIPOPROTEIN LIPASE AND PLASMA HDL (HDL_2)

Lipoprotein lipase (LPL) is a lipolytic enzyme bound to endothelial surface of capillaries in extrahepatic tissues, mainly adipose tissue, skeletal muscle and myocardium. It is released by heparin and can be assayed in postheparin plasma or in heparin eluates of tissues in vitro. The enzyme hydrolyzes triglycerides in chylomicrons and VLDL. During the course of this hydrolysis the surface phospholipids, cholesterol and apoproteins of these lipoproteins become fused with HDL increasing the total HDL mass and causing a shift from HDL_3 to less dense HDL_2 particles.[2] The rate of both hydrolysis of triglyceride-rich particles and of the formation of HDL or HDL_2 is dependent on the activity of LPL which enzyme thus is in key position in the metabolism of all these lipoproteins. In the presence of LPL deficiency (genetic or experimentally induced by specific antiserum) chylomicron and VLDL concentrations are much elevated while HDL is reduced to very low level and increases upon reactivation of LPL.[9] In normal human subjects the concentrations of total HDL cholesterol and of HDL_2 cholesterol and phospholipid are positively correlated with LPL activity measured either from tissue eluates or from postheparin plasma.[10,11] In contrast, no such relationship can be found between HDL_3 cholesterol and the LPL activity.[11] The metabolic linkage between VLDL and HDL at the LPL reaction explains well the known inverse correlation between the plasma concentrations of these two lipoproteins.

Many of the known influences on HDL cholesterol levels by physiological variables, pharmacologic agents or disease states can

be accounted for by corresponding changes in LPL activity. A number of these factors are listed in Table 1.

Following the puberty women have higher levels of HDL than males at all ages. The sex difference is present only in the HDL_2 subfraction. Women have also higher LPL activity than men in adipose tissue and in postheparin plasma but not in skeletal muscle.[10] Aerobic type exercise training increases both LPL activity of tissues and plasma HDL[12,13] whereas anaerobic training does not influence either LPL or HDL.[12] During immobilization both HDL_2 and HDL_3 are decreased[14] and it is possible that lack of physical activity is a major underlying cause of the low HDL levels reported in patients with ischemic cerebral disease or claudication and other conditions which restrict mobility. It is possible that also the IHD patients of prevalence studies have shown reduced HDL levels because of poor physical activity.

The low HDL cholesterol associated with obesity is hardly explained by a decreased LPL activity or low removal of VLDL. Even though the LPL activity in adipose tissue of obese subjects is at low side the total body LPL activity is normal or even increased. The production rate of VLDL triglycerides is increased in obesity but the turnover of non-triglyceride components of VLDL may be normal or low thus leading to reduced transfer of material from VLDL to HDL. For a similar reason the production of HDL through the lipolytic pathway may be diminished during low fat - high carbohydrate diet when little chylomicrons are being synthesized and therefore the HDL level falls.

HEPATIC ENDOTHELIAL LIPASE AND PLASMA HDL (HDL_2)

Hepatic lipase (HL) is a phospholipase located at the endothelial cells surrounding the liver sinusoids. It is probably bound to the luminal surface in a similar fashion as the LPL since it is also released by heparin and can be determined in postheparin plasma. This enzyme was formerly thought to be active in the hepatic uptake of chylomicron remnants but more recent findings of several laboratories have favored the view that the physiological function of HL is hydrolysis and uptake of HDL_2. The evidence is based on following data. The HDL and HDL_2 cholesterol and phospholipid concentrations of experimental animals are increased after blocking the HL activity in vivo by a specific antiserum.[15,16] Purified HL hydrolyses HDL_2 phospholipids in vitro at a much higher rate than phospholipids of other lipoproteins.[17] In normal human subjects an inverse correlation can be demonstrated between the HDL_2 and the postheparin plasma HL activity.[18]

Some of the variations of HDL and HDL_2 cannot be related to changes of LPL but seem to be associated with alterations of HL activity. These are listed in Table 2.

Table 1. Factors and conditions modulating plasma HDL or selectively HDL_2 levels presumably by changing lipoprotein lipase activity

Variable	Lipoprotein Lipase	HDL or HDL_2
Sex: Female/Male	High/Low	High/Low
Endurance training	Increase	Increase
Immobilization	Decrease?	Decrease
Regular use of alcohol	Increase	Increase
Caloric restriction	Decrease	Decrease
Clofibrate	Increase	Increase
Nicotinic acid	Increase	Increase
Probucol	Decrease	Decrease
Interferon	Decrease	Decrease
Insulin deficiency (diabetes)	Low	Low
Insulin resistance (diabetes, obesity)	Normal or low	Low
Insulin treated diabetes	Normal or high	Normal or high
LPL deficiency or apo C-II deficiency	Very low	Very low
Type 4 or 5 hypertriglyceridemia	Normal or low	Low
Uremia	Low	Low
Septic conditions, exdotoxins	Low	Low

The HL is evidently regulated by sex steroids. Women have significantly lower HL activity than men which difference may contribute to the higher HDL levels observed in women. Upon administration of estrogens the HL activity falls and the HDL is increased. Progestins with androgenic activity have an opposite effect increasing HL and decreasing the HDL and HDL_2 levels while medroxyprogesterone acetate which is weakly androgenic does not significantly influence HL activity and causes also little change in HDL concentration.[19] An anabolic steroid oxandrolone is a powerful inducer of HL[20] and its administration is followed by decrease of HDL.[21]

THE IMPORTANCE OF THE LPL AND HL IN TRANSPORT FUNCTION OF HDL

In spite of some similarities between LPL and HL the two enzymes appear to be regulated independently and to function separately without any mutual feedback system. In such situation where two enzymes responsible for production and removal of one metabolite vary independently, it may be difficult to demonstrate correlations between the concentration of the metabolite and the activity of either enzyme. This can be done either by selecting subjects who have almost similar activity of one of the enzymes but show a wide range of variation in the other. It can be shown that the correlation coefficient between HDL and LPL in different cohorts increases with decreasing variability of HL activity. For example, in obese subjects who show a very wide range of HL activities there is no significant correlation between LPL activity and HDL level whereas in hypothyroid patients who have a consistently low HL activity the correlation between LPL activity and HDL cholesterol concentration is of the order of + 0.85. Analogously, comparing individuals at similar LPL activity level a rather close correlation between the HDL_2 concentration and HL activity can be revealed.

It is important to realize that HDL and HDL_2 levels are determined by several factors other than the activity of the two enzymes. However, both LPL and HL appear to be important for the cholesterol transport function of HDL and for development of atherosclerosis. We have previously formulated a hypothesis on HDL cycle.[22] This concept involves, (1) secretion of new HDL particle by the liver, and rearrangement in circulation to HDL_3, (2) addition of phospholipid, protein and cholesterol to this (HDL_3) as products of breakdown of triglyceride-rich lipoproteins by LPL, (3) uptake of cholesterol from peripheral cells by the phospholipid-enriched HDL_2 (not excluding a similar function for HDL_3) and, ultimately, (4) degradation and uptake of HDL_2 by HL in the liver with possible (not established) re-entry of HDL particle (HDL_3?) into circulation and excretion of cholesterol into bile.

It seems that a high activity of LPL is advantageous in leading to accelerated production and increased concentration of HDL and

Table 2. Factors causing reciprocal changes in plasma HDL or HDL_2 and hepatic lipase activity

Variable	Hepatic lipase	HDL or HDL_2
Sex: Female/Male	Low/High	High/Low
Exercise	Decrease	Increase
Obesity	High	Low
Estrogenic hormones	Decrease	Increase
Progestational hormones	Increase	Decrease
Anabolic steroids	Increase	Decrease
Androgens	Increase?	Decrease

HDL_2 but it is more difficult to conclude whether a high HL is also antiatherogenic by increasing the hepatic uptake of HDL_2 cholesterol but by simultaneously leading to decrease of HDL concentration. It remains for future studies to explore whether there are two separate entities of hypo-HDL-emia, one caused by low production rate and another by a rapid removal of HDL, and whether these have different impact on atherosclerosis.

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PECULIARITIES OF COMPOSITION AND PROPERTIES OF HIGH-DENSITY LIPOPROTEINS IN PATIENTS WITH CORONARY ATHEROSCLEROSIS

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Epidemiological studies in various countries of the world have shown that lowering of HDL Ch level is a risk factor for coronary heart disease.[1,2,3,4] It has been found that the incubation of HDL with a smooth muscle cell culture results in the decrease of Ch level in the latter, while HDL Ch level increases.

At present an opinion has formed that antiatherogenic HDL properties are linked to the ability of these blood plasma proteins to remove Ch excess from membranes, including the membranes of smooth muscle and endothelial cells.[4,5] This unique property of high density lipoproteins is due to at least two characteristics of their composition.[6,7]

Firstly, HDL contain much more protein and phospholipids than other lipoproteins;

Secondly, apo-A-I apoprotein accounts for 70% of the protein while in other lipoproteins only traces of it can be found.

About 70% of all HDL phospholipids at normal HDL Ch are made up by lecithin and 12% - by sphingomyelin.

In model systems lecithin liposomes effectively bind cholesterol; that is also characteristic of apo-A-I, and the degree of binding increases if lecithin is added.[8]

It was also demonstrated that spirality of A-I apoprotein increases from 55 to 69%[9] if it is reconstructed with lecithin.

The results of these model experiments made it possible to assume that organization of HDL particles and performance of their antiatherogenic functions may also depend on phospholipid composition, amount of apo-A-I, and interaction of apo-A-I and phospholipids in the surface monolayer of the particles.

However, to perform continuously their cholesterol acceptor functions HDL need free Ch esterification. Free Ch comes to the surface monolayer from the membranes of smooth muscle and endothelial cells as well as from VLDL in the course of their catabolism in blood plasma. During esterification fatty acid is transferred only from lecithin to cholesterol by lecithin-cholesterolacyltransferase (LCAT).[10] The enzyme is activated by apo-A-I.

HDL consist of two main subclasses - HDL_2 and HDL_3. During the incubation of HDL_3 with lecithin and/or apo-A-I free Ch is esterified and particles of HDL_2 density are formed.[11]

It has been shown in a number of studies including our own that a reduction of HDL Ch in the blood plasma of patients with coronary heart disease is usually due to the decrease of HDL_2 Ch.[12,13] That is especially important since Ch esters contained by HDL_2 are transported into the liver for subsequent excretion from the organism. Though, molecular mechanisms responsible for the reduction of HDL antiatherogenic properties remained unclear.

Thus, we were interested whether in HDL_2 and/or HDL_3 of CDH patients with low HDL Ch there are:

1) any peculiarities is phospholipid composition
2) differences in apo-A-I amount;
3) changes in physico-chemical parameters and, specifically, in the surface monolayer fluidity of the particles, and whether
4) activity of LCAT-reaction, which transforms free Ch into esters and facilitates the transformation of HDL_3 into the particles of HDL_2 density, is changed in the plasma.

Finally, in connection with the data on the interplay between the platelet Ch content and their ability to aggregate, we tried to find out whether:

5) aggregational characteristics of platelets change after the incubation with HDL.

To clarify these questions all the patients observed (n=60) were divided into two groups: with normal and low HDL Ch. In both groups total plasma Ch and TG levels were within the limits of normal values. Reduction of HDL Ch was accompanied by the stat-

istically significant increase of plasma TG, though their level remained within normal values.

In each of these groups we determined the amount of main phospholipids (lecithin and sphingomyelin), apo-A-I content in HDL_2 and HDL_3, and the activity of LCAT-reaction according to Stokke and Norum.

In patients with coronary atherosclerosis as compared with the control a decrease of HDL Ch was accompanied by the reduction of lecithin percentage, lecithin/sphingomyelin ratio, and the increase of sphingomyelin percentage in HDL_2. The same changes were registered in HDL_3 but they were less pronounced; lecithin/sphingomyelin ratio was significantly decreased only at low HDL Ch.

Then we selected the groups of patients with a strongly marked TG level increase (IV type of hyperlipoproteinemia-HLP) as well as patients with an increase of total plasma Ch only, i.e. with HLP of IIA type.

At low HDL Ch and HLP of IIA type small differences in HDL_3 and HDL_2 phospholipid composition were detected as compared with the control. Considerable changes were registered in the properties of main phospholipids of patients both with low HDL Ch and IV type HLP and those with the normal lipid plasma level: decreased lecithin %, increased sphingomyelin % and decreased lecithin/sphingomyelin ratio in HDL_2 and HDL_3. Since in type IV HLP lipoprotein lipolysis is considerably lower than in IIA type, one can assume that changes in the composition of the particles are not due to the association of secondary remnants formed in the course of lipoprotein lypolysis.

Comparison of the discovered differences in phospholipid composition with apo-A-I level shows that at low HDL Ch in HDL_2 and HDL_3 the decrease of lecithin/sphingomyelin ratio in subjects both with normal plasma Ch and TG levels, and with type IV HLP is accompanied by the decrease of apo-A-I, which is contained both by HDL_2 and HDL_3. Along with these changes, we found a decrease in the amount of free Ch, esterified Ch, and LCAT-reaction activity in the blood plasma of patients with low HDL Ch.

Thus, in CHD patients with low HDL Ch we have found the changes in phospholipid composition and apo-A-I amount, i.e. the parameters which determine molecular organization of HDL_2 and HDL_3 surface monolayer.

Next part of this study was aimed at investigation of HDL_2 and HDL_3 physico-chemical parameters in CHD patients with low HDL Ch. The parameters which reflect physico-chemical characteristics, specifically, HDL surface monolayer fluidity, were measured using ESR-spectroscopy of spin labels.

The paper gives the parameters of spin-mobility of hydrophobic spin labels incorporated into HDL_2 and HDL_3. The values of these parameters for HDL_2 and HDL_3, isolated from the blood plasma of CHD patients with low HDL Ch, reflect the decrease in spin-label surroundings in both particles as compared with the control. These differences are more pronounced in HDL_2 and HDL_3. The alterations were also found in the values of critical temperatures, which characterize phase transitions or phase separations of the label lipid surroundings in HDL_2 and HDL_3 surface monolayer. The alterations indicate the decrease of fluidity of HDL_2 and HDL_3 surface monolayer in CHD patients with low HDL Ch.

The differences in spectral parameters of spin-labels registered in HDL of patients with low HDL Ch are likely linked to the changes in lipid and protein content of HDL_2 and HDL_3, which were given in the first part of the paper. Evidently, they are accounted for by the alterations in the character of protein-lipid interactions in the surface monolayer of these particles.

ESR-spectroscopy analysis of HDL (isolated from the plasma of healthy subjects with normal HDL Ch) after incubation with spin-labeled Ch showed that there were two components in ESR-spectrum of HDL_2 and HDL_3, which indicates the incorporation of spin-labeled Ch into two loci of these particles with different organization of microsurroundings.

At low HDL Ch one component was missing in ESR-spectrum of HDL_2, which indicates a decrease of its cholesterol-acceptor functions, while HDL_3 completely failed to incorporate spin-labeled Ch.

Comparison of the results obtained by chemical methods with ESR-spectroscopy data made it possible to assume that changes in HDL_2 and HDL_3 composition and characteristics likely result in the decrease of cholesterol-acceptor properties of these particles. That is manifested by the lowering of HDL Ch in blood plasma of most IHD patients.

Subsequently, we made a simple test which allows to detect alterations in HDL cholesterol-acceptor characteristics. HDL_2 and HDL_3 were incubated with platelets whereupon aggregational ability of the latter was measured. The in vitro experiments (14), which showed that the accumulation of Ch in platelet membranes is accompanied by the alteration in adenilatcyclase activity and ATP-induced aggregation, were a prerequisite for this part of our study. HDL were isolated both from the plasma of control group subjects and IHD patients.

Necessary amounts of lipoproteins (LP) were added to the suspension of washed platelets from the plasma of normal subjects to obtain the final concentration similar to a normal LP concentration

in blood plasma. After 10, 20, 30 and 60 min of incubation at 37° platelets were precipitated by centrifugation and resuspended in autologous plasma. In the obtained suspension ATP-induced platelet aggregation was measured.

HDL_2 and HDL_3 of patients with coronary atherosclerosis in concentrations similar to those of the HDL subfractions of healthy subjects did not inhibit aggregation.

On the contrary, the incubation of platelets with low and very low density lipoproteins (LDL and VLDL) isolated from plasma of IHD patients enhanced platelet aggregation.

Degree of the discovered effects both for HDL_2/HDL_3, and LDL/VLDL was proportional to the concentration of Ch in these lipoproteins.

A decrease in aggregational ability of platelets following the incubation with HDL of healthy subjects may be linked to their ability to accept Ch from the plasma membrane of platelets.

The absence of any effect of HDL_2 and HDL_3 isolated from blood plasma of IHD patients is of special interest. Likely, that may be accounted for by the decrease in HDL cholesterol-acceptor ability due to an alteration of chemical composition of HDL surface monolayer in IHD patients; lecithin/sphingomyelin is decreased due to reduced lecithin percentage and increased sphingomyelin percentage. The latter makes the structure of HDL surface monolayer more rigid.

Thus we have found that decreased HDL Ch level in blood plasma of patients with coronary atherosclerosis is accompanied by the alteration in a number of parameters of HDL_2 and HDL_3 surface monolayer molecular organization, namely, in phospholipid composition and amount of apo-A-I. These alterations result in the decrease of HDL antiatherogenic function and manifest themselves in:

1) decreased ability of HDL to accept Ch from the membranes of smooth muscle and endothelial cells, from platelets and VLDL in the course of their metabolism in blood plasma;

2) reduced activity of LCAT-reaction, whereby free Ch is transformed into esters and HDL_3 - into the particles of HDL_2 density, which transport Ch into the liver for excretion from the organism.

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BLOOD LIPIDS AND ANTIHYPERTENSIVE DRUGS: THE OSLO STUDY

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Epidemiologic studies have repeatedly shown a positive correlation between blood pressure and both coronary and cerebral mortality. Moreover, there is also a positive correlation between serum cholesterol and blood pressure. Nevertheless, randomised, controlled drug treatment hypertension trials have failed to achieve a favorable reduction of the incidence of coronary heart disease (CHD), in contrast to cerebral artery disease which has been significantly reduced by drug treatment.

The reason for this challenging observation is not known. There might be many explanations. Recently attention has been drawn to blood lipids and the effects of antihypertensive drugs on the metabolism of lipoproteins.

In the Oslo Study we have undertaken some studies of the effect on blood lipids in drug treatment of hypertension. Most of these studies are of short duration and involve few patients, which calls for some caution when drawing conclusions. On the other hand, the studies were done in homogeneous groups of persons, all men in a steady metabolic state. All were healthy and at work, only suffering from mild, symptomfree hypertension. No other drugs than the test drug were taken - given in a pre-fixed, unchangeable dose - and a randomized drug approach was used, only with one exception - the 3-year study in which the test groups were matched. We have used both cross-over designs and parallel groups.

In this way we have studied 4 beta-blockers, the alpha-blocker, prazosin, and a thiazide, separately, and 5 combinations of two drugs.

Of the beta blockers (Table 1) only pindolol was lipid neutral, probably due to its pronounced intrinsic symptomimetic activity, while propranolol, atenolol and oxprenolol lower HDL cholesterol and increased serum triglycerides. Hydrochlorothiazide (HCTZ) did not influence blood lipids. Prazosin lowered serum LDL + VLDL cholesterol and total triglycerides.

Table 1. Oslo Study. Metabolic effects (%change) of some antihypertensive drugs. Hypertension WHO 1. Men 47-55.

	Praz n=23 8 w	HCTZ n=10 10 w	Pind n=10 10 w	Aten n=9 16 w	Oxpren n=10 16 W	Prop n=23 8 w
Total chol	-8.9***	ns	ns	ns	ns	ns
LDL+VLDL chol	-10.1***	ns	ns	ns	ns	ns
HDL chol	ns	ns	ns	-16.7*	-11.5*	-13.0***
Chol ratio (1)	+7.0*	ns	ns	-19.2**	-13.7*	-15.2***
Total trigl	-16.2***	ns	ns	+17.9ns	+19.4ns	+24.0***
Uric acid	ns	ns	ns	ns	ns	+10.4**

*** $p < 0.001$
** $p < 0.01$
* $p < 0.05$
ns not significant
(1) $\frac{\text{HDL chol}}{\text{Total chol - HDL chol}}$

The combination of pindolol + prazosin lowered LDL + VLDL cholesterol by 12% (Table 2). Propranolol + HCTZ lowered HDL cholesterol and increased triglycerides. Propranolol + prazosin lowered HDL cholesterol. Methyldopa + HCTZ, and HCTZ + amiloride had no effect on blood lipids.

The clinical consequences of these metabolic changes are uncertain. The lipid effect might be of importance in long-term treatment of hypertension, especially for young people.

Table 2. Oslo Study. Metabolic effects (% change) of some antihypertensive drug combinations. Hypertension WHO 1. Men 40-55

	Praz Pind n=10 14 w	Praz Prop n=22 8 w	HCTH Amil n=10 14 w	M.dopa HCTH n=33 3 yrs	Prop HCTH n=33 3 yrs	Untr contr n=33 3 yrs
Total chol	ns	ns	ns	ns	ns	ns
LDL+VLDL chol	-12.0*	ns	ns		**	
HDL chol	ns	-7.5**	ns	ns ⊛	-18.1 ⊛	ns ⊛
Chol ratio	ns	ns	ns			
Total trigl	ns	ns	ns	ns	+44.3**	ns
Uric acid	+7.3*	+13.6**	+13.9*	ns	+21.8*	ns

** $p < 0.01$
* $p < 0.05$
ns not significant
⊛ Difference from mean of two treatment and a control group.

ENDOGENOUS REGULATION OF PROSTACYCLIN SYNTHESIS IN ARTERIAL SMOOTH MUSCLE CELLS

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The physiological and pathological roles of prostacyclin* (PGI_2) and thromboxane A_2 have attracted much attention in atherosclerosis.[1] Many investigations support the hypothesis that the loss of balance between these two prostaglandins is involved in vascular disease.[2] The capacity of the vascular wall to produce PGI_2 was reported originally for the intimal surface,[3] but medial smooth muscle cells also produce significant quantities both in vivo[4,5] and under culture conditions.[6] This capacity seems to be of importance especially after endothelial injury, such a situation probably involved in the atherosclerotic process.[7] Using arterial smooth muscle cells in culture, we have previously demonstrated that, in comparison with healthy cultured cells, cells originating from atherosclerotic aortas have a decreased capacity to produce PGI_2.[8] Such a reduced prostacyclin formation has been reported in aged aortic smooth muscle cells[9] associated with an increased PGE_2 synthesis. Despite this, the regulative mechanisms of PGI_2 generation in arterial cells remains unclear.

In the present report, evidences are presented which indicate that:

1. Several prostaglandins are produced by cultured smooth muscle cells.

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*Abbreviations used:- PGI_2 : Prostaglandin I_2 (prostacyclin); $PGF_{2\alpha}$, PGE_2 : Prostaglandins $F_{2\alpha}$, E_2; 6 $KPGF_{1\alpha}$: 6-Keto Prostaglandin $F_{1\alpha}$; A.A. : Arachidonic Acid; HETE : Hydroxyeicosatetraenoic acid; TLC : Thin Layer Chromatography; HPLC : High Performance Liquid Chromatography; GC-MS : Gas Chromatography-Mass Spectrometry; SMC : Smooth Muscle Cells.

2. The release of A.A. from phospholipids is not a limiting factor for the prostaglandins secretion by smooth muscle cells.
3. Both cycloxygenase and PGI_2 synthetase activities are depressed in atherosclerotic cells.
4. Significant amounts of HETEs were formed in arterial smooth muscle cells.

On the basis of these findings, it is suggested that endogenous lipoxygenase activities, either by some competitive mechanism with cyclooxygenase or, by the way of endogenous hydroperoxydes intermediates, or both, may be implicated in the regulation of the vascular PGI_2 synthesis.

METHODOLOGICAL APPROACH

Rabbits were made atherosclerotic using an alternate, moderately hypercholesterolemic diet (500 mg cholesterol/day) over a period of 1 year. At the end of the experimental period, the animals had developed fibrous lipidic plaques.[10] Aortic SMC were obtained from medial explants of both normal and atherosclerotic animals essentially as described by Ross[11] and used in subcultures, at confluency, as previously described.[8] These cells retained a number of metabolic features concerning growth, collagen synthesis and arachidonic acid metabolism which were in good agreement with those observed in vivo.[12,13]

Arachidonic acid conversion was studied either using intact 1.^{14}C A.A. prelabeled SMC or cell homogenates incubated with the labeled precursor as described.[14] Incubation media were acidified and extracted with ethyl acetate, then analysed either by TLC for prostaglandins, or HPCL for lipoxygenase derivatives. In each case, results were confirmed using GC-MS determination.

PROSTAGLANDIN PRODUCTION BY AORTIC SMOOTH MUSCLE CELLS

Rabbit vascular SMC in subculture synthesized radiolabeled prostaglandins from both endogenous (Table 1) or exogenous (Table 2) substrates. PGI_2 (measured as 6 Keto $PGF_{1\alpha}$, its stable degradation product in vitro) appeared as the main derivative formed. The relative distribution of the 3 metabolites (PGI_2, $PGF_{2\alpha}$, and PGE_2) correlated well with that obtained from freshly isolated aortic tissue (Table 3), despite the unexpected finding, that, in cell homogenates, PGE_2 constituted a minor component as compared to intact cells.

INHIBITION OF PROSTAGLANDINS PRODUCTION IN ATHEROSCLEROTIC SMOOTH MUSCLE CELLS

Rings of aortic tissue originated from the media of atherosclerotic rabbits showed a reduce capacity to produce PGI_2 and $PGF_{2\alpha}$ (Table 3), accordingly, estimation of prostaglandin production in subcultures of SMC originated from atherosclerotic media showed that both PGI_2 and primary prostaglandin formations were significantly depressed (Table 4). When compared to healthy cells, the ratios of Prostaglandin formation by intact SMC incubated with labeled A.A. were found to be 0.37, 0.43, 0.54 for 6 Keto $PGF_{1\alpha}$, $PGF_{2\alpha}$ and PGE_2 respectively, in atherosclerotic cells. These results were confirmed in cell homogenates. So, despite an enhanced percentage of primary prostaglandins (50 vs 37%), and a relative increase of PGE_2 concentrations as compared to total prostaglandins formed in normal cells (40 vs 28%), a significantly lower prostaglandin biosynthetic capacity appeared in atherosclerotic smooth

Table 1. Prostaglandin production from endogenous arachidonic acid

	Percent of the Total Radioactivity Released			
	6 K $PGF_{1\alpha}$	$PGF_{2\alpha}$	PGE_2	Free A.A.
Healthy cells	15.3 ± 0.7	2.4 ± 0.5	6.0 ± 2.7	15.1 ± 3.1

1h. prelabeled cell layers were washed then activated by scraping in 5 ml of Tris NaCl buffer pH 7.5. The mixture was centrifuged and supernatant was extracted. Results expressed as percent of total radioactivity recovered for each compound are the mean of three experiments.

Table 2. Exogenous arachidonic acid conversion

Cell Type	Percent Transformation of A.A.*		
	6 K $PGF_{1\alpha}$	$PGF_{2\alpha}$	PGE_2
Intact S.M.C.**	8.87 ± 1.54	1.61 ± 0.56	4.20 ± 1.76
S.M.C. Homogenate***	2.58 ± 0.34	0.80 ± 0.27	0.36 ± 0.05

* 10nM 1.^{14}C A.A.;
** 2 ± 0.2 10^6 cells maintained in the cultured flask;
*** 1 mg protein.
Results are expressed as mean ± SD from five different experiments.

Table 3. Exogenous arachidonic acid conversion by freshly isolated aortic tissue

	Percent Transformation of A.A.*		
	6 Keto $PGF_{1\alpha}$	$PGF_{2\alpha}$	PGE_2
Healthy aortic Media**	7.8 ± 1.1	1.03 ± 0.4	0.8 ± 0.3
Atherosclerotic aortic Media***	3.4 ± 1.1	0.4 ± 0.3	0.8 ± 0.5

* 10 nM of 1.^{14}C A.A.;
** 100 mg (w/w);
*** Atherosclerotic area defined by histologic examination after incubation.
Results are expressed as mean ± SD from ten different experiments.

muscle cells, without any compensatory activation of PGE_2 formation, clearly suggesting that PGI_2 synthetase and cyclooxygenase activities were depressed.

These results indicate that, either the rate of formation of endoperoxydes or the activity of the prostaglandin I_2 synthetase or both are critical steps in atherosclerotic cells. Lipid peroxydes resulting from the lipid accumulation in atherosclerotic arteries have been previously shown as specific inhibitors of the prostacyclin synthetase activity[15] and are candidates to explain the difference observed. However, under the experimental conditions described, neither the lipid concentrations in the culture or incubation media nor the lipid composition of the two types of cells were different. The previous identification of a lipoxygenase activity in vascular tissue[16] led us to perform experiments to investigate the possibility of such an activity in cultured SMC.

LIPOXYGENASE ACTIVITIES IN CULTURED SMOOTH MUSCLE CELLS

Homogenates from cultured SMC were incubated under the presence of 1.^{14}C A.A. After ethyl acetate extraction, the extracts were analysed by TLC. A significant lipoxygenase activity was observed. The rate of formation of monohydroxylated derivatives (HETEs) expressed as percent of the total A.A. conversion raised from 0.8 in healthy cells to 2.4 in atherosclerotic cells (Table 5). HPLC and GC-MS determinations indicated that several mono-HETEs derivatives are formed, namely 15 HETE and 12 HETE. Both of them have been previously recognized as efficient inhibitors of PGI_2 synthesis.[17]

Table 4. Arachidonic acid conversion by cultured smooth muscle cells originated from atherosclerotic aortas

Type of Experiment	Percent Transformation of A.A.		
	6 Keto $PGF_{1\alpha}$	$PGF_{2\alpha}$	PGE_2
Prelabeled cells	7.0 ± 1.0	3.5 ± 2.0	3.2 ± 3.3
Intact cells	3.31 ± 0.94	0.70 ± 0.35	2.70 ± 0.52
Cells homogenates	0.94 ± 0.21	0.47 ± 0.08	0.39 ± 0.07

Experimental conditions are identical as described in Table 1 and 2. Results are expressed as mean ± SD from 3 (prelabeled cells) or 5 different experiments.

Table 5. Lipoxygenase activity in cultured arterial smooth muscle cells

	Lipoxygenase Activity*	HETEs 6 K $PGF_{1\alpha}$
Healthy cells	0.85 ± 0.42	0.22 ± 0.03
Atherosclerotic cells	2.42 ± 0.61	1.20 ± 0.24

Homogenates of cultured smooth muscle cells (1 mg Protein) were incubated with 10 nM 1.^{14}C A.A. as described.

* Lipoxygenase activity is expressed as percent A.A. transformation.
Results are expressed as mean ± SD from three different experiments using the same cellular types.

CONCLUDING REMARKS

The capacity of the vascular wall to produce prostacyclin seems to be of importance as a protective mechanism against atherosclerosis. The decreased generation of PGI_2 by arterial SMC may be significantly involved in the progression of the atherosclerotic process. However, the regulative mechanisms of PGI_2 production in arterial cells remains unclear as yet. Since the participation of extracellular hydroperoxydes (HPETE) to the inhibition of the vascular PGI_2 formation appears unlikely,[18] an enhanced lipoxygenase activity in SMC leading namely to the formation of monohydroxy derivatives (15 and 12 HETE) may contribute either by some competitive mechanism with cyclooxygenase or by the way of endogenous hydroperoxy intermediates (HPETE) or both, to the progression of the atherosclerotic process.

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PLATELETS, MEGAKARYOCYTES AND VASCULAR DISEASE

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There is now much evidence from man and animals that platelets play an essential role in the production of atheroma. Pigs suffering from von Willebrand's disease have a defect in platelet function. In normal pigs atheromatous plaques can be produced in arteries by high cholesterol diet, but if the same diet is fed to pigs with von Willebrand's disease no atheroma is produced. Platelet dysfunction protects the pigs against atheroma[1]. Likewise atheroma may be produced in rabbits by high cholesterol diet but they may be protected against it by thrombocytopenia produced in them by injection of antiplatelet antibodies[2]. If baboons are injected intravenously with homocistine they suffer endothelial damage firstly and later atheromatous lesions in arteries. If the baboons are pre-treated with dipyridam, which reduces platelet reactivity, then the baboons still suffer endothelial damage from homocistine but atheromatous plaques do not develop.[3] Although Eskimos are heavy smokers they have a very low incidence of myocardial infarction and atheroma. They also have a long bleeding time and decreased platelet aggregation. The explanation for this probably lies in the large amount of eicosapentaenoic acid in their diet[4]. There is therefore evidence from both animal experiments and the Eskimo that platelets are necessary for the production of the atheromatous lesion no matter what other conditions may also be necessary.

Despite its importance, much concerning the physical nature of the platelet is still under dispute. One view holds that the platelet is produced from the bone marrow as a dense cell which becomes lighter as it circulates[5]. Another that platelets are produced in a variety of densities and volumes from the bone marrow and circulate without undergoing significant change in normal function[6]. Clearly resolution

of this problem is important since if the differences between platelets in volume, density and reactivity are a consequence not of ageing but of production then there is the possibility that if different types of platelets can be produced from the bone marrow then disease states may be related to alterations in platelet production. To observe changes in platelet density and volume platelets must be separated by using density gradients for the former, or countercurrent centrifugation or cell sorting for the latter. Workers holding different views about the relationship between platelet density and ageing have all used density gradient centrifugation as evidence for their conclusions. Table 1 shows that a variety of different conditions have been used by various workers yet it has recently been demonstrated that for a particular type of gradient only one set of conditions can give actual separation by density[15]. It is therefore possible that different conclusions about platelet density in the literature may arise from methodological problems.

Table 1*

Author		Date	Medium	+D/C	g	t(mn)	gxt
Booyse	(7)	1968	Sucrose	D	14,650 35,200	45 150	1,600,000 2,200,000
Karpatkin	(8)	1969	Oil	D	5,900	4	23,600
Minter	(9)	1971	Silicone	D	9,500	2	19,000
Ginsberg	(10)	1972	Oil	-	micro-haematocrit centrifuge	10	-
Bonneu	(11)	1973	Sucrose	D	55,000	60	3,300,000
Busch	(12)	1973	Ludox PVP	C	800	60	4,800
Charmatz	(13)	1974	Albumin	D	7,000	20	140,000
Penington	(6)	1976	Ludox PVP	C	10,180	5	50,900
Corash	(5)	1977	Stractan	D	52,951	30	1,600,000
Cieslar	(14)	1979	Stractan	D	2,000	45	90,000

* Conditions used by authors to separate platelets by density
+ D = continuous gradient
C = discontinuous
gxt = measure of the force used in the separation procedure

PLATELET DENSITY

If density is measured by continuous gradients of low viscosity media under conditions of isopicnic centrifugation then a bell-shaped distribution is observed that approaches the normal distribution (Figure 1). The mean density is approximately 1.062 gml^{-1}. The range of density is very small indicating that platelet density may not be an important physiological variable. The shape of the curve is argument that platelets are produced in all densities from megakaryocytes. If they were produced only as light cells that became dense or dense cells that became light then they would have to alter their rate of change of density in one direction and then the other along lines demanded by the normal distribution. It is more reasonable to assume that the shape of the curve is produced by random distribution of granules and mitochondria that are known to be denser than the rest of the platelet constituents[16].

^{75}SE Selenomethionine is an isotope-labelled aminoacid which is rapidly cleared from the blood, does not label circulating platelets and is taken up by megakaryocytes and incorporated into cytoplasmic protein. Platelets released into the circulation thereafter are labelled with selenomethionine. This tool could therefore be used to investigate whether platelets are produced from megakaryocytes in all densities. Figure 2 illustrates that if platelets are produced in all densities from megakaryocytes then radioactive platelets will be found in all densities as soon as radioactive platelets are produced from the megakaryocytes; but if only light cells or dense cells are produced then radioactivity will only be found in the appropriate area of the gradient. When such an experiment was carried out using rats, radioactive platelets were indeed found among all densities from one to five days after selenomethionine was injected intravenously.

In a second experiment a representative platelet population was taken from adolescent male monkeys and labelled with chromium (^{51}CR) and then re-injected. The following day blood was taken from the animals, fractionated into density subpopulations and radioactivity and number measured. It was observed that the ^{51}CR labelled platelets were distributed throughout all density subpopulations. Platelet life span in the monkey is similar to that of man (8 days) and when blood was again taken on the 5th day after labelling it was seen that the ^{51}CR labelled platelets were again distributed throughout all density subpopulations but there had been a significant yet small increase in platelet density among the ^{51}CR labelled platelets. The increase is approximately 5%, (Figure 3). The reason for this increase is not clear but it may be that platelets take up a heavy constituent as they circulate. These two experiments add evidence to the argument that platelets are produced from the megakaryocyte in all densities and circulate with only very slight increase in density as they do so.

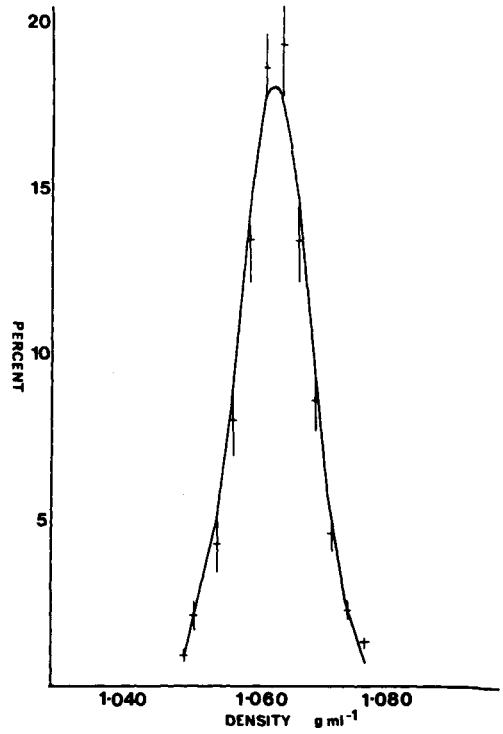
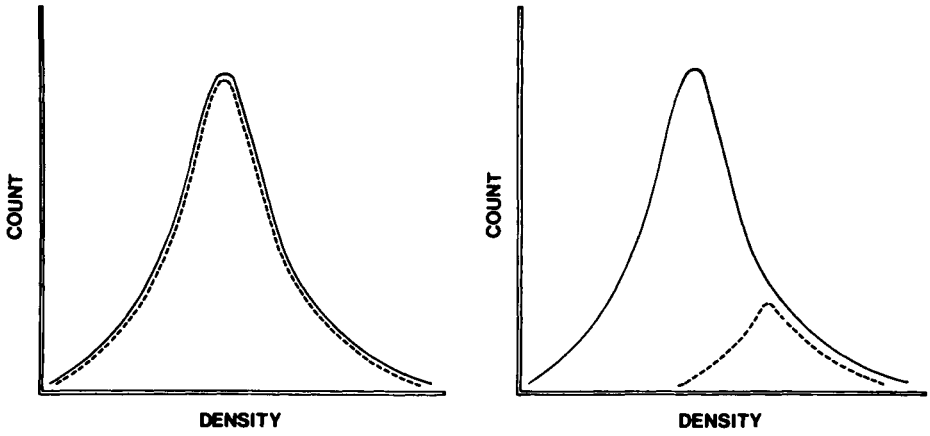


Fig. 1. A single platelet volume distribution from a normal male. The crosses indicate the position of the curve had it been normal (or Gaussian). The approximation is very close. A single curve is shown as the mean of many curves would tend towards the normal as predicted by the central limit theorem.

PLATELET VOLUME

Platelet volume distribution is unique among cellular volume distributions in that it is log normal (Figure 4). The platelet is unique in that it is not produced by mitosis. The two phenomena are linked in the physical fragmentation theory of platelet production[18,17]. Unlike platelet density, platelet volume is a variable of wide range and therefore may have physiological significance. The origin of changes in the volume distribution is found in the volume distribution of megakaryocyte cytoplasm[18]. If density determined subfractions of platelets have their volume distribution measured then it is seen that platelets of all volumes are found within each density subpopulation. Applying this knowledge to the experiments above it can be deduced that platelets of all volumes are produced at thrombopoesis and that they do not substantially change their volumes as they circulate under normal conditions.



Day 2

Fig. 2. A sketch plan of possible results on the second day following the injection of ^{75}SE selenomethionine as described in the text. If platelets are produced from megakaryocytes only as dense cells, then radioactivity would only be found in the dense part of the platelet density distribution curve (on the right). If platelets are produced in all densities then, on day 2, radioactivity would be found in platelets of all densities (as on the left). If the radioactivity curve and the platelet number curve fit exactly then their index of concordance is 1. The actual indices of concordance for days 1-5 of the experiment were 0.992, 0.998, 0.995, 0.995 and 0.993.

If thrombocytopenia is induced very large platelets are produced immediately afterwards[18]. Large platelets are more reactive than small[19], and it could be that the major response to thrombocytopenia is the production of large platelets and secondarily of production of an increased number of platelets.

MEGAKARYOCYTE POLYPLOIDY

Apart from the normal ploidy number of a 2N complement of chromosomes megakaryocytes exhibit polyploidy: they can increase their nuclear DNA concentration up to 128N. This phenomena, rare in mammalian biology, allows rapid increase in cell size without the need to undergo mitosis which is time and energy consuming. Larger megakaryocyte cytoplasm then gives larger platelets[18]. The steady state mean ploidy number in the mammal is 16N; after thrombocytopenia this can increase to a mean of 32N with the appearance of ploidy numbers not usually seen. This mechanism is probably under hormonal control[20].

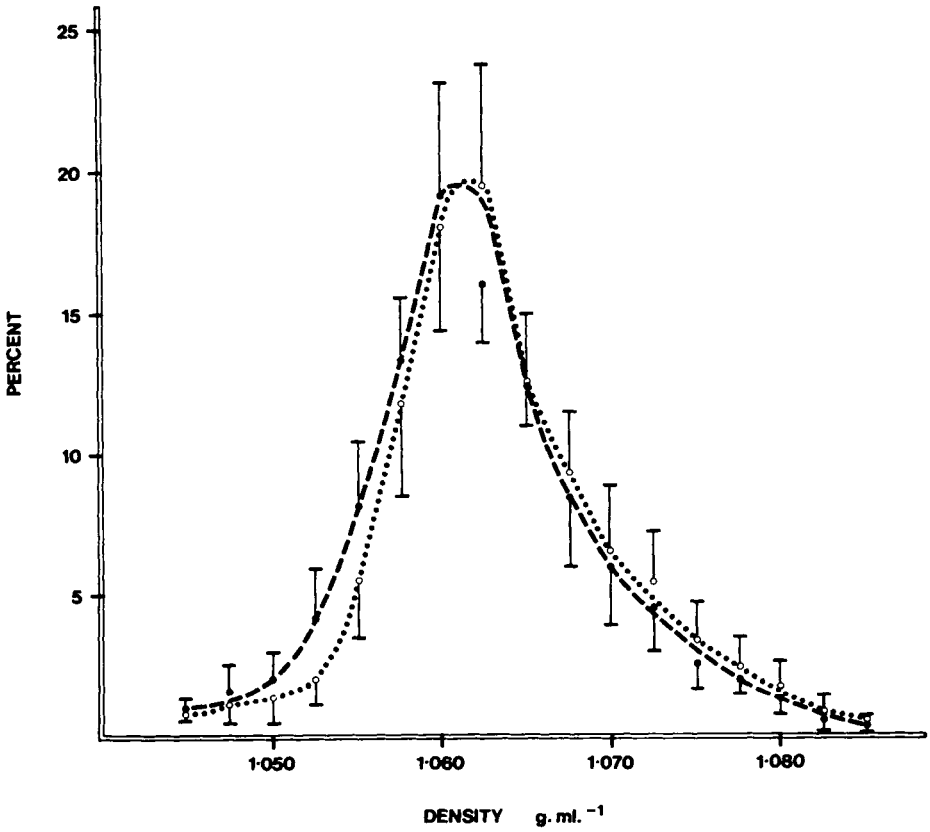


Fig. 3. Platelet number (---) and ^{51}CR radioactivity (●●●) (mean ± SEM, n = 12) against platelet density in blood taken from monkeys 5 days after the injection of an autologous representative population of ^{51}CR labelled platelets. There has been a very small, yet significant, increase in platelet density since the radioisotope labelled platelets were re-injected. ($p<0.001$ X^2 test).

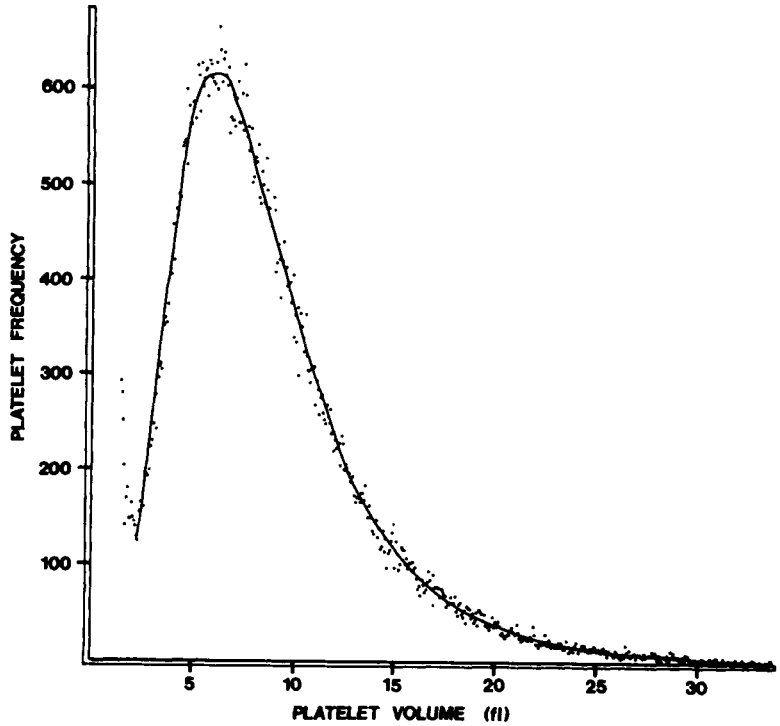


Fig. 4. The platelet volume distribution from a representative platelet population from a normal male. The curve is log normal i.e. when the log of platelet volume is plotted the curve becomes normal (Gaussian). Cells produced by mitosis have a normal volume distribution. The log normal platelet volume distribution is unique and reflects the platelet's unique mode of production.

MYOCARDIAL INFARCTION

A malfunction of the physiological system described here may be involved in disease, either as cause or effect. Platelet volume and density were therefore measured in men suffering myocardial infarction. If platelets are heterogeneous cells then any study must isolate a truly representative population. Since platelets are separated from whole blood by physical means, their particular biophysical variables may be preselected by the separation method. In this study >93% of the platelet population in whole blood was isolated by use of self generated sigmoidally shaped gradients of polyvinyl pyrrolidone-coated silica (Percol) onto which whole blood was layered and then subjected to velocity centrifugation. If the rate of recovery of platelets from whole blood was any less than 90% then the volume differences reported below were not seen, as the large platelets were lost in the velocity separation procedure. Mean platelet volume and density in 15 men with evidence of myocardial infarction from history (of less than 12 hours duration) and classical electrocardiographic and cardiac enzyme changes were compared to an age matched control group admitted to the same coronary care unit with chest pain but without evidence of myocardial infarction. Mean platelet volume was increased in the myocardial infarct group ($p < 0.0005$ Mann & Whitney u test) Figure 5. Platelet density was slightly increased ($p < 0.005$), probably as a consequence of the increase in platelet volume Figure 6. Eleven of the test group and 22 of the control group were again tested 6 weeks later. The myocardial infarction group still had a significant increase in mean platelet volume compared to controls (Figure 7). It may be argued that mean platelet volume is increased in men with myocardial infarction before the infarct occurs because, 1) since the platelet survival in man is approximately 8 days most of the platelets circulating 12 hours after infarct would have been produced before the infarct occurred 2) the increase is still present at 6 weeks when the infarct might be expected to have healed, 3) because the log normality of the platelet volume distribution was maintained in the test group (Figure 8).

CONCLUSIONS

Platelet volume (and density) are determined at the time of platelet production. There is evidence that platelets are involved in atherogenesis and arterial thrombosis. Platelets are larger than normal in myocardial infarction and this change may have a causal role in the vascular pathology. Men suffering myocardial infarction may have altered megakaryocytes that determine changes in their circulating platelets.

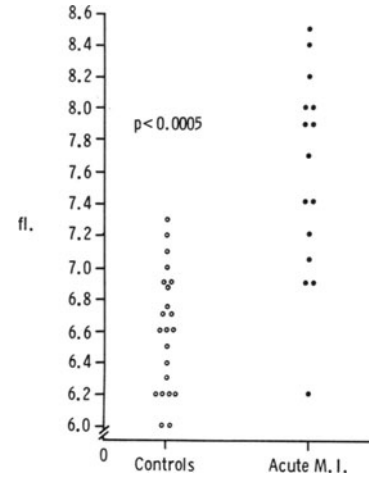


Fig. 5. Mean platelet volume in men suffering acute myocardial infarction (MI) and in controls. Blood was taken within 12 hours of onset of symptoms.

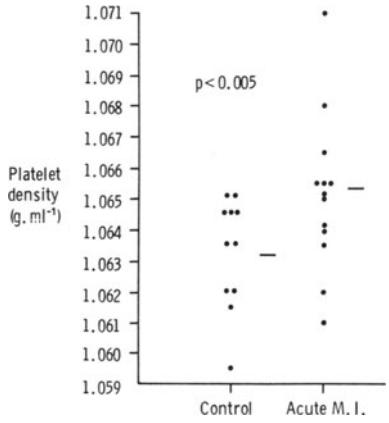


Fig. 6. Mean platelet density in the acute MI and control group.

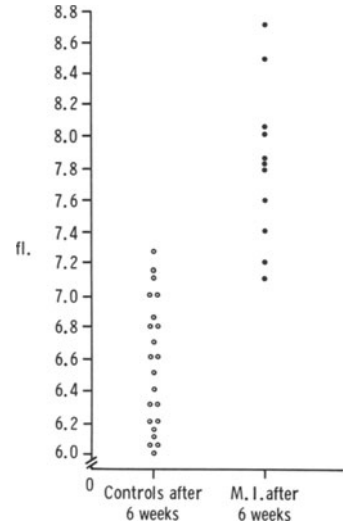


Fig. 7. Mean platelet volume in men 6 weeks following MI (●) and controls (○). Platelets are larger in MI group ($p<0.001$, Mann & Whitney u test).

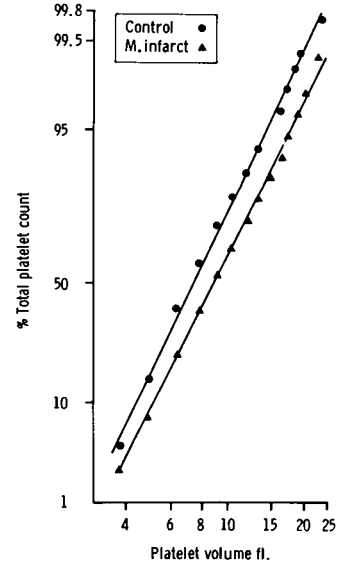


Fig. 8. Log probability plot of mean platelet volume in men suffering myocardial infarction (▲) and in the control group (●). The log normality of the platelet volume distribution is maintained after myocardial infarction.

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Experimental work referred to here was done both with Professor D. G. Penington and his colleagues, in the University of Melbourne Department of Medicine, St. Vincent's Hospital, Fitzroy, Australia and with my colleagues in the University of Sheffield Department of Medicine.

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EFFECTS OF BETA RECEPTOR BLOCKING DRUGS ON PROSTACYCLIN (PGI_2) AND THROMBOXANE A_2 (TXA_2) BIOSYNTHESIS AS A NEW ASPECT OF THEIR MODE OF ACTION

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The discovery of TXA_2 and PGI_2 with their antagonistic actions on vessel wall and platelets were unquestionably milestones that fundamentally altered the prevailing ideas on pathophysiology and therapy of cardiovascular diseases.

In animals with acute coronary ligation and in patients with ischemic heart disease and angina pectoris the ratio PGI_2/TXA_2 is altered in favor of TXA_2.[1,2] If disorders in this ratio are considered as a general pathogenetic mechanism the clinically used antianginal drugs should normalize this disturbed ratio.

In earlier investigations[3,4] could be shown that some clinically used antianginal drugs increase the PGI_2 biosynthesis, thus restoring the normal PGI_2/TXA_2 ratio. A second possibility is the selective inhibition of the TXA_2 synthesis with a concomitant promotion of the PGI_2 synthesis. These effects show some other antianginal drugs.[5,6] Open was the question whether beta receptor blocking drugs act only as adrenolytic substances or influence also the synthesis of eicosanoids.[7]

In 1981 we[5] reported on an inhibition of the malondialdehyde generation by pindolol whereas this effect could not be evoked by propranolol even in a dose one power of ten higher. In a comparative study with propranolol and pindolol both substances enhanced the PGI_2 release from guinea pig Langendorff hearts when applied in concentrations evoking a negative inotropic effect (Figure 1).

Considering the arachidonic acid (AA)-induced aggregation in rabbit PRP 0.1 - 1.0 mmol/l propranolol diminished the aggregation in a dose dependent manner (Figure 2) whereas the biosynthesis

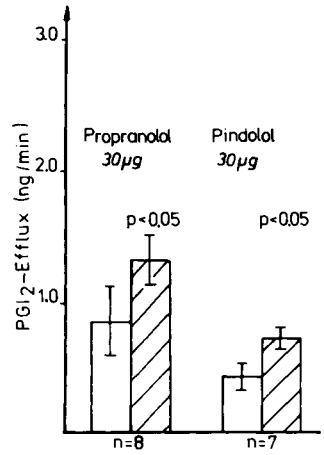

Fig. 1. Influence of Propranolol and Pindolol on the PGI_2-Efflux from Langendorff Hearts of Guinea Pigs.

of TXA_2 was not influenced. Higher concentrations up to 5 mmol/l - not shown in the figure - inhibited also the TXA_2 formation. Experiments with human PRP revealed a complete inhibition of the AA-induced aggregation already with about 0.6 mmol/l. The lowest TXA_2 inhibiting concentration was 1 mmol/l. Both experiments show that an inhibition of the TXA_2 biosynthesis could be proved only with concentrations distinctly higher than those which completely inhibited the AA-induced aggregation. Contrary to propranolol, pindolol inhibited the AA-induced aggregation as well as the TXA_2 synthesis in a dose dependent manner (Figure 3). Effective concentrations were 0.1 mmol/l or more. Both isomers of pindolol had approximately the same antiplatelet and TXA_2 synthesis inhibiting effects although in all concentrations used the (-) isomer was somewhat more effective than the (+) isomer. There were no distinct differences between both isomers of propranolol, too.

The cause of the inhibition of TXA_2 by pindolol may be found in its relatively potent inhibition of the cyclooxygenase: 10 uM inhibited it by about 60% and 200 uM by 76-83%. By comparison, indomethacin was about one power of ten more potent. In contrast to pindolol, propranolol even in the concentration of 200 uM did not significantly inhibit the cyclooxygenase and showed with 1 mM only a slight inhibition.

Other differences of pindolol and propranolol were seen in different incluences on platelet aggregation induced with the PG endoperoxide analogue U-46619. Propranolol exerted an inhibition one power of ten more potent than pindolol (Figure 4).

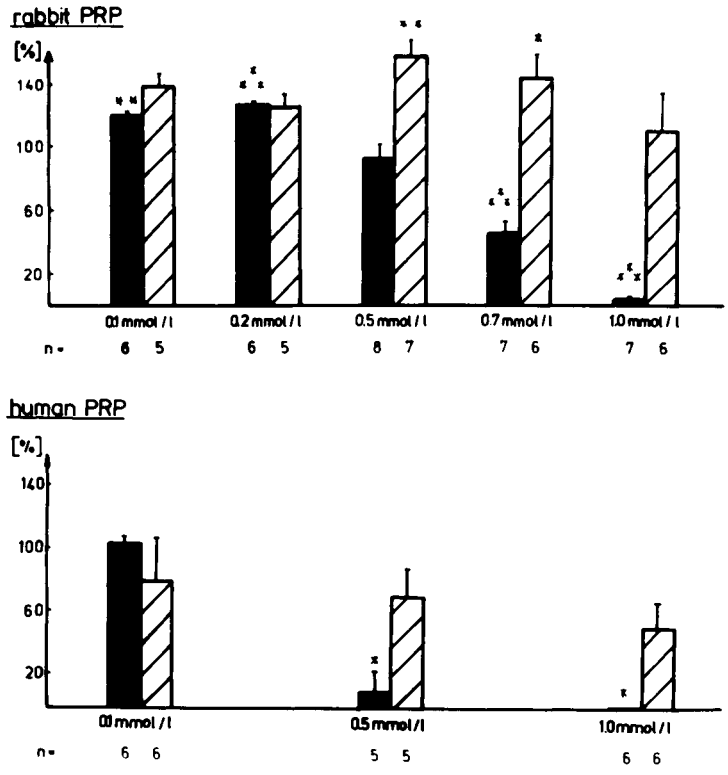

Fig. 2. Effect of Propranolol on Arachidonic Acid-induced Aggregation and TXA_2 Formation (Rabbit and Human PRP, TXA_2-Bioassay) Aggregation ■ TXA_2 □.

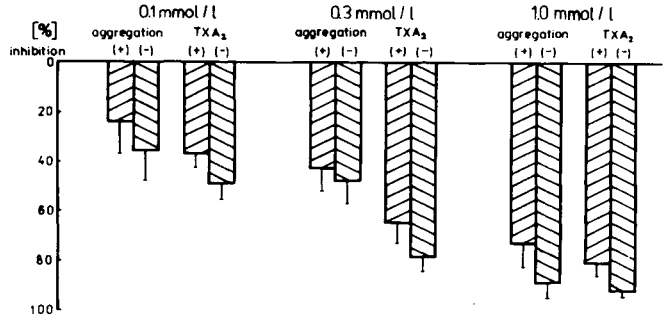

Fig. 3. Effect of Pindolol-Isomers on Aggregation and TXA_2 Formation (Rabbit-PRP, AA-induced Aggregation, TXA_2 Bioassay).

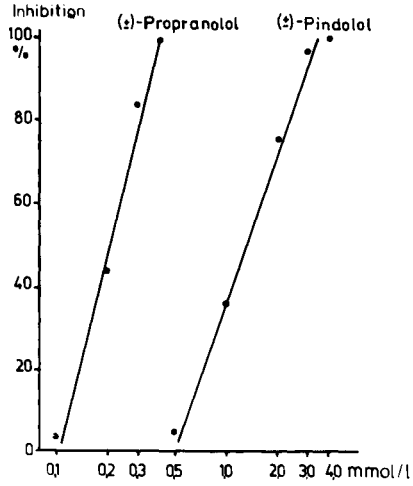

Fig. 4. Effect of (±)-Propranolol and (±)-Pindolol on U-46619-induced Aggregation (Human-PRP).

SUMMARY

Only one part of the mechanism of action of beta receptor blocking drugs can be explained via their β-adrenolytic effect. The antihypertensive and antiplatelet action is induced by influences on the formation of eicosanoids. Pindolol seems to be a prototype with a potent TXA_2 synthesis inhibiting effect probably via a relatively potent cyclooxygenase inhibition. Propranolol has a more potent antagonistic effect on "TXA_2- receptors" than on TXA_2 synthesis. Both drugs stimulate the PGI_2 synthesis in coronary walls. In this combination of the adrenolytic effect with the antiplatelet and anti-TXA_2 actions we might have found the clue for the potent clinical efficacy of the beta adrenergic blocking drugs.

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PLATELET SPREADING AND THROMBI-FORMATION IN VITRO

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INTRODUCTION

The present paper deals with three main problems. In the first section we consider platelet interaction with surfaces coated with genetically distinct types of human collagen, which makes a part of various vessel wall structures. The main stages of platelets-collagenous substrates interaction are described: a) initial attachment of platelets to the substrate, b) platelet spreading on the substrate, and c) formation of thrombi-like aggregates adherent to the substrate.

In the second section we dwell in more detail on the process of platelet spreading and a possible role of spread platelets in mural thrombi formation.

Finally, in the third section we consider: a) the induction of spreading and adherent thrombi-like aggregates formation on collagen by arachidonic acid and stable prostaglandin endoperoxides analogues, and b) the inhibition of the processes by antithromboxane agent Trapidil and stable prostacyclin analogues.

PLATELET INTERACTION WITH COLLAGEN SUBSTRATES FORMED BY GENETICALLY DISTINCT TYPES OF HUMAN COLLAGEN

Four types of collagen have been found in human and animal vessels: I, II, IV, and V. These collagens differ in chain compo-

sition, the ability to form fibrils, and localization in the vessel wall. Many researches have investigated the ability of different collagen types to induce platelet aggregation in suspension. It was concluded that the above mentioned types of collagen in fibrillar form induce platelet aggregation while those in amorphous or monomeric form are not capable of being aggregation inducers.

Platelet interaction with collagen-coated surfaces differs from platelet-collagen interaction in suspension by a number of parameters. Thus, we studied how platelets would interact with various types of human collagen if the collagens are immobilized on the surface.

Monomeric collagen of different types was isolated from human placenta and immobilized on the surface in two different ways (Figure 1). Procedure I: collagen was added into the wells of multiwell tissue culture plates; fibril formation was performed in suspension at 37°C and neutral pH with subsequent immobilization of fibrils on well bottoms. Procedure II: collagen in monomeric form was directly adsorbed on the well bottoms at 4°C. Thus, two types of surfaces were formed; one coated with fibrillar collagen, another with monomeric collagen. Human platelets separated from plasma by gel-filtration were added into thus prepared wells and incubated with rotation for 40 min. at 37°C. After removing nonadherent platelets and washing the wells, adhesion was analyzed by scanning electron microscopy (SEM) (Figure 1).

Figure 2 shows the interaction of platelets with a surface coated with type III collagen. The coating was produced in two ways: 1) collagen fibrils with an average diameter of 100 nm, which covered well bottoms by a dense multilayer network, were formed (Figure 2,a); 2) fibrillogenesis was not carried out and well bottoms were covered with amorphous collagen (Figure 2,b). Though, regardless of absence or presence of collagen fibrils adherent platelets form on the substrate large multilayer aggregates, which resemble thrombi (Figure 2,a,b).

Platelet interaction with the surface covered with type III collagen also results in intensive platelet spreading (Figure 3). Platelets spread on collagen are a highly attractive substrate for the platelets from suspension; they readily bind to the upper surface of spread platelets (Figure 3). Sheets of confluent spread platelets form the basis of thrombi-like structures (Figure 4).

Type I collagen used as a substrate behaves in a similar way. Both in amorphous and fibrillar form this type of collagen stimulates thrombi-like aggregate formation and platelet spreading.

Completely different picture of adhesion is observed when type V collagen is used as a substrate (Figure 5,a). In this case platelet

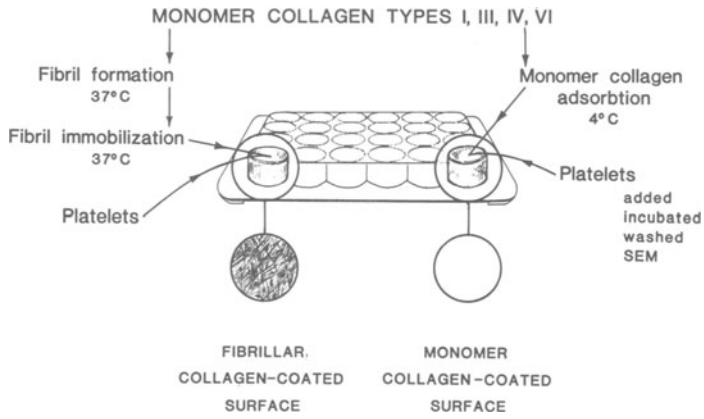


Fig. 1. General scheme of experimental studies of platelet interaction with surfaces covered with genetically distinct types of human collagen. Well bottoms of Multiwell culture plates were covered with fibrillar or monomeric collagens (0.6 - 0.8 mg per well) of type I, II, IV, and V, isolated from human placenta. Gel-filtered human platelets were added into the wells (I-I,5 x 10^8/ml, 0.2 ml per well) and incubated for 40 min. at 37°C with rotation in a horizontal incubator-shaker at 36 rev./min. Then, nonadherent platelets were removed, the wells washed, and the adhesion was studied by scanning electron microscopy (SEM).

adhesion level is 20-100-fold lower for type I and III collagens. Adherent platelets are usually at the stage of initial attachment and have discoid or spheroid shape. There is no platelet spreading and adherent aggregates formation on this substrate (Figure 5,a). If type I and type V collagens are mixed in the ratio 4:I the resulting substrate looks like the one made of pure type I collagen (Figure 5,b), while platelet adhesion is practically the same as that to the purified type V collagen (Figure 5,b). Likely, type V collagen when bound to type I collagen blocks its sites responsible for the formation of thrombi-like aggregates and platelet spreading.

While interacting with the surface covered with type IV collagen in amorphous form, platelets spread (Figure 6,a) and make up small aggregates, which are localized on the upper surface of spread platelets (Figure 6,b). Similar picture of adhesion is observed when type IV collagen is immobilized in fibrillar form.

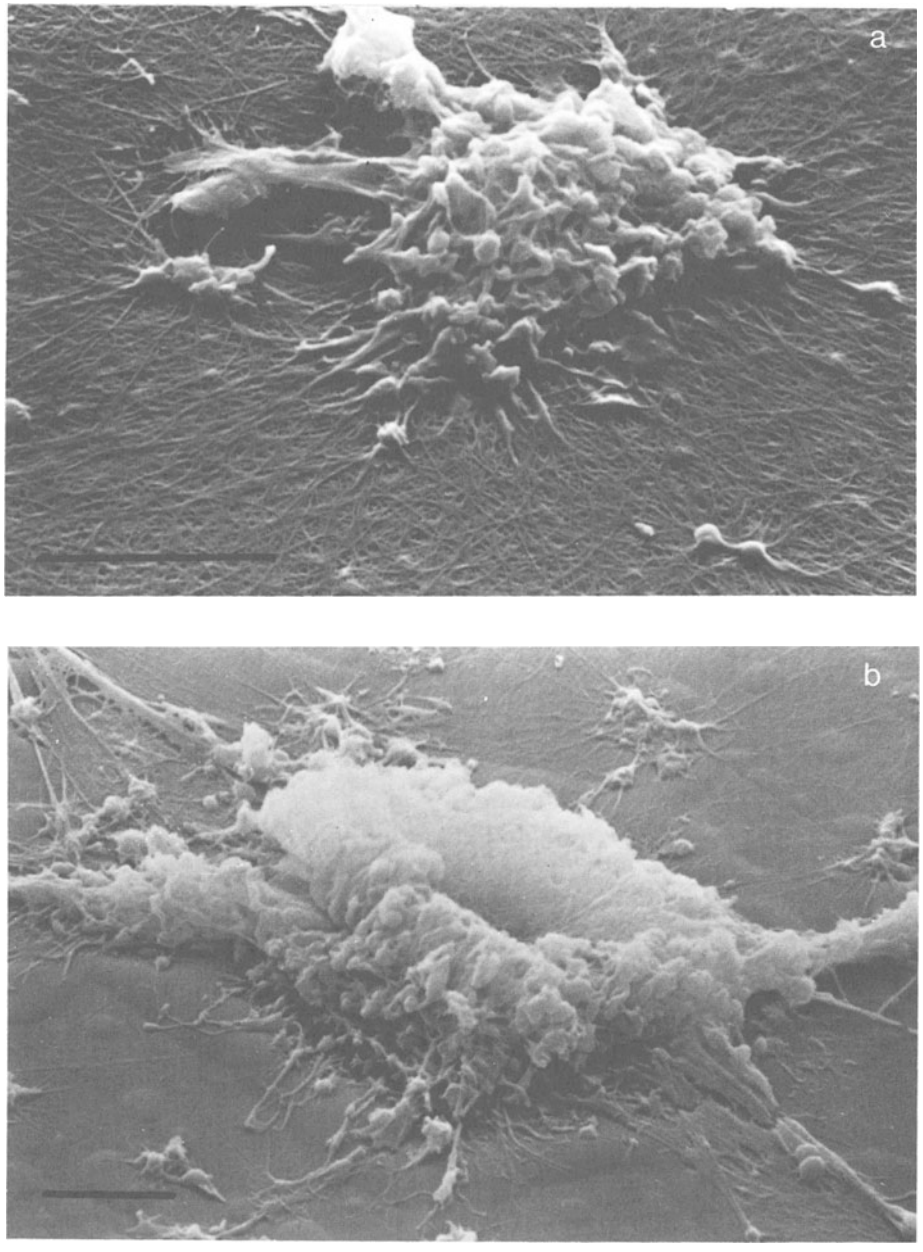


Fig. 2. Formation of thrombi-like aggregates on surfaces covered with type III human collagen in fibrillar (a) or monomeric form. Irrespective of the presence (a) or absence (b) of fibrils, large multilayer thrombi-like platelet aggregates are deposed on the surface. Scale bar - 10μm.

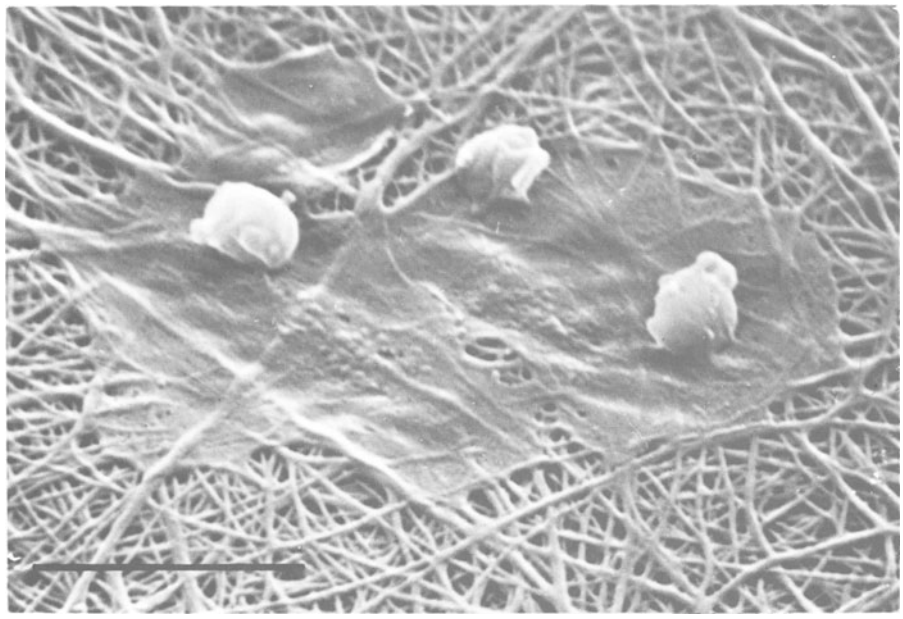


Fig. 3. Platelet spreading on the surface covered with type III fibrillar collagen and adhesion of platelets from suspension to the upper surface of spread platelets. The micrograph shows three completely spread platelets; two of them have adherent spheroid platelets from suspension on the upper surface. Scale bar - 5 μm.

Thus, various types of collagen substrates have different thrombogeneity with respect to platelets (Table 1). Type V collagen is an athrombogenic substrate characterized by low level of adhesion, and adherent platelets are nonactivated or low activated. Type IV collagen is a substrate of moderate adhesiveness; platelets spread on it and form small aggregates. Type III and I collagens are thrombogenic substrates whereon all stages of platelet activation, even thrombi-like structures formation, are observed (Table 1).

It should be underlined that during the interaction of platelets with immobilized collagen surface the degree of substrate thrombogeneity is mainly determined by the genetic type of collagen and does not depend on whether collagen is in fibrillar of amorphous form (Figure 2,6). On the contrary, it is only fibrillar forms of collagen that induce platelet aggregation in suspension. The reason for these differences is likely accounted for by the fact that the substrate formed both by fibrillar and monomeric collagen offers a

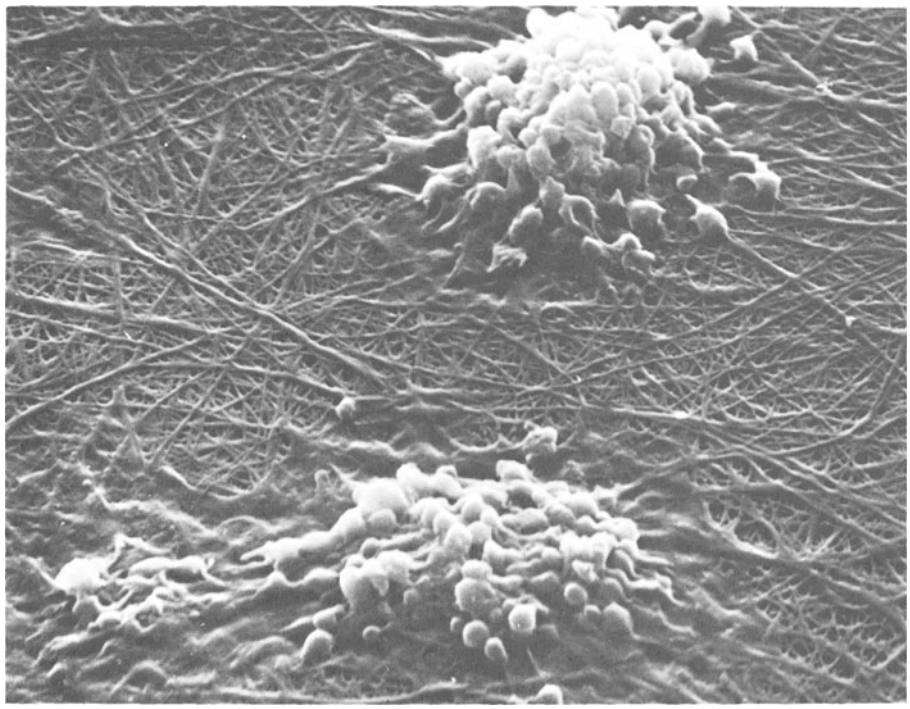
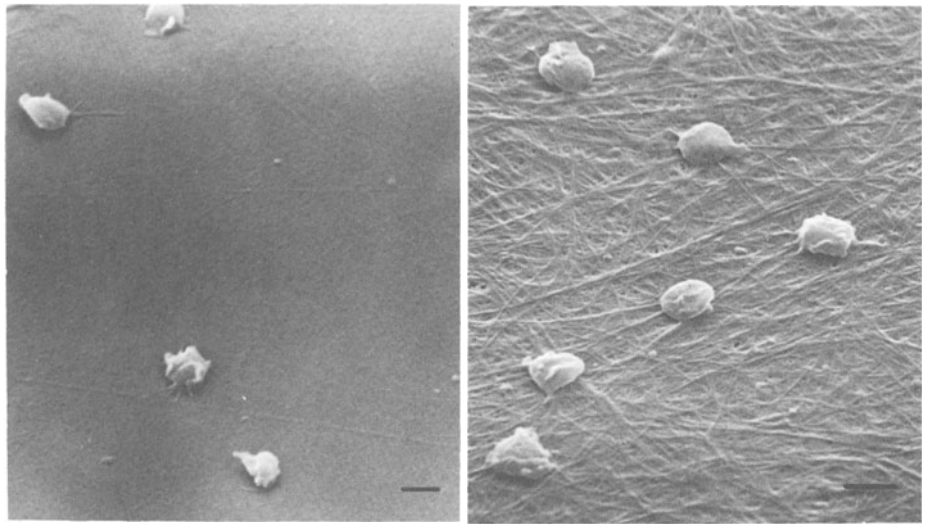


Fig. 4. Sheets of confluent spread platelets at the basis of thrombi-like aggregates adherent to type III fibrillar collagen. Scale bar - 10 μm.

great number of platelet-binding sites concentrated on the surface available for the simultaneous interaction with platelets. In suspension a similar "concentration" of platelet-binding sites can be obtained only if monomeric collagen is organized into fibrils.

The obtained data elucidate the differences in thrombogenic characteristics of various anatomic structures of the in vivo vessel wall. Taking into account the localization of the above mentioned collagen types with respect to the luminal surface one can better understand: athrombogenity of the endothelial lining of undamaged vessel, moderate thrombogeneity of basal membrane in shallow vessel wall injuries, and high thrombogeneity of subendothelial intimal layer, media and adventitia, which are exposed in deep injuries and frank trauma of the vessel (Table 1). The interaction of thrombogenic and athrombogenic collagen types may also play an important role in mural thrombi formation (Figure 5).



(a) (b)

Fig. 5. Platelet adhesion to the surfaces covered with type V collagen (a), and a mixture of type I and type V collagens in the ratio 4:1 (b). The first substrate is amorphous (a), the latter is fibrillar (b). In both cases adherent platelets are at the stage of initial attachment; there is no spreading and thrombi-like aggregate formation. Scale bar - 2 μm.

SPREAD PLATELETS AND THEIR POSSIBLE ROLE IN MURAL THROMBI FORMATION

In the series of experiments described we used commercial acid soluble calf skin collagen (CSC) to cover the well bottoms of multiwell tissue culture plates (Figure 1). Reactivity of the collagen with respect to platelets strongly resembles human type IV collagen. Similar to type IV collagen platelet interaction with the surface covered with CSC involves three main processes: 1) platelets from suspension attach to the collagen substrate, 2) then they spread, and 3) platelets from the suspension attach to the upper surface of spread platelets. That results in the formation of small aggregates on spread platelets; large multilayer thrombi-like aggregates are not formed. Thus, calf skin collagen is a fitting substrate to study *in vitro* the initial stages of thrombogenesis, i.e. platelet spreading and attachment of platelets from suspension to the upper surface of spread platelets.

Figure 7 shows one of the mechanisms of platelet spreading on the substrate. It is clearly seen as membranous web spreads between two pseudopods of a spheroid platelet (Figure 7,a). At later stages

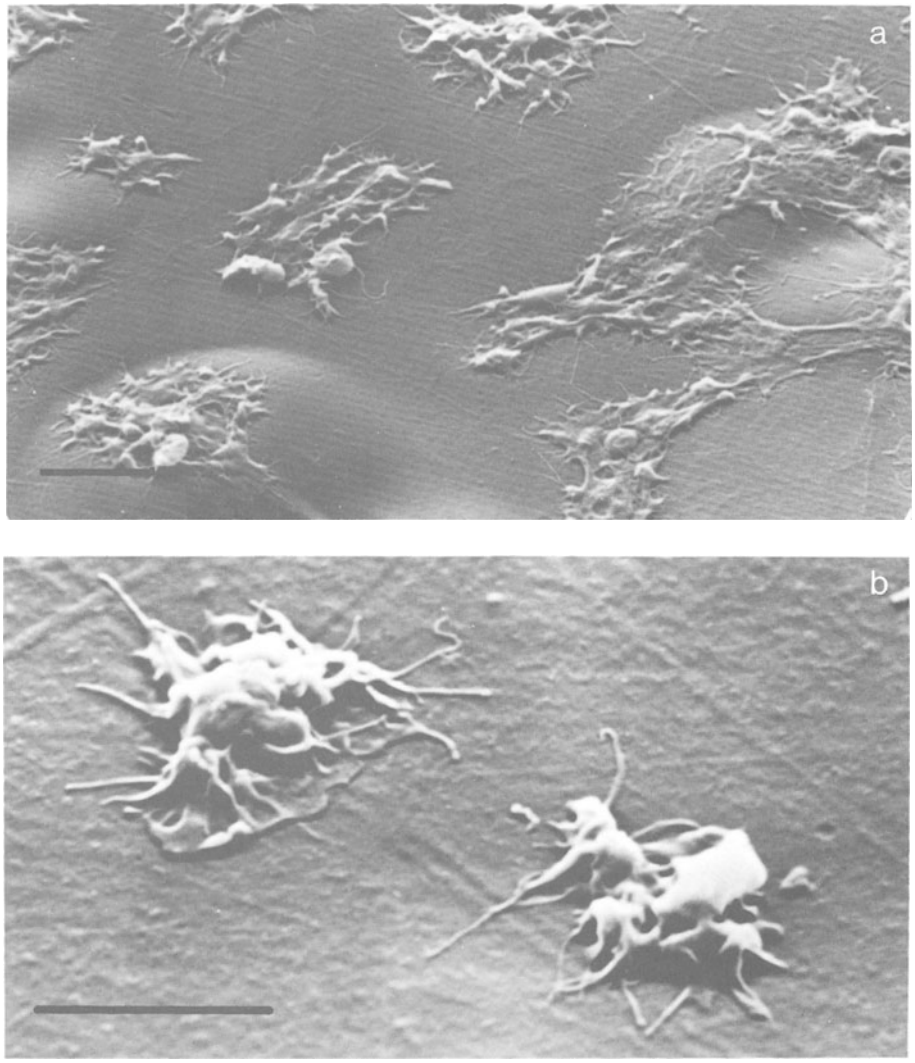


Fig. 6. Platelet adhesion on the surface covered with amorphous type IV collagen. Platelets actively spread on the collagen substrate; single platelets (a) and microaggregates (b) bind to the upper surface of spread platelets. Scale bar - 5 μm.

Table 1. Summation of Experimental Data on Platelet Interaction with the Substrates Formed of Genetically Distinct Types of Human Collagen (Fig. 2-6)

Collagenous substrate	Platelet-collagen interaction		Main localization of collagen in the vessel wall (I - 3)
	Adhesion level	Shape change, Aggregation	
V	low	Initial attachment, formation of pseudopods	Liminal surface of endothelial cells
IV	moderate	Spreading, moderate aggregation	Basal membrane
III	high	Spreading, aggregation, formation of thrombi-like structures	Subendothelium
I	high		Media, adventitia

of spreading the platelet comes into close contact with the substrate, radial spreading of webs between other pairs of pseudopods takes place (Figure 7,b) and thus a well-spread platelet is formed (Figure 7,c). The analysis of spreading kinetics shows that in our model system this process takes about 10 min.

Another stage of platelet interaction with a collagen-covered surface is the adhesion of platelets from suspension to the upper surface of spread platelets (Figure 8,a). Adherent platelets may be organized into aggregates (Figure 8,b). Sometimes several spread platelets fuse forming local cell sheets whereto the platelets from suspension are attached (Figure 8,c,d). The density of adhesion of platelets from suspension per an area unit of the spread platelet exceeds that of fibrillar collagen regions by 10-30, and in certain experiments, by 50-100-fold. These data demonstrate that spread platelets can be a substrate for mural thrombi formation.

To what extent does the nature of a substrate determine the processes of platelet spreading and subsequent adhesion of platelets to their surface? To answer the question important for the understanding of thrombogenetic mechanisms we have studied how these processes develop on two substrates: calf skin collagen and collagen treated with plasma fibronectin - a glycoprotein, which actively stimulates the attachment of various nucleated cells to a collagen substrate.

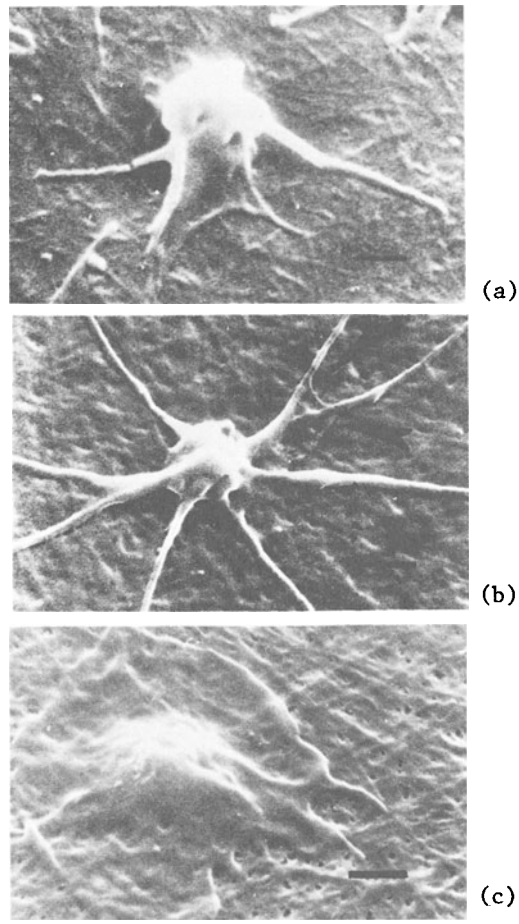


Fig. 7. The mechanism of platelet radial spreading on collagen substrate. For the experiments shown in Figure 7-16 wells covered with CSC (Sigma, C-35II) were used. (a) - a spheroid platelet at early stage of spreading. A membrane "web" is being formed between the two pseudopods. The edges of pseudopods and the web are in contact with the substrate; (b) subsequent stage of spreading. Radial spreading of webs between three pairs of pseudopods (arrows). Close contact of platelets with the substrate along the whole lower surface of pseudopods and webs; (c) - platelet spreading is coming to an end. There is a flat circle of lamellar cytoplasm (hyalomer) on the periphery of the platelet and granulomer hillock in the center which indicates the transfer of platelet granules from the periphery to the center. Granulomer hillock disappears with complete platelet spreading and exocytosis of granules (Figure 3,8,a). Scale bar - I μm.

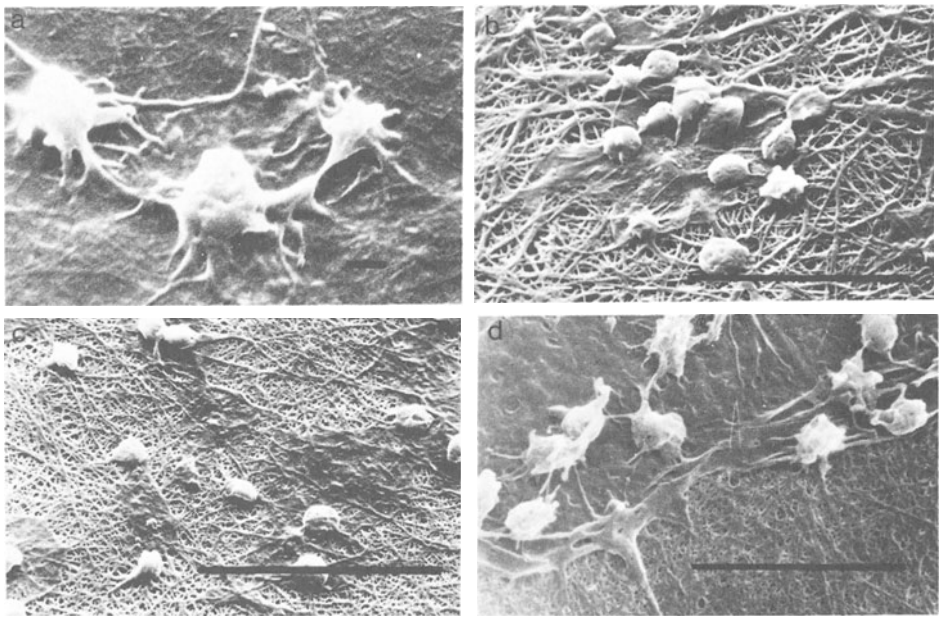


Fig. 8. Adhesion of platelets from suspension on the upper surface of spread platelets. (a) - three single spheroid platelets adherent to the upper surface of a completely spread platelet with irregular jagged shape (scale bar - I μm); (b) - formation of microaggregates of spheroid and discoid platelets on the spread platelet surface; (c,d) - local cellular layers of confluent spread platelets with single adherent platelets and micro-aggregates. Collagen covered regions neighboring spread platelets have considerably fewer adherent spheroid and discoid platelets (a-d). Scale bar - 10 μm.

It was found that pretreatment of collagen substrate with fibronectin increases platelet spreading by 8-9-fold (Figure 9). Such stages of platelet activation as disc-to-sphere transformation and formation of pseudopods, which precede platelet spreading, are stimulated by fibronectin to a considerably lesser extent (Figure 9).

It is very important that fibronectin treatment sharply reduces the adhesion of platelets from suspension to the upper surface of spread platelets (Figure 10). We estimated this effect by measuring the percentage of the so-called adhesive spread platelets, i.e. the platelets, which have adherent platelets on their upper surface. It has been established that after fibronectin treatment the relative number of adhesive spread platelets decreases from 45% to 10-12% (Figure 10).

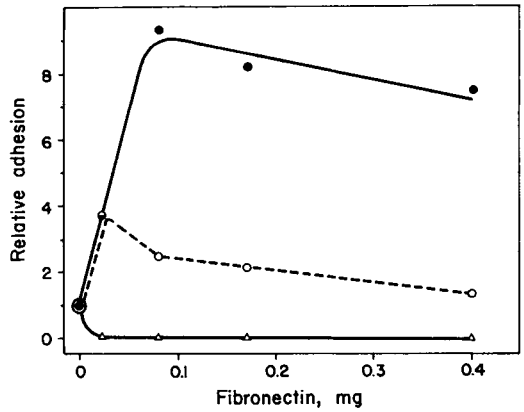


Fig. 9. Fibronectin effect on the spreading of platelets on collagen substrate. CSC covered wells were pretreated with different doses of fibronectin and washed, and adhesion was measured by SEM. ●——● - spread platelets; ○——○ - activated unspread platelets (discs with pseudopods, spheres and spheres with pseudopods); △——△ - inactivated platelets - discs.

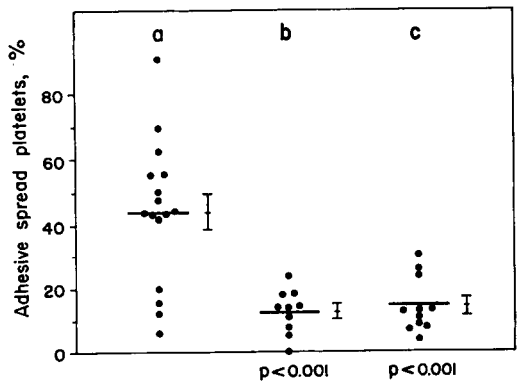


Fig. 10. Fibronectin effect on the binding of platelets from suspension with the upper surface of platelets spread on collagen. Adhesion was carried out in CSC-covered wells non-treated (a) or pretreated (b,c) with fibronectin. Prior to the addition of platelets unbound fibronectin was either removed from the incubation mixture (b) or not (c). The ability of spread platelets to bind the platelets from suspension was determined by measuring the percentage of adhesive spread platelets, i.e. the platelets with adherent platelets on the upper surface. The total number of spread platelets was taken for 100%. Mean values ± standard errors are given. Statistic significance of differences between the means (p) in experiments (b) and (c) as compared with the experiment (a) was calculated using Student's t-test.

The obtained data indicate that while increasing with fibronectin the ability of a collagen substrate to spread platelets one can simultaneously decrease the adhesiveness of the upper surface of spread platelets for the platelets from suspension. It is possible that one of the physiological functions of fibronectin is to stimulate the formation of "pseudoendothelial" carpet of unadhesive spread platelets in deendothelialized zones of the vessel wall by binding to these zones.

Is it a specific effect of fibronectin or a general phenomenon accounted for by the fact that platelets better spread on attractive substrates and become less adhesive for the platelets from suspension? To investigate the problem we have compared platelet spreading and adhesion on spread platelets in two substrates: 1) collagen and 2) activated polystyrole, a material used for multi-well culture plates, which has high adhesiveness for a wide variety of cells. It turned out that platelets spread on polystyrole more actively than on collagen. This is manifested by a 2-fold increase of the spread platelets fraction and mean area of one spread platelet (Table 2). Besides, there is a considerable difference in the shape of spread platelets. The calculation of the so-called two-dimensional form parameter, i.e. the ratio of figure area to its squared perimeter, showed that on polystyrole the shape of spread platelets approximates a circle, while on collagen they have irregular jagged shape (Table 2). The result demonstrates that polarization of spreading, i.e.

Table 2. Binding of Platelets to CSC and Activated Polystyrole. Platelet Spreading and Adhesion of Platelets from Suspension to the Upper Surface of Spread Platelets.

Substrate	Spread platelets			Adhesion on spread platelets
	per cent*	mean area, μkm^2 +	form parameter+, F	x $10^{-3}/nm^2$
Collagen	30.6 ± 6.0	13.4 ± 0.7	0.42 ± 0.01	22.4 ± 5.2
Plastic	62.1 ± 12.8	25.6 ± 1.0	0.85 ± 0.01	3.5 ± 1.2
	$p < 0,05$	$p < 0,01$		$p < 0.001$

Platelets were added into uncovered wells of Multiwell (Falcon) and CSC-covered wells. *Spread platelets - % of the total number of adherent platelets. + Mean area and two-dimensional form parameter (F) of spread platelets were measured in MOP-3 (Reichert-Jung, Austria), $F = \frac{4TS}{U^2}$, where S - figure area, U - figure perimeter. For a circle F = I; for markedly irregular figures (elongated or jagged) F approximates to 0. Mean ± standard error is given for 7 experiments. Statistic significance of differences between the means (p) was calculated using Student's t-test.

selection of major directions for spreading is quite different on collagen and plastic.

A 6.5-fold fewer number of platelets from suspension attach to the platelets spread on polystyrole as compared with the platelets spread on collagen (Table 2). That seems to be the most important result of these experiments. Thus, just as in case of fibronectin the platelets spread on attractive substrate are characterized by a decreased adhesiveness for platelets from suspension. This phenomenon may be possibly accounted for by the existence of certain common proteins within receptor complexes responsible for the interaction of spread platelets both with the substrate and platelets from suspension. An increase in collagen substrate adhesiveness conditioned by fibronectin or the use of a more attractive substrate (polystyrole) results in the migration of these proteins from the upper surface of spread platelets onto the surface facing the substrate. Such a redistribution leads to the decrease of spread platelets adhesiveness for the platelets from suspension, which in its turn reduces a possibility of mural thrombi formation on spread platelets.

These data make it possible to broaden the approach to selection of artificial biomaterials for vascular grafts and prosthetic heart valves. Probably, along with the search for new low adhesive materials researchers should turn to those that facilitate intensive platelet spreading. In this way surfaces covered with a carpet of nonadhesive spread platelets may be obtained.

MODULATION OF PLATELET SPREADING AND OF FORMATION OF THROMBI-LIKE AGGREGATES ADHERENT TO COLLAGEN SUBSTRATE

Platelet spreading and adherent thrombi-like aggregate formation were stimulated by exogenous platelet activators - arachidonic acid and stable methano-epoxy-analogue of prostaglandin endoperoxides (U46619). Inhibition of these processes was studied using Trapidil, U46619 antithromboxane agent, and stable prostacyclin analogues - carbacyclin and 6β - PGI_I.

Calf skin collagen was used as a substrate. In the absence of platelet activators an ordinary picture of adhesion to this type collagen is observed: adhesion of single platelets, moderate spreading and adhesion to spread platelets (Figure 11,a). Adhesion in the presence of arachidonic acid or U46619 results in surface accumulation of thrombi-like aggregates alternating with thrombi-free areas (Figure 11,b). Such distribution pattern makes it possible to quantitate thrombi formation by counting the number of adherent thrombi-like structures.

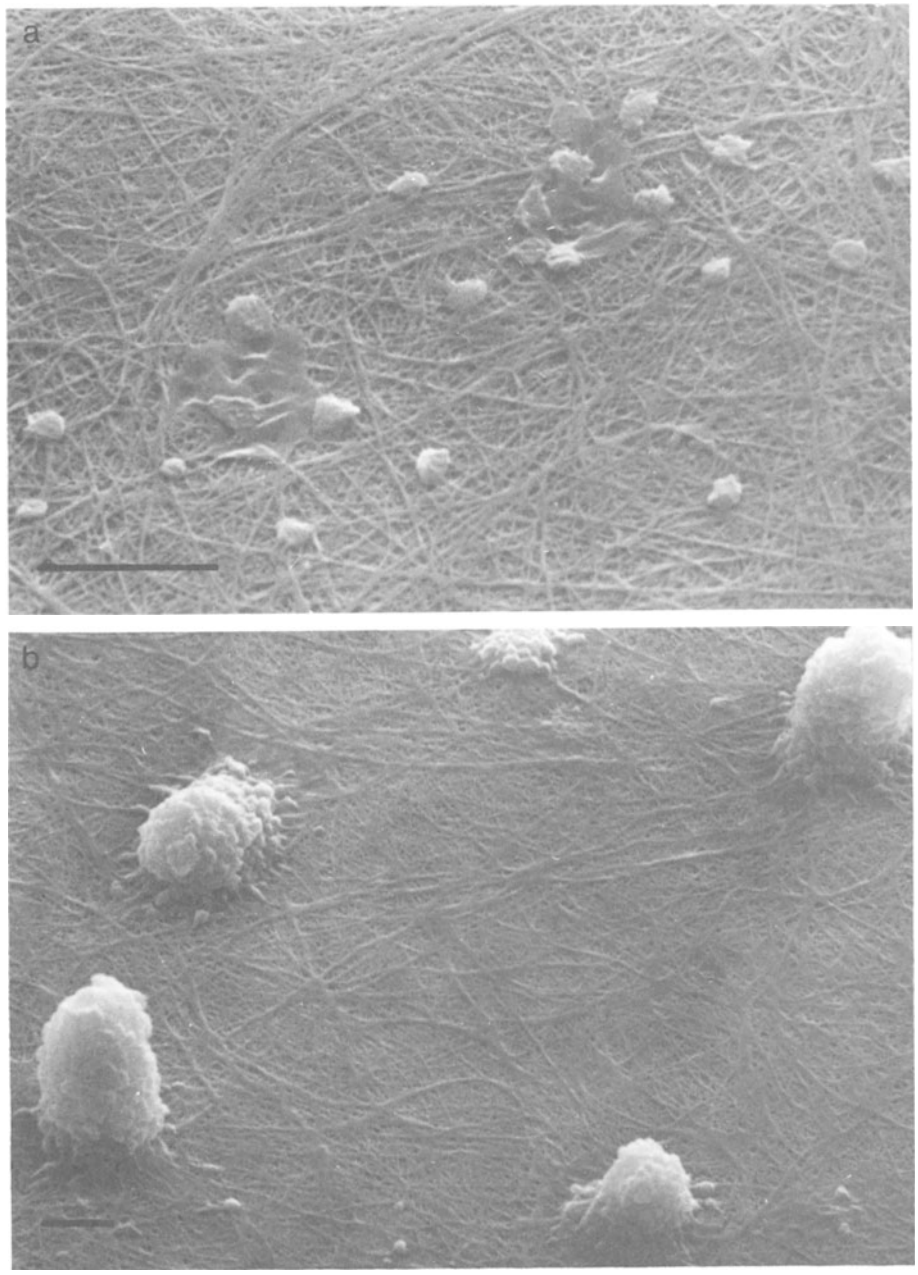


Fig. 11. U46619-induced formation of thrombi-like aggregates adherent to collagen. The binding of platelets with CSC-covered surface was performed in the absence of the inducer (a) or with I μm of U46619 (b). U46619 was added

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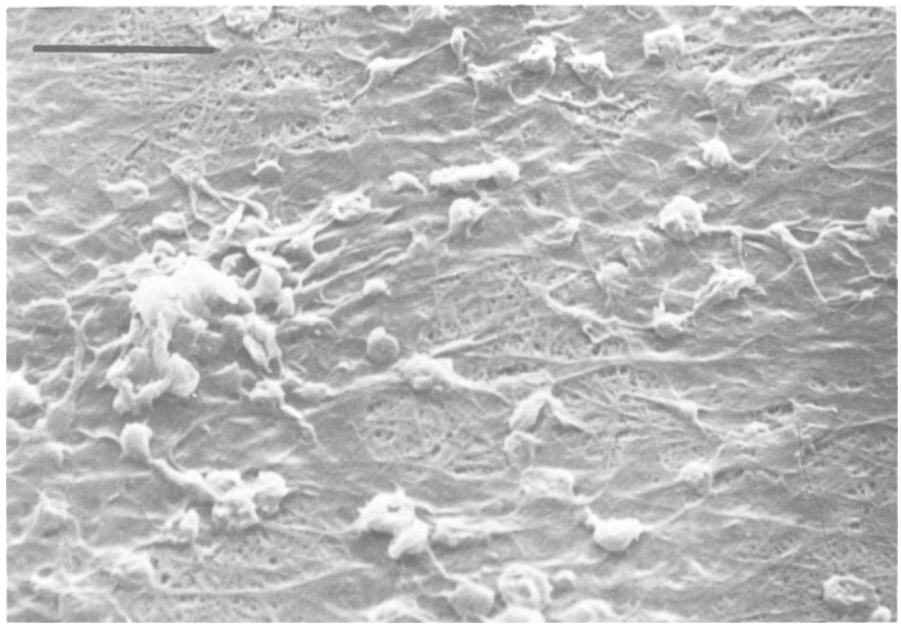

Fig. 12. Induction of platelet spreading on collagen substrate by arachidonic acid. Arachidonic acid (200 μM) was added into CSC-covered wells 3-5 min. prior to platelets. Intensive platelet spreading results in the formation of vast cell sheets on the substrate whereto single platelets from suspension, microaggregates and thrombi-like structures are binding. Scale bar - 10 μm.

Fig. 11 (continued)

into the wells 3-5 min. before the platelets and directly prior to stirring. In the absence of the inducer platelet interaction with CSC-substrate is characterized by initial attachment to the substrate, moderate spreading, and the adhesion of platelets from suspension to spread platelets (a). The inducer action leads to formation of thrombi-like structures discretely distributed on the substrate (b) and fields of confluent spread platelets (Figure 12). Scale bar - 10 μm.

The above mentioned activators also induce intensive platelet spreading (Figure 12). Spread platelets fuse and form the sheets covering up to 50% of collagen substrate. Single platelets, aggregates and thrombi-like structures bind to the upper surface of spread platelets (Figure 12).

Antithromboxane agent Trapidil (Rocornal) completely inhibits platelet spreading and thrombi-like aggregate formation on CSC (Figure 13). Trapidil also has antithrombotic effect when thrombogenic human Type I collagen is used. At the same time Trapidil inhibits arachidonic acid-induced thromboxane A_2 synthesis in platelets only by half (Figure 14). These data indicate that antithrombotic Trapidil action cannot be fully accounted for by the inhibition of thromboxane A_2 snythesis in platelets.

We have studied antithrombotic action of carbacyclin and 6β - PGI_I, chemically stable prostacyclin analogues. According to the data of other authors, carbacyclin and 6β - PGI_I prevent platelet aggregation induced by U46619 and arachidonic acid in human platelet-rich plasma. 10 ng/ml dose of carbacyclin and 50 ng/ml dose of 6β-PGI_I produce a half inhibiting effect. Complete inhibition is caused by 30 and 100 ng/ml, respectively (Figure 15). The effect of prostacyclin analogues on adherent thrombi-like structures formation

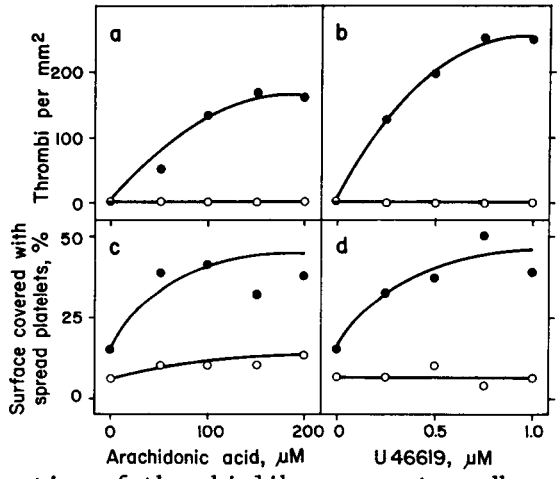

Fig. 13. Formation of thrombi-like aggregates adherent to collagen (a,b) and platelet spreading (c,d) induced by arachidonic acid and U46619. Inhibitory Trapidil effect. Trapidil, 1 μM (○——○) or the buffer (●——●) were added into CSC-covered wells. Then, after 3-5 min. intervals platelets and different doses of arachidonic acid (a,c) or U46619 (b,d) were added following each other. The formation of thrombi-like aggregates and spreading were measured by SEM.

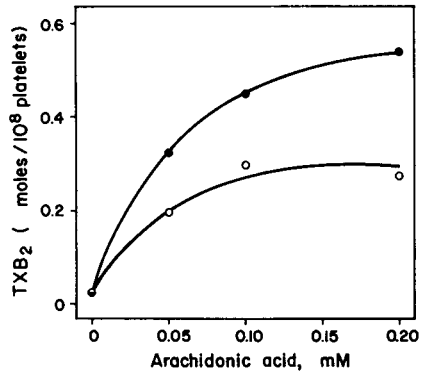

Fig. 14. Trapidil effect on arachidonic acid-induced thromboxane B_2 synthesis in platelets. The conditions under which the adhesion was carried out are given in the legend to Figure 13. Thromboxane B_2 was determined by radioimmunoassay in the unadherent platelets fraction in the absence (●——●) and in the presence (○——○) of 1 mM Trapidil.

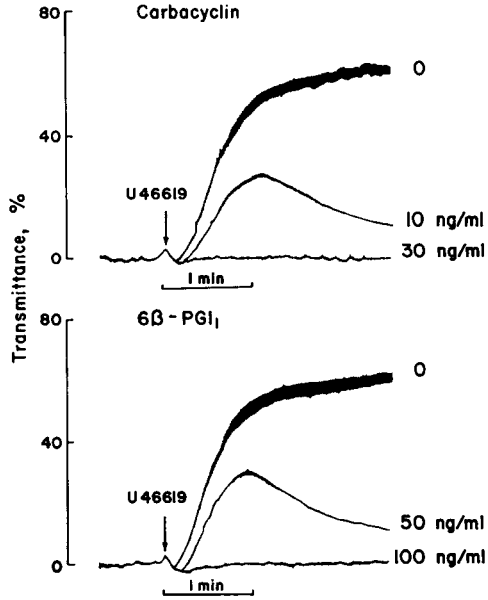

Fig. 15. Inhibition of U46619-induced platelet aggregation in suspension by stable prostacyclin analogues. First U46619 (1 μM) and then (after 1 min) the above mentioned doses of carbacyclin and 6β - PGI_I were added to the platelet-rich human plasma, and platelet aggregation was measured in a Payton aggregometer.

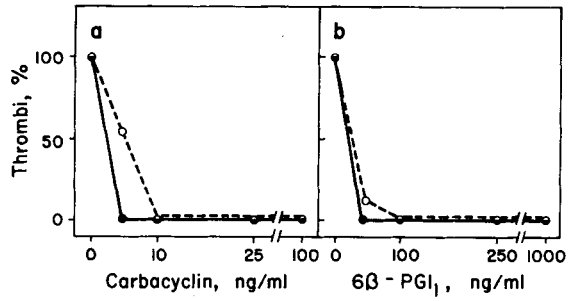

Fig. 16. Inhibition of adherent thrombi-like aggregate formation by stable prostacyclin analogues. Different doses of carbacyclin (a) or 6β - PGI_I (b) were added into CSC-covered wells; 3-5 min after they were followed by platelets and, finally, after the same interval - by 1 μM U46619 (○——○) or 200 μm arachidonic acid (●——●). Thrombi-like aggregates were measured by SEM.

induced by arachidonic acid and U46619 has been studied. It was demonstrated that both analogues fully inhibit thrombi formation on collagen substrate, but to produce the effect a 10-fold lower concentration of carbacyclin is needed in comparison with 6β - PGI_I (Figure 16).

GENERAL CONCLUSIONS

The study suggests a simple model system, which makes it possible to study thrombogenesis on collagen-coated surfaces, namely:

1) to quantitate adherent thrombi-like platelet aggregates;
2) to study platelet spreading and their role in thrombogenesis;
3) to study the contribution of various collagen types to platelet-vessel wall interaction;
4) to study the modulation of adhesion and thrombi formation by humoral factors;
5) to perform the screening of antithrombotic and thrombolytic drugs.

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IMPAIRMENT OF FIBRINOLYSIS AS A RISK FACTOR FOR THROMBOSIS

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PATHOGENESIS OF ARTERIAL AND VENOUS THROMBOSIS

The factors generally accepted to be important in thrombus formation are known as Virchow's triad:[1] the vessel wall, the composition of the blood and the flow of the blood in the vessel. The components in the blood which play a role in the pathogenesis of thrombosis are the blood platelets, the coagulation system and the fibrinolytic system.

The contribution of the three factors of Virchow's triad to the pathogenesis of thrombosis is different in arterial as compared to venous thrombosis. The main cause of arterial thrombosis appears to be intimal atheroma. However, other factors also interfere since severe atheromatosis may exist without thrombosis while thrombosis may occur in the presence of minimal vessel lesions. There is a considerable overlap in the extent of atherosclerosis between patients who died from myocardial infarction and patients who died from other causes, although the myocardial infarction group as a whole, showed more coronary artery disease.[2] Atheroma may lead to platelet adherence to the exposed subintimal layer, leading to formation of a platelet plug which is then covered by fibrin (white thrombus). Platelet aggregation on a diseased vessel wall appears to be the primary event in arterial thrombosis; the formation of fibrin is secondary although it may largely contribute to the occlusion of the artery.

Astrup[3] suggested that impaired fibrinolysis may lead to the persistence of fibrin deposits at sites of luminal injury, the incorporation of the fibrin into the vessel wall, and ultimately, the further development of degenerative atherosclerotic changes.

The key event in myocardial or cerebral infarction is not the development of the arterial lesions themselves, but the occlusion of an artery by a thrombus. There is indeed little correlation between infarction and the severity of atherosclerotic plaques in the brain or coronary vessels, but a close correlation between infarction and thrombosis of a major supply vessel.

Venous thrombosis mainly occurs in areas with stasis and acccumulation of procoagulants. Venous thrombi essentially consist of fibrin and entrapped red cells (red thrombus). "Hypercoagulability" of the blood, such as encountered in the postoperative period, during pregnancy or in women on oral contraceptives greatly increases the frequency of venous thromboembolism. Deficient fibrinolysis may result in delayed clearance of fibrin and thus predispose to overt thrombosis.

Thus all three commonly encountered vascular lesions, namely cardiac infarction, cerebral infarction and venous thromboembolism have as their underlying pathological process thrombosis of critically situated vascular segments.

REGULATION AND CONTROL OF FIBRINOLYSIS

The blood fibrinolytic system consists of three main components: the pro-enzyme plasminogen, which can be activated by limited proteolysis to plasmin; plasminogen activators which may be of different origin; and inhibitors which rapidly neutralize plasmin or which may interfere with the activation of plasminogen.

During the past few years, specific molecular interactions between plasminogen activator and fibrin, between plasminogen and fibrin and between plasmin and α_2-antiplasmin have been described, on the basis of which a molecular model for the regulation of fibrinolysis *in vivo* was proposed.[4,5]

Extrinsic plasminogen activator has a weak affinity for plasminogen in the absence of fibrin (K_M = 65 μM) but a much higher affinity in the presence of fibrin (K_M between 0.15 and 1.5 μM).[6,7] This increased affinity appears to be the result of a "surface assembly" of plasminogen activator and plasminogen on the fibrin surface. In this reaction plasminogen binds to fibrin primarily *via* specific structures called the "lysine-binding site". Thus one way of regulating fibrinolysis is at the level of plasminogen activation localized at the fibrin surface.

Plasmin is extremely rapidly inactivated by α_2-antiplasmin ($k_1 \approx 10^7\ M^{-1}s^{-1}$ [8]; the half-life of free plasmin in the blood is therefore estimated to be approximately 0.1s. Plasmin with an occupied lysine-binding site is however inactivated 50 times slower

by α_2-antiplasmin. Reversible blocking of the active site of plasmin with substrate also markedly reduces the rate of inactivation by α_2-antiplasmin. From these findings one can extrapolate that plasmin molecules generated on the fibrin surface, which are bound to fibrin through their lysine-binding sites and involved in fibrin degradation, are protected from rapid inactivation by α_2-antiplasmin. Plasmin released from the fibrin surface would however be rapidly inactivated by α_2-antiplasmin.

IMPAIRMENT OF FIBRINOLYSIS AS A RISK FACTOR FOR THROMBOSIS

Impairment of fibrinolysis at several levels has been identified and shown to be associated with an increased tendency to thrombosis.

Deficient Synthesis and/or Release of Plasminogen Activator from the Vessel Wall

Isaacson and Nilsson[9] found a defective release of plasminogen activator from the vessel walls during venous occlusion and/or a decreased plasminogen activator content in walls in superficial veins in about 70% of a large series of patients with idiopathic venous thrombosis. This association between non-acute venous thrombosis and a defect in the fibrinolytic system is closer than that between the former and any other known disturbance of the hemostatic balance.

Phenformin combined with ethylestrenol stimulated the release of the fibrinolytic activity in the vessel wall in these patients and was found to be associated with a diminished frequency of thrombotic episodes.[10]

Johansson et al.,[11] found a defective release but a normal content of plasminogen activator in the vessel wall in members of a large family with a high incidence (37 percent) of deep vein thrombosis. Thus, this family appeared to suffer from a hereditary thrombotic tendency in association with a defective release of plasminogen activator; the pattern of heredity was however not clear.

Clayton and coworkers[12] determined several clinical and laboratory parameters preoperatively in women undergoing gynecological surgery and measured the occurrence of postoperative deep vein thrombosis. On the basis of three clinical parameters (age, percentage overweight for height, presence of varicose veins) and two laboratory tests (the euglobulin clot lysis time and the level of fibrin(ogen) related antigen in serum) they could construct a preoperative index which allowed to make a clinically useful separation between patients subsequently developing deep vein thrombosis or not. Of these parameters, a prolongation of the euglobulin clot lysis time was the most discriminating to identify patients who would develop thrombosis.

Rákóczi and coworkers[13] have applied Clayton's index to a comparable group of gynecological patients and confirmed that a low level of plasma plasminogen activator preoperatively is associated with an increased risk of developing postoperative deep vein thrombosis.

Deficiency or Functional Defects of the Plasminogen Molecule

Congenital plasminogen deficiencies have so far not been described and may be incompatible with life. Plasminogen production is however delayed in the human fetus; an infant will reach adult levels at the age of 7-10 months. Ambrus et al.,[14] have therefore performed a double-blind, randomized study in which 500 premature infants were treated with plasminogen or placebo intravenously within 60 minutes of birth. They found a substantial decrease in severe clinical respiratory distress, death caused by hyaline membrane disease, and total mortality in the plasminogen-treated infants as compared to the controls. This study may attest to a vital function of the fibrinolytic system, at least during the first hours after birth.

Abnormalities in the plasminogen molecule resulting in defective activation to plasmin have been described by several authors.[15,16] Some of the affected individuals presented with thromboembolic disease, which became prominent only during their adult life.

Increased Levels of Inhibitors

Some reports suggest that impaired fibrinolytic activity due to the presence of increased levels of inhibitors of fibrinolysis is associated with a thrombotic tendency. In these studies the identity of the inhibitors was not established.

CONCLUSION

It appears that thrombosis is a disease of mosaic etiology, caused by multiple interacting factors such as lesions of the vascular wall, stasis of blood, hypercoagulability and deficient fibrinolysis.

Although the role of deficient fibrinolysis in the pathogenesis of arterial thrombosis and of atherosclerosis is not firmly established, there is ample evidence that impairment of the fibrinolytic system which may be localized at the level of deficient synthesis or release of plasminogen activator from the vessel wall, abnormalities in the plasminogen molecule or increased levels of inhibitors predisposes to venous thromboembolic disease.

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BLOOD TESTS FOR THE DIAGNOSIS OF THROMBOEMBOLISM

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In the past ten years, there has been an explosion of knowledge in the field of hemostasis and thrombosis. During this time, there has been a great deal of interest both for the clinician and the investigator in finding blood tests that can be used to predict thrombosis in high-risk patients or to confirm or exclude the diagnosis of thrombosis when it is clinically suspected. In addition, attempts have been made to find tests that can be applied in the investigation of the role of thromboembolism in the genesis and the complications of ischaemic heart disease, peripheral and cerebrovascular disease and in the vascular complications of diabetes.

Numerous reports of the relationship between blood tests and thrombosis have appeared in the world literature and have been the subject of a number of recent reviews.[1-6] The interpretation of the reports has been fraught with many difficulties because the tests have lacked sensitivity and specificity and because the experimental design of the studies has been adequate. For the most part they have provided a stimulus to perform more rigorously designed studies.

The methodology is now available to carry out properly designed and controlled trials. Firstly, sensitive and specific biochemical and immunochemical tests have been developed which have allowed the detection of very small concentrations of the products of intravascular thrombin generation, intravascular fibrin formation and products of platelet activation and release. In addition, methods to detect circulating activated clotting factors and complexes of activated clotting factors with their inhibitors have been developed. The second major advance has been the development of readily available objective techniques to diagnose thrombosis, both arterial and venous. Finally, advances in the field of epidemiology and in the

design of clinical trials have enabled more definitive studies on the laboratory diagnosis of thrombosis to be carried out.

There are a number of criteria that should be fulfilled to provide evidence of a relationship between an abnormal blood test and a clinical thrombotic event. These criteria include an adequate study design, consistency, biologic gradient and biologic plausibility.

The relationship between an abnormal test and thrombosis might be either causal or incidental and in practice, it may be difficult to differentiate between these. In addition, it may be difficult to determine whether an abnormal test precedes or occurs as a consequence of the thromboembolic event. However, if a relationship can be demonstrated, it is of practical importance because it may be used as a marker or predictor of a thrombotic event and may provide valuable information on the pathogenesis of thrombosis.

Although the pathogenesis of arterial and venous thrombosis is similar, there are a number of important differences. In arterial thrombosis, the interaction between platelets and damaged blood vessel wall may be the initiating event whereas in venous thrombosis, activation of blood coagulation and the formation of a mass of intravascular fibrin is the predominant feature. It is likely therefore, that these differences will be reflected in changes in different blood tests. In addition, arterial thrombosis frequently complicates arteriosclerosis and changes in the blood may be secondary to the arteriosclerotic process itself, whereas in the case of venous thromboembolism, the blood changes are probably more directly related to the occurrence of the thrombotic event.

In venous thromboembolism, transient abnormalities in blood may occur in association with tissue injury which predisposes to thrombosis. The systemic response to injury includes a non-specific acute phase reaction and a more specific response of activation of blood coagulation. The non-specific acute phase reaction includes elevation of a number of plasma proteins including fibrinogen, factor VII, alpha 1-antitrypsin and other alpha globulins, leukocytosis, thrombocytosis and fever. Increase of fibrinogen and factor VIII results in a shortening of coagulation tests such as the activated partial thromboplastin time although there is no evidence that this change predisposes to thrombosis. An increase in alpha 1-antitrypsin concentration is associated with an increase in antiplasmin activity which results in a decrease in blood fibrinolytic activity which may be causally related to the development of thrombosis. Tissue injury is also associated with systemic activation of blood coagulation. This may be caused by release of tissue thromboplastin or in the interaction of plasma coagulation factors and platelets with injured vessel walls. When thrombin is generated changes in peripheral blood which can be detected by laboratory tests may occur.

Thrombin releases fibrinopeptide A and B (FpA, FpB) from fibrinogen and through a feedback mechanism which activates factor V and VIII. Thrombin also stimulates prostaglandin synthesis (Thromboxane A_2, TXA_2) by platelets and stimulates the platelet release reaction (Betathromboglobulin, BTG; Platelet factor 4, PF4). These products of thrombin action can be detected by sensitive radioimmunoassays in blood. The fibrinolytic enzyme system may also be activated which produces soluble fibrin degradation products (FDP) from fibrin which can be detected in plasma or in serum.

Many of these tests have been applied for the diagnosis of venous thromboembolism but none has been found to be sufficiently sensitive or specific.

In deep vein thrombosis (DVT), the core peptide of fibrinogen, Fragment E, has been found to be useful to exclude a diagnosis of deep vein thrombosis in high risk patients.[7] Betathromboglobulin was originally introduced as a new blood test for the diagnosis of deep vein thrombosis and has been critically evaluated and found to be of no diagnostic value although increased urine BTG concentration may predict occurrence of DVT in high risk patients.[8]

In arterial thrombosis, which usually occurs as a complication to arteriosclerosis or in the presence of prosthetic surfaces, the initial process involves the reaction of platelets with damaged vessel walls or arterial surfaces whereupon they adhere, release the contents of their granules and aggregate. Thus, tests that reflect platelet activation, aggregation or release may be abnormal in arterial thromboembolism. These include measurement of platelet release proteins, betathromboglobulin and platelet factor 4, or measurement of products of prostaglandin synthesis including thromboxane B_2. If the process is extensive or continuous, platelet survival measured by isotope techniques may be used. If occlusive thrombi occur in the arterial circulation, blood changes may also occur as a consequence of tissue infarction which includes typical acute phase reaction response. The process of blood coagulation may be activated both locally and systemically leading to changes similar to those described for venous thrombosis.

The use of blood tests for the diagnosis of acute arterial thrombosis has been of limited value but they have given useful information on the pathogenesis of the process.[9]

A number of abnormal blood tests have been shown to be associated with an increased risk of thrombosis and some of these have been reported to be predictive of thrombosis.

The best documented of these is antithrombin III deficiency. Antithrombin III is a protein that inhibits activated factors XII, XI, X and IX and thrombin and there are a number of reports of

idiopathic and secondary venous thrombosis occurring in families with Antithrombin III deficiency, which is transmitted as an autosomal dominant trait. Secondary Antithrombin III deficiency occurs in patients with liver disease, patients on estrogen therapy and in premature infants. In some cases of Antithrombin III deficiency, unusual episodes of arterial thrombosis occur.

Dysfibrinogenemia is a disorder which results in a prolonged thrombin clotting and reptil times in the presence of normal or decreased levels of plasma fibrinogen and is associated, paradoxically, with reports of recurrent venous thrombosis. There is one specific fibrinogen abnormality Fibrin Oslo in which there is a familial incidence of recurrent venous thrombosis.

There are anecdotal reports that patients with persistent thrombocytosis associated with myeloproliferative disorders or after splenectomy have an increased risk of thrombosis but moderate postoperative thrombocytosis and in most patients with splenectomy, there is no increased risk. Patients with polycythemia have an increased risk of arterial and venous thrombosis which is contributed to by increased plasma viscosity and thrombocytosis.

These is one report of recurrent venous thrombosis in a patient with reduced functional but normal immunological levels of plasminogen.

A number of studies have reported an association between defective fibrinolysis and thrombosis. Defective fibrinolytic activity has been reported in patients on oral contraceptives, during the last trimester of pregnancy, in patients with malignant disease and in obese patients. There is, however, no conclusive evidence that the predisposition to thrombosis in these clinical states is associated with a decrease in fibrinolysis. There is a specific condition of recurrent superficial or deep vein thrombosis which has been shown to be associated with reduced fibrinolytic activity measured by vessel wall plasminogen activator activity or as circulating plasminogen activator.

Increased levels of coagulation factors such as factor VIII accelerate blood coagulation in vitro but there is no evidence that increased levels lead to increased rate of in vivo fibrin formation. There have been several reports of increased concentrations of factors VII, VIII and V and accelerated thromboplastin generation in patients with a history of thrombosis. There is, however, no evidence that these changes have a causal role. Levels of coagulation factors are increased in pregnancy and in patients on oral contraceptives but there is no evidence that these changes are associated with an increased risk of thrombosis.

Platelet survival has been reported to be reduced in patients with arterial thromboembolism, vasculitis, homocystinemia, chronic valvular heart disease, prosthetic heart valve replacement, prosthetic arterial grafts, arteriovenous shunt, recurrent venous thrombosis and ischaemic heart disease. However, the reports are inconsistent and often conflicting. This may be related to differences in the experimental technique used to perform the test and in the method used to calculate the results. Abnormalities in platelet adhesion, aggregation and coagulant activity have been reported in various thrombotic disorders but the results are variable and the tests are insufficiently sensitive or specific to be of clinical value. Circulating platelet aggregates are measured either by a screen filtration method or by platelet aggregate ratios which have been reported in patients with ischaemic heart disease, transient cerebral ischaemia, recurrent venous thrombosis and myocardial infarction, but it is uncertain whether these techniques actually measure aggregates in vivo or whether it is an in vitro artefact.[9]

The development and application of specific tests for thrombin generation, fibrin formation and its dissolution, and for activation of platelets have provided valuable information about the pathogenesis of thrombosis. However they have only a limited role in the management of patients with thromboembolic disease. There are currently no blood tests which are appropriate for use as predictors of post-operative deep vein thrombosis. High-risk patients can be recognized by well-established clinical criteria although there is some evidence that measurement of fibrinolytic activity may improve the predictive power but the benefit is marginal and does not justify its routine use. As far as the investigation of patients with proven thrombosis is concerned, the measurement of Antithrombin III concentration is the only test that should be performed routinely. On the arterial side of the circulation, increased platelet turnover has been reported with arterial thromboembolism but there is considerable overlap of patients and controls and the results cannot be used in clinical practice. Elevated levels of platelet specific proteins have been reported in arterial thromboembolism but the significance of these findings is as yet uncertain.

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HEART MICROCIRCULATION

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It is not by chance that our Symposium is dedicated to the peculiarities of the macro- and microcirculation in the heart work in normal and pathological conditions. The heart blood supply is a complex of mechanisms and it is provided by the coronary vessels, though the heart microcirculation is a subsystem of the general heart circulation. It is important to note, that the epicardium layers and the myocardium middle layers are supplied with the blood by other branches, as compared with the endocardium layers. The branches of the coronary vessels, entering the endocardium, ramify and form the anastomosing arcades, which interlace above the endocardium. Smaller arteries continue into the arterioles and capillaries and then venules, which gather in the veins. About 85% of the blood from the left coronary artery outflows from the myocardium into the coronary sinus and then into the right auricle. About 15% of the blood enters the right ventricle via Tebesia's veins. The rest small portion of the blood outflows into the left ventricle. The blood outflowing from the right coronary artery and flowing through the capillaries, enters the right auricle and ventricle via the anterior cardiac veins. The blood filling rate in the vascular bed makes up 6-14 ml per 100 g. of the myocardium and it comes for the account of the venous vessels. The microcirculatory part of the coronary circulation begins from the small arteries (150÷250 mkm) and continues by the collecting venules. Thus, the microcirculatory bed is organically involved in the coronary (regional) circulation of the heart and represents its integral part, reflecting the functional and morphological peculiarities of the heart circulation. This corresponds to the general proposition, that the organ microcirculatory unit - the so-called modules (W. Fransher and H. Wayland, 1972; V. V. Kuprianov

et al., 1975) - composing the microcirculatory bed, are intimately connected with the organ surrounding tissues. They organize peculiar "quantums" of the organs and tissues, therefore we have called them "functional elements" of organs. The functional elements of the organs and tissues are intermediate in their transition from the cellular level of the organism integration to the organ-tissue one. Our investigations have shown, that in the primary lesions of the nervous system (neuritis, cutting), significant disturbances were developed not only in the muscular tissue, but in the capillaries and venules, in which the direct (synaptic) connections with the nervous system were not visible. Due to the functional elements the heart microhemocirculatory system is closely connected with the organ-tissue physiology and pathology. The myocardial tissues have the functional elements, consisting of the formations specific for these tissues. The microcirculatory unit, finely adapted by its architectonics to the heart function, is the center of such an element. The blood vessel capillaries form a complicated network, passing via the fissure-like spaces between the heart muscular cells. Thus, the latter are oriented around the microcirculatory unit. The myocardial cells (fibers) are known to be represented by two types: muscular cells and specialized cells of the conductive system. Therefore there are two types of the heart functional elements. The fibrous formations and cells of the connective tissue (mast cells included) are oriented around the microvessels in the space between the myocardial cells. They play equally a supporting and exchange-physiological role. The lymphatic capillaries are related hereto. The nerve formations (parasympathetic and sympathetic efferent and afferent nerve endings) are interlaced with the architectonics of the heart functional element. So, the structure of the heart functional element is optimally adapted to its activity. The heart function, in its turn, is realized by participating of physiologically active substances (first of all, bioamines - catecholamines as well as prostaglandins, metabolites a.o.), which are organically involved in the microcirculation regulatory mechanisms.

As a part of the whole blood circulation system, the heart microhemocirculation has numerous and significant connections with the general hemodynamics. The state of all the parts of the microcirculatory bed may affect, in any case, on the main homeostatic parameters of the general hemodynamics and, first of all, on the arterial pressure.

The blood flow in the cardiac muscle, which is necessary for its effective metabolism, is regulated, in the end, at the microcirculatory level. Of a particular significance is the unity of metabolism, blood supply, regulation and activity, which is realized at the level of the heart functional element. Hence, we may conclude, that the heart microcirculatory system plays the important role as

in the myocardium physiological conditions (i.e. in the healthy state) and in the ischemic lesions. In this case, of a great importance in the normal heart activity is the oprimal correlation between the lumen size, blood pressure and blood flow velocity in the microvessels in the cardiac cycles, when the perfusion of the exchange part of the heart microcirculatory bed is realized.

For a long time, all these peculiarities of the heart microcirculation had been rather difficult for being studied. However, for the recent years, it has become possible to elaborate principally new methods for studying the heart microcirculation in the normal and pathological conditions. There appeared the techniques for measuring the microhemodynamics by using the isotopic technics, radioactive microspheres. The methods of the heart biomicroscopy have been considerably developed. The electromicroscopic examinations, including the cardiac muscle biopsy during the heart surgery, became of a great value. Though there is much to be clarified, the ways have been outlined and the valuable data obtained in the following trends of investigations:

1. The myocardial blood flow, its regulation at the microcirculatory level, transmural blood distribution and optimal perfusion of the heart microvessels, including blood rheological properties.

2. The capillary-cellular metabolism in the cardiac muscle, role of the capillary-venular departments, ways and mechanisms of the transcapillary transport.

3. The optimal coronary circulation and its correlation with the dynamics of the myocardial metabolic processes.

A great attention is also paid to the study of the heart microcirculation disturbances.

The work in these directions have been made in many countries, and some results were summarized at the 1st Symposium held in September 1980 in Garmisch-Partenkirchen (FRG) at the XI European Conference for Microcirculation (see Biblioteca anatomica, N. 20, 1981).

I would like to cite some examples from our data. The regulation of the myocardial blood flow at the microcirculation level, is realized mainly by changing the tonus and, hence, the size of the muscle arterioles lumen. It is disputable, whether the microvessels contractile endothelial cells (particularly in the capillary wall) are involved in this process. Although in our laboratory there are available some data, evidencing such a possibility. In the heart vessels the precapillary sphincters are less pronounced (or absent) and they do not regulate the myocardial

blood flow. It was shown by us, that under the effect of histamine, EDTA and the denervation, produced in the cytoplasma, the formed fascicles, consisting of the actin-like microfibrils (by diameter not more than 8 mm), appeared in the exchange vessels endotheliocytes. Using cytochalasin B - a specific polymerization blockator of the actin proteins - it is possible not only to prevent the formation of the fibrillar actin, but to dissolve the early formed actin gel. However, it was observed, that the "endothelial contraction" was fragmentary and differently pronounced. In this case, there takes place, without fail, the structuralization of the contractile apparatus in the endothelial apparatus, in distinct from the contraction of the muscles cells. The nervous system and local physiologically active substances are involved in the regulatory mechanisms of the myocardial blood flow.

We have specially studied the myocardial microvessels in the adult intact rats and mice by the electronmicroscopic method. Due to the use of serial sections, we have convinced in a number of cases, of the presence of the nerve endings not only at the arterioles, but also on the capillary wall. We have revealed some variants of the interrelation between the nerve terminals and effectory structures: similar terminals were observed directly at the endothelium and pericytes; at the myocardial cells; sometimes they "contacted" simultaneously with the capillary and muscular cell; however, more often they were disposed in the interstitial space at some distance from the vessels and muscle cells, reminding the "free endings". Both, the cholinergic and adrenergic endings were found. It appeared, that the microvessels regulation of a capillary type, was realized, most likely, by their innervation, according to the dissynaptic type, with the neuromediators free diffusion towards these microvessels. Depending on the distance, passed by the neuromediators, the nervous regulation of the capillary wall may be the "direct and fast", "direct and delayed" and conjugated with the myocardium cells regulation. It is clear, that the mediator, releasing from free terminals, diffuses in all sides and effects on the whole "microregion" with its cellular and non-cellular elements. For the account of such terminals, the indirect regulation of the microvessels function may be realized immediately the vasoactive substances, if, under the neuromediators action, these terminals are liberated from the connective tissue cells, particularly from the mast cells. Thus, our data and those of other authors evidence that the significance of the nervous, in particular, adrenergic regulation of the heart microvessels, was under-valued. The use of such powerful blockators as α or β adrenoreceptors allowed to give a favourable value in this respect.

The coronary blood flow is directly connected with the oxygen consumption and, therefore, such factors as the oxygen tension (P_{O2}), carbonate gas (P_{CO2}), hydrogens (H^+), kalium (K^+), osmotic pressure, lactate, prostaglandins, serotonin and adenosin are under

the special intensive study. Adenosin is known to provide the metabolic dilatation of the coronary vessels and to easily penetrate through the myocardial cells membrane. Its concentration rapidly increases at any stage of the sensitivity to the hypoxia (for instance, in ischemia), thus promoting the vessels dilatation.

However, it should be taken into account, that the coronary vessels perfusion depends on many other factors and, first of all, on the aorta blood pressure and then on the difference in the arterio-venous pressure, peculiarities of the heart muscle contraction, metabolic processes, myocardium intramural pressure and blood rheological properties (viscosity, erythrocytes aggregability and other blood elements). It is especially important with regard, that the size of the heart capillaries lumen makes up 3-5 mkm. It is generally known, that the difficulty in the myocardium vessels perfusion by the ventrical contractions, is a complicating factor of the blood flow in the heart microvessels. Due to the micro-circulation peculiarities during the cardiac cycle, the study of the myocardial regional blood flow with the aim to elucidate the transmural blood distribution in different parts of the myocardium, has become an important advance of the recent years. With this purpose, we have widely used the mobile radioactive microspheres (10 mkm), which being administered in the left auricle, mixed up with the blood and then penetrated into the coronary vessels. In the left ventricle, the intramyocardial pressure was found to be higher in the subcardial layers and it fell almost up to zero in the subepicardial layers. Due to such an unequal distribution of the pressure in the left ventricular wall, the blood flow in the subcardial layers was realized only during the diastole, and in the subepicardial layers - during the systole and diastole.

In this respect, the study on the interrelation of such blood flow values as : pressure-diameter-velocity in the heart micro-vessels is of a great interest.

We have studied the ways and mechanisms of the capillary-cellular exchange in the cardiac muscle as in the experiment and in the clinic. These experiments allowed us to reveal a significant role of the microcirculation capillary-venular department in this exchange. It should be taken into account that there are two barriers between the blood and myocardial cell: capillary wall and sarcolemma. The exchange is realized via the capillary and venular walls by several ways, depending on the molecule size and lipo-solubility degree of the penetrating substance. These are the diffusion, filtration, microbubbling and passage of substances via the interendothelial intervals. The penetration is easy for the liposoluble substances and limited for the water soluble and large molecules. The electromicroscopic equivalents of small and large pores have been already studied. The substances passage via the sarcolemma is supposed to be more complicated.

Now briefly about the generally-pathological characteristics of the heart microcirculation disturbances. In my opinion, they may be divided into three groups:

1. Vascular wall disturbances. They include the destructions and deformations of the endothelial cells, changes of the wall permeability, adhesion of the blood elements, cells diapedesis via the microvessel walls and, finally, microhemorrhages. By the frequency, the disturbances of the vascular wall permeability are to be put in the first place.

11. Intravascular disturbances. Hereto should be related three processes, playing an important role in the heart pathology - blood rheology disorders, coagulation and thromboembolism disturbances, changes of the blood flow velocity, connected with the difficulties in the blood perfusion (up to a complete microvessel occlusion).

111. Intravascular changes. They include the injuries of the perivascular components of the functional element, and, first of all, the mast cells activation, microlymphatic circulation disorders and, finally, the microvascular bed involvement in the tissue dystrophic process.

A particular emphasis should be made on the important role of the blood rheological disturbances, which impede the perfusion through the microvessels lumen. In this case, two parameters are of a large importance: blood viscosity and critical radius value of the vessel lumen, both depending on the following factors: the appearance of thrombi in the blood liquid part, aggregation of thrombocytes and erythrocytes; deformation and changes of the erythrocytes rheological properties, hematocrit, blood plasma viscosity. The factors, resulting in the erythrocytes aggregation, may be divided into two groups: 1) influence of the environment, i.e. the shift rate and tension, concentration of the plasma proteins, plasma ion composition and cells concentration; 2) the erythrocytes tendancy to aggregate (deformability of cells, affinity of cellular surfaces to the macromolecules, surfaces electrical potentials).

As it was shown in our experiments, the erythrocyte aggregates might be of a different form and structure in the pathological conditions, depending on the organism state and peculiarities of the pathological process (shock, hypoxia, trauma etc).

The development of some forms of the cardiovascular diseases (heart coronary disease, hypertension, kidney failure, atherosclerosis, thrombosis, peripheric vascular diseases, diabetes) is known to depend not only on the environmental factors, but on the genetic ones, which, in their turn, are likely to influence on the

peculiarities of the intravascular changes of the blood rheology and, hence, on the degree of its perfusion via the microvessels lumen.

Finally, some words about the correlation between the coronary blood flow and myocardial metabolism. This is an important, but, at the same time, difficult problem. In the healthy heart, the blood flow rate in the microcirculatory bed is proportional to the oxygen consumption. Hence, the vasomotor processes are dependent directly on the myocardial metabolism. It is also likely, that there exists a direct interdependence between the rich in energy phosphatides and the liberation velocity of the vasoactive metabolites. It should be noted, that the mechanisms of this interconnection have not been yet clarified even for the norm. The matter is that a lot of factors, which act simultaneously and cooperatively, participate in these processes. Between these factors there is not a direct (linear), but probabilistic (cybernetic) interconnection. The interaction processes between these factors should be determined quantitatively, and not qualitatively.

We have made special comparative studies on the myocardium metabolic and functional changes in connection with the microcirculation disturbances. These disturbances have been produced by a simultaneous administration of high-molecular dextran and vasopressin in the rabbits and rats. The heart ischemia and hypoxia, caused by such a way, resulted in a change of the mitochondria oxidative metabolism, a decrease of the respiration and phosphorylation, and of the ATP concentration under an increase of the creatine-phosphate level. The glycogen content and phosphorylase "a" activity were unchanged.

Of a great importance are our data evidencing that the disturbances of the myocardial microcirculatory bed in hypercholesterinemia represent an initial link of the coronary atherosclerosis. Hence, the microcirculation disturbances of a dystrophic nature may be the starting points in the further development of disturbances in the microvessels lipoid metabolism.

Further studies on the heart microcirculation will promote a successful development of this important field of the modern cardiology.

DYNAMIC CORONARY STENOSIS: THE ELUSIVE LINK BETWEEN CORONARY ATHEROSCLEROSIS AND CLINICAL MANIFESTATIONS OF ISCHAEMIC HEART DISEASE

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SUMMARY

Recent concepts on the roles of dynamic coronary stenosis in ischaemic heart disease are reviewed, with particular reference to episodes of transient myocardial ischaemia and their implications for patient management and therapy. The possible contribution of spasm and other obstructive mechanisms, such as platelet aggregation and thrombosis, to the various forms of angina pectoris, to myocardial infarction and sudden death are analysed in the light of clinical experience and clinical investigations. Based on these conceptual and clinical considerations, guidelines are suggested for future research and therapy. Long-term treatment with nitrates and calcium antagonists, which appear to be effective in preventing dynamic stenoses, resulted in low mortality and infarction rates over a period of two to four years, in a group of patients with ischaemic heart disease at high risk.

INTRODUCTION

Our understanding of coronary artery disease is rapidly expanding as a result of a major revision of traditional concepts. This revision is based on new observations which have disproven the deeply endowed theory that increased myocardial demand in the presence of critical coronary atherosclerotic stenosis was the only respectable cause of angina pectoris.[1-2]

Objective measurements[3-5] have convincingly proven the hypothesis that coronary vasoconstriction, or other factors interfering with coronary blood supply, are usually responsible for nocturnal angina,[6]

for angina at rest,[3-10] for cold-induced angina,[11] for "variant" angina caused by exertion,[12-13] and for the variable threshold of exertional angina, frequently observed in some patients.[14] It is important to stress the fact that "variant" angina is by no means the only electrocardiographic manifestation of coronary vasospasm but rather it represents only the most striking electrocardiographic change.[15] Indeed S-T segment depression or T wave changes can be caused by coronary vasospasm when ischaemia is not transmural.[3,5,16-18] It was also objectively demonstrated that coronary vasospasm may occur in vessels with an extremely variable degree of coronary atherosclerotic obstructions.[5,15] In those patients with severe critical atherosclerotic obstructions angina from vasoconstriction occurring at rest or on cold exposure or for variable levels of exertion may be associated with the traditional form of exertional angina occurring any time the patient exercises beyond a rather fixed, critical level of effort.[15] These views seem to be gaining acceptance[19,20] and find experimental confirmation.[21,22,23,24]

Furthermore the studies of patients with frequent anginal attacks at rest have shown that coronary vasospasm may cause sudden death[15,25] and, probably in association with platelet aggregation and thrombosis may be one of the causes of myocardial infarction.[26,27]

In this lecture I wish to discuss the role of coronary spasm and vasoconstriction in ischaemic heart disease with particular reference to episodes of transient myocardial ischaemia and its implications for patient management and for research

THE NEW UNDERSTANDING OF ANGINA PECTORIS

It is now well established that angina may be caused by different pathogenetic mechanisms even in the same patient: it can be secondary to increased demand beyond supply according to the traditional textbook theory, or it can be primary*: i.e. caused by other mechanisms not directly related to the presence of a coronary atherosclerotic stenosis which, in this case, is the by-stander or a favouring element rather than the culprit.[1,2,28] Coronary vasoconstriction is the only

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* The term "primary" borrowed from other fields of medicine is only meant to provide an immediate clear-cut separation between "secondary" angina, for which the diagnostic and therapeutic approaches are well established, from all other types of angina which will require specific diagnostic and therapeutic approaches. It is expressly used to emphasize that this differentiation, rather than the proof of the existence of coronary spasm, is the most important message of the new understanding of angina: thus, rather than bask in the now accepted concept of spasm, we will keep our minds open to the possible existence of other causes of angina.

cause of "primary" angina identified so far, possibly because it was more carefully searched for than other mechanisms such as transient platelet aggregation,[29] inappropriate vasodilatation,[30] small vessel disease[31] and alterations of myocardial metabolism.[32]

Furthermore, systematic study of angina patients has demonstrated that angina pectoris should be now considered only one of the possible manifestations of acute transient myocardial ischaemia,[1,2,15] which can be caused by different pathogenetic mechanisms. Indeed, acute transient myocardial ischaemia might manifest itself with typical chest pain (which usually follows minutes after its onset) or only with signs of left ventricular failure and/or with arrhythmias or it may remain completely asymptomatic (in spite of severe left ventricular function impairment and, often, of arrhythmias.[3,9,16,17]

The myth of coronary stenosis and of excessive increase of myocardial demand

The concept that an excessive increase of demand in the presence of atherosclerotic obstructions of the large coronary branches is the only respectable cause of angina has conditioned our diagnostic and therapeutic management of patients during the past three decades. By extrapolation, without objective evidence, it was commonly accepted that rest angina was caused by a sudden increase of heart rate and/or blood pressure, decubitus angina by an increased venous return, nocturnal angina by increased heart activity associated with dreaming. The common observation that these same patients sometimes had a good exercise tolerance was totally overlooked. Accordingly, cold-induced angina and the variable threshold of exertional angina so often observed, were attributed to variable levels of demand for the same external work of the heart.

Again by extrapolation, the identification of a coronary obstruction was automatically accepted as proof of the patients' symptoms. Conversely, in the absence of this proof, the ischaemic symptoms of the patient or the coronary arteriogram was questioned.[33] The attitude was similar to that of the police officer who incriminates an individual found on the scene of the crime just because he is well known for his previous crimes. Monitoring of the variables that control myocardial oxygen demand has clearly disproven this hypothesis.

Coronary Flow Reserve

According to the traditional mechanism, angina occurs because the possibilities of coronary flow reserve are transiently exceeded by myocardial demand. Although this is certainly a frequent event in patients with critical coronary obstruction, now it must be ob-

jectively proved in order to be accepted as the actual cause of the ischaemic episodes.

Animal experiments indicate that an acute reduction of the lumen of a major coronary branch by 85% still allows a three fold increase of myocardial flow[34] sufficient to allow a physical exercise of about one half the maximum working capacity. In chronic condition the stenosis, being compensated at least in part by collaterals, will cause an even smaller impairment of flow. These data are consistent with the rather frequent observation of patients with severe, proximal triple vessel disease and remarkably good exercise tolerance. Thus, even when acute ischaemia ensues during increased heart activity it should be considered "primary" unless it is proven that it occurs because coronary flow reserve has been exceeded and often the role of organic stenosis, per se, may be more important prognostically than pathogenetically.

Since organic stenosis, per se, are directly responsible for ischaemic attacks only when they reduce coronary flow reserve below the levels required by the patient, it becomes essential to evaluate practically the coronary flow reserve of the patient. Stress testing should establish the level of exertion that the patient can never exceed without signs of symptoms of ischaemia (as a percentage of his theoretical maximal working capacity): this gives an indication of the limitation to the increase of coronary flow caused by organic stenosis. The reproduction of the test may be a reasonable guarantee that the limitation is caused by organic rather functional transient obstructions.[2] Indeed, the positivity of the test, per se, may not give us a clue as to the actual cause of the patients' symptoms:

1. if the test is positive, but only at a maximal or submaximal level of work (which he may never reach in his ordinary daily life) it fails to explain anginal attacks occurring at much lower levels of cardiac work.
2. When the threshold is variable or the patient presents the phenomenon of walk through angina, ischaemia can be caused by a transient, dynamic stenosis which causes a reversible reduction of blood supply. This case is described by a recent typical case report of a patient with a variable threshold of exertional angina documented by stress testing: coronary arteriography performed during exercise revealed a transient increase of the severity of a stenosis in the LAD from 75% to over 90%.[14] A recent report[36] indicates that ischaemic changes occurring in walk through angina may be caused by coronary vasospasm. Transient ischaemia during ordinary activity occurring well below the maximal heart rate tolerated during exertion was documented by Holter monitoring.[37,38]

Reduction of coronary blood supply

Continuous haemodynamic monitoring has shown that the attacks of angina occurring at rest during the day, the early or late part of the night or after meals are not caused by increased demands.[3, 6-10] Furthermore, they usually occur at much lower levels of heart rate-blood pressure product than achieved without symptoms during exertion or during a pacing test.[39,40] In the majority of the cases the temporal sequence of events is remarkably similar to that observed in dogs following ligation of a coronary artery.[41] Thallium^{201} myocardial scintigraphy consistently shows a reduction of tracer uptake during angina at rest, transmural when the S-T segment is elevated[4] and diffuse when it is depressed.[18] Continuous monitoring of coronary sinus oxygen saturation has shown that the onset of the attacks is preceded by a pronounced drop of saturation thus providing the missing link for the demonstration that a reduction in flow preceded, and hence caused, the ischaemic episodes.[16,17] A reduction of coronary flow has been also shown by thermodilution during cold pressor test induced anginal episodes,[11] and following ergonovine.[42] Thus, dynamic transient coronary stenoses appear to play an important role in causing angina.

Role of coronary vasospasm and of platelet aggregation

In this era of transience of our understanding of the mechanism of ischaemia, reduction of coronary flow caused by vasoconstriction can be provisionally grouped together under the broad term of "spasm". As soon as we can identify different mechanisms for vasoconstriction in larger branches free from organic stenosis and in branches with severe stenosis, or as soon as we can identify constriction of smaller vessels, it will become appropriate to develop an appropriate nomenclature. In the present discussion we use "spasm" as a broad equivalent of vasoconstriction of epicardial coronary arteries.

Several pieces of evidence converge to indicate that vasospasm of the large epicardial coronaries is responsible for the transient reduction of myocardial blood supply in the presence of an extremely variable degree of atherosclerotic narrowing.

1. The angiographic demonstration of transient obstruction reversible by sublingual or intracoronary nitrates.
2. The reproducibility of the spasm by drugs such as ergonovine.[5, 43,44]
3. The prevention of anginal attacks by nitrates.[45]

These elements, although indicative of the role of vasospasm, do not allow any inferences on the role of platelet aggregation as a triggering factor or as a possible consequence of the blood stagnation and intimal damage caused by coronary spasm. Nor do they

allow us to exclude that transient aggregation of platelets may sometimes, per se, be responsible for a reduction of flow. The conflicting interpretation of the cyclic coronary flow variations across an experimental stenosis[46,47] confirms the complexity of the factors responsible for transient reduction of coronary flow. The demonstration that prostacyclin and thromboxane A_2 affect both platelets and smooth muscle, provides the ground for the possible causal role of both intravascular and of vascular wall obstruction in large arterial branches in the genesis of transient ischaemia.

Angiographic demonstrations of coronary vasospasm, once limited to "variant" angina at rest and to angina at rest in general, have now extended to "variant" angina during exertion,[12,13] to angina occurring in the post-exercise period[36] or with a variable threshold.[14] Ischaemia caused by reduced myocardial blood flow was demonstrated with the cold pressor test and attributed to increased alpha tone.[11]

Continuing investigations on a large scale will contribute to clarify the relative role of exhaustion of coronary reserve, limited by fixed organic lesions, and of transient dynamic stenoses, suddenly and transiently interfering with myocardial blood supply in the genesis of angina pectoris. Our experience indicates that the role of transient dynamic stenoses plays a large contributory role in the genesis of angina, together with atherosIcerotic obstructions. When carefully questioned angina occurring only and exactly for the same level of exercise is not common.

Is typical angina pectoris only the tip of the iceberg?

The traditional definition of angina as of "characteristic chest pain brought about by exertion and relieved by rest"[48] is certainly far too restrictive to be still acceptable. This conclusion is derived from a series of studies where the presence of transient myocardial ischaemia could be objectively assessed by electrocardiography, continuous haemodynamic monitoring and myocardial perfusion studies.[3-18]

This proposition demands a redefinition of angina pectoris, and the study of the natural history and epidemiology of the disease in this new, broader conception. We believe that the following points deserve careful consideration and further investigation.

Angina with preserved exercise tolerance

It was reported some years ago for patients with "variant" angina[49] but according to our experience, it can be observed quite frequently if electrocardiographic recordings are taken at the time

when patients experience the episodes of chest pain. Since these recordings may be difficult to obtain, these patients are usually dismissed with the assurance that their pain is not of cardiac origin. Indeed, the diagnostic problem in these patients is not an easy one. Unless they had some previous objective demonstration of ischaemia, such as unequivocal electrocardiographic ischaemic changes recorded after one episode, which would alert the physician, the normality of the stress test usually is considered a sufficient element to discard the ischaemic nature of pain. Even when more modern approaches are taken, it may be difficult to arrive at the diagnosis if no episodes occur during the Holter monitoring period or if the inappropriate leads were monitored. Accordingly, provocative tests[41,42] may be negative if the patient is in a relatively "cool" phase of the disease.[50]

Thus a careful follow up of the patient and a prolonged observation during the waxing phase of his symptoms may often be the only means to arrive at the diagnosis.

Painless myocardial ischaemia Continuous electrocardiographic and haemodynamic monitoring showed that pain is quite a late marker of ischaemia occurring well after obvious signs of left ventricular function and electrocardiographic abnormalities.[51] Furthermore, it may be completely absent.[3,7-9,52] Not only episodes of ischaemia may spontaneously resolve before the pain occurs, but sometimes, in some patients, also prolonged episodes of ischaemia with severe impairment of left ventricular function may remain completely asymptomatic or be accompanied only by dyspnoea, although often they may cause severe arrhythmias, including ventricular tachycardia and fibrillation.[15]

Painless episodes in patients with recurrent episodes of angina at rest often find their equivalent in stress testing when diagnostic S-T segment changes occur before, or in the absence of chest pain.

The observation of asymptomatic transient myocardial ischaemia should not be surprising since it is known that even myocardial infarction may be asymptomatic in about 10% of cases.[53] Continuous electrocardiographic ambulatory monitoring suggest that asymptomatic myocardial ischaemia may be quite frequent in some individuals.[46,47]

Atypical pain Having been forced to accept that transient myocardial ischaemia can occur in patients with preserved exercise tolerance and that it can sometimes be completely asymptomatic, we began to explore the possibility that atypical pain, such as sharp, inframammary or dull, localized, continuous precordial pain may be in some patients occasionally caused by transient myocardial ischaemia. Once more continuous electrocardiographic monitoring showed unequivocal changes indicative of myocardial ischaemia during these atypical symptoms. In one of those patients who presented

repeatedly with dull, mild inframammary pain with transient electrocardiographic changes, during angiography we were able to document repeatedly a transient complete spasm of the left anterior descending coronary artery.[54]

Thus it seems reasonable that an electrocardiogram taken at the moment of atypical pain, when clearly positive, may be useful for the diagnosis of atypical chest pain.

Anginal pain without electrocardiographic changes The absence of transient electrocardiographic changes during angina in patients with a history of ischaemic heart disease and with a grossly abnormal resting tracing is rather common. However, we were repeatedly unable to detect electrocardiographic changes in any of the 12 standard leads during typical severe anginal pain at rest, even in patients with normal resting electrocardiograms.

This puzzling observation poses two questions:

1. the sensitivity of the electrocardiogram in detecting ischaemia on certain occasions;
2. the possible extracardiac origin of typical anginal pain. Although the possibility of oesophageal spasm deserves consideration,[55] techniques capable of detecting ischaemia such as measurement of regional myocardial flow, metabolism or contractile function are required for a correct diagnosis in these cases. Anginal pain without ECG changes was shown to be associated with the appearance of transient impairment of ventricular function similar to those occurring during transient ischaemic episodes.[56]

CORONARY VASOSPASM AND MYOCARDIAL INFARCTION

One of the major difficulties in the study of the pathogenetic mechanisms of myocardial infarction is the inability of the physician to witness the onset of the event. The study and the close observation of patients who are under high risk of developing infarction allowed us to make some interesting observations[26] that appear to find support.[57]

The possible role of spasm

As recently reported[26] it appears remarkable that infarction developed in all cases in the same territory previously undergoing repeated transient episodes of acute myocardial ischaemia shown to be caused by vasospasm. Furthermore, in the patients in whom the infarction appeared to be small, transient episodes of ischaemia often recurred in the same territory. These observations indicate

a remarkable analogy between the pathogenetic mechanisms of angina at rest and of infarction.

In addition, in all patients the onset of the ischaemic episode which evolved in infarction was indistinguishable from the previous ones which were reversible, in particular it was not caused by any increased heart activity.[26] Thus, the demonstration that anginal attacks preceding the infarction were indeed caused by coronary vasospasm and that myocardial infarction clearly followed the development of spasm in one patient,[26] suggest that coronary spasm appears to be an initiating mechanism of myocardial infarction and perhaps of reinfarction in patients with recurrent anginal episodes. Since often myocardial infarction occurs at rest and since premonitory episodes of chest pain are frequently reported in carefully collected histories, it is conceivable that spasm may be frequently involved in the genesis of infarction. The recent observations of coronary spasm in some patients with acute myocardial infarction[60] supports this hypothesis.

Prolonged vascular occlusion

We were able to document a prolonged vascular occlusion at the onset of acute infarction in all the three patients in whom we performed coronary arteriography within a short time of the onset of pain. The occlusion could not be reversed by repeated intracoronary injection of nitrates in any of the cases. The first case has been described already.[26,27] Two other patients were investigated following the report of vessel reopening following intracoronary nitroglycerin injection in the early phase of acute infarction.[60]

A protocol was approved for emergency coronary artery injection of nitrates within one hour of the onset of an irreversible episode of myocardial ischaemia. Contrast injections were performed into the right coronary of two patients with persisting inferior ST-segment elevation and intractable pain, 30 and 40 minutes after the onset of symptoms and repeated after intracoronary administration of isosorbide dinitrate up to 5 mg. The vessel was occluded without distal filling respectively at the origin of the posterior descending and at the proximal third of the right coronary and remained occluded also after nitrates. According to the established protocol, after heparine infusion, on the next day a single contrast injection was repeated: the previous occlusion appeared partially reopened with the aspect of recanalized vessel and with good distal filling. Unfortunately, we have no information as to whether this aspect of recanalized thrombus was already present before the onset of MI (which was documented by typical electrocardiographic changes and serum enzymes elevation in both cases). At present it is impossible to tell to what extent the prolonged occlusion documented in these two patients and in the patient previously reported[26] was sustained

by an irreversible spasm, by platelet aggregation and/or by thrombosis which subsequently underwent a process of lysis. The temporal sequence of events observed in the patient previously described[26] indicates that spasm may lead to platelet aggregation of initiate thrombus formation. In this patient spasm was documented during an anginal attack shortly before the irreversible episode and also at its onset; at post mortem a mural platelet thrombus was observed distal to the site of the occlusive spasm. Reopening of coronary arteries after MI was previously reported in 3 patients following angiographically documented occlusion attributed, without clear proof, to embolus.[58,59] Accordingly the recurrence of episodes of angina in patients in whom the infarction was small, with the same location of transient electrocardiographic changes and with the same characteristics of the episodes preceding the infarction, indicates that the vessel was not permanently occluded and that vasospasm may recur in the same vessel.[26,27] A similar finding was recently reported.[61]

Conversely, in patients who sustained a large infarction, we observed a permanent vascular occlusion with an organized thrombus documented at post mortem in one case.[26]

An unifying hypothesis

The thrombotic hypothesis has been challenged by several pathologists[62,65] on the basis of the inconsistent relationship between thrombotic occlusion and infarction, in particular, small infarctions coming to autopsy were much less frequently associated with occlusive thrombosis than large infarctions coming to autopsy relatively late. Accordingly, data with labelled fibrinogen suggest late or progressive thrombus formation occurs in acute myocardial infarction.[66]

Our observations suggest that a combination of spasm and platelet aggregation and thrombosis rather than coronary spasm by itself,[67] may result in a vicious circle leading to prolonged vascular occlusion. The subsequent balance between positive and negative feedback mechanisms controlling the duration of vasospasm and the dynamic equilibrium between thrombotic and fibrinolytic processes which is likely to be influenced by the size of infarction,[64] may be responsible for the reopening, permanent closure or recanalization of the vessel.

The occlusive role of spasm which usually involves a long segment of the vessel, and therefore may prevent also flow from collaterals, may explain the occurrence of large infarctions distal to thrombotic occlusion occurring in vessels with old stenoses greater than 90% in the presence of well developed collaterals which cannot be explained only by the small further reduction of the lumen caused by thrombus alone.[64]

The hypothesis of spasm as a possible mechanism of infarction is consistent with the frequent finding of plaque haemorrhage and explosion of plaque debris into the lumen sometimes observed at post mortem. These events, associated with blood stagnation caused by spasm, are likely to greatly enhance local thrombosis with thromboxane A_2 liberation from platelets and potentiation of vasospasm. This hypothesis is also consistent with the observation of radio-fibrinogen presence in the thrombus, early after the onset of symptoms.[68]

Following vascular occlusion the final infarct size is probably determined by a complex combination of humoral, nervous and haemodynamic factors and by the basic anatomic alterations of the coronary bed and of the ventricle. The different types of cellular damage observed by Baroldi[69] suggest that differences in metabolic response of the myocardium may play a major role in determining the final size of the infarction. The recent demonstration that fibrinolytic agents directly infused into the occluded coronary artery may reopen the vessel adds conclusive proof to the role of thrombosis in infarction.[70,71] However, the frequent occurrence of reversible ischaemic episodes prior to the onset of irreversible ones suggest that spasm may play the initiating role.[26,27]

SUDDEN DEATH

The epidemiological definition of sudden death is too broad to allow a pathogenetic classification. Thus, we prefer to adopt a clinical definition: death resulting from ventricular fibrillation or asystole. These fatal arrhythmias may occur apparently as primary rhythm disturbances or they may develop during clinically recognizable episodes of acute myocardial ischaemia. Since coronary spasm may be responsible for acute aschaemia, it obviously may be one of the causes of sudden death resulting from ischaemia-induced arrhythmias.

Ventricular fibrillation was often observed during "variant" angina, a reasonable hallmark of vasospastic angina and in our series it was much more frequent than asystole.[15] It showed the tendency to recur in the same patient during the central phase of the ischaemic attack (independent of the presence of pain) or at its end.[15,25]

We have yet no clue as to why potentially fatal arrhythmias during transient ischaemic attacks are frequent in some patients and rare in others.

In our patients arrhythmias could not be prevented by antiarrhythmic therapy with lidocaine, procainamide, disopyramide but were not observed when attacks were prevented by antianginal drugs.

INDICATIONS FOR FUTURE RESEARCH AND THERAPY

These expanding views on ischaemic heart disease, based on a series of objective demonstrations, offer new lines of research and indicate new therapeutic approaches.

Historical perspectives

In the process of trying to outline research lines it is quite instructive to look back and find that most of the conclusions that we are arriving at now are mere rediscoveries. H. Huchard in 1889 noted that attacks of spontaneous angina were less frequent than those for which a provocative cause could be identified, and suggested that spasm superimposed on organic lesions could explain the paroxysmal nature of anginal attacks. He also admitted that angina could occur in the absence of organic stenosis, in particular, in smokers.[72] W. Osler in 1910 suggested the existence of a circulating, perverted internal secretion which favors spasm of the (coronary) arteries. He also described sudden death during angina with normal coronary arteries at autopsy.[73] But the most clear and comprehensive report is that of Gallavardin who concluded that spontaneous angina at rest was much more rare than typical exertional angina, however, he was unable to separate all angina patients into two distinct categories because both syndromes so frequently occurred in the same patient. He believed that a variety of angiospasmodic perturbations, reflexes, and coronary spasmogenic influences superimposed their effects on the coronary stenotic lesions to produce the variety of clinical anginal syndromes he saw.[74]

The "scientific" approach, according to which only "demonstrated" hypotheses can be accepted, lead to the simplistic dismissal of careful observation and deduction, simply because it could not (yet!) be proved. This sequence of events must be kept in mind.

Lines of Research

A major effort should be directed towards the development of methods for the objective detection of myocardial ischaemia since it appears to be much more polimorphic than traditionally thought and since the presence or absence of coronary stenosis cannot be any longer considered at all a diagnostic criterium.

It is also necessary to define the natural history and epidemiology of I.H.D. in the new broader conception where myocardial ischaemia is not only caused by increased demand, can present itself also with varied symptoms or remain asymptomatic.

The awareness of the relevant role of spasm and platelets in the genesis of myocardial ischaemia must stimulate investigations on the predisposing and precipitating mechanisms of coronary spasm as well as the feed-back mechanisms which control its duration and reversibilities.

The observation of anginal pain in patients with myocardiopathy cannot be induced by ergonovine prompt the search for the pathogenetic mechanisms of these clinical syndromes.

Indication for Therapy

Since often ischaemia is not caused by excessive increase of demands, reduction of myocardial oxygen consumption cannot be considered any longer the basic therapeutic approach would require the identification of the pathogenetic mechanisms responsible for the ischaemic attacks. Unfortunately, these mechanisms are probably varied, complex and still largely speculative. Therefore, our treatment remains empirical. The hypothesis of alpha-sympathetic and mediated spasm[75,76,77,78] lead to the reappraisal of plexectomy[79] with some encouraging results.[80] However, medical treatment is usually adequate for management of these patients. Drugs that are known to raise the threshold for smooth muscle stimulation, like nitrates and calcium antagonists, are so far the only medication that, in controlled clinical trials, were consistently effective in reducing drastically the number of ischaemic episodes.[45,81,82,83,84]

In preliminary trials we were unable to obtain comparably consistent responses with the use of atropine,[50] phentolamine,[85] aspirin (140 mg every 72 hours)[86] and prostacyclin.[87] Finally, the common clinical observation that onset and waxing of anginal symptoms often coincide with psychological problems and breakdown in adaptation should not be understressed.[88]

For the evaluation of the response to treatment the frequent spontaneous waning and waxing of symptoms and the quite frequent presence of asymptomatic ischaemic episodes suggests:

1. the design of trials preferably according to a double cross-over scheme[45,81] or with interruption of treatment to check the reappearance of symptoms when off the drug.
2. the continuous monitoring of at least two ECG leads for an objective detection of the ischaemic episodes, preferably lead III and V3.[89]

In our hands, long term treatment with nitrates and calcium antagonists resulted in a low mortality (1.7% per year) and infarction rate (0.8% per year) over a two to four year period in a group of 120 patients who presented with severe angina at rest.[90]

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FATE OF THE CORONARY MICROVASCULATURE IN INFARCTING CANINE MYOCARDIUM: ROLE OF COLLATERAL BLOOD FLOW

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It has been shown in many studies (for review see[1]) that occlusion of a left coronary descending artery in an initial phase produces ultrastructural symptoms of myocardial ischemia that finally pass the phase from reversible to irreversible injury, i.e. infarction. Duration of coronary artery occlusion, actual oxygen consumption of the left ventricle, and absence or presence of collateral perfusion have been shown[2] to be factors modifying the speed of development of infarction, but after 24-48 hrs the final infarct size of 80% of the area at risk will, nevertheless, be present. Collateral blood vessels apparently do not influence very significantly final infarct size, but they greatly modify the process of infarction, as will be described in the following text. All data relate to ultrastructural observations made on cardiac tissue from dog hearts subjected to experimental myocardial infarction by occlusion of a coronary artery.

Electron microscopic investigation of needle biopsies taken from the center of the infarcted area or from a normally perfused part of the left ventricle revealed the following changes in the myocardial microvessels:

1. Microvessels from nonischemic myocardium contained an abnormally large number of neutrophil granulocytes even after a relatively short occlusion time such as 90 min (Figure 1). The fact that the surrounding myocardial cells were of normal appearance lead to the conclusion that these leucocytes were "on their way" to the ischemic zone. It is well known from histologic pathology that leucocytes accumulate between the 6th and 24th hour of infarction in afflicted tissue to form the characteristic demarcation line between infarcted and noninfarcted tissue.

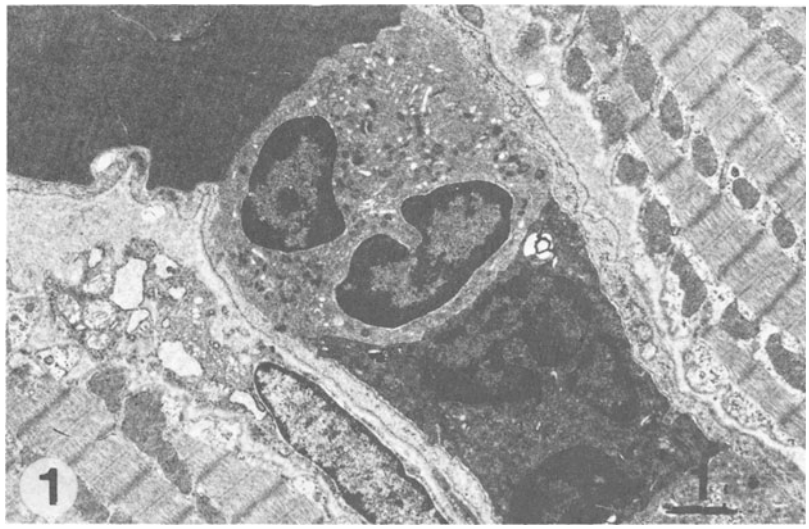

Fig. 1. Nonischemic myocardium. Intact microvessels containing neutrophils and erythrocytes.

2. On the other hand neutrophil granulocytes were already observed in microvessels of ischemic or already infarcted myocardium (Figure 2) as early as 45-90 min after occlusion. They frequently showed close adhesion to endothelial cells, i.e. the early inflammatory phenomenon of "stickiness", was observed. The endothelium in this case may either be intact (Figure 2) or it may show extensive cellular swelling (Figure 3). These granulocytes most probably leave the microvessels because they frequently showed adhesion to irreversibly injured myocardial cells (Figures 4, 5, 6). Accumulation of neutrophils in intact or damaged microvessels of ischemic myocardium and their immigration into the injured tissue were observed at early stages of infarction and without any reperfusion of the tissue by loosening of the arterial ligature. Since by numerous careful measurements the collateral perfusion rate has been shown to vary between 5-20% of preischemic coronary blood flow,[2] it has been assumed that these blood cells entered the ischemic myocardium via this route but not by capillary proliferation. It may be possible that ischemic myocardial cells provoke neutrophilic movements by releasing a potent chemotactic agent, such as histamine or other substances.
3. Some microvessels with either intact or slightly injured endothelium showed an accumulation of erythrocytes, neutrophils and especially of platelets (Figure 7) indicative of formation of microthrombi during ischemia.

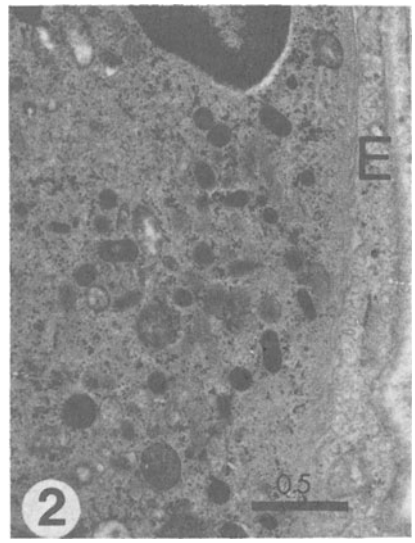

Fig. 2. Part of a neutrophil adheres closely to endothelial cell (E) of microvessel from ischemic zone.

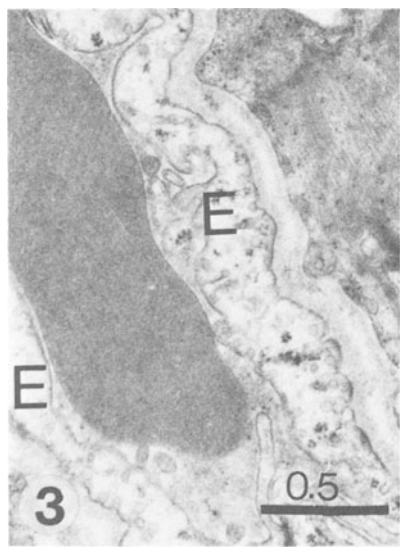

Fig. 3. Capillary endothelial cells (E) from ischemic myocardium.

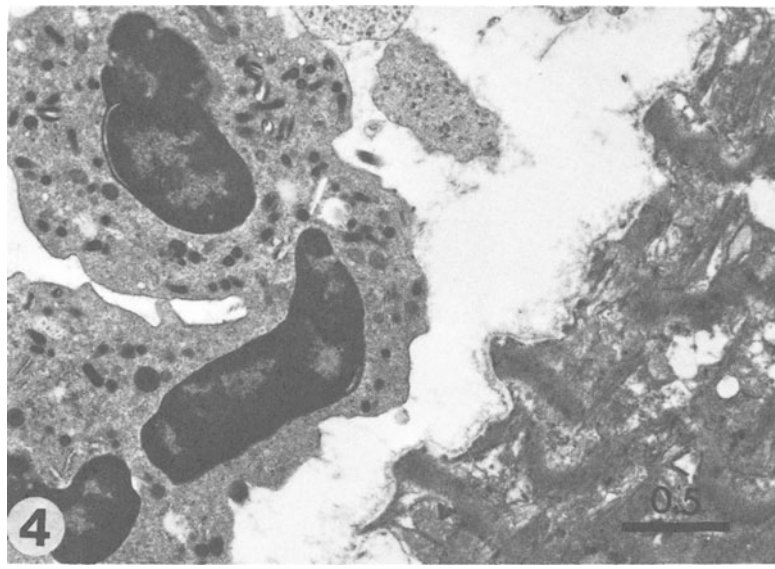

Fig. 4. Neutrophil in the interstitial space next to an irreversibly injured myocardial cell (left).

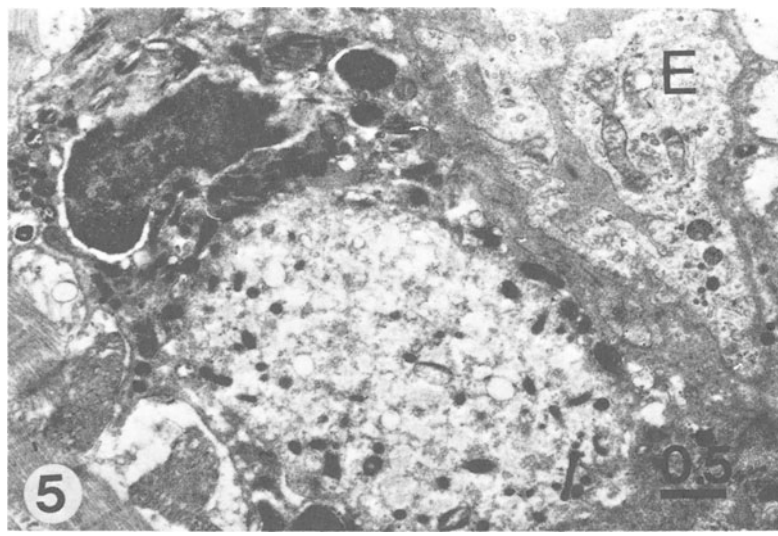

Fig. 5. Extravasation of neutrophils from capillary with swollen endothelium (E).

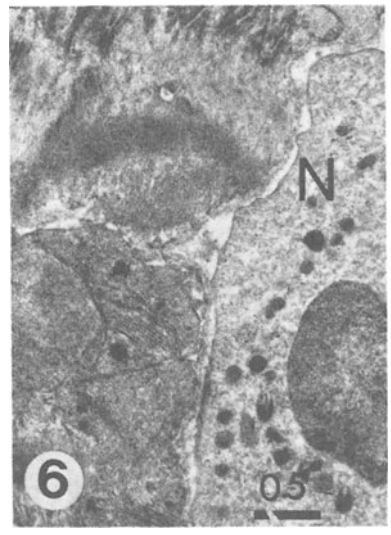

Fig. 6. Neutrophil (N) closely adhering to irreversibly injured myocardial cell.

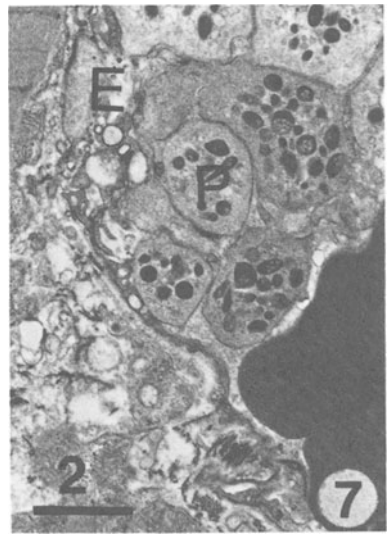

Fig. 7. Intact venule from ischemic zone (E) containing platelets (P) and erythrocytes.

4. Microvessels in ischemic myocardium that did not receive any collateral blood flow contained "old" erythrocytes (ghosts) and cellular debris (Figure 8). Destroyed microvessels were numerous even in hearts with high collateral blood flow, a fact indicative of inhomogeneous distribution of persisting coronary flow and thereby also indicative of an insufficient total blood supply.
5. First symptoms of tissue reparation were observed already during the first 6 hrs of the infarction process. Ultrastructurally, this was evidenced by the occurrence of monocytes besides erythrocytes and neutrophils, within as well as outside of the microvessels, and by the presence of macrophages (originated from blood monocytes), fibroblasts and histiocytes in the extracellular space (Figures 9, 10).

Large parts of the cardiac microvasculature are destructed by ischemic injury leading to infarction of the myocardium. Numerous small vessels, however, that were supplied by collateral blood flow, usually stay intact from a morphological point of view. These contain neutrophil granulocytes, erythrocytes, occasionally platelets, and later on monocytes. These blood cells most probably are contributing to an early unspecific inflammatory reaction that may accelerate the destruction process of irreversibly injured myocardial cells. In later stages of ischemia, these blood vessels facilitate scar formation by providing blood cells and substances for proliferation of interstitial cells.

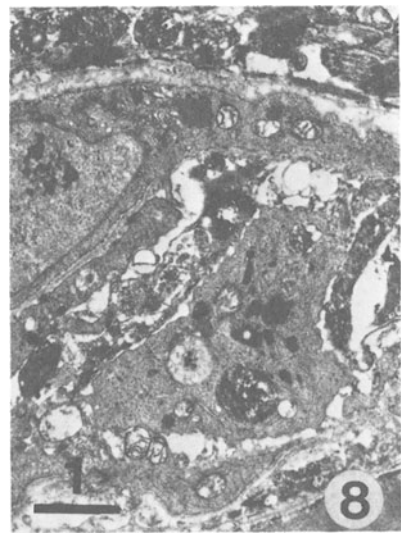

Fig. 8. Completely destroyed microvessel from ischemic zone without collateral flow.

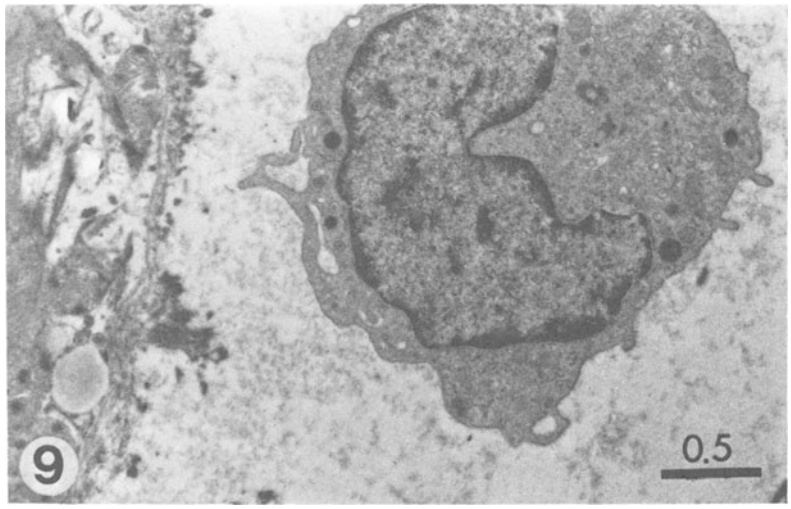

Fig. 9. Monocyte transforming into a macrophagic cell is situated next to irreversibly injured myocardial cell (left).

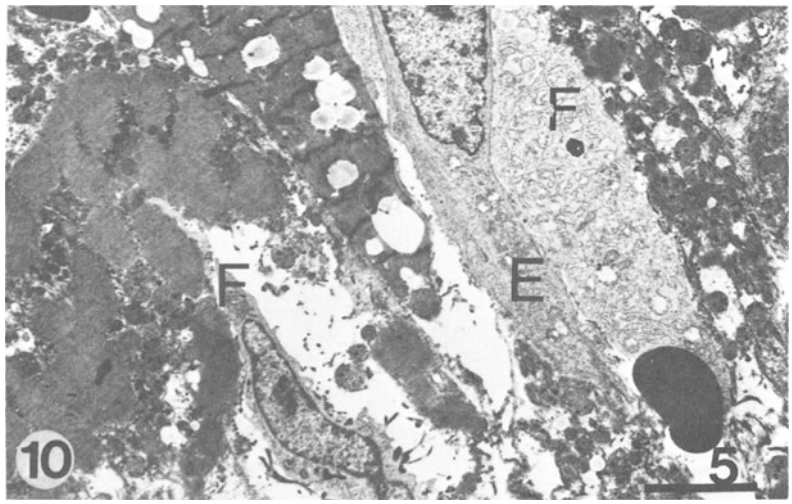

Fig. 10. Destruction of injured myocytes is accompanied by simultaneous proliferation of fibroblasts (F) and new microvessel (E).

In the absence of collateral blood flow, on the other hand, an inflammatory reaction is absent, destruction of myocytes occurs by processes resembling autolysis, and scar formation, an essential

prerequisite for the survival of the heart as an intact entity, is an extremely slow process. The presence or absence of collateral perfusion of ischemic myocardium may therefore be regarded as an important factor contributing to the process of ischemic injury but also to scar formation and therefore to survival of the whole heart.

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EPIDEMIOLOGY OF SUDDEN CARDIAC DEATH

William Ruberman and Eve Weinblatt

Health Insurance Plan
of
Greater New York

Although deaths due to coronary heart disease (CHD) have declined in the United States in the past decade, they still constitute about one third of all deaths. Further, of all CHD deaths, one third occur in persons under age 65, and about 60% occur outside the hospital. Sudden cardiac death remains one of the major problems facing medicine today.

In 1970 Lown and Ruberman[1] suggested that sudden death outside the hospital was in large measure due to ventricular fibrillation. They noted that such deaths occurred in persons with variable amounts of acute ischemic damage and constituted a public health problem requiring a broad range of strategies. But because such deaths for the most part occurred shortly after onset of symptoms - or without any overt symptoms - they concluded that early identification of potential victims offered the best hope for prevention. Drawing from experience in coronary care units,[2] they reasoned that evidence of electrical instability of the myocardium in patients with CHD might identify those at relatively high risk for sudden death.

In 1972 we began a study focused on this issue at the Health Insurance Plan of Greater New York, a large prepaid group-practice plan offering comprehensive medical care. From a population of 120,000 men aged 35-74, those with recent myocardial infarction (MI) or effort angina were offered a special examination which included one hour of single-lead ECG monitoring for ventricular arrhythmia, as well as a standard ECG, clinical and laboratory observations. Over a period of almost four years, 2155 men who met study criteria[3] for MI (1739 men) or angina without prior MI (416 men) were examined and monitored for ventricular premature beats (VPB). Patients were followed for mortality for periods up to 5½ years, with a minimum

observation of 2 years. Mortality status as of the cut-off date of April 1, 1978 was known for all patients. There were 411 deaths from all causes during this period, with 167 meeting study definition of sudden cardiac death - occurring within minutes of the patient's usual state of health in the absence of symptoms or findings suggesting acute MI. Non-sudden cardiac deaths were predominantly due to recurrent infarction, but also included some ascribed to congestive heart failure.

Of the 1739 MI survivors monitored at baseline, 50% had experienced acute MI within the preceding 3 months and 30% within 4 to 8 months, while 9 or more months had elapsed in the remaining 20%. Slightly over half of these patients showed one or more VPB during the baseline monitoring hour, and these cases were in turn divided about evenly between men with simple VPB only and those who had one or more of the qualitative features defined as complex VPB - R on T, runs of two or more, bigeminal or multiform beats. There is a strong association between VPB of high frequency and the presence of these complex features.[4] Of men with simple VPB only, less than one fourth demonstrated 10 or more VPB in the hour, compared with over three fourths of men with complex VPB.

Cumulative probability of death over a 5-year period is shown in Figure 1 for men with and without complex VPB in the baseline monitoring hour. Among the patients with complex VPB, 5-year mortality risk from any cause is almost twice (35% vs 20%) and risk of sudden cardiac death is more than twice that of the men free of such forms (18% vs 8%, Figure 2). For non-sudden cardiac deaths, the corresponding rates are 15% and 7%. Figure 3 examines differentials in cardiac death with respect to the presence of ventricular arrhythmia in greater detail. In the upper half of the figure, we see a very large risk for sudden cardiac death associated with the presence of runs of 2 or more VPB or R on T during the monitoring hour. Five years after baseline, the age-adjusted probability of sudden death in these men was 25%, compared with 13% in men with other forms of complex VPB, 12% in men with simple VPB only, and 6% in men with no VPB in the monitoring hour. In contrast, the curves for non-sudden cardiac death (lower half of Figure 3) show no difference between men with runs or early VPB and those with other complex forms; nor is there any difference associated with the presence of simple VPB in comparison with men free of any ventricular ectopic activity. The age-adjusted relative risk for sudden cardiac death over the 5-year period in the men with runs or R-on-T forms is 4 to 5 times the risk of the men without VPB in the hour. But the corresponding relative risk for non-sudden cardiac death in the men with runs or early beats (or with other complex forms) is only twice that of the men without any ventricular arrhythmia during monitoring.

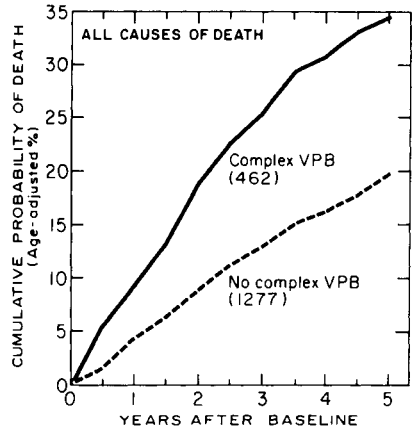

Fig. 1. Mortality over a 5-year period among male survivors of myocardial infarction, in relation to presence of complex ventricular premature beats (VPB) during 1 hour of baseline monitoring.

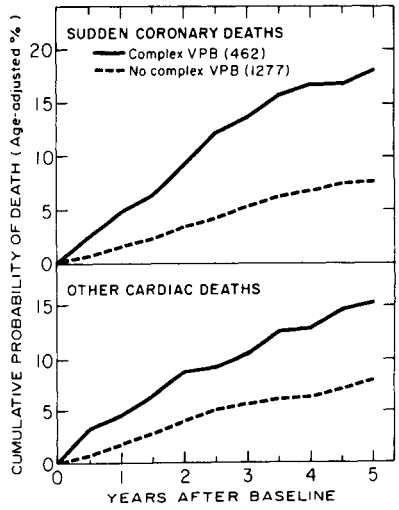

Fig. 2. Sudden and non-sudden cardiac death over 5 years among male MI survivors, in relation to presence of complex ventricular premature beats (VPB) during 1 hour of baseline monitoring.

Cross-classification of our data with respect to three strong prognostic variables in this study - VPB, ST depression, and ex-

perience of congestive heart failure - and performance of age-adjusted survival analyses reveal important mortality differences in relation to how many of the three factors are positive. Table 1 shows that in moving from patients free of ST depression, congestive heart failure and VPB during the monitoring hour to patients with all three of these characteristics present, the 5-year cumulative probability of death from any cause increases almost fivefold, from 11% to 52%. The gradient in risk in relation to the number of positive characteristics suggests that each of these factors contributes independently to mortality risk. Although complex VPB are indeed more likely to be encountered in the presence of other factors associated with severe myocardial damage, multivariate analyses establish that the presence of such beats makes a contribution to poor prognosis that is independent of these other features.[5,6]

In developing our first mortality data for the HIP studies on VPB and sudden death, we examined the influence of social and personal characteristics on prognosis of the 1739 male MI survivors over a period of three years after baseline examination.[7] Estimates were developed, controlled for age, with respect to color, religion, marital status, place of birth, work status, education, occupation, alcohol and coffee consumption, smoking habits, and weight/height ratio. With one important exception - educational attainment - none of these characteristics showed any ability to identify men at

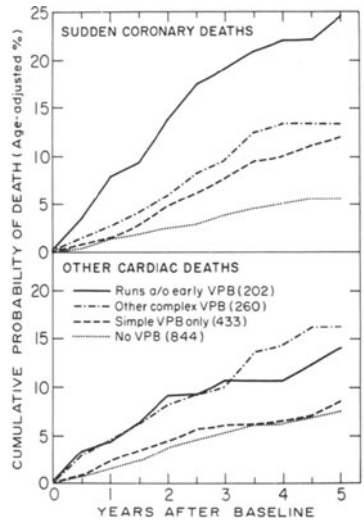

Fig. 3. Sudden and non-sudden cardiac death over 5 years among male MI survivors, in relation to type of ventricular premature beats (VPB) during 1 hour of baseline monitoring.

Table 1. Five-year mortality among male MI survivors in relation to combinations of ST depression (STD), congestive heart failure (CHF), and presence of one or more ventricular premature beats (VPB) in one hour of baseline monitoring

No. of positive factors	STD	CHF	⩾ 1 VPB	No. of patients	5-yr cumulative probability of death (age-adjusted %) All causes	Sudden coronary
None	-	-	-	421	11.1	4.5
One only	+	-	-	178	20.2	6.1
	-	+	-	145	18.6	4.2
	-	-	+	327	18.5	10.1
Two	+	+	-	100	35.6	11.6
	+	-	+	224	30.3	16.5
	-	+	+	177	31.0	16.3
Three	+	+	+	166	51.9	28.4

relatively high, or low, risk of death. But among men who had completed only 8 years of schooling or less, a large disadvantage became apparent in comparison with the better educated men. Men in the low education group who showed complex VPB in the monitoring hour showed a far higher risk of sudden death than comparable men in the high-education group. But in the absence of complex beats, almost identical survival curves were found for the two education categories (see Figure 4). Extending the follow-up period to produce 5-year mortality estimates did not change these relationships. The excess risk for sudden death among low-education men demonstrating complex VPB in the baseline monitoring hour persists in multivariate analyses that control for other factors of importance. We are currently exploring the hypothesis that low education may be a marker for relatively high levels of psychosocial stress that favor conversion of a chronic stable arrhythmia - complex VPB - to lethal ventricular fibrillation.

Identification of ventricular ectopic activity in one hour of ECG monitoring of men with effort angina but no antecedent MI also served to select a group at elevated risk of death over the ensuing 5 years.[8] Among the 416 men with angina in the absence of prior MI, those with VPB in the hour of monitoring showed a 5-year mortality risk of 28% compared with 12% for the men free of VPB. Thus, among men with ischemic heart disease, ventricular ectopic activity detected in one hour of monitoring serves to mark a group at elevated risk of death, especially sudden death. Among patients with prior MI, our findings with respect to sudden death define a gradient of risk, from patients with no VPB in the hour, through those with simple VPB only, to those at higher risk - men with complex VPB. Among the latter, the subgroup with runs and/or early VPB is at

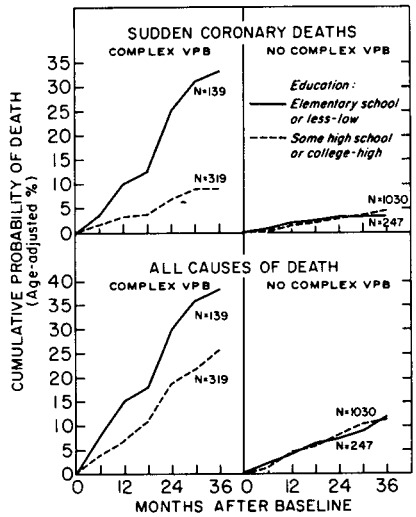

Fig. 4. Mortality over a 3-year period among MI survivors, according to educational level and presence of complex ventricular premature beats (VPB) in the baseline monitoring hour.

extremely high risk for sudden death, and this finding is consistent with the hypothesis that such ectopic forms are uniquely related to ventricular fibrillation. These findings emphasize the importance of continued controlled trials of antiarrhythmic drugs in patients with CHD to determine whether such use reduces the risk of sudden cardiac death. Such an effect has recently been documented in two large, controlled trials of beta-blockers.[9,10] Additional trials of antiarrhythmic drugs in subsets of CHD patients could potentially increase our ability to protect against the threat of sudden death.

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NEURAL FACTORS IN SUDDEN DEATH

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To understand the relationship between the nervous system and ventricular fibrillation, the leading cause of sudden death, is a key factor to comprehend the underlying pathophysiologic mechanisms, to establish rational preventive measures, and to improve the early identification of high risk patients. Considerable progress has been made during the last decade[1,2,3] and some of the main points will be mentioned here, including the problem of adequate animal models for sudden death, the role of psychologic stress, of the cardiac sympathetic nerves and of the vagi, and the potential for autonomic reflexes to be used for the identification of high risk subgroups.

A recent realization is the need for appropriate animal models for sudden cardiac death; this is important both for understanding the different roles of the main factors involved and for a meaningful assessment of antiarrhythmic interventions. Too often drugs are claimed to have a major antiarrhythmic effect on the basis of results obtained in conditions too different from those relevant to the clinical problem. The following is a model specifically designed to explore the role and interaction of a few factors, all clinically relevant, in the genesis of sudden cardiac death, with special care for the autonomic nervous system. Briefly, dogs undergo implantation for chronic measurement of various hemodynamic variables and balloon occluders are placed around the left descending and left circumflex coronary arteries. The dogs are subjected to submaximal exercise on a motor-driven treadmill for 18 minutes. At the 17th minute a balloon occluder is inflated for 2 minutes and acute myocardial ischemia is produced. Thus, this short-lasting ischemia episode affects the last minute of exercise and the first minute after exercise, and this sequence of events allows separation between

arrhythmias dependent on cessation of exercise or on release of occlusion. This protocol begins 3 weeks after initial surgery and is repeated after production of an anterior or inferior myocardial infarction.[4] Grossly, this model resembles what may happen to a patient with a prior myocardial infarction who engages in physical activity and has a brief reduction in coronary flow (spasm?) leading to acute myocardial ischemia, cardiac pain and arrest of exercise. A critical point is represented by the fact that such a brief myocardial ischemia does not induce ventricular arrhythmias at rest; however, when it is coupled with exercise, it results in a high incidence of life-threatening arrhythmias, which are particularly frequent immediately after cessation of exercise. In this new model, the ventricular tachyarrhythmias depend on the interaction between acute myocardial ischemia, level of heart rate, exercise and its cessation, and vagal and sympathetic reflexes. Using this protocol, ventricular tachyarrhythmias occurred in 8 of 15 control dogs (53%) culminating in ventricular fibrillation in 6 (40%). Ten dogs were studied 3 weeks after production of an anterior myocardial infarction and in this group the incidence of ventricular arrhythmias and of ventricular fibrillation was higher (70% and 60%, respectively). It is noteworthy that most instances of ventricular fibrillation occurred immediately after cessation of exercise. The underlying mechanism for this still unexplained specific temporal relationship is under investigation; in any case, it bears a striking similarity with what happens in most sudden deaths in athletes. Do autonomic interventions affect the susceptibility to ventricular fibrillation in this setting? Preliminary data suggest an affirmative answer, because in 10 dogs with an anterior myocardial infarction that were studied after left stellectomy, ventricular arrhythmias occurred in only two cases (20%) and ventricular fibrillation in none, despite the fact that heart rate was even higher compared with that of control animals.

Psychologic stress has been shown to precipitate life-threatening arrhythmias, both experimentally and clinically. Lown, Verrier and their associates have demonstrated that the exposure to a stressful environment is sufficient to induce ventricular tachyarrhythmias in a dog with a recent myocardial infarction.[5] Very recently they have shown that psychologic stress can increase coronary artery resistance and lower the threshold for repetitive extrasystole, an index of vulnerability to fibrillation.[6] These changes are mediated by increases in sympathetic activity and can be prevented by antiadrenergic interventions. The idiopathic long QT syndrome is a clinical entity in which ventricular fibrillation frequently occurs following a psychologic stress and unquestionably represents the most provocative example of neurally mediated non-coronary sudden death. Experimental and clinical evidence supports the concept that it depends on a congenital imbalance in cardiac sympathetic innervation with a dominance of left sided nerves, secondary to a lower-than-normal right cardiac sympathetic activity.[7]

Our data on almost 600 patients with the idiopathic long QT syndrome indicate that mortality in untreated patients is 78% and with beta-adrenergic blockade is reduced to 7%; 41 patients, who continued to have syncopal episodes despite full dose beta-blockers, thus constituting a subgroup at a even higher risk, underwent left stellectomy and only 3 of them (7%) died subsequently. If these patients had not received this surgical treatment the mortality figure for beta-adrenergic blocking agents would undoubtedly be higher. Thus, anti-adrenergic interventions seem to have radically modified the prognosis of these patients.

A new and important concept is that right and left cardiac sympathetic nerves exert a different influence on cardiac arrhythmias and, more specifically, that left sided nerves have a particularly high arrhythmogenic potential.[8] This is partially due to their quantitative dominance at ventricular level. Stimulation of the left stellate ganglion may induce ventricular tachyarrhythmias, even in the absence of acute myocardial ischemia, in dogs and in man. In conjunction with myocardial ischemia activation of the sympathetic nervous system, or of the left sided nerves alone, easily induces ventricular fibrillation.

The high arrhythmogenic potential of left sided cardiac sympathetic nerves, discussed earlier, has unavoidably led to the realization of the antiarrhythmic effect of their removal. We have indeed shown that left stellectomy has several effects which, directly or indirectly, increase the electrical stability of the heart.[9] It reduces the ventricular arrhythmias associated with acute myocardial ischemia and increases the threshold for ventricular fibrillation. It lengthens the ventricular refractory period, thus decreasing cardiac excitability. It halves the incidence of ventricular fibrillation in conscious dogs undergoing an episode of acute myocardial ischemia one month after an anterior myocardial infarction. It reduces to zero (0 out of 10) from 60% (6 out of 10) the incidence of ventricular fibrillation in dogs subjected to a brief episode of acute myocardial ischemia superimposed at the end of an exercise stress test, also in this case one month after an anterior myocardial infarction. Also, left stellectomy by increasing the capability of coronary bed to dilate, both at rest and during exercise, may limit the ischemic area by improving the perfusion of the border zone and contribute to the preservation of ventricular function.

It is noteworthy that the antiarrhythmic effect of left stellectomy, at variance with the most of antiarrhythmic drugs, is not accompanied by a negative inotropic effect. Thus, myocardial contractility is not reduced by left stellectomy neither at rest nor during an exercise stress test. Of particular clinical relevance is the fact that this lack of negative effects on the contractility of the left ventricle is present also in dogs with a prior myo-

cardial infarction performing an exercise stress test. Also in man left stellectomy does not induce the appearance of dyspnea on effort during an exercise stress test at variance with what happens with bilateral stellectomy, which indicates the opportunity of not entirely depriving the heart of adrenergic support.

These data have led to a multicenter trial, ongoing in Northern Italy since 1979, in which a subgroup of patients with an anterior myocardial infarction at high risk for sudden death is randomized between placebo, the beta-blocker oxprenolol and high thoracic left stellectomy.

A growing body of evidence, largely obtained by Lown and Verrier, suggests that enhancement in vagal efferent activity, when is not excessive, has a salutary effect in protecting from sympathetically induced decreases in cardiac electrical stability. This effect partly depends on the maintenance of an optimal heart rate, preventing excessive tachycardia, which preserves underperfused tissue from advancing ischemia and partly depends on an electrophysiologic effect which seems to be mediated by a cholinergic antagonism of adrenergic effects.

The knowledge of both the arrhythmogenic potential of high sympathetic activity and of the protective action of increased vagal activity led us to investigate the possibility that the analysis of autonomic reflexes could identify subgroups at high risk for sudden death.[10] Baroreceptor reflex mediated changes in heart rate can provide a meaningful way to assess autonomic neural control of the heart. Seventeen dogs were chronically instrumented and studied four weeks after an anterior myocardial infarction. The animals were given i.v. injections of phenylephrine and nitroprusside to raise or lower systolic arterial pressure by 30-55 mmHg. The R-R intervals versus the systolic pressure of the preceding beats were plotted and the baroreflex slope was determined. On a subsequent day a 2 min. occlusion of the left circumflex coronary artery was initiated at the beginning of the last minute of an exercise stress test and was continued for 1 min. after the cessation of exercise, as discussed above. The animals were divided into two groups on the basis of their response to this test; 11 animals had ventricular fibrillation (65%, susceptible), whereas 6 dogs (35%) did not and survived (resistant). When the baroreflex slopes of the two groups were compared it was evident that they were strikingly different because the slopes of the resistant dogs were much steeper (11±4.5 vs 4.5±2 msec/mmHg); also the heart rate reduction for a 30 mmHg increase in arterial pressure was significantly different for the resistant (-40±12 b/min) and for the susceptible dogs (-13±5 b/min). This indicates that the resistant animals have a greater capability to react with strong vagal reflexes, which has been shown to reduce vulnerability to ventricular fibrillation. Also, among dogs with a prior myocardial infarction, an almost flat baroreceptive slope

identifies a subgroup at very high risk for ventricular fibrillation. The possibility that these results may be applied to man and that may help in identifying high risk patients after a myocardial infarction is currently under investigation. This study stresses further the tight relationship between the autonomic nervous system and the susceptibility to sudden death.

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MANAGEMENT OF THE PATIENT AT HIGH RISK FOR SUDDEN CARDIAC DEATH

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Can one protect the subject at high risk for sudden cardiac death? To be able to answer this question requires identification of the specific pathologic derangement contributing to the sudden fatality. However, in most victims of sudden cardiac death (SCD) no acute lesions are demonstrable. Autopsy studies document severe occlusive atherosclerotic disease of major epicardial coronary arteries.[1,2] But acute arterial events such as thrombosis of diseased vessels, hemorrhage into a plaque, fracture of atherosclerotic lesion are observed in a minority of cases.[3] Even if coronary occlusion is not demonstrable, this does not preclude the occurrence of myocardial infarction. But even with sensitive stains, a majority of patients dying suddenly exhibit no evidence of myocardial infarction. Reichenback and coworkers[4] noted such findings in only 5% of sudden death victims.

How then is one to identify the subject at risk; and once identified, how is the potential victim to be protected? It is this latter question that we want to concern ourselves with, namely, management of the patient at high risk. The incidence of sudden cardiac death in a population with coronary heart disease is about 1% to 2% annually. It is therefore difficult to determine efficacy of any particular measure except as a multicenter trial involving many thousands of participants. While the use of beta-adrenergic blocking drugs is associated with a substantial reduction of mortality in the immediate period following acute myocardial infarction, treatment with antiarrhythmic drugs, as yet, has not been proven efficacious in such a population.

We have attempted to determine drug effectiveness somewhat differently, namely, by focusing on the patient who is exhibiting life threatening ventricular arrhythmias. Increasing numbers of patients are now presenting who have been resuscitated from sudden death. In fact they present an explicit syndrome (see Table 1). Such resuscitated patients are at high risk for having recurrence.[5] Thus, the mortality was 26% at 1 year and 36% at 2 years. Most of those dying had a second episode of ventricular fibrillation. The high risk for recurrence indicates that only modest numbers of patients are required to ascertain drug effectiveness.

Another group of patients that can be profitably studied involve those who have potentially malignant ventricular arrhythmia. The availability of cardioversion and lidocaine have contributed to the survival of many patients with recurrent episodes of hemodynamically compromising ventricular tachycardia. We have designated as malignant arrhythmia, those patients who have experienced either ventricular fibrillation or have had repeated episodes of sustained ventricular tachycardia with either syncope or hemodynamic decompensation.

The essential hypotheses actuating our work is that sudden cardiac death is due to ventricular fibrillation. The putative background condition is electrical instability of the myocardium. By that, we mean a predisposition of the myocardium to respond to stimuli of threshold intensity with repetitive reentrant arrhythmia. Furthermore, we believe that the presence of certain ventricular premature beats (VPBs) constitute markers for electrical instability of the myocardium. These concepts have been extensively reviewed.[6,7]

VPBs AS RISK FACTORS

Coronary risk factors, either individually or in combination, do not select patients predisposed to sudden cardiac death. Other identifying factors need to be found. It has been our view that the electrical instability of the myocardium long antedates the terminal event which results from an electrical accident. Because the mechanism of sudden death is invariably ventricular fibrillation, it is logical to regard arrhythmias as potential harbingers.[8] We have therefore hypothesized that advanced grades of ventricular premature beats in patients with ischemic heart disease constitute markers of susceptibility to ventricular fibrillation.[9,10] Numerous reports associate the presence of ventricular ectopic activity during longer monitoring intervals with increased occurrence of out-of-hospital sudden cardiac death.[11-17]

The mere presence of VPBs in a single hour of monitoring, however, has limited prognostic significance. The fact that ventricular premature beats are noted in a majority of patients with ischemic

Table 1. Syndrome of Sudden Cardiac Death

Sex	Male (4:1)
CHD	75%
Acute Myocardial Infarction	Unusual
Prodromes	None
Death	Instantaneous
Mechanism	V.F.
VPBs	Present and of advanced grade
Recurrence after resuscitation	30% annually

heart disease argues that if ectopic beats augment the risk for sudden death, this property must be ascribed to some special attributes rather than mere occurrence. While VPBs are prevalent in a population without evident heart disease, complex forms are noted in less than 2% of such patients.

The grading of VPBs is compelled by some of the consideration discussed below and is presented in Table 2.

EARLY CYCLE VPBs

Patients in whom an extrasystole interrupts the T wave, the so-called R-on-T phenomenon, are predisposed to major ventricular arrhythmias. This conclusion finds support in animal work as well as in clinical observations. In early days, when pacemakers were not synchronized, but discharged randomly, a number of reports documented the occurrence of ventricular fibrillation when the pacer pulse accidentally discharged during the vulnerable period occurring at the apex of the T wave. Recently the significance of R-on-T VPBs has been questioned, this is based on several misunderstandings:[18]

1. Ventricular tachycardia and ventricular fibrillation are two distinct electrophysiologic disorders. T-wave interruption is of significance in the genesis of ventricular fibrillation but not in ventricular tachycardia. Campbell and coworkers[19] have demonstrated that R-on-T VPBs triggered ventricular fibrillation in 16 of 17 episodes. Such early ectopics initiated arrhythmia in only 4 of 250 odd patients with ventricular tachycardia.
2. Early cycle VPBs are rarely encountered except in the presence of acute myocardial ischemia or infarction or in patients with severe heart disease, these are the very subjects with increased susceptibility to sudden death. The study by Campbell[19] indeed confirmed that the incidence of early VPBs was highest in the first 3 hours after onset of myocardial infarction. Thereafter, mid and late cycle VPBs were preeminent.

Table 2. VPB Grading System

Grade	Characteristics
0	No ventricular beats
1A	Occasional, isolated VPBs (less than 30 per hour) less than 1 per minute
1B	Occasional, isolated VPBs (less than 30 per hour) more than 1 per minute
2	Frequent VPBs (more than 30 per hour)
3	Multiform VPBs
4A	Repetitive VPBs Couplets
4B	Repetitive VPBs Salvos (ventricular tachycardia)
5	Early VPBs (i.e. abbutting or interrupting the T-wave)

This grading system is applied to a 24-hour monitoring period and indicates the number of hours within that period that a patient has VPBs of a particular grade, which is expressed in the resulting "equation" as a superscript. Subscripts are used to indicate particular aspects of the VPBs of a given grade. In the equation below, for example, the subscript of grade 2 indicates the approximate total number of grade 2 VPBs over the 24-hour period; for grade 3 it denotes the number of different forms observed in any single hour; for grade 4B the two subscripts indicate first the largest number of paroxysms of tachycardia in a single hour and the second denotes the maximum number of successive cycles; for grade 5 the subscript represents the largest number of early ectopic beats in any single hour. A complete translation is given below for this particular equation:

$$0^{3}\ \ 1A^{0}\ \ 1B^{4}\ \ 2^{6}_{760}\ \ 3^{6}_{2}\ \ 4A^{2}_{3}\ \ 4B^{2}_{4-7}\ \ 5^{1}_{3}$$

Grade	
0	Occurred during 3 hours
1A	No infrequent VPBs
1B	Infrequent VPBs but greater than 1 per minute observed during 4 hours
2	Occurred during 6 hours (with a total of 760 VPBs)
3	Occurred during 6 hours and exhibited two forms
4A	Occurred during 2 hours and their greatest frequency in any 1 hour was 3
4B	Occurred during 2 hours, there were 4 paroxysms, the longest duration was 7 cycles
5	An early VPB was observed 3 times during a single hour in the 24-hour monitoring session

3. The prolonged QT interval has been recognized over many years as predisposing to sudden death, especially in patients who have frequent VPBs. This combination increases the likelihood of an ectopic beat discharging during the vulnerable period of the T wave and thereby triggering ventricular fibrillation.

4. When a late cycle ectopic beat initiates a paroxysm of tachyarrhythmia there is no certainty that the succeeding early cycle VPB was not the culprit. Several decades ago, Smirk[20,21] called attention to this phenomenon and designated it V-V' in which V is the late extrasystole and V' is the early one which interrupts the T wave of the preceding ectopic cycle and launches the tachyarrhythmia.

REPETITIVE VENTRICULAR PREMATURE BEATS

Animal studies have shown that discharging a stimulus during the vulnerable period does not provoke ventricular fibrillation unless a large electric current is delivered. Since extrasystoles carry but miniscule electric charges, how is ventricular fibrillation provoked? Significant is the fact that when extrasystoles are repetitive, the threshold of the vulnerable period is progressively reduced. Thus, in the ischemic heart a sequence of 3 early cycle extrasystoles lower the vulnerable period threshold to the point wherein fibrillation can be induced by current just sufficient for mid-diastolic depolarization.[22] Clinicians have long been aware of this physiologic relationship that salvos of ventricular ectopic beats, especially if they occur in accelerating sequences, are harbingers of ventricular fibrillation.

It has been our view that these categories of VPBs, namely, early ectopics interrupting T waves and those that fire in salvos, constitute the essential attributes which impart risk for ventricular fibrillation. These advanced grades of VPBs therefore help in identifying those predisposed to sudden cardiac death.

SEVERITY OF HEART DISEASE WITH VPBs IN RELATION TO THE RISK OF SUDDEN DEATH

It may be that the VPB is not an indicator of risk for SCD, but merely an expression of the type and severity of the underlying heart disease which is the decisive variable. A number of studies have shown an association between the severity of heart disease and the prevalence as well as grade of ventricular ectopic beats. Calvert et al.,[23] reported that the presence of advanced grades of VPBs correlated with the extent of coronary artery involvement, with elevation of left ventricular end-diastolic pressure and with the occurrence of assynergy. Schulze and coworkers,[24] who studied

patients with myocardial infarction in the late convalescence phase, found that advanced grades of VPBs occurred predominantly among those having an ejection fraction of less than 40%. However, in a later follow-up, sudden death afflicted only those who had advanced grades of ectopic activity and spared those who had a low ejection fraction, but were free of such complex VPBs. Ruberman and coworkers[25] noted that men with congestive heart failure who did not exhibit advanced grades of VPBs during a one hour period of monitoring showed a lesser risk of sudden cardiac death compared to men who had exhibited congestive heart failure. The poor prognosis among those with advanced grades of VPBs was largely due to sudden cardiac death. Graboys and coworkers[26] studied a small exceptional group of patients who had no heart disease but experienced ventricular fibrillation. These patients showed the same constellation of VPB grade distribution as those with coronary heart disease who had experienced sudden death. The pattern of advanced grade was quite dissimilar among patients merely having ischemic heart disease. The VPB of advanced grade thus emerges as a predictor of mortality independent of a host of hemodynamic variables.

WHO NEEDS TO BE TREATED?

At the present time predisposition to sudden death is indicated in the following subsets of patients: (1) among those resuscitated from ventricular fibrillation in the absence of myocardial infarction; (2) when potentially malignant ventricular tachycardia recurs frequently; (3) when ventricular tachycardia and multiple ventricular responses are precipitated by intracardiac electrophysiologic techniques; (4) when certain advanced grades of VPBs are exposed by monitoring and exercise testing in patients with ischemic heart disease (see Table 3).

The vast majority of patients with VPBs require no therapy other than reassurance about the benignity and ubiquity of their disorder. Therapy is prescribed for a minority in whom arrhythmias prove symptomatically disabling. It needs to be emphasized that patients are generally unaware of irregular heart beats. This is true even when ectopic activity is abundant and renders the underlying rhythm seemingly chaotic.

HOW TO TREAT

The physician confronts a number of clinical questions. How does one select a drug that is both effective and safe? How is an appropriate antiarrhythmic program to be instituted with economy of time and effort? How is one to know that a drug selected will be effective long range? How can one be certain that a particular dosing schedule will not result in chronic toxicity? Neither

Table 3. Indications for Treatment of VPBs

1.	Primary ventricular fibrillation in the absence of acute myocardial infarction.
2.	Postmyocardial infarction < 6 months and grade 4 or 5 VPBs.
3.	New onset of angina pectoris and grade 4 or 5 VPBs.
4.	In patients with coronary heart disease accelerating salvos of ventricular tachycardia on 24-hour monitoring or exercise stress testing.
5.	Peak exercise > 1 mm ST ↓ with grade 4 or 5 VPBs.
6.	Advanced grades of VPBs during angina pectoris.
7.	Prolonged QT syndrome with syncope and VPBs.
8.	Flail mitral leaflet with symptomatic paroxysms of ventricular tachycardia.
9.	Symptomatic arrhythmia.

electrophysiologic classification of drugs into categories based on attributes determined in isolated Purkinje fibers nor pharmokinetic concepts relating to the so-called therapeutic blood level have helped the clinician in selecting a drug for the individual patient. In effect present day therapy is largely pragmatic.

If pragmatism is to be the order of the day, there is need to admit this honestly and to systematize the approach. To accomplish a systematic approach, we have developed a strategy consisting of 4 phases: Phase 0 involves data acquisition, Phase 1 is based on acute drug testing, Phase 2 consists short-term drug maintenance and Phase 3 includes long-term chronic therapy.

PHASE 0 - DATA ACQUISITION

The objective of this Phase is to define the patient's level of spontaneous dysrhythmia. This constitutes a control period against which drug action is compared. When the patient enters the hospital, all cardioactive drugs including beta-adrenergic blocking agents, unless being employed for unstable angina, are discontinued. This has been mandated by our finding that life-threatening arrhythmias may result from the very agents used for their suppression.[27] We have not encountered rebound accentuation of an arrhythmia by abruptly stopping any drug. Digitalis drugs are maintained only if overt cardiac decompensation is present. During the drug-free period of 48 hours, continuous Holter monitoring and maximal symptom-limited exercise testing is carried out. Two 24 hour monitoring sessions provide adequate data relating to VPB variability and to the reproducibility of advanced grades. An exercise test is maximal, fatigue-limited and is accomplished on a motorized treadmill accord-

ing to a Bruce protocol. Phase 0, in addition to providing baseline data relating to the frequency and grade of VPBs, also permits acquisition of insight as to psychologic and social factors which may have contributed to the genesis of arrhythmia.

PHASE 1 - ACUTE DRUG TESTING

Crucial to the effectiveness of many antiarrhythmic drugs is the establishment of a significant so-called therapeutic blood concentration.[31] This can be achieved by the oral administration of a single large dose. The purpose of acute drug testing is to induce a therapeutic blood level during a brief period to observe the course of drug action and the extent of VPB suppression, and to determine promptly whether any toxic complications ensue. Since only a single dose is used, side effects, if any, are short-lived and risk to the patient is brief (Table 4). Moreover, highly trained personnel are in attendance at all times and are well equipped to deal with threatening arrhythmias. By a series of such tests with different drugs during phase 1 studies, one establishes in a relatively short period of time which drugs are most effective. The therapeutic objective is to eliminate grade 4 and 5 VPBs. When a patient has been experiencing malignant ventricular arrhythmias, the aim of phase 1 studies is to test efficacy of several antiarrhythmic drugs. If untoward effects emerge during long range maintenance therapy, another effective agent can then be substituted immediately without requiring costly hospitalization for retesting and without exposing the patient to the hazard of interrupted therapy.

Acute drug testing involves four essential elements:

1. Administration of a single large oral dose of a selected antiarrhythmic drug.
2. The use of programmed trendscription to display the time course of drug action.
3. Exercise on a bicycle ergometer to help define drug action and reproducibility.
4. Sampling of blood for drug concentration to permit correlation with the onset and dissipation of antiarrhythmic or toxic effects.[7]

Testing for Digitalis Effect

During this phase, the effect of digitalis drugs on VPB frequency and grade is determined as well. This is accomplished by using acetyl strophanthidin, an ultra rapid acting digitalis-like drug. We have found that acetyl strophanthidin will ameliorate ventricular arrhythmia in nearly half the patients who have ectopic activity.[28] If the patient's response is favorable, digitalization

Table 4. Antiarrhythmic Drugs and Dosages during Phase 1 and Phase 2 Studies

Drug	Phase 1 Acute Drug Test (Single oral dose, mg)	Phase 2 Short-term maintenance (mg/day)
Quinidine	600	1200-1600
Procainamide	1500	3000
Disopyramide	300	400-600
Propranolol	80	120-160
Metoprolol	100	100-200
Mexiletine	400	600-1200
Tocainide	---	1200-2400
Aprindine	200-300 (i.v.)	100-200
Pindolol	20	40-80

with digoxin is carried out. In 142 patients with VPBs who underwent testing with acetyl strophanthidin, the frequency and grade were diminished in 65 or 46%. In this group, VPBs were reduced by 82% and in nearly half, all ectopic activity was abolished. The antiarrhythmic action of acetyl strophanthidin did not appear to depend on its positive inotropic effect. In 30 patients without demonstrable heart disease, but with frequent VPBs, acetyl strophanthidin reduced or eliminated arrhythmia in 60%. It has been suggested that the action of digitalis glycosides to diminish or abolish VPBs is due to an indirect reduction in Purkinje fiber automaticity resulting from augmented vagus nerve tone. Such vagal enhancement induces a lessening in adrenergic effects on the myocardium.

Electrophysiologic Studies

In about 20% of patients who have experienced malignant arrhythmias, ventricular ectopic activity is either scanty or non-reproducible. In such patients invasive techniques are required to expose the presence of myocardial electrical instability and assess its response to antiarrhythmic drugs. These techniques utilize programmed electrical stimulation (see Podrid & Lown paper presented at this Congress).

Two approaches have been pursued: one involves the provocation of ventricular tachycardia as an endpoint and was developed independently by Durrer et al.,[29] and Coumel et al.[30] The second approach is based on the induction of multiple ectopic beats following single, dual and triple pulses, and have been termed the multiple repetitive response or (MRVR) or nonsustained ventricular tachycardia.

When sustained ventricular tachycardia is the endpoint employed, complications are frequent. Ventricular fibrillation is provoked in 10-15% of such studies. Not uncommonly the ventricular tachycardia does not respond to burst pacing and the arrhythmia results in hemodynamic impairment, one has to resort then to cardioversion. Our approach using nonsustained VT or MRVR as a marker of electrical instability has significantly reduced the risk of provoking sustained ventricular tachycardia or ventricular fibrillation.

PHASE 2 STUDIES

Short Term Drug Maintenance

After completion of phase 1 testing during which one or more drugs have been shown to suppress advanced grades of arrhythmia, the patient enters phase 2. In the minority of patients who require electrophysiologic studies, the 2 phases are amalgamated. The aim of this phase is to determine efficacy of antiarrhythmic drugs with a dosing program simulating chronic drug administration. A further aim is to assess patient tolerance of the antiarrhythmic agent with chronic therapy. Antiarrhythmic drug efficacy is evaluated by means of 24 hour Holter monitoring and maximal exercise testing.

In monitoring and exercise the criteria for therapeutic response involve:

1. Total abolition of grade 4B and 5 VPBs
2. Reduction of more than 90% in grade 4A VPBs
3. Greater than 50% reduction in the number of VPBs per 24 hours and/or a reduction of more than 50% in the number of hours during which grade 2 VPBs occurred compared to monitoring during phase 0 and during exercise a 50% reduction compared to the control preexercise stage.

If the underlying arrhythmia has been ventricular fibrillation, more than one drug is employed. The use of drug combinations is the rule - "a fail-safe" system of drug protection is mandated by the high risk of recurrence.[32] Unlike other disorders wherein the physician may have opportunity to remedy a therapeutic error, this is rarely the case with ventricular fibrillation. In addition to the use of drugs, proper management of the patient requires attention to the psychologic problems that abound in nearly every patient afflicted with malignant ventricular arrhythmias. The use of relaxation technique as well as other psychologic support has proven invaluable in contributing to amelioration of the refractory rhythm disorder.

PHASE 3 - CHRONIC THERAPY

This is post hospital follow-up phase and defines long range drug efficacy. Furthermore, information is provided concerning the prevalence of complications with chronic use. In the majority of patients tested as indicated above, adverse reaction can generally be overcome by minor adjustments in daily dose. The type of screening we have outlined here prevents serious toxicity from surprising the physician and upsetting the sensitive apple cart of therapeutic management. At 3 to 6 month intervals, Holter monitoring and exercise stress testing is repeated to determine whether the arrhythmia remains under adequate control.

Drug-induced Arrhythmic Complications

The methods of management outlined here, especially phase 1 and phase 2 studies, alert the physician to drug-induced complications before serious injury has been incurred. We have encountered signigicant aggravation of arrhythmia with nearly all the drugs in current use.[27] A systematic examination of this problem has been conducted in 155 patients who were subjected to 722 drug studies. In 53 patients having 80 drug tests, aggravation of arrhythmia was observed in 11%. The criteria used to judge aggravation of the arrhythmic disorder included a four fold increase in VPB frequency, a ten fold increase in grade 4 and the first occurrence of ventricular tachycardia. In all cases blood drug levels were within the established therapeutic range. Thus, antiarrhythmic drugs in a substantial number of patients aggravate ventricular arrhythmias. Phase 1 and Phase 2 studies permit early detection of such susceptibility.

Long Range Therapeutic Results

If advanced grades of VPBs are indicators of electrical instability, their suppression should afford protection against sudden cardiac death. Our studies now encompass nearly 300 patients having malignant ventricular arrhythmias. When advanced grades of VPBs were controlled, the annual occurrence of sudden death was less than 3%. In 20% of patients in whom such an objective could not be achieved, sudden death exceeded 40% during the follow-up period.[32] Even when patients were matched for extent of hemodynamic deficit, survival was far better for those in whom evidence of myocardial electrical instability had been eliminated.

CONCLUSIONS

The present studies extending for more than 10 years indicate that the VPB hypothesis has been validated and is of practical

significance. Advanced grades of ventricular ectopic beats, namely those of grade 4B and 5 are the targets of therapy. In 20% of patients recourse to invasive electrophysiologic techniques are required. There is need for multiple drug screening. Therapy must be individualized. A systematic approach involving acute drug testing as well as phase 2 studies facilitate identification of these drugs which are effective as well as those which are potentially hazardous. Ventricular fibrillation can be prevented by currently available antiarrhythmic drugs.

The problem now on the frontier of research is to identify more precisely the patient at risk for sudden death so as to justify interventions with antiarrhythmic agents.

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STROKE AND CARDIAC DISORDERS

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Combined lesions of the heart and the brain with circulatory disorders in them are observed very often due to their common etiology (atherosclerosis, hypotensive disease, other arterial hypertensions) and frequent interdependence. The relations between circulatory disorders in the both organs may be various.

Cardiocerebral pathology in which heart lesions produce disorders of cerebral circulation frequently with the development of infarct in the brain occurs most often and is studied better than other pathology. Copious literature deals with infarcts. I'd like to mention two Symposia "Infarcts of Heart and Brain" held in Cologne. Prof. Zülch who is present here was the chairman of these Symposia.

Sharp fluctuations of coagulative and rheological properties of the blood in the presence of changed vessels of the heart and the brain can lead to simultaneous circulatory disorders in them with the development of infarcts.

Another possibility when brain stroke produces disorders of the coronary circulation and metabolism in the myocardium is studied to a less extent. It is these cerebrocardial changes that are the subject of the present communication.

The possibility of cerebrogenic disorders of the coronary circulation and cardiac activity leaves no doubts although the question about the possibility of cerebrogenic infarct remains to be discussible.

Numerous experiments and observations of neurosurgeons demonstrated that actions on some areas of the brain cause disorders of the coronary circulation and cardiac activity. The limbico-hypothalamo-reticular complex and the hypophysial-adrenal system related to it are of primary importance.

Clinical manifestations of the cerebrocardiac syndrome in an acute stage of stroke are usually little pronounced and do not differ greatly depending on the character of stroke. They show up mainly as rhythm disturbances (tachycardia, transient arrhythmias). Pains in the region of the heart as a rule do not occur. Pronounced manifestations of cardiac or cardiovascular insufficiency are observed only when stroke is associated with myocardial infarction, pneumonia or when the terminal state develops.

Much has been already written about very common ECG changes in stroke but they still attract attention of investigators.

According to our data, 76.5% of patients in an acute stage of stroke exhibited in addition to ECG changes which are usual for ischemic heart disease and arterial hypertension other changes in ECG of the cerebrogenic character.

The most characteristic changes of these are sinus tachycardia, an increase of P wave, an elongation of Q-T interval, a displacement of S-T segment downward of the isoelectrical line, an inversion of T wave, presence or absence of U wave.

The most dynamic cerebrogenic changes in the ECG were disorders of cardiac rhythm and T wave which could rapidly change their form, disappear for some time and appear again, sometimes within several hours. On improvement of the patient's condition in most cases first of all heart rate became normal, arrhythmias disappeared, then displacements of S-T segment disappeared. Changes in T and U waves and an elongation of Q-T interval could be recorded for a longer period.

Individual cerebrogenic changes in the ECG as a rule are found in various combinations that enabled us to identify three variants of them depending on the degree of severity.

Variant I is characterized by cardiac rhythm disturbances, a moderate elongation (by 10-20%) of Q-T interval. Insignificant changes of T wave, presence of U wave can take place.

Variant II is characterized by a displacement below the isoelectrical line of S-T segment, an elongation of Q-T interval, changes of T wave up to inversion. There can be disturbances of cardiac rhythm and present of U wave.

Variant III observed in severe strokes is characterized by an increase of T wave in the right or right and medial chest leads, often in its combination with an inversion in the left chest leads; T wave is not infrequently wide, U wave is often increased.

The cerebrogenic character of the above described changes in the ECG is confirmed by their dynamics related to the course of the process in the brain. They occur in the first days following stroke, sometimes later - during deterioration of the patient's condition, in the terminal state they become particularly pronounced. In a favourable course of stroke they disappear already within the first week but more often they persist for several weeks.

Also, we could often observe cases when the ECG recorded not long before stroke was normal and following stroke it changed according to the type described above.

Cerebrogenic changes in the ECG were seen in hemorrhagic strokes more frequently than in ischemic strokes (in 96.5% and 68.5%, respectively) and were more pronounced- variants II and III were encountered twice more often than in brain infarct.

In an acute stage of brain stroke the ECG changes are very similar to those in subendocardial myocardial infarction.

The ECG changes in stroke reflect disorders of the coronary circulation and metabolism of the cardiac muscle resulting from direct, neurogenic, and indirect, humoral influences which we shall dwell on later.

Recently Dimant and Grob (1977) and then Norris et al., having carried out very thorough investigations found an increased content of cardiac enzymes in the blood of stroke patients, in particular - a significant elevation of the level of cardiac fraction of creatine phosphokinase (CPK). Creatine phosphokinase was found already in the initial phase of stroke and its level gradually increased. Thus lesion of the myocardium is caused by stroke. We also noted a considerable rise of CPK activity in an acute stage of stroke. These studies show that organic changes in the heart in brain stroke occur much more frequently than it has been considered until recently.

Post-stroke disorders of systemic hemodynamics show up as changes in the cardiac output value and a reduction of the myocardial contractile function.

In most cases marked changes were found in hearts of patients who had died from stroke. Light and electron miscroscopy showed scattered dystrophic changes of myocytes in the form of eosinophylia of plasma, disappearance of cross striation, fragmentation, vacuo-

lization and globular disintegration as well as disseminated foci of myolysis and coagulative necrosis. Uneven and focal character of damage of myocytes is combined with dystonia of vessels of the microcirculatory bed. In some areas capillaries were paralytically dilated with stases from erythrocytes and "coin columns"; in other areas - thickened capillaries. There were plasmorrhagias, hemorrhages, infiltrates from lymphoid elements and monocytes around some vessels. Microvascular aneurysms observed so often in the brain in arterial hypertension were not found.

Such changes which can be considered as "cerebrogenic" were noted by us in 20.5% of stroke patients who had no pronounced atherosclerosis of the coronary arteries.

Recent myocardial infarctions in an acute period of stroke occur usually on the background of already existing atherosclerotic changes of the cardiac arteries. In such situations it is difficult to establish cause and effect relations. However it is possible to determine according to morphological signs which focus (in the brain or in the heart) developed earlier.

The possibility of the development of extensive myocardial infarction following stroke in the absence of pronounced atherosclerosis of the coronary arteries is in doubt and continues to be discussed.

We recognize such possibility. I shall present two observations to illustrate this. The both patients were elderly (aged 60 and 58) and suffered from hypertensive disease. They both developed extensive hemorrhage into deep areas of the hemispheres, the blood gradually went inside and eventually rushed into the ventricles. In the first days the ECG showed only slight changes indicating slowing of intraventricular conduction. Sudden death occurred some days after stroke. Recent infarctions were found in the heart. The coronary arteries proved to be unchanged: their ostia were wide, there were no plaques in the intima along the course of the arteries.

One can suppose that in these patients myocardial infarction resulted from spasm of the coronary arteries (like in subarachnoid hemorrhage extensive infarcts develop in the brain due to spasm of the arteries supplying the brain). But such cases are encountered rarely. Among our 600 autopsy cases of stroke myocardial infarction which can be considered as cerebrogenic occurred only in 3% of them. Although in these cases (apart from the observations mentioned above) atherosclerotic changes were found in the cardiac coronary arteries, they did not cause stenosis of their lumina.

In all cases of cerebrogenic myocardial infarctions there were either extensive brain infarcts damaging the hypothalamus or the brain stem directly or as a result of edema and dislocation, or

deep hemorrhages. Cardiac infarction developed simultaneously or several days after stroke if the volume of the focus in the brain gradually increased.

Pathogenetic mechanisms responsible for disorders of the coronary circulation and myocardial metabolism in stroke are very complicated and have not yet completely been understood. By the present the following conception is the most common.

Damage of the brain, mainly of the limbico-hypothalamo-reticular region, leads to cardiac disorders in two ways: the direct one through the cardiac nerves, and the indirect one - through changes in the adrenal glands' functions.

The direct neurogenic action produces both neurotrophic and vasomotor changes.

Recently V. Golubykh (1981) showed in experiments on dogs that destruction of the hypothalamus in the region of the lateral mamillary nucleus caused sympathetic constrictory reactions of the coronary vessels. Earlier Melvill et al. by stimulating the hypothalamus in cats produced lesions of the myocardium including the development of infarction; interruption of sympathetic tracts in the spinal cord eliminated this effect.

Stress in the form of stroke leads to hyperfunction of the sympathetico-adrenal system with an increase of serum level of catecholamines whose "cardionecrotic effect" is well known. Excretion of catecholamines by nervous elements located in the heart increases. Greenhoot and Reichenback (1969) demonstrated that the most pronounced changes in the heart are found just near the endings of the adrenergic nerve fibers excreting noradrenalin into the extracellular space. An increased corticosteroid content in the blood leads to a loss of intracellular potassium and thereby enhances the damaging action of catecholamines. Catecholamines also increase platelet aggregation that worsens microcirculation and contributes to formation of ischemic foci of necrosis.

Products of lipid peroxidation also have the damaging action on the myocardium. In an acute period of stroke we found a sharp rise of the content of the final product of peroxidation - malondialdehyde.

The data on disturbances of electrolyte balance in stroke are extremely contrary and it is yet difficult to determine what concrete role these shifts play in disorders of cerebral circulation.

During recent decade the attention of investigators was focused on such highly active substances as prostaglandins and cyclic nucleotides. Their role in disorders of cerebral circulation began to be studied as well (Welch, Fukuda et al.).

The influence of prostaglandins (PG) on the frequency and rhythm of cardiac contractions, value of stroke volume, permeability of myocardial cell membranes, contractility of the cardiac muscle and on the coronary blood flow.

An important role in the mechanism of cerebrocardial interaction belongs also to the system of cyclic nucleotides responsible for realization of intracellular effects of catecholamines, serotonin, prostaglandins and other neurohumoral substances.

Our workers studied the contents of some prostaglandins and cyclic nucleotides in the blood and cerebrospinal fluid of patients with stroke. It turned out that concentrations both of prostaglandins and cyclic nucleotides elevated in the blood and CSF particularly in the first days of stroke, irrespective of its character. In the peripheral blood an increase of PG of E series was greater than that of $F_2\alpha$. A considerable elevation of PG level as well as some increase of cGMP took place in strokes with damage of the brain stem. In these patients the PGE level was three times increased and the level of PG $F_2\alpha$ was almost twice increased. These changes were much less pronounced in lesions of cerebral hemispheres. Only in two very severe cases of stem stroke causing death of the patients a predomination of PG $F_2\alpha$ was noted on the background of low general content of PG.

Concentration of both cyclic nucleotides (cAMP and cGMP) in the first days of stroke also increased, the cAMP concentration being greater so that their ratio noticeably changed. Subsequently dysbalance between individual fractions of PG and cyclic nucleotides reduced.

It should be noted that the content of cyclic nucleotides (especially cAMP) in the venous blood flowing off the brain was significantly greater than in the arterial blood that confirms their cerebral genesis.

The available data on the influence of prostaglandins and cyclic nucleotides on the cardiac activity and circulation make it possible to conclude that the change of their quantitative composition in the blood and cerebrospinal fluid of stroke patients exerts a certain influence on formation of cerebrocardial disorders. Dysbalance in ratio of pressor and depressor prostaglandins and their "mediators" - cGMP and cAMP, which in particularly severe cases of stroke manifests itself in predomination of vasoconstricting prostaglandins can lead to myocardial ischemia.

Activation of the prostaglandin and cyclic nucleotide systems in stroke patients can probably be of the compensatory and adaptative character; however in some cases as a result of hypercom-

pensation shifts in these systems can turn into a pathologic factor exerting an unfavourable effect on myocardial blood supply and metabolism.

Summing up, it should be stressed first of all that cerebrogenic pathology of the cardiac activity and the coronary circulation in brain stroke is observed often and can be very serious up to the development of myocardial infarction. It aggravates the course of stroke and sometimes determines its outcome. Its presence should be taken into consideration by a therapeutist and a neurologist during elaboration of the programme of treatment.

The intimate mechanisms responsible for the development of this pathology are not quite understood and their further investigation is of great importance. The new methods of research - study of myocardial blood supply by using isotopes, ultrasound, echography, computed tomography, etc. will permit to evaluate more objectively and precisely cerebrogenic pathology of the heart and this can exert a significant influence on the character of therapeutic measures.

THE PATHOGENESIS OF VASCULAR DISORDERS OF BRAIN AND HEART - ARE THERE SIMILARITIES OR DISSIMILARITIES?

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INTRODUCTION

This review is based on the discussion of two international symposia in Köln/Cologne 1976 and 1978 and our own experience. It will be understood that only a few problems of the discussion can be pointed out, particularly since the proceedings of both symposia are extensively published with Springer, Heidelberg-New York 1977/1979,[1,2] and summarized later.[3]

I. Terminology

First some semantic misunderstandings in the nomenclature have to be clarified: the angloamerican term "atherosclerosis" corresponds to "arterioclerosis" in German. "Arteriosclerosis" in English apparently sums up atherosclerosis and "small vessel disease" which corresponds in German pathology to "arteriolosclerosis" or "hyalinosis". The latter is subsequent to long-standing hypertension and responsible for mass hemorrhage in the brain and granular nephrosclerosis in the kidney.

II. Risk Factors

It seems of importance to distinguish between two sets of different risk factors, namely 1) for the causation of stenosing atherosclerosis and 2) for the acute promotion of the clinical events of stroke and myocardial infarction. Although most risk factors involve the whole arterial system, there are differences in the grade of arteriosclerotic manifestation in the different

body organs, either on the base of constitutional or environmental factors, disregarding the fact that normally cerebral atherosclerosis follows the coronary disease with a delay of one decade. One coincidental observation could be that risk factors for atherosclerosis have a different rank order in brain and heart.

For instance, in brain infarcts hypertension is accused particularly on epidemiological grounds to be the risk factor number one, although certain morphological observations do not always support such a concept.

In myocardial infarction the most important risk factor is hypercholesterolemia followed by cigarette smoking, while hypertension is only on third place. Interesting to remember that in peripheral vascular arterial disease the most important risk factor is undoubtedly cigarette smoking.

On the other hand, the cholesterol level is only weakly related to atherothrombotic brain infarct (ABI) as is also cigarette smoking.

Diabetes is an important factor in both disorders.

For the clinical manifestation the risk factor hypertension is certainly of prime importance for both types of stroke, namely (a) cerebral mass hemorrhage (via hyalinosis in the striatal arteries etc.) and (b) cerebral infarct.

Meanwhile it is undoubtedly proven that by a strict regime with antihypertensive treatment both the number of strokes and of myocardial infarctions have decreased remarkably. This has been proven by several multi-center-studies, e.g. the project of the National Heart, Lung, and Blood Institute Bethesda, and the Finland project.

III. Predilection site of atherosclerosis

Cerebral atherosclerosis has a pronounced local predilection and follows here the local factors of increased wall stress as at bifurcations, branchings, curves, bony fixations, and outer strangulations of the artery. The first atherosclerotic plaque is observed at the bifurcation of the basilar artery already sometimes as early as 25 years of age, yet it is hardly impeding. In contrast marked stenosis in the coronaries has been observed in the same age group already during World War I and later in the Korean War. Decisive stenoses usually occur in the brain at the origin of the middle cerebral arteries and less commonly in the posterior cerebral arteries and in the third place only in the anterior and basilar arteries. The vertebral arteries are liable to total occlusion particularly at the point of penetration of the dura.

The predilection sites of stenoses for the heart are the following:

a) the left descending artery 2 cm after its origin
b) the right main artery 2-3 cm distally to its origin, however, also in 8-10 cm distance, and
c) the left circumflex artery again 2-3 cm from the ostium.

Where do we find the total occlusions and what is their pathogenesis? Most frequently they develop on and occlude a highly stenosed vessel segment in the brain; only rarely the thrombosis of a youthful vessel occurs.

Since early coronary angiography is now more often performed in acute myocardial infarction for subsequent intracoronary fibrinolysis it has become clear that in almost 80% of acute infarctions a complete thrombotic occlusion is demonstrable: following lysis of the occluding clots atherosclerotic stenoses of less than 50% are found in only about 6% of patients, 75-90% stenoses in around 50% of patients, and sub-total stenoses in 30%.

Clinical important investigations of 1000 stroke patients of our unit have proven that in more than 80% the very cause of cerebral infarction was stenosing atherosclerosis. Only in 60% macroembolism was the cause.

From the patient group with complete occlusion, whereby those with macroembolism were disregarded, 4/5 of these cases showed a very pronounced atherosclerosis with a final thrombosis on top, whereas a totally occlusing primary thrombosis comprised only 20% of the cerebral arteries.

IV. Anastomoses and collaterals

In the brain the possibility of a collateral supply in emergency is favorable, because of a great number of preexisting anastomoses:

1. Transverse anastomoses between the two carotid arteries and between the two vertebral arteries.
2. Anastomoses between the external carotid and (a) the internal carotid via ophthalmic artery and (b) the vertebral artery via its occipital branch.
3. Circle of Willis.
4. Intracranial meningeal anastomoses of Heubner from 200 up to 1000 micra(!) in diameter. This proves that the general pattern of the vascular supply of the brain has an almost web-like net, allowing collateral pathways at any level.

Of these the role of the meningeal anastomoses may be so effective that in proximal occlusion of the middle cerebral artery the defect can be completely compensated for by retrograde flux through the meningeal anastomoses from the anterior and posterior arteries. Clinically up to 20% of middle cerebral artery occlusions are free from neurological symptoms. Most of these anastomoses are usually sufficiently developed as to be able to operate immediately in emergency. They can be further dilated during increasing functional demand, however, a new-growth of functional vessels occurs in the brain only on the capillary level and will hardly be of any advantage for a defected circulation.

The significance of the functional role of the collateral network of the heart is controversial, although there exists a vast network of precapillary collaterals especially in the subendocardium and the middle layers of the myocardium: these are small vessels with a diameter of approximately 50 micra which are not able to restore a sufficient coronary blood flow following an acute occlusion of a coronary artery. In acute myocardial infarction collaterals seem to be of minor importance, yet a smaller action of collaterals - if present - may be operative and the actual territory of infarction is then smaller than the real distribution area of the artery. No convincing explanation has so far been found for the considerable variability of development of collaterals in the heart. It may be secondary to a different genetic disposition or due to different responses to the various stimuli capable to increase the anastomotic network. The pressure gradient is the decisive stimulus for the development of efficient collaterals by increasing of the diameter. A close correlation is therefore found between the extent of the collateral network and the severity of coronary disease (degree of stenosis and number of involved vessels). To illustrate the extraordinary importance of existing collaterals we want to point to the survival times of the various organs. Cerebral tissue in man has a survival time still oscillating at about 5-8 minutes and only in animal experiments, this can be prolonged to 1 hour. This limited survival time inhibits any successful action due to a growth of collaterals.

The same is true for the heart although there the survival time may be higher up to 30-45 minutes.

However, for the brain we have to point out to two facts which allow to understand the process of infarction in man better. The human brain has normally already a reserve circulation of 200% of the demand. Loss of function follows only after decrease to 50% of the normal circulation. However, necrosis is only caused by a further fall beneath 15-20%. There is a safety mantle then of 30% between the 50 and 20% borders. Here function is lost yet tissue can recover. While in the center of an infarcted area the brain may already be necrotic, the surroundings may be functionless, yet

recoverable. This afunctional tissue is liable to recovery by further improvements of circulation and metabolism which can be achieved either by conservative treatment or even by surgical means such as extraintracranial anastomoses etc. Although it has been shown that local cerebral blood flow may increase markedly according to the local demand, overall cerebral blood flow is rather stable. On the other hand, overall coronary blood flow has to increase extensively on exercise. The so-called "coronary reserve" corresponds to 300-400% of coronary blood flow at rest. However, since wall stress is higher in the subendocardium the reserve for vasodilation is also less in these layers and overall limitation of perfusion pressure and augmentation of extrinsic compressive factors comprises the subendocardial tissue first.

V. Metabolic changes leading to brain edema

We now know from many hundreds of experiments of microembolization in the cat that a pericapillary edema in the cortex arises already after a few minutes due to blood brain barrier damage. However, this damage disappears already after four hours and then the barrier will be tight again.

Thereafter a second form of edema occurs extracellularly. Pathogenetically this is a transudation through the larger veins. This extracellular edema may be volume taking and has a detrimental action on the myelin. This form of edema is mainly located in the white matter and may diffusely propagate from the perivenous base through the whole white substance, leading to necrobiosis of the myelin sheaths in the course of weeks, causing both an acute impairment of the tissue oxygenation and finally by secondary uptake of the protein-rich edema into the myelin sheaths a necrosis. Only sometimes, particularly in the hemorrhagic forms of infarcts the increase of volume can be so enormous as to lead to remarkable mass shifts and a fatal symptomatology.

Experimental work is only rarely focussed on the problem of edema in myocardial infarction, since the pericardial sac is large enough not to counteract the minor expansion of the myocardium by edema. The role of edema should, however, not be underestimated. Ischemic edema first adds to the extrinsic factors increasing coronary resistance such as the systolic compressive effects and secondly increases the distance of oxygen diffusion in the tissue.

VI. Pathogenesis of infarction

Cerebral infarction is widely dependent upon two factors, first the general systemic circulation, e.g. "vis a tergo" and secondly local impediments to the blood flow in the arteries.

For the clinical promotion of stroke hypertension may participate 1) by acute elevation of blood pressure, the "hypertensive crisis" inducing a "break-through" of autoregulation and blood brain barrier damage, the vessels assuming a pseudospastic image which is only the highest degree of autoregulation according to the Bayliss principle. 2) By the acute fall of chronic hypertensive pressure values to "normal" or even "hypotensive" levels with subsequent impairment of local circulation.

It seems to be proven now that at least during the "hypertensive crisis" real spasm "sensu strictu" does not follow the cerebral arterial tree. That spasm may occur in the brain by mechanic causes is undoubted.

In the heart coronary spasm is widely accepted to be the cause of Prinzmetal angina and may even lead to myocardial infarction.

VII. Transient ischemic attacks and angina pectoris

To pass over from the more morphological phenomena to the functional processes a comparison between the transient ischemic attacks (TIA) in the brain and angina pectoris in the heart will be tried. The most favored explanation of TIAs is now a speculated microembolism from ulcerated arteriosclerotic plaques either at the carotid bifurcation or at any other proximal point of the arterial tree. We admit that such emboli have been photographed in the retina in amaurosis fugax, e.g. transient blindness. Yet, 1) a statistical proof that these retinal microemboli do occur also in TIAs is still lacking. 2) In Toole's series of carotid endarterectomies of patients with TIAs only 13 of the 123 surgical specimens showed such an ulcerated thrombotic surface, however, the patients were cured after removal of the obstacle in the arterial wall. 3) According to the symptoms of TIA the emboli should always go to a particular circumscribed clear-cut vessel territory. Yet, in the experiment smaller microemboli of less than 80 micron, injected into the carotid artery, are spread evenly over the total distribution area. Only emboli of 1 mm - 10-15 times larger - remain in the central blood stream, when injected into the carotid artery, and have therefore a local predilection for the middle cerebral artery. 4) Another important observation is that in patients with retinal emboli neurological symptoms never occur which would have been expected if the microemboli would be evenly distributed over the whole carotid artery. We personally favor therefore the hemodynamic theory, e.g. a local stenosis in an artery and a temporary decrease of flow as the two promotors of a local disorder and triggered by circadian blood pressure fall, heart arrhythmias, orthostasis, etc.

VIII. Therapy

Most interesting would be a discussion of implications for therapy. This may be only introduced by one statement: The interpretation of an "ideal" blood pressure for the patient in a given situation may be different for the cardiologist and neurologist. The brain specialist will keep the blood pressure as high as possible emphasizing that this organ may secondarily suffer an irreversible infarct following lowering of the blood pressure distal to a vascular stenosis. The internist on the other hand tries to protect the heart from any unnecessary demand by maintaining a low systemic blood pressure. Many other implications of this kind are worthy of a serious discussion. Symposia of this kind, where neurologists and cardiologists are discussing these problems together, may intensify our knowledge for the benefit of the patients.

Acknowledgement

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ATHEROSCLEROSIS AND THROMBOSIS OF THE CEREBRAL AND CORONARY ARTERIES

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An association between cerebrovascular diseases and cardiac diseases has been recognised.[1,2] Three mechanisms are considered to contribute to the relation between them:

1. Cardiac diseases and cerebrovascular diseases are linked through common risk factors, e.g. hypertension, serum lipids and cigarette smoking.
2. Cerebral hypoperfusion due to impaired cardiac function contributes to the development of cerebral infarction and cerebral ischemia.
3. Thromboembolization from the heart associated with myocardial infarction, rheumatic heart disease, etc.

Prevalence of cerebrovascular diseases associated with organic heart diseases and the cases with abnormal ECG findings was investigated by reviewing 1162 autopsy cases (20 or over 20 years of age) at Department of Pathology, Kyushu University from Nov. 1971 to Oct. 1981. Forty-eight cases with cerebral infarction (CI) were found among 86 cases with myocardial infarction (MI) and its prevalence was higher than that of the control group (the cases without organic heart diseases, collagen disease, amyloidosis or chronic renal failure) according to sex and age adjusted statistical analysis (Table 1). Eighteen out of 40 cases with rheumatic heart disease (RHD) were associated with CI, which was higher in prevalence than the control group. It was considered that cerebral embolism contributed to these CI in some cases (Figure 1). Ten cases of CI seemed to be embolic cerebral infarction. Intracardiac thrombi and atrial fibrillation (af) were frequently found in these cases. Eleven out of 18 cases with RHD and CI were believed to be embolic. Eight cases were associated with af and intracardiac thrombi and

Table 1. Cerebrovascular Diseases among the cases with Cardiac Diseases

	No. of cases	Cerebral infarction	Non-embolic cerebral infarction	Cerebral hemorrhage
Myocardial infarction	86	48**	38*	1
relative risk		2.83	2.10	0.27
Rheumatic heart disease	40	18**	9	1
relative risk		3.38	1.16	0.52
Control	994	238	231	41

* $p<0.05$
** $p<0.01$ vs. control group (the cases without organic heart diseases) by Mantel-Haenszel χ^2

one case with af only. It was further studied whether the cases with MI were frequently associated with non-embolic cerebral infarction (Table 1). Prevalence of non-embolic CI among the MI group was significantly higher than the control group. But there was no difference in the prevalence of non-embolic CI in the RHD group and the control group. The cases with abnormal ECG findings (af, left ventricular hypertrophy, non-specific ST-T change) showed higher prevalence of CI than the cases without each abnormal ECG findings (Table 2).

The results of autopsy studies in Hisayama town in Japan[3] revealed that the severity of atherosclerosis showed variety in each case and each organ. Cerebral atherosclerosis was usually less severe than atherosclerosis of the aorta and coronary arteries. The severity of cerebral atherosclerosis was promoted statistically significantly by hypertension, but it showed less dramatic increase by serum cholesterol level.

Hypertension not only promoted cerebral atherosclerosis but also affected the pathogenesis of cerebral artery thrombosis. Thirty-nine cases of occluded cerebral arterial segments and 54 cases of occluded coronary arteries were investigated histopathologically by serial sections (Table 3).[4,5] The most conspicuous and frequently observed finding in the thrombosed segments of the cerebral arteries was intramural hemorrhage, which was present in 28 out of 39 segments. Hypertension was associated with 76.0% of the cases with intramural hemorrhage. Five cases showed fibrinoid necrosis of the small blood vessels in the atheroma. On the other hand, it was found that major cause of thrombus precipitation in

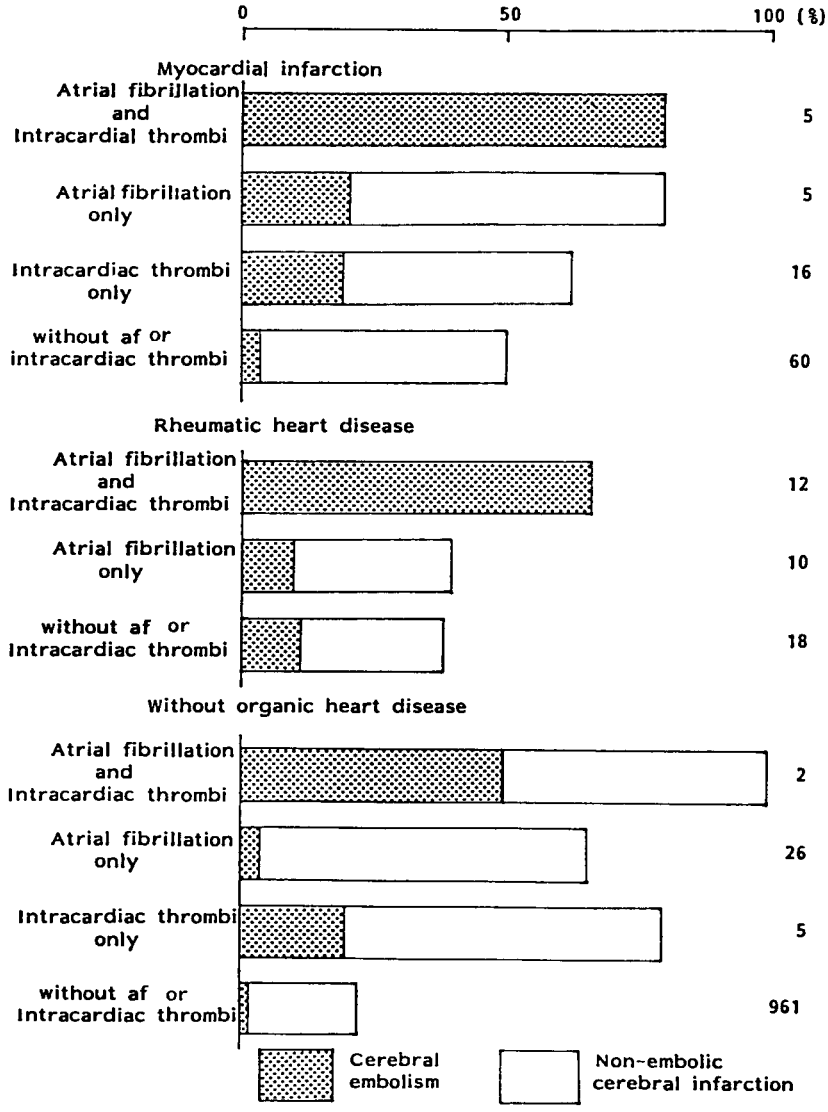

Fig. 1. Cerebral infarction among the cases with atrial fibrillation and intracardiac thrombi

the pathogenesis of coronary artery thrombosis was rupture or fissure formation of atherosclerotic plaque occupying 55.6% (30 cases). These characteristics in the pathogenesis of the cerebral atherosclerosis and the cerebral artery thrombosis are important in considering the relationship between cerebrovascular diseases and cardiac diseases.

Table 2. Cerebral Infarction among the cases with Abnormal ECG Findings

in the cases without organic heart diseases	No. of cases	Cerebral infarction No.	relative risk
Atrial fibrillation	28	19**	4.36
Left ventricular hypertrophy	170	82**	2.25
Non-specific ST-T change	248	123**	3.41

** $p<0.01$ vs. the cases without each abnormal ECG findings by Mantel-Haenszel χ^2

Table 3. Histological Characteristics of Coronary and Cerebral Arteries with Occluding Thrombus

	Cerebral arteries	Coronary arteries
Hemorrhage in plaque	28 (71.2%)	20 (37.0%)
Rupture of plaque	1 (2.6%)	30 (55.6%)
Edema of plaque	4 (10.3%)	0
Liquefaction of intimal surface	0	1 (1.8%)
Plaque alone	6 (15.4%)	3 (5.6%)
Total	39	54

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MANIFESTATIONS OF CORONARY DISEASE PREDISPOSING TO STROKE: THE FRAMINGHAM STUDY

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The relation of coronary heart disease to development of strokes in general and brain infarction in particular has been examined prospectively over 24 years of follow-up in the Framingham Study cohort. In the course of 24 years of biennial surveillance, there were 169 strokes in men and 175 in women aged 45-84; brain infarctions occurred in 100 men and 107 women, comprising 60% of strokes. Routine ECG's chest X-rays coronary heart disease and cardiac failure status were ascertained biennially on regular examinations and risk of strokes determined in relation to these. Age and other stroke risk factors (including blood pressure, diabetes, cigarettes and lipids were also routinely measured and were taken into account in multivariate analysis of the net and joint effects of CHD manifestations as precursors of strokes. The incidence of stroke was lower and stroke occurred later in life than coronary heart disease. Stroke incidence in men lagged that of myocardial infarction by more than 10 years. In women, the incidence of brain and myocardial infarction was similar. In men the average annual incidence of myocardial infarction (8.5/1000) was three times that for brain infarction (2.7/1000).[1] In both sexes, stroke incidence rose with age, more than doubling each successive decade above age 45 (Table 1). In contrast to myocardial infarction, where there is a striking male predominance, the incidence rate for brain infarction is only about 30% greater in men than women, and even this small sex differential is seen chiefly below age 65. Some 20% of brain infarctions occurred below age 65.

Since the underlying pathological features of atherosclerosis in the cerebral, cardiac and peripheral circulation are virtually identical, it is not unexpected that they share precursors. Atherosclerosis commonly affects all three areas simultaneously, and

Table 1. Occurrence of Stroke vs. Myocardial Infarction by Age and Sex. 24 yr. follow-up. Framingham Study. Average Annual Incidence per 10,000

	Brain Infarction		Stroke All Types		Myocardial Infarction	
Age	Men	Women	Men	Women	Men	Women
45-54	10	7	20	11	54	8
55-64	23	16	40	27	95	25
65-74	55	48	90	83	122	52
75-84	139	94	176	127	192	98
All Ages	26	21	44	34	83	25

persons with one clinical manifestation are at increased risk of the others. Also, asymptomatic carotid bruits predicted myocardial infarction as well as a brain infarction (Figure 1).[2] Brain and myocardial infarction often coexist, particularly in advanced age and coronary heart disease is the chief cause of death in stroke and brain infarction survivors. This is also the case in patients with transient ischemic attacks or carotid bruits.[1,2]

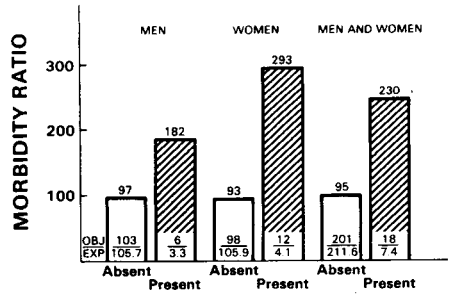
Coronary heart disease frequently occurs in persons who appear well. Strokes and brain infarctions more often occur on a background of illness: coronary heart disease (30%), occlusive peripheral arterial disease (30%), cardiac failure (15%), and diabetes (15%). Established hypertension is present in 70%.[1]

CARDIOVASCULAR RISK FACTORS

Although some non-trivial differences exist in their impacts on the incidence of each disease, blood pressure, serum cholesterol, ECG-LVH, glucose intolerance and cigarette smoking are precursors common to brain infarction and myocardial infarction. When all five major CHD risk factors are considered jointly, they are actually more highly predictive of brain infarction than coronary heart disease. Blood pressure and ECG-LVH are the chief determinants of this predictive capacity. Hyperlipidemia and the cigarette habit are less important than for coronary heart disease. Nevertheless, the coronary risk profile predicts stroke risk over a wide range, identifying in the upper decile of risk a tenth of the population from which half the strokes evolved compared to only 25% of CHD events (Table 2).

The dominant predisposing risk factors for brain infarction are hypertension and various cardiac impairments, including clinical

(a)



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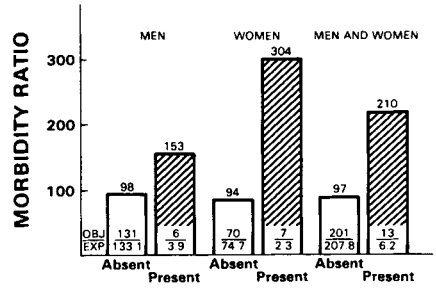

Fig. 1. (a) Age adjusted 2-year incidence of stroke by carotid bruit. Men and women 50-79 years. Framingham Study.
(b) Age adjusted 2-year incidence of myocardial infarction.

manifestations of coronary heart disease, cardiac failure, atrial fibrillation and preclinical evidence of a compromised coronary circulation. In candidates for coronary heart disease with an ominous cardiovascular risk profile, suggesting accelerated atherogenesis, the appearance of ECG abnormalities and X-ray evidence of cardiac enlargement not otherwise explained were hallmarks of a compromised coronary circulation, associated with an increased risk of myocardial infarction.[3]

CARDIAC IMPAIRMENTS

For stroke in general, an abnormal ECG indicated a greater than 3-fold increased risk of an event in men and 4-fold in women; the excess risk persisting on adjustment for age and other risk factors including blood pressure. ECG-LVH was the strongest ECG precursor with an associated 5 to 6-fold increased risk (Table 3). Chronic atrial fibrillation carried a 6-fold excess risk, persisting at 3-fold after adjusting for coexistent risk factors.[4] X-ray cardiac enlargement was not as powerful as ECG abnormalities,

Table 2. Percentage of cases of Coronary Disease, Brain Infarction, and Intermittent Claudication in the Upper Decile of Multivariate Risk* among Men and Women 45-74 years of age who participated in the Framingham Study: 16-year follow-up

	Coronary Disease		Brain Infarction		Intermittent Claudication	
Age	Men	Women	Men	Women	Men	Women
45-54	25.9	20.0	54.5	44.4	30.0	60.0
54-64	26.7	25.7	52.3	42.9	46.7	42.9
65-74	21.3	40.9	57.1	45.5	26.7	50.0

* Based upon systolic blood pressure, serum cholesterol, number of cigarettes smoked, electrocardiographic evidence of left-ventricular hypertrophy, and glucose tolerance.

carrying only a 2-fold increased risk which also increased uniformly with heart size. For brain infarction in particular, ECG-LVH was also the most powerful ECG predictor (greater than 4-fold) and was more ominous when there were associated ST-T wave repolarization abnormalities. Again, X-ray left ventricular hypertrophy was a less powerful predictor, but more important than generalized enlargement (Table 3). In men, there was no independent contribution of X-ray enlargement when ECG-LVH and hypertension were taken into account. Nonspecific ST-T wave abnormalities carried an increased risk in both sexes, independent of age and associated risk factors. Intraventricular block was a significant contributor to brain infarction risk only in women.

Thus, even asymptomatic cardiac impairments associated with a compromised coronary circulation were clearly associated with an increased risk of stroke. Cardiac impairments ranked third, following age and hypertension, as risk factors for stroke in general and for brain infarction in particular. At any age, in either sex, and at any level of blood pressure, persons with cardiac disease, whether overt or subclinical, have more than a doubled risk of a stroke (Figure 2). These predisposing cardiac impairments include not only overt coronary heart disease, but occult diseases such as left ventricular hypertrophy by ECG or X-ray and atrial fibrillation.

OVERT CARDIAC DISEASE

Subjects with overt cardiac disease had almost a tripled risk of a stroke (Table 4). This applied equally in both sexes. As

Table 3. Risk of Stroke according to Left Ventricular Hypertrophy by ECG vs. X-Ray. Subjects 45-84. The Framingham Study. 24-year follow-up. Age-adjusted average annual incidence per 10,000

	Stroke (all types)				Brain Infarction			
	ECG-LVH		X-Ray LVH		ECG-LVH		X-Ray LVH	
Abnormality	Men	Women	Men	Women	Men	Women	Men	Women
None	39	28	41	28	23	17	24	15
Possible	68	86	54	29	62	39	34	18
Definite	206	182	68	54	102	133	42	41
Risk Ratio	5.3	6.5	1.7	1.9	4.4	7.8	1.8	2.7
Multivariate Z-Value	3.57	5.09	0.90	2.39	2.39	3.72	0.78	3.21

might be expected, the risk associated with a myocardial infarction (5-fold) was substantially greater than that associated with angina (2-fold).

Cardiac failure which is predominantly due to hypertension and coronary heart disease, increased the risk of stroke 5 to 6-fold for strokes in general and 2 to 4-fold for brain infarction in

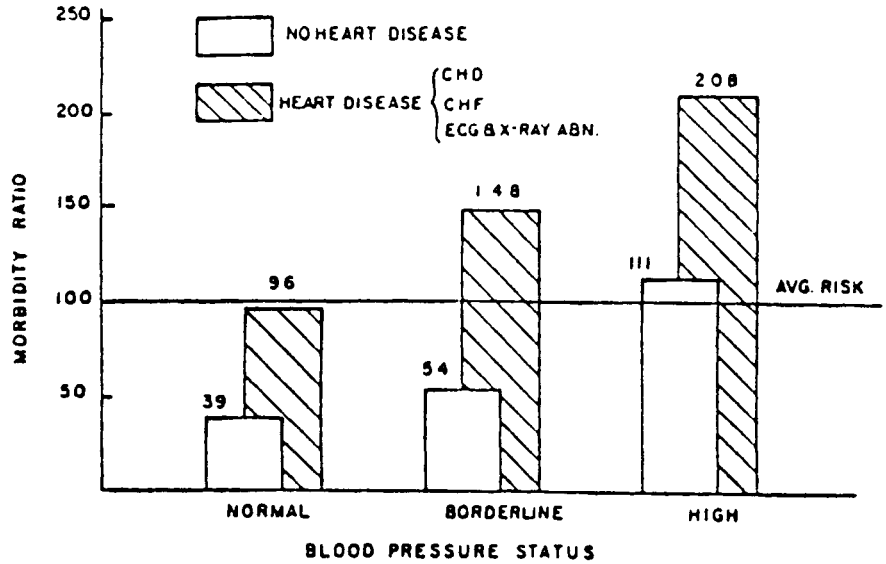

Fig. 2. Risk of stroke according to blood pressure and cardiac impairment. 24-year follow-up. Framingham Study.

Table 4. Risk of Stroke comparing ECG-LVH, Cardiac Failure and Coronary Disease. 24-year follow-up. Framingham Study. Subjects 45-84. Age-adjusted average annual incidence per 10,000

Abnormality	CHF		CHD		ECG-LVH	
	Men	Women	Men	Women	Men	Women
Negative	41	31	37	28	39	28
Definite	221	192	102	78	206	182
Risk Ratio	5.4	6.2	2.8	2.8	5.3	6.5
Multivariate Z-Value	5.13	3.59	4.04	3.59	3.87	5.09

particular (Table 4). The risk appears to be greater in women than men. Both for congestive heart failure and coronary heart disease the excess risk, particularly for men, persists on taking associated risk factors, including hypertension, into account. Coronary disease, when associated with hypertension or cardiac failure, carried a greater risk than when not complicated by these features (Figure 2).

Thus, strokes are a part of a larger problem of cardiovascular disease and once coronary heart disease, congestive heart failure appear, or even asymptomatic evidence of a compromised coronary circulation appears, risk of stroke is greatly escalated (Figure 2). Also, survival and recurrences following a stroke are greatly influenced by comorbidity; notably by the coexisting coronary heart disease and congestive heart failure (Figure 3). Prevention of strokes requires not only control of hypertension, but prevention and relief of coronary heart disease, congestive heart failure and atrial fibrillation.

SUMMARY

Based on 24 years of biennial examinations, during which time 344 strokes occurred, the role of CHD was examined as a precursor of stroke. The five major risk factors for CHD were jointly even more predictive of brain infarction than myocardial infarction, identifying a tenth of the asymptomatic population from which half the strokes evolved. The dominant stroke risk factors were hypertension, clinical manifestations of CHD, cardiac failure and preclinical evidence of a compromised coronary circulation. CHD more than doubled the risk and cardiac failure was associated with a 5-fold increased incidence of stroke. Chronic atrial fibrillation increased the stroke risk 6-fold and ECG-LVH was the most powerful ECG predictor. All cardiac impairments added to the stroke risk associated with hypertension.

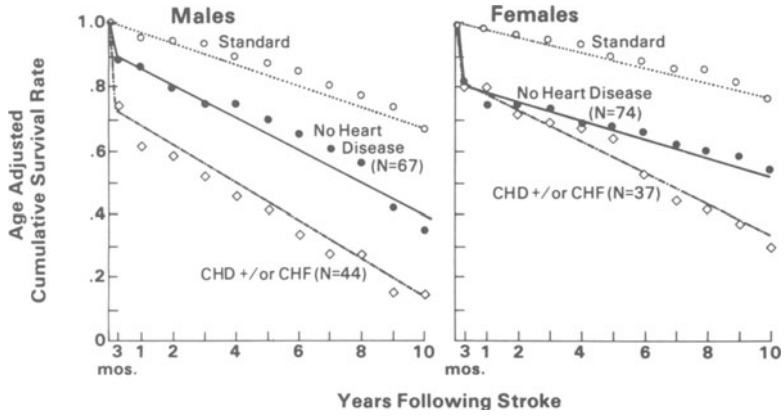

Fig. 3. Survival following ABI, effect of prior Cardiac Comorbidity. Framingham Study. 26-year follow-up.

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LIMITATION OF INFARCT SIZE: THEORETICAL ASPECTS

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The basic postulate is that cardiac infarct size is directly related to clinical complications and death. The consequent therapeutical implication is to reduce the "theoretical" size and/or to prevent its progression (or expansion) in the surrounding myocardium. The latter is considered more prone to ischaemia and therefore at high risk of necrosis ("jeopardized border zone").

In the present controversy on both the existence of such a border zone and the possibility to limit the infarct size, the definition of infarct necrosis and its natural history as well as the definition of the type of changes in the normal myocardium at the periphery of the infarct in man seem opportune.

Human infarct necrosis

The first functional disorder is the loss of contraction. The myocardial cells are dying in relaxation phase ("atonic death"). Thinning of the dead cells, elongation of sarcomeres and nuclei are the earliest histologic signs which reflect the passive distension of the irreversibly relaxed elements by the intraventricular pressure (systolic paradoxical bulging). Centripetal infiltration of polymorphonuclear leukocytes, secondary wall degeneration and thrombosis of the vascular structures, removal by macrophages and fibrous replacement are the main progressive changes of the repair process. A regular, periodic order of the myofibrils is maintained even in the last remnants of necrotic tissue.

This necrosis is usually termed "coagulation necrosis" and represents the pathognomonic postmortem finding in patients dying from typical clinical pattern of cardiac infarct.

Table 1. Infarct size and age in 200 consecutive acute cases

Age in Days	Infarct Size (%) less 10	11-20	21-30	31-40	41-50	50	Total
2	34	11	11	9	1	4	70
3-10	17	9	24	10	9	5	74
11-30	9	17	9	8	8	5	56
Total	60	37	44	27	18	14	200

In the majority of the cases the lesion is monofocal. In 200 consecutive and selected (no other cardiac and non cardiac disease, no surgery) patients its size ranged from less than 10 per cent to more than 50 per cent of the left ventricular mass[1,2]. The survival time (from clinical onset to death) varied from less than two to 30 days and was not related to the infarct size (Table 1). In no one instance of these 200 acute infarct cases progression (or expansion) of the primitive coagulation necrosis was documented. In other words the association of an earlier phase of the coagulation necrosis external to an older one was never observed.

Little is known on the time relation between reversible and irreversible infarct damage in man. The only available data are from experimental coronary occlusion models. In dog it has been calculated that after an occlusion lasting 20 minutes there is a total recovery, while after one hour most of the involved myocardial cells are dead. Due to the obvious differences among various species and overall between experimental models and human pathology, we may only induce that approximately in a similar period of time coagulation necrosis is also settled in man.

Structural changes in the "normal" myocardium around the infarct

More precisely by normal myocardium we should intend the myocardium preserved by the coagulation necrosis. In fact in most of the fatal cases two other types of myocardial necrosis can be detected[3].

From the morpho-functional standpoint the first one is the opposite pattern of the coagulation necrosis. The myocardial cells are contracted, or better hypercontracted, with rhexis of the myofibrils and anomalous cross-band formation. These structural changes suggest that the myocells die in irreversible contraction state ("tetanic death"). The subsequent fragmentation is likely due to the mechanical action of the normal acting myocardium on the rigid, hypercontracted elements. This type of lesion is present around an

infarct in about 80 percent of the cases. It may involve single elements or foci formed by few myocells or may become massive by confluence of several foci. Apparently is not related to infarct size and coronary damage, and is mainly located at the lateral-external sites of the infarct. This necrosis never shows specific inflammatory infiltration or vessel degeneration and its repair process progresses again by macrophagic phagocytosis, followed by collagenization of the empty salcolemmal tubes ("alveolar pattern"). Present in many other conditions (pheochromocytoma, stone heart, transplanted human heart etc.) is reproduced experimentally by catecholamine infusion. Variously called (contraction band necrosis, myofibrillar degeneration, coagulative myocytolysis) may be simply defined as "Zenker necrosis". The first structural changes can be detected within few minutes and consist in alteration of the myofibrillar system.

The other type of necrosis is still a primary damage of the myofibrils, but with a completely different pattern from the previous one. This process is characterized by a progressive "edematous" vacuolization with disappearance of the myofibrils. Histologically the myocardial cell appears as an "Empty" clear element with an apparently normal nucleus. Found in any cardiomyopathy with low output syndrome, has been variously named (sarcolysis, colliquative myocytolysis) and may be called "myocytolysis", using this term for this specific lesion without confusion with the previously described Zenker necrosis. The histologic findings associated with cardiac failure suggest a progressive diminution in strength and velocity of contraction as basic morpho-functional disorder ("failing death"). In more than 40 percent of the 200 acute infarcts this lesion was seen in the preserved myocardium around the survived vessels and beneath the endocardium mainly in cases with longer survival and extensive myocardial fibrosis (Table 2).

Table 2. Myocytolysis in Relation to Survival Time and Myocardial Fibrosis.

	Survival time - days		
200 AMI	2	3-10	11
Total	70	74	56
With myocytolysis	20 (28.5%)	28 (37.8%)	28 (50%)

	Myocardial fibrosis	
	no or minimal	extensive
Total	145	55
With myocytolysis	46 (31.7%)	30 (54.5%)

A last observation has to be mentioned to complete the whole histologic pattern seen in the acute myocardial infarct. Practically in all earliest cases a rim formed by a thin layer of hypercontracted myocardial cells with anomalous cross band, at the lateral and external surface of the coagulation necrosis is visible. Already described in the first reports as part of the latter, such a well distinct rim may vary in depth and extension. Furthermore, since the polymorphonuclear infiltration originates in the outer site where blood flow is present, this rim of Zenker necrosis is associated with leukocytes, and may be easily confused with the coagulation necrosis.

Finally it should be mentioned that in 208 selected cases of sudden coronary death (apparently healthy people who died out-of-hospital without any medical intervention and resuscitation attempts) in 72 per cent Zenker necrosis was the only documented acute lesion, while coagulation necrosis associated with Zenker necrosis was demonstrable in 15 per cent and myocytolysis was practically absent. In 97 "controls" (normal subjects dying suddenly by accidental death) no one showed coagulation necrosis and myocytolysis while 19 presented minimal foci of Zenker necrosis.[4,5]

Theoretical aspects in relation to the morpho-functional findings

From the previous findings several aspects should be considered. First the clear-cut morpho-functional difference indicates that the three types of myocardial necrosis are distinct entities and not stages of the same damage. Human and experimental data show that each type represents a specific metabolic disorder. This implies different pathogenesis in accordance with the general rule that each pathogenic mechanism determines pathogonomic dysfunctional and structural changes. The assumption that the three types of damage may correspond to different degrees of ischaemia should imply that any focus of infarct necrosis is a "mixed" lesion since topographical and chronological gradients of ischaemia exist in the evolution of the damage (total reversibility within 20 min, dyshomogeneous irreversibility before one hour). Again Zenker necrosis is experimentally established in a few minutes and increasing doses of catecholamines produce a larger, but not different type of lesion. Finally, according to the structural changes it is unlikely that a fragmented, hypercontracted myocell may revert in a normal distended cell or vice versa.

At present etiology and pathogenesis of each single myocardial necrosis in man is controversial. Infarct necrosis (coagulation necrosis) is likely due to reduction of the nutrient flow, even if the cause-effect relation for many, if not all, the proposed causes is still questionable. On the other hand Zenker necrosis and myocytolysis appear to be non ischaemic, primary "metabolic" myocellular disorders in which excess or depletion of catecholamines or catechol-

amine-like substances seem to have an important role in determining the specific cellular dysionism. However what seems not to be overlooked is the presence and the frequent association of these different types of damage in the natural history of the so-called ischaemic heart disease. In the latter complications and death seem more linked with metabolic disorders at myocellular level than to blood flow reduction. In particular in people predisposed by congenital and/or acquired factors, sympathetic overstimulation leading to malignant arrhythmias may occur as primary event (sudden death in absence of an infarct); or following an infarct. In this condition at a critical point of myocardial mass with loss of contraction (infarct), the normal myocardium is stimulated to compensatory hyperfunction mediated by the sympathetic nervous system, particularly at the border of the infarct where there is maximal mechanical tension.

From the other site exhaustion or depletion of catecholamines may be responsible for cardiac failure secondary to an acute infarct. The histologic hallmark of this dysfunction (myocytolysis) is mainly found in cases with longer survival and with associated myocardial fibrosis.

The infarct size concept

A correct determination of the infarct size should be limited to the measurement of the tridimensional extension of the coagulation necrosis. Contradictory results, some misunderstanding and "delayed" necrosis concept are likely due to the lack of discrimination between the primitive coagulation necrosis and the other subsequent necrotic processes by the various methods in vivo and by post-mortem enzymatic tissue stain.

With this in mind, the first question is whether or not the infarct size is related to the various clinical courses characterizing this nosologic entity (uneventful recovery or recovery after more or less severe complications or death both sudden during a regular course or after complication). At present the only way to establish the infarct size is a direct measurement of the necrotic tissue controlling by microscope the exact type of necrosis. In turn this means that our information is confined to the small percentage of patients who die; and therefore they may not be representative of the whole population of infarcted patients. Nevertheless the finding that death, as end result of major complications, does not correlate with the infarct size, strongly suggests that the latter may, in general, not correlate with the different clinical courses. In other words the clinical course may be benign or malignant independently of the size of coagulation necrosis (in thirty per cent of the fatal acute infarcts the size is less than ten per cent and about half of these patients had a size less than twenty per cent; while only in 16 per cent the size was greater than 40 per cent).

In these circumstances one may argue if any attempt to reduce the size of the coagulation necrosis is appropriate. The establishment of infarct necrosis requires from 20 to 60 minutes. There is little time for any intervention to restore the nutrient flow in the ischaemic area. Last but not least reflow after 20-40 minutes aggravates the myocardial damage and lowers the threshold for ventricular fibrillation. Therefore any attempt to reduce the extension of the coagulation necrosis may be ineffective (small infarct) or unrealistic (too late) if not harmful (reflow damage); keeping in mind that there is no evidence that expansion of the primitive coagulation necrosis may occur in man. Since other non ischaemic types of irreversible damage linked with complications are present, the concept of limitation of infarct size should be substitutive by the concept of protection of the normal myocardium from damaging mechanisms other than blood flow reduction. For instance, betablockade before experimental coronary occlusion protects against ventricular fibrillation and Zenker necrosis[6]. Many therapeutical interventions thought to be effective in reducing the infarct size, are likely acting in this direction, and patients with uneventful recovery are self-protected by this damage.

In accordance therapeutical interventions and preventive actions should be related to etiology and pathogenesis of the metabolic damage, discovering effective signals capable of discriminating the patients at risk of complications.

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QUANTITATIVE ASSESSMENT OF INFARCT SIZE AND ITS INFLUENCE BY THROMBOLYSIS

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INTRODUCTION

Conventional nuclear cardiology employs gamma-emitting tracers such as technetium-99 (^{99m}Tc) or thallium-201 (^{201}Tl). Such tracers are not physiological metabolites and accordingly behave somewhat differently from the physiological constituents being traced. Single-photon emitters decay by liberating energy in the form of photons characteristic of the parent radio-nuclide, much of which is absorbed (attenuated) before it reaches the detector. Because of the variability of attenuation it is difficult, if not impossible, to accurately define both the amount of radiation emitted and its location. Thus, even though the reconstructed images may be aesthetically satisfying, quantitative limitations preclude definitive correspondence between the reconstructed image and the actual distribution of tracer in the organ of interest in absolute terms.[1]

Tracers employed in positron emission tomography (PET) such as carbon-11 (^{11}C), nitrogen-13 (^{13}N), oxygen-15 (^{15}O), and others decay by emitting positively charged particles, positrons, which traverse only a short distance before encountering a negatively charged electron and undergoing annihilation with liberation of two 511 kev photons directed approximately 180^{o} apart. Accordingly, coincidence counting provides electronic collimation and spatial localization of the emitted positron with attenuation compensated regardless of the locus of the source with respect to each member of each detector pair. [2]

Instrumentation employed at Washington University, developed by Dr. Michel M. Ter-Pogossian and his colleagues, permits reconstruction of seven 16 mm thick transaxial sections of the heart with an inter-section distance of 10 mm from apex to base. Repeat imaging after the patient has been shifted 9.5 mm provides a total of 14 tomographic reconstructions after a single injection of tracer. Since most positron-emitting radionuclides have short half-lives (for example, 20.4 min for ^{11}C), they can be utilized sequentially without imposing excessive radiation burdens on the patient.

Regional Myocardial Metabolism

During the past several years, we have demonstrated that regional myocardial metabolism can be defined *in vivo* with the use of positron-emitting radionuclides despite influences of residence time and altered extraction fractions as a function of flow.[3-11]

Because fatty acid is the primary substrate for energy production by myocardium *in vivo*, we have utilized carbon-11 labeled palmitate. Time-activity curves permitted calculation of extraction fraction and the monoexponential rate of metabolism of ^{11}C-palmitate incorporated into cellular lipids in isolated perfused rabbit hearts, verified by chemical and radio-chemical analyses. Subsequently, normal canine myocardium was shown to accumulate the tracer homogeneously *in vivo*. Tissue rendered ischemic exhibited markedly decreased uptake demonstrated tomographically correlating closely (r=.97) with regions of necrosis verified histologically and enzymatically at autopsy. When imaging was performed prior to the development of irreversible injury but in the face of ischemia, an analogous "cold-spot" was evident. However, under these conditions reflow led to accumulation of intravenously administered ^{11}C-palmitate in the previously ischemic zones.[3] With a multi-slice system (PETT IV) and delineation of the intracardiac blood pool and the endocardial border of the heart in patients with the use of inhaled ^{11}CO-labeled hemoglobin, close correspondence between the electrocardiographic locus of infarction and persistent impairment of fatty acid metabolism was well demonstrated and infarct size estimated by PET correlated closedly (r=.92) with infarct size estimated by the serial creatine kinase method.

Others have utilized agents such as glucose labeled with fluorine-18 and obtained qualitatively similar images. On the other hand, the introduction of a halogen alters the metabolism of glucose. Thus, it is difficult to interpret such images unequivocally since analysis of carbohydrate metabolism in normal and ischemic tissue cannot be extrapolated directly to metabolism of congeners with altered affinity for enzymes involved.

The present study was performed to determine whether PET with ^{11}C-palmitate provides a potentially useful approach for assessment of the efficacy of thrombolysis in restoring myocardial metabolism and to delineate the temporal dependence of its efficacy.

METHODS

Conditioned dogs anesthetized with sodium thiopental (12.5 mg/kg) and α-chloralose (60 mg/kg) were studied after left femoral artery and vein cannulation and administration of lidocaine. Coronary thrombus was induced with a copper coil (5 to 7.5 mm in length) inserted into the left anterior descending coronary artery.[12] An occlusive thrombus developed within 5 to 15 minutes, heralded by typical electrocardiographic signs of ischemia including ST elevation and ventricular dysrhythmia and confirmed angiographically. Thirty minutes after induction of thrombus, and at selected intervals from 1 to 14 hours after occlusion each dog was given 15 to 20 mCi of ^{11}C-palmitate ($t_{1/2}$=20.4) intravenously and studied tomographically.

Immediately after tomography had been completed, streptokinase (4000 U/min) dissolved in saline (2000 U/min) or saline alone was administered through the intracoronary catheter. In control and treated dogs, positron emission tomography was performed again after a second intravenous injection of ^{11}C-palmitate 1.5 hr after the initial tomographic study, coronary arteriography was repeated, and a catheter was advanced into the left ventricle for injection of approximately 4 million ^{85}Sr microspheres (50 μCi, 15 μm diameter spheres) for measurement of myocardial blood flow.

RESULTS

Coronary thrombi was lysed successfully, judging from sequential angiograms, in approximately 85% of all attempts. Approximately 30 minutes after the initiation of thrombolysis, reperfusion arrhythmia occurred (ectopic ventricular beats often progressing to accelerated idioventricular rhythm).[13] All dogs with induced coronary thrombus exhibited tomographic defects of ^{11}C-palmitate accumulation in initial transverse tomograms. In the absence of lysis, tomographic defects were stable (Table 1). After reperfusion tomographic changes indicative of restored metabolism in these previously identified zones were markedly dependent on the duration of the preceding occlusion (Figure 1). Longer intervals of occlusion were associated with persistence, rather than of the defects.[14]

Occlusion for one to two hours followed by reperfusion led to a 51.1± 6.3% <u>decrease</u> in the overall extent of metabolically

Table 1. Tomographically Estimated Infarct Size in Dogs with and without Coronary Thrombolysis

Experimental Group and Interval After Occlusion	n	Tomographically Estimated Jeopardized Zones[a] Tomogram I	Tomogram II[b]
Controls; 1 to 6 hours	6	24.7 ± 1.5	25.6 ± 2.7
Thrombolysis; 1 to 2 hours	4	24.6 ± 3.8*	12.0 ± 2.3*
Thrombolysis; 2 to 4 hours	6	25.7 ± 2.8*	20.3 ± 2.3*
Thrombolysis; 4 to 6 hours	4	24.7 ± 3.2	21.6 ± 4.0
Thrombolysis; 12 to 14 hours	3	23.9 ± 3.1	24.5 ± 3.7

* $p < .01$ within the same experimental group.

a) Values are means ± SE of the number of pixels exhibiting less than 50% of the maximum count rate/total left ventricular wall pixels (thus expressed as % of left ventricle).

b) The second tomographic study was initiated 1.5 hours after the first scan. In each case in experimentally treated animals, thrombolysis was initiated after tomogram I had been obtained.

Table 2 Tomographically Estimated Metabolic Activity in the Jeopardized Zone Before and After Coronary Thrombolysis

Experimental Group and Interval After Occlusion	n	Metabolic Activity in the Initial Tomographically Defined Jeopardized Zone[a] Tomogram I	Tomogram II[b]
Controls; 1 to 3 hours	6	31.0 ± 2.5	33.6 ± 3.5
Thrombolysis; 1 to 2 hours	4	30.1 ± 2.6*	61.8 ± 4.6*
Thrombolysis; 2 to 4 hours	6	39.1 ± 2.8*	55.6 ± 5.0*
Thrombolysis; 4 to 6 hours	4	28.2 ± 2.8	37.6 ± 6.2
Thrombolysis; 12 to 14 hours	3	31.3 ± 3.9	33.8 ± 1.6

* $p < .01$ within the same experimental group.

a) Values are means ± SE of ^{11}C-radioactivity/pixel in the initial jeopardized zone (defined as the region in which count rate was less than 50% of maximum normal left ventricular wall count rate in the initial tomogram) normalized by expressing the values as % of average counts/pixel in the normal zone.

b) The second tomographic study was initiated 1.5 hours after the first scan. In each case in experimentally treated animals, thrombolysis was initiated after tomogram I had been obtained.

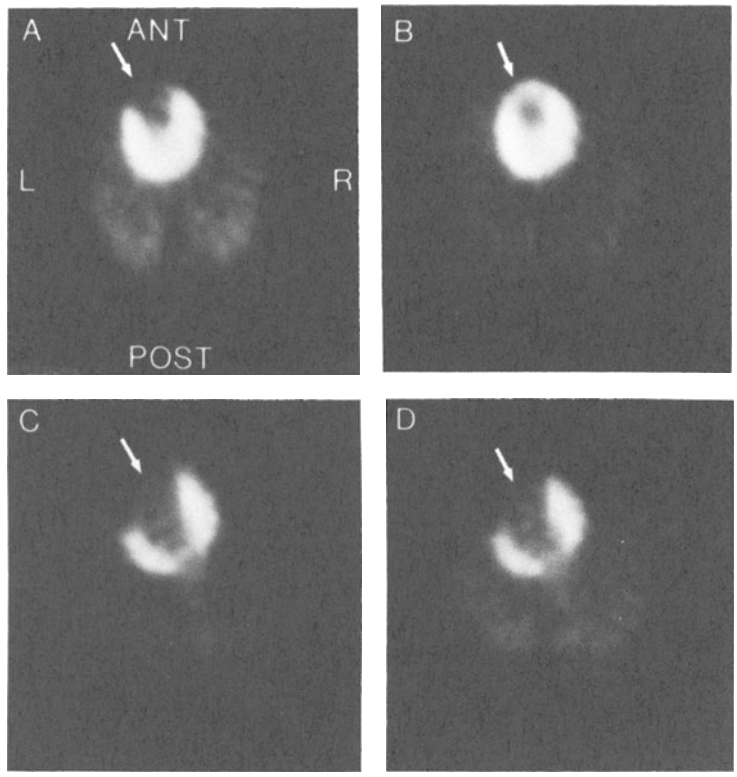

Fig.1 Transverse cardiac positron emission tomographic reconstructions obtained after intravenous administration of ^{11}C-palmitate in dogs. Reconstructions depicted are those obtained 1 hour after experimentally induced left anterior descending coronary artery thrombosis (A) and again after thrombolysis in the same dog (B). Normal myocardium extracts palmitate uniformly, whereas the ischemic zone exhibits diminished accumulation of tracer (arrow). The tomogram in panel B demonstrates substantial restoration of metabolism in the previously compromised anterior myocardium. In the lower panel, a tomogram 6 hours after onset of thrombosis and prior to the administration of streptokinase is shown (C) with a repeat tomogram (D) from the same dog 1 hour after intracoronary thrombolysis with streptokinase (confirmed angiographically). In contrast to the restoration of metabolism observed in dogs in which reperfusion was induced early after thrombosis, animals subjected to thrombolysis later than 6 hours after occlusion exhibited no significant restoration of metabolism despite angiographically documented lysis of coronary thrombi. (After Sobel et al, reprinted with permission from the Am. J. Med.)

compromised zones ($p < .01$) (Table 2). Occlusion for two to four hours prior to thrombolysis led to a more modest decrease in the overall extent of the metabolically compromised zone (by 21 ± 1.8%) and to a more modest increase in apparent metabolic activity (by 51.8 ± 12.6%) ($p < .01$). With occlusion of longer duration prior to thrombolysis no statistically significant change in the distribution of impaired metabolic activity was evident and dogs with twelve hour occlusions prior to thrombolysis showed no alteration in the distribution or magnitude of metabolic compromise (Table 3). Hearts of all dogs subjected to reperfusion exhibited transmural myocardial hemorrhage, contraction bands, and interstitial edema. After both early (≤ 6 hours) and late (6 to 14 hours) reperfusion, transmural flow was substantial in previously ischemic zones (>80% and >60% of normal).

Table 3 Percentage Change in Tomographically Estimated Jeopardized Zone and Metabolic Activity After Coronary Thrombolysis

Experimental Group and Interval After Occlusion	n	Changes as a Function of Time With and Without Thrombolysis: Jeopardized Zone[a]	Metabolic Activity[a]
Controls; 1 to 6 hours	6	2.5 ± 7.0%	9.5 ± 10.1%
Thrombolysis; 1 to 2 hours	4	-51.1± 6.3%	111.0 ± 24.3%*
Thrombolysis; 2 to 4 hours	6	-21.0± 1.8%	61.8 ± 12.6%*
Thrombolysis; 4 to 6 hours	4	-13.9± 7.5%	40.1 ± 30.2%
Thrombolysis; 12 to 14 hours	3	1.0 ± 10.0%	10.1 ± 9.0%

* $p < .01$ within the same experimental group.

a) Values represent means ± SE expressed as the percentage change in comparisons between results of the two tomographic studies for each of the parameters expressed in absolute terms in Tables 1 and 2.

Tables 1, 2 and 3 are from S. R. Bergmann, R. A. Lerch, K. A. A. Fox, P. A. Ludbrook, M. J. Welch, M. M. Ter-Pogossian, and B. E. Sobel, The temporal dependence of beneficial effects of coronary thrombolysis characterized by positron tomography, published with permission of the American Journal of Medicine.

DISCUSSION

These results indicate that the metabolic status of jeopardized ischemic myocardium can be assessed objectively by sequential positron emission tomography after administration of ^{11}C-palmitate — a modality already employed successfully for other purposes

in patients.[8,11,15]. When reperfusion was initiated within four hours after coronary occlusion, restoration of myocardial metabolism was substantial. However, with longer intervals of occlusion prior to reperfusion, salutary effects on metabolism were less pronounced.

Thus, the interval during which reperfusion will be beneficial to myocardium appears to be limited sharply. Its boundaries will obviously depend on the extent of the collateral circulation, the completeness of occlusion, hemodynamic and metabolic demands, and other factors.

Although intracoronary thrombolytic therapy early after the onset of apparent infarction and as late as 18 hours after the onset of infarction in patients has been reported to be helpful, conventionally available criteria of efficacy may be misleading. When thrombolytic therapy is initiated prior to evolution of unequivocal signs of infarction and ST-segment elevation is present with prolonged chest pain but plasma CK values are still normal, results are difficult to interpret.[16] Infarction does not occur invariably in this setting, as shown in a large, well controlled multi-center study of preinfarction angina which demonstrated that documented infarction accompanies such episodes in the absence of thrombolysis only infrequently.[17] Thrombolysis initiated as late as 18 hours after the onset of infarction may be accompanied by amelioration of chest pain and ST-segment changes; accelerated evolution of Q waves; elevated plasma CK activity; and improved left ventricular ejection fraction analogous to changes reported after early thrombolysis. However, reperfusion may fail to restor viability or exacerbate injury after such prolonged intervals.[18,19]

This study demonstrates the potential value of positron emission tomography as a tool for objectively assessing effects of thrombolysis on myocardial viability in patients. Results indicate that salvage of jeopardized myocardium by early coronary thrombolysis in experimental animals *in vivo* is detectable readily. They underscore the dependence of its efficacy on the interval available for effective reperfusion and may be of particular relevance to the design of longitudinal, clinical trials employing objective endpoints which can be interpreted unambiguously in the continuing effort to elucidate this potentially promising intervention.

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LIMITATION OF INFARCT SIZE BY PHARMACOLOGICAL INTERVENTIONS

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By the introduction of coronary care units the mortality in acute myocardial infarction decreased from about 40% to about 15-20%. This was mainly due to:

(1) good monitoring of patients with early detection of arrhythmias,
(2) effective antiarrhythmic drugs (e.g. lidocaine),
(3) well trained staff with early and skilled handling of cardiac standstills.

For these reasons a further gain in mortality reduction due to arrhythmias could not be expected. The other main cause of death is left ventricular failure, which is correlated to myocardial infarct size. To further reduce mortality in acute myocardial infarction, it seems logical to try to limit infarct size.

A number of experimental studies show that if therapy is started early enough infarct size may be limited. Many clinical studies have demonstrated the correlation between infarct size and mortality (Table 1).[1-12] In 9 out of 10 studies there is a good correlation between peak enzyme values and hospital mortality. In 3 out of 5 studies where the patients were followed for 3 to 5 years high serum enzyme values were positively correlated to high mortality. It can thus be assumed that limitation of infarct size would decrease mortality after a myocardial infarction. Very few data on this subject have been available hitherto.

Table 1. Correlation Between Peak-enzyme Value and Mortality

Authors	Numbers of pat	Percentage of 1st infarction	Enzyme	Hospital mortality	1-5 years mortality
Kibe and Nilsson 1967	155	Not given	LD, GOT	+	+
Kluge 1969	84	Not given	CK	+	
Chapman 1971	376	Not given	LD,GOT	+	
Hofvendahl 1971	271	70%	GOT	-	-
Sobel et al. 1972	33	Not given	CK	+	
Scheinman and Abbot 1973	230	60%	LD, GOT	+	
Helmers 1973	606	74%	GOT	+	-
Henning et al. 1975	2008	63%	GOT	+	
Vedin et al. 1977	292	100%	GOT		+
Nordlander and Nyguist 1979	194	67%	CK, GOT	+	
Thompson et al. 1979	560	72%	CK, ASAT		+
Thanavaro et al. 1980	745	100%	GOT	+	

+ peak enzyme value and mortality correlated

- not correlated

METHODS FOR ESTIMATION OF INFARCT SIZE

There has been an intensive debate on the accuracy of the clinical methods for infarct size measurements. It is obvious that no existing method is ideal. The most widely used method is serum enzyme measurements, as introduced by Shell et al.[13] Using an enzyme with a flat disappearance curve, such as lactate dehydrogenase (LD), there is a good chance to hit the curve near its maximum. This means that LD_{max} could be used instead of enzyme curves. Creatine phosphokinase (CK) appears in serum and reaches maximum very early with a rapid disappearance rate. It is therefore more difficult to get a serum sample at the maximum serum concentration. Using this enzyme, serum concentration curves are more accurate.

Precordial ECG mapping, as described by Maroko et al,[14] using 24-28 electrodes, gives an estimate of infarct size in anterior and lateral wall infarctions.

Myocardial scintigraphy is a newly developed method for infarct imaging with very promising results. The best method in the future, however, will probably be the use of nuclear magnetic resonance (NMR) techniques.

CLINICAL STUDIES OF INFARCT SIZE LIMITATION

The prerequisite for effective limitation of infarct size is the concept that the acute phase of myocardial infarction is a dynamic process evolving over several hours (Figure 1).[15] Only then there is a chance that the patient can be treated before the completion of the infarction. This is confirmed by the results of the Swedish metoprolol trial[16,17] (Figure 2) where the best clinical effect was found among those patients who came into treatment early, i.e. within 12 hours.

The infarct evolves because of an imbalance between myocardial oxygen and substrate demand and metabolite production on one hand and oxygen and substrate supply and metabolic washout on the other. Different techniques have been used to restore this balance. This can be done either by optimizing the hemodynamic situation or by reducing heart work, optimizing coronary flow and increasing diffusion in the ischemic tissue. Utilization of glucose consumes less oxygen than that of free fatty acids (FFA) per mole ATP produced. It is thus advantageous in the ischemic situation to increase glucose and decrease FFA utilization. Moreover, intermediary metabolites of fatty acid metabolism are accumulated in the ischemic myocardium and will negatively influence adenine nucleotide metabolism.

Hyaluronidase was already tried in the 1950's to limit infarct size. This method was further studied by Dr Maroko both in dogs and

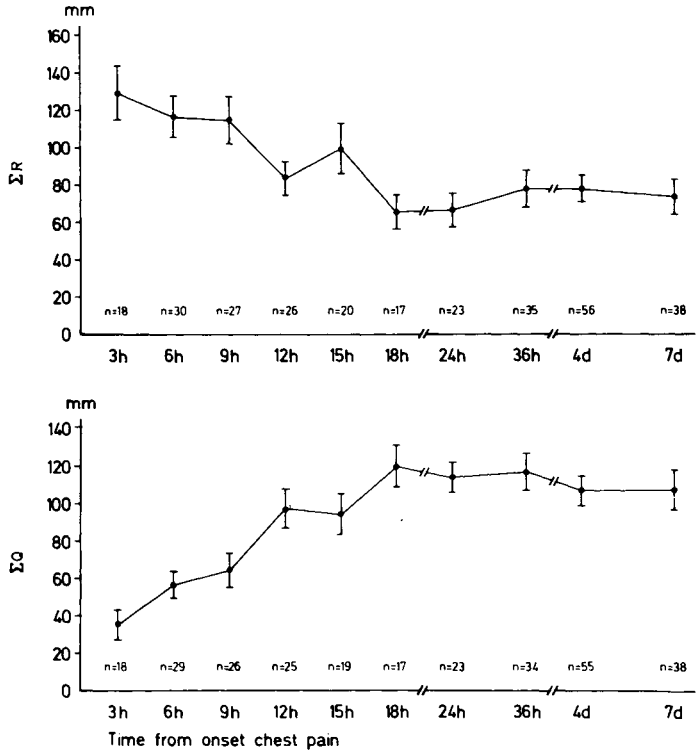

Fig. 1. Time course for reduction of ΣR (above) and elevation of ΣQ (below) in anterior myocardial wall infarction.[15]

man. In 1977 a randomized study of 91 patients with anterior infarction was presented.[18] Only patients with infarcts not older than 8 hours were included. The patients were treated with hyaluronidase

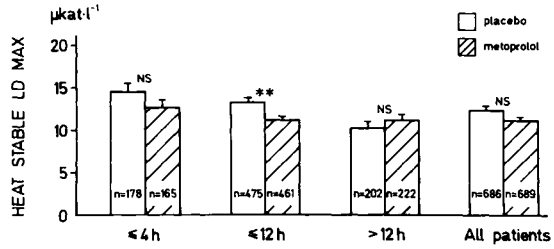

Fig. 2. Mean max heat stable LD activities in all patients and in those given blind injections within different time intervals after onset of pain. Missing data in 15 of the patients in whom enzyme values were obtained. *P < 0.05; **P < 0.01. Mean ± SEM.[15]

i.v. for only 48 hours. The ECG changes consistent with myocardial necrosis were reduced in the treatment group when a 35-lead precordial ECG mapping was performed.[18] In 1982 three studies were published in the same issue of the Lancet.[19-21] In a study by Saltissi et al.,[19] 79 patients with suspected acute myocardial infarction were randomly treated with placebo or i.v. hyaluronidase within 6 hours after onset of chest pain. Precordial electrocardiographic indices of infarct size (R-wave loss and Q-wave appearance) were reduced in the treatment group.[19] Flint et al.[20] demonstrated a reduction in mortality in 483 patients (Figure 3) at 6 months when all patients irrespective of trial diagnosis were analyzed. Henderson et al.[21] randomized 192 consecutive patients arriving within 12 hours after onset of symptoms suspected of myocardial infarction. Patients with definite acute myocardial infarction in the treatment group had significantly less QRS changes and development of Q-waves than those in the placebo group.

Positive effects of infusion of glucose-insulin-potassium (GIK) was first reported by Sodi-Pallares in 1962.[22] Rogers et al.[23] reported lower CK-B leakage in 23 GIK-treated patients when compared to 27 controls. At this congress the same authors reported the results of a larger series comprising 190 patients admitted within 12 hours after onset of chest pain. No effect on infarct size could be confirmed but there was a trend towards lower hospital mortality. The data on GIK infusion are thus not conclusive.

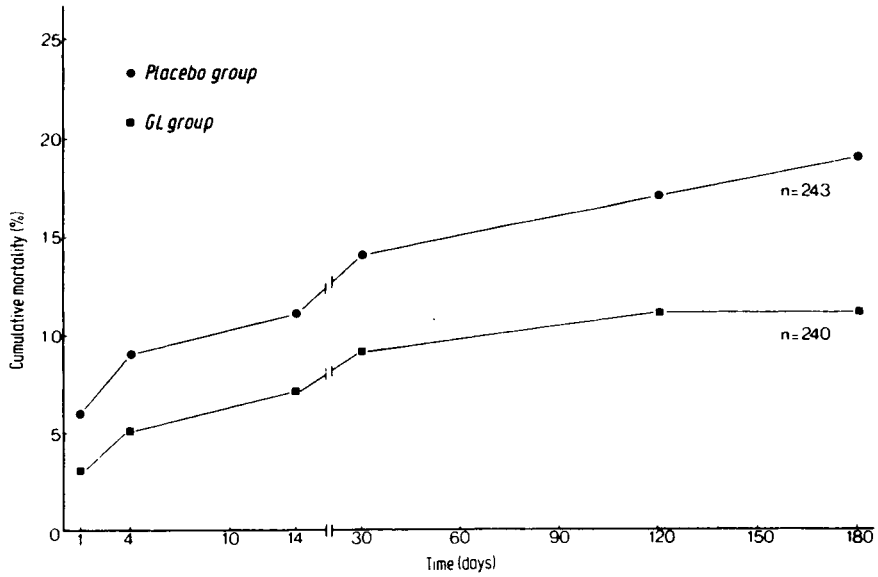

Fig. 3. Cumulative mortality over a 6-month period for all patients entered into the study.[20]

Table 2. Effect of Beta-pyridylcarbinol in AMI. Treatment within 6 Hours of Clinical Onset Cumulated CK (ukat/l)

Hrs	-1	0	2	6	24	48
Control (9)	1.9	4.4	10.4	19.7	40.2	43.4
Drug (9)	1.0	1.9	5.2	12.9	46.8	54.1

Reduced evolution of QRS infarct vector during the 15 hours of betapyridylcarbinol infusion when FFA were lowered. Overshoot after stop of infusion.

Kjekshus and Grøttum 1977

Methyprednisolone has proved effective in experimental myocardial infarction but when used in a clinical trial adverse effects with higher mortality in the treatment group was reported by Roberts and co-workers.[24]

During acute myocardial infarction serum levels of FFA are elevated. This might be deleterious for above mentioned reasons. In 1977 Kjekshus and Grøttum[25] published a study where patients were treated within 6 hours of onset of symptoms (Table 2). Beta-pyridylcarbinol - a lipid-lowering drug - was infused for 15 hours. CK release and ECG changes were reduced but after completion of infusion there was an overshoot of CK release and more marked ECG changes. The effect of this treatment thus seems to be controversial.

Lowering of myocardial work has also been done by nitroglycerine infusion. Derrida et al.[26] used this treatment in a series of 74 patients with acute myocardial infarction. In the 39 treated patients mortality was less than among the 35 controls. Also indices of infarct size, such as Q and R-waves were favourable in the treatment group.

In 1979 Bussman et al.[27] presented another prospective randomized trial. When nitroglycerine infusion was started within 8 hours after onset of chest pain a positive effect was shown. When treatment was started after 8 hours the signs of infarct size measured as CK leakage were less marked. This was, however, a small trial with only 15 patients.

In 1982 two trials were published in the same issue of New England Journal of Medicine on effects of nitroprusside treatment in acute myocardial infarction patients.[28,29] Durrer et al.[28] included 328 patients with ECG signs of acute myocardial infarction. One hundred and sixty-three patients were randomly allocated to nitroprusside treatment. Myocardial infarct size was estimated by the use of CK-MB levels and mortality was studied after one week.

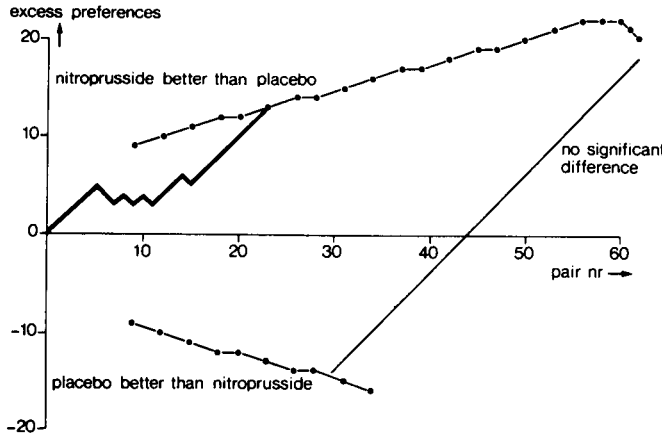

Fig. 4. Sample path (thick line) and predesignated boundaries (thin line with dots) in armitage closed sequential plan.[28]

These authors could demonstrate smaller infarcts and lower death rate (Figure 4)[28] in the treatment group. In a larger study by Cohn et al.[29] no effect on mortality could be seen after 13 weeks in the 812 patients included in the trial. This was also a double-blind placebo-controlled trial. Peak CK-B values indicated smaller infarcts in the treatment group irrespective of early or late treatment. The conflicting data of these trials are even more confusing as nitroprusside appeared to have a deleterious effect on mortality in patients treated early, although CK-B values tended to be lower in this group. Both trials were prospectively randomized, placebo-controlled and including appropriate trial number of patients. In the study by Cohn et al.[29] all patients were male with elevated left ventricular filling pressures and treatment was started later than in the study by Durrer et al.[28] where both sexes were included, but without any clinical signs of elevated filling pressures. As the two trials are not totally comparable, no firm conclusion can be drawn. Probably the treatment will not affect mortality when given late to patients in failure but nitroprusside may reduce mortality when given early to well compensated patients.

Positive effects of beta-blockers in patients with acute myocardial infarction was first reported by Snow in 1965.[30] Since then four randomized trials with beta-blockers for infarct size limitation have been published.[17,31-33].

In a small trial by Peter et al.,[31] 18 patients were treated with propranolol within 4 hours and compared to controls. CK-release was smaller in the treatment group. In another group treated after 12 hours after onset of chest pain, no difference in CK could be seen

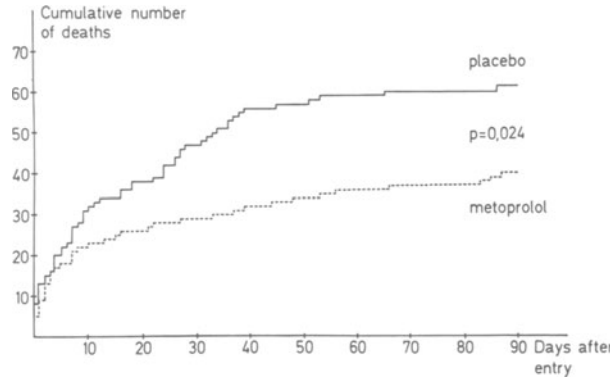

Fig. 5. Cumulative number of deaths in all patients randomly allocated to treatment with metoprolol and placebo. P-value is calculated according to Mantel-Haenzel.[17]

between the two groups. Alprenolol was used in a study from Copenhagen[32] where the patients were treated within 12 hours after onset of symptoms. A lower CK leakage was noted in the treatment group. One-year mortality was reported to be lower in the treatment group among patients $\leq$ 65 years of age. In patients > 65 years there was a trend towards a higher mortality in the treatment group[33] and the study was stopped prematurely in this age group.

In the Göteborg metoprolol trial,[17] 1395 patients were randomly allocated to placebo or beta-blockade. The patients were followed for 3 months. "Infarct size" was smaller when metoprolol was given $\leq$ 12 hours after onset of pain, as judged from enzyme curves and from Q and R-wave changes. The reduction in mortality was significantly lower after 3 months (Figure 5). In this study, the number of analgetic injections and need for furosemide were reduced among patients treated $\leq$ 12 hours in whom "infarct size" was limited. In a study by Sleight et al.,[34] including 504 patients with myocardial infarction, atenolol was found to limit "infarct size" measured as cumulative CK-B.

CONCLUSION

In Table 3 are listed regimes where a positive effect on infarct size has been reported. For most of the drugs listed the results are inconclusive. Beta-blockade seems to be the only drug today with reliable positive effects in acute myocardial infarction. Heart work is reduced and thereby oxygen consumption, lactate production is decreased, and supraventricular and ventricular arrhythmias are repressed. Even ventricular fibrillation is prevented by beta-blockade.

Table 3. Drugs with Documented Positive Effects on Infarct Size in Man.

o Beta blockade
o Hyaluronidase
o Nitroglycerin
o Nitroprusside
o Trimethaphan (in hypertensive patients)

The important question today is whether infarct size can be limited, and if so, which effects of beta-blockade are directly related to such limitations. Although there is a well documented reduction in mortality, the above mentioned question cannot be satisfactorily answered. There is good circumstantial evidence that "infarct size" is limited as CK and LD leakage is decreased, and this is true only for patients treated early. This should rule out the possibility that beta-blockade per se due to changes in washout and metabolism would alter the enzyme curve, which would otherwise be the case also for patients treated late. Another support for this idea is that less patients went into failure in the treatment group. It is reasonable to believe that this was because of salvage of muscle mass. ECG mapping also indicated smaller infarcts in treated patients. However, many questions remain to be answered: How much of the myocardium may maximally be jeopardized and therefore salvageable? Is this amount of tissue compatible with the clinical effects? There is an old observation that noradrenaline in the "normal" myocardium is depressed also when a coronary artery is ligated in a dog.[35] This situation will last for up to six weeks. Is this decrease of noradrenaline the reason for cardiac failure in acute myocardial infarction and is this loss of noradrenaline prevented by beta-blockade?

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SUBSTRATE EFFECTS IN MYOCARDIAL ISCHEMIA

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It is now just 20 years ago since Sodi-Pallares (1962) produced his provocative idea that myocardial ischemic damage could be minimized by replacing potassium loss from ischemic cells by a solution of glucose-insulin and potassium. Although his ideas are still being evaluated, it is interesting to see how even controversial concepts can lead to the evolution of new and important hypotheses.

EFFECTS OF EXTERNAL GLUCOSE

In the late 1960's and early 1970's two lines of reasoning linked substrate supply to the heart with the outcome of ischemia. First, the ideas of Sodi-Pallares were carried further by supposing that the critical factor was not replacement of potassium but the enhanced uptake of glucose. It was argued that, according to the Pasteur-effect, increased anaerobic energy provision should help protect the ischemic myocardium. In regional ischemia there tended to be a swing in the substrate metabolism of the ischemic zone from fatty acid to glucose; it is now known that such data were based on local venous sampling techniques which must have drained largely cells of only a modest severity of ischemia. The idea that increased glycolysis should benefit the ischemic myocardium received a major setback with the discovery of Neely's group (Rovetto et al., 1975) that glycolysis was inhibited at at least two levels (phosphofructokinase and glyceraldehyde-3-phosphate dehydrogenase activity) by the products of glycolytic flux, namely protons and lactate (Rovetto et al., 1975). It has even been thought that increase in glycolytic flux might damage the ischemic myocardium by accumulation of protons.

Yet the possible role of glycolysis in minimizing ischemic damage can by no means be ignored. There is, as yet, no true evi-

dence that increased glycolysis has worsened ischemia, and there is increasing evidence that increasing glycolysis might decrease ischemic damage in zones of less severe ischemia. Recently, Liedtke et al., (1982) have proposed that increased glycolytic flux might protect the ischemic myocardium chiefly by acting on the non-ischemic zone.

Abnormalities of glucose metabolism have been linked to electrophysical abnormalities early in myocardial infarction in an important article by Russell and Oliver (1979). A proposed and still speculative mechanism of action is that glucose can in some way, perhaps by provision of glycolytic ATP, help maintain action potential duration.

Whatever the result of conflicting experimental studies might be (compare for example Dalby et al., with Heng et al.,) two other points deserve emphasis:

1. In patients, administration of glucose-insulin potassium leads to decreased fatty acid levels, and on present evidence this can be seen as a beneficial effect on the myocardium in patients with acute myocardial infarction according to the randomized study of Rackley et al., (1979).
2. Administration of glucose to isolated rat hearts with constant circulating free fatty acid levels and with experimental infarction, can lessen fatty acid-induced damage (de Leiris et al., (1975).

FREE FATTY ACIDS AND ARRHYTHMIAS

Another very important and even more seminal idea was being evolved towards the end of the 1960's. Oliver et al., (1968) in Edinburgh showed that in patients with high blood-free fatty acids and myocardial infarction, the survival of these patients was limited when compared with controls. There are many explanations for the finding, of which probably the simplest is that patients with more severe infarctions have greater release of catecholamines with higher blood-free fatty acids. The question of "toxicity" of free fatty acids (Kurien et al., 1971) has been much debated with the present consensus of opinion seeming to favor the view that elevated free fatty acids in acute myocardial infarction are on the whole an undesired phenomenon.

The possible mechanisms of free fatty acid toxicity are manifold and include: (a) a direct damage of the cell membrane as suggested for example by increased rates of enzyme release; (b) increased intracellular metabolism with accumulation of acyl CoA and acyl carnitine; (c) an as yet unspecified mechanism whereby free fatty acids induce increased oxygen uptake and "oxygen wastage".

Recently, the focus has swung away from possible abnormalities induced by circulating free fatty acids to abnormalities in myocardial lipid metabolism induced even in isolated myocardial tissue by ischemia. The very stimulating and novel idea has been proposed that accumulation of certain specific lipids could induce electrophysiological abnormalities which in turn could promote arrhythmias. The work of Sobel and Corr (1978) is therefore the original Oliver-Kurien hypothesis updated.

PHOSPHOLIPIDS AND ARRHYTHMIAS

It now becomes necessary to evaluate whether the changes in myocardial phospholipids can actually cause arrhythmias. Katz (1982) has adopted a theoretical analysis suggesting that in certain specified circumstances, lysophospholipids can reverse some of the effects of membrane hydrolysis associated with ischemia. Katz's proposals, provocative as they be, are not necessarily in conflict with the proposals of Corr, Gross and Sobel (1982). Katz is essentially looking at the effects of ischemia on membrane structure and arguing that membranes may be better off with, than without, their liberated lysophospholipids. Corr et al., on the other hand are looking at the results of the liberated lysophospholipids on arrhythmogenesis.

COMMENT

The original concepts that circulating substrates could influence the outcome of myocardial ischemia have undergone considerable refining. Especially in the case of lipids, the focus is now on the myocardial cellular damage which is largely seen as the consequences of ischemia in altering lipid pathways in the ischemic zone. The early seminal ideas of Sodi-Pallares and Oliver have led to further investigations which are now focussing on the effect of myocardial ischemia on the cell membrane.

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PREVENTION OF CATECHOLAMINE-INDUCED MYOCARDIAL DAMAGE

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The improvement of prognosis of the acute myocardial infarction has mainly been due to detection and treatment of arrhythmias and congestive heart failure.

To further improve treatment of the acute stage of myocardial infarction we have to influence the myocardial infarction process. There is ample circumstantial evidence for this process being dynamic for at least 12 hours (Figure 1)[1]. which would be long enough for interventions to start. But to do this we have to know more about the factors of importance for this process. How important is platelet aggregation?[2] How important is the plaque per se? Is spasm important? This paper will deal with the possible role of catecholamines for initiation and expansion of the infarction process.

EXPERIMENTAL STUDIES

The cardiotoxic effects of catecholamines have been known since the beginning of this century[3,4], but it was not until the works of Raab[5] that this field was extensively investigated and the clinical relevance was discussed. In 1962 Raab and co-workers[5] could induce ST-changes of the ECG of anesthetized and vagotamized cats by stimulation of the sympathetic nervous system, or by intravenous injection of adrenaline or by anoxia. This could be only be demonstrated when a coronary artery dilatation was prevented by the use of a special restriction device. No ST-changes were shown when heart rate was increased by atrial pacing or blood pressure elevated by the use of angiotensin 11. Their theory was that ischemic attacks may start because certain areas of the myocardium with restricted coronary supply are made hypoxic by e.g. noradrenaline release.

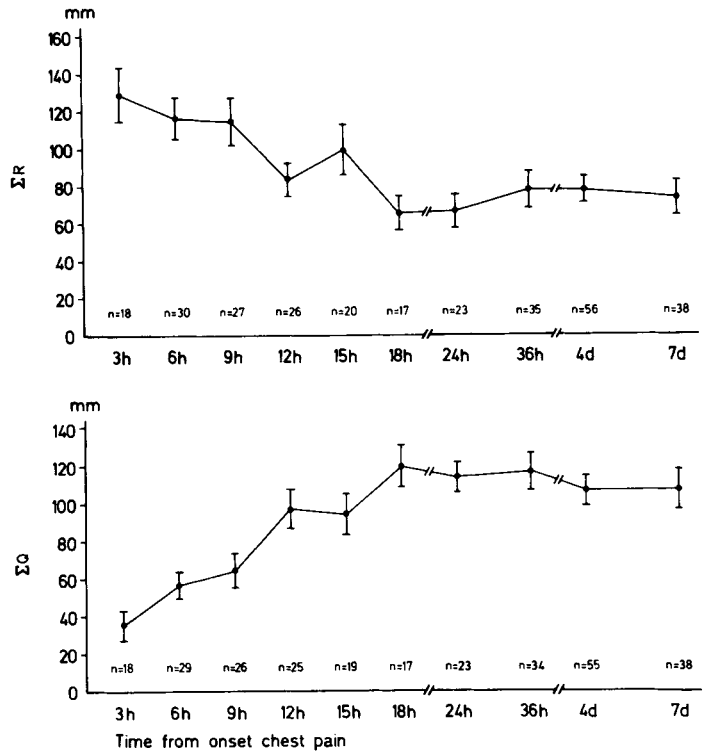

Fig. 1. Time course for reduction of $\sum R$ (above) and increase of $\sum Q$ (below) in anterior myocardial infarction.[1]

Rona et al[6] were able to demonstrate the cardiotoxic effects of isoproterenol when it was injected into healthy rats. In these animals myocardial necrosis appeared without any restriction in coronary flow. There are many possible explanations why catecholamines exert this damage. One possibility is that platelet aggregation is enhanced with subsequent thrombosis formation in a coronary artery.[2,7]

When isoproterenol is given to turtles, the response is somewhat different. The epicardial layer of the myocardium is supplied by vessels but the inner layer is spongy with lacunar supply. Isoproterenol induces necrosis preferentially in the spongy part which suggests that a vascular mechanism is most important.[8] When large doses are used ventricular aneurysms develop. This could, however, be prevented by keeping the animals at +4°C.[9]

During the 1970's many studies were made in order to find factors that changed myocardial infarct size. Thus it was found that isoprenaline infused to a dog subjected to coronary ligation would increase the zone of ischemia (Figure 2)[10] whereas beta-blockade would decrease infarct size.[11,12]

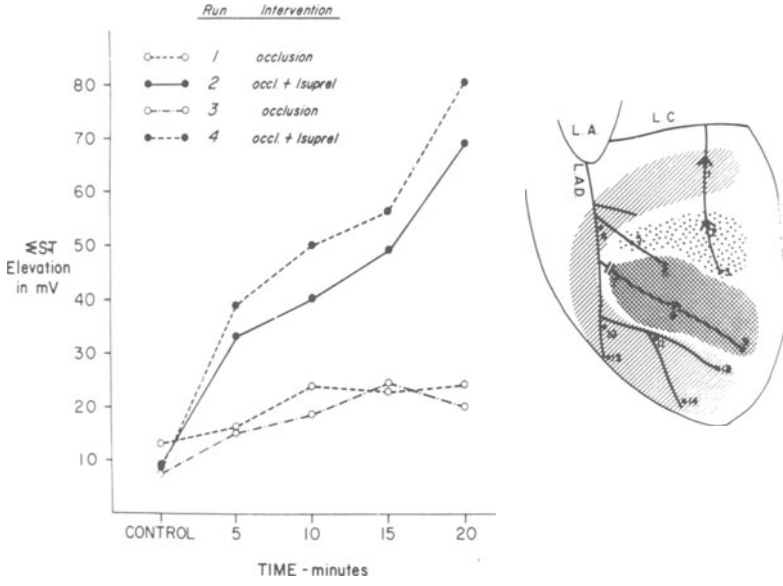

Fig. 2. Effects of occlusion alone and occlusion after the infusion of isoproterenol (0.25 ug/kg/min). Right panel: Schematic representation of the anterior surface of the heart. The coronary arteries and branches, and sites of epicardial electrograms are marked. LAD = left anterior descending coronary artery; LA = left atrial appendage; LC = left circumflex coronary artery; Cross-hatched area: area of injury after 15 minutes of occlusion. Stippled area: increase of area of injury when the occlusion was performed under the influence of isoproterenol. Lined area: area that showed no ST segment elevation under any circumstances. Left panel: ΣST in the same experiment after three simple occlusions and after two occlusions under the influence of isoproterenol. Time = minutes after occlusion.[10]

The concept that acute myocardial infarction is caused by coronary artery thrombosis or occlusion has been debated lately.[13-17] Different types of cellular lesions in acute myocardial infarction

were described by Baroldi.[17] One type, coagulative myocytolysis, is mainly seen in the border zone of an acute myocardial infarction but also in patients with pheochromocytoma.[18] It is thus reasonable to believe that this type of lesion is induced by catecholamines. In the light of these observations we started our investigations in isolated rat hearts. When the hearts were perfused with increasing concentrations of adrenaline in the perfusion buffer, a myocardial damage could be demonstrated (Figure 3) as a release of creatine phosphokinase (CK) and aspartate aminotransferase (ASAT) to the buffer and as typical histological changes.

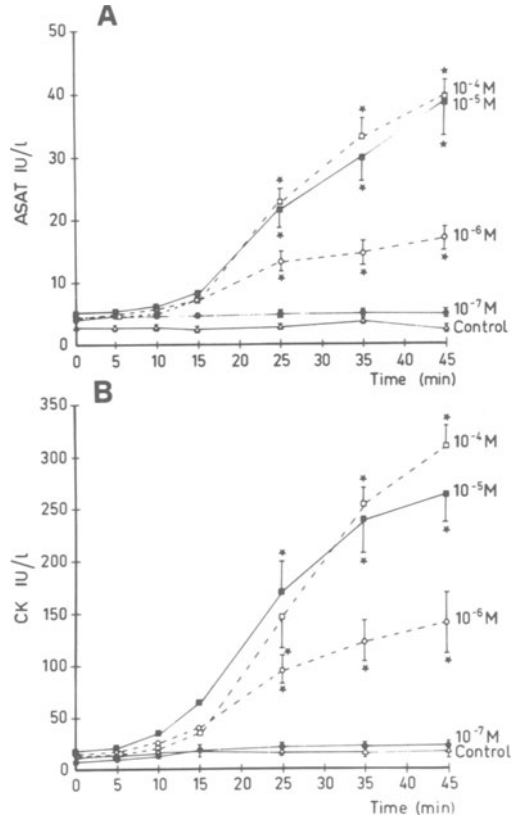

Fig. 3. A (upper panel): Release of ASAT from the hearts to the perfusion medium when perfused with different concentrations of noradrenaline. After 15 minutes of anterograde perfusion 30 minutes of retrograde perfusion followed.
B (lower panel): Release of CK under the same conditions as in panel A. Each value represents the mean ± SE for 6-8 hearts. x = significantly different from initial value ($p<0.05$).[19]

A concentration of noradrenaline of 10^{-6} M seemed to be needed to induce necrosis in this experimental model. And this is a concentration not unlikely to occur in real life. To test this hypothesis further, the noradrenaline stored in myocardial nerve endings was released by adding tyramine to isolated perfused hearts. Again a damage was induced with enzyme leakage and histological changes. This damage could be prevented by beta-blockade (Figure 4)[19] It could be shown that myocardial contents of noradrenaline, if extensively released, would induce infarct-like lesions and these lesions could be prevented by beta-blockade as well as by the calcium antagonist verapamil. More studies on the protective effects of calcium antagonists in ischemia have been performed by Nayler et al[20] where isolated perfused rabbit hearts were used. During ischemia ATP and CK levels were much better preserved in verapamil-treated animals.

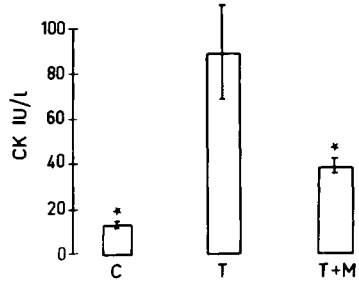

Fig. 4. Release of CK from hearts perfused anterogradely for 60 minutes with tyramine 250 ug/ml (T) with or without metoprolol 10^{-3}M (M). Controls are perfused without addition of tyramine or metoprolol. Each bar represents the mean ± SE for 6-8 hearts. x = significantly different from tyramine-treated hearts ($p < 0.05$).[19]

Locally released catecholamines may thus induce myocardial damage and under very special conditions, such as pheochromocytoma, circulating catecholamines may produce myocardial necrosis as well. Stress is also a "well known" important etiological factor for myocardial infarction but real proof is scarce. Some very interesting experiments were performed in the early 1960's by Melville and coworkers[21]. Based on the fact that patients with cerebrovascular accidents may show ECG changes indicative of myocardial ischemia or infarction they electrically stimulated hypothalamus of anesthetized cats. By doing so they could induce a rise in blood pressure, ST-T deviations on the ECG and ventricular arrhythmias including ventricular tachycardia.[21] The same authors could also show that injections of picrotoxin into the lateral cerebral ventricle of rabbits and cats evoked intense sympathetic cardiovascular effects. It is well known that domestic pigs sometimes die spontaneously during transport to the slaughter house. The hearts of such animals often show myo-

cardial damage from microscopical necrosis to macroscopical visible areas of necrosis. And yet these animals are without signs of coronary atherosclerosis or thrombosis. In some very interesting experiments, Johansson et al[22,23] subjected pigs to restraint stress. This was performed by i.v. infusion of succinylcholine chloride resulting in paralysis for 12 minutes. Respiratory insufficiency was avoided.

The amygdaloid part of the limbic system is of importance in eliciting emotions of fear and aggression. Stimulation of this area can cause flight and fight behaviour whereas amygdalectomy suppresses such behaviour. When restraint stress was induced in control pigs as well as in amygdalectomized ones, myocardial degeneration was found in all controls. No myocardial changes were found in the operated animals. These changes correlated to plasma catecholamines so that amygdalectomized animals had significantly lower levels than controls.[23] As this stress-induced myocardial necrosis seems to be mediated via catecholamines, the same authors studied the protective effect of propranolol. Interestingly, these authors find that low i.v. doses of propranolol will not prevent from damage. A very high dose of i.v. propranolol will protect the myocardium, but in this case no increase in catecholamine release could be seen. Only with oral treatment with clinical doses for a week stress-induced necrosis could be totally prevented even though catecholamine concentrations in blood were elevated (Table 1).[24] Maybe long-term treatment modifies the myocardium and/or beta-receptors in a way that makes it more efficient than acute beta-blockade! These mechanisms are complicated and still obscure. We have some recent data on selective beta-perfused rat hearts where contractility seems to be more sensitive than chronotrophy.[25]

CLINICAL TRIALS

The cardiotoxic effects of catecholamines are now experimentally well established. Possibly a clinical infarct could start by a sudden release of noradrenaline in an area of restricted perfusion/relative ischemia. A vicious circle may start ending up in myocardial necrosis. During this stress, catecholamines are peripherally released accentuating the already compromised heart and the infarct may increase. Following this hypothesis, the Göteborg metoprolol trial[26] was designed in the mid 70's. Four well designed studies have now been published with positive effects on mortality using metoprolol[26], timolol[27], alprenolol[28] and propranolol.[29] It seems clear that beta-blockers reduce mortality in acute myocardial infarction, and if used early data suggest that myocardial infarct size is restricted as evidenced by ECG mapping and enzyme curves. These findings have been found mainly in patients who were treated within 12 hours after onset of chest pain.[30] It is very reasonable to assume that the positive effect of beta-blockade in acute myocardial infarction is due to blockade of toxic effects of catecholamines.

Table 1. Effect of various pretreatments with propranolol on extent of heart lesions after stress

Different groups of pigs		Propranolol pretreatment	Total number of pigs	Number of pigs with myocardial cell necrosis graded according to point scale 0	1	2	3	4	5	6	Statistical sign*
Controls A		-	5		1		1	2	1		
Controls B		-	3				1	2			
	acute	1 mg/kg i.v., single dose	3				2	1			n.s.
		3 mg/kg i.v., single dose	3					1	2		n.s.
		10 mg/kg i.v., single dose	5	1	1	3					p<0.02
	long-term	120 mg orally, 3x6 days	3	2	1						p<0.005
		120 mg orally, 3x6 days	3	2	1						

*The myocardial changes in the different groups were evaluated statistically versus those in the controls (A+B) by the Mann-Whitney U-test (n.s. = non-significant).

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MAINTENANCE OF Ca^{2+} HOMEOSTASIS IN THE MYOCARDIUM

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SUMMARY

Maintenance of intracellular homeostasis with respect to Ca^{++} during prolonged episodes of ischaemia and upon reperfusion is critical to the survival of the myocardium. Loss of Ca^{++} homeostasis results in massive ultrastructural damage, associated with excessive phospholipase and proteinase activation. Aggravating factors include hyperthermia, excessive cardiac work and hyperthyroidism. Conversely hypothermia, a reduction in sympathetic drive and a diminution in work load are all protective. Since the release of intracellular constituents, including enzymes and myoglobin, into the extracellular phase during post ischaemic reperfusion occurs as a secondary response to the Ca^{2+}-induced tissue damage the quantitation of protective procedures that are based simply on the measurement of serum enzymes may not provide an accurate assessment of the degree of protection that has been achieved. In some instances it is possible to dissociate the gain in tissue Ca^{2+} from the release of intracellular constituents into the extracellular phase.

Recently there has been a rapid growth of ideas concerning the involvement of Ca^{2+} in the progression of events that are precipitated by an ischemic episode and which become exacerbated upon reperfusion. Shen and Jennings (1972) first noted a possible association between the occurrence of the tissue damage which occurs under these conditions and the massive increase in Ca^{2+}. However measurements of total tissue Ca^{2+} provide little information as to why ischemic heart muscle loses its capacity to maintain ionic homeostasis with respect to Ca^{2+}. Loss

of ionic homeostasis with respect to Ca^{2+} is not the prerogative of hearts that have been made ischemic and then reperfused. Thus, for example, the administration of large doses of isoproterenol (Fleckenstein, 1971) and the re-introduction of Ca^{2+} after a relatively brief period of Ca^{2+}-free perfusion (Zimmerman and Hulsman, 1966) both cause mammalian heart muscle to become overloaded with Ca^{2+}. The consequences of a raised tissue Ca^{2+} are far reaching and include irreversible loss of mitochondrial function (Nayler, Ferrari and Williams, 1980; Nayler, 1982), massive ultrastructural damage, ATP wastage, a raised end diastolic resting tension (Nayler, Poole-Wilson and Williams, 1979) and the development of membrane defects (Jennings and Reimer, 1981). Possibly these membrane defects reflect the activation of the phospholipases, some of which are under calmodulin-Ca^{2+} control. Despite its importance comparatively little is known about the precise sequence of events that ultimately results in the reperfused ischemic heart becoming overloaded with Ca^{2+}. Probably several distinct phases are involved.

Initially there may be a small modest gain in Ca^{2+} due, possibly to an enhanced entry of Ca^{2+} in exchange for Na^{+} (Figure 1). The aetiology of this may be as follows:- as the result of an ATP-depletion induced failure of the $Na^{+}K^{+}$ ATPase, the ischemic myocardium fails to extrude Na^{+} against the prevailing concentration gradient. As a result the tissue Na^{+} rises (Regan, Broisman, Haider, Eaddy and Oldewurtel, 1980). Under these circumstances we can expect to see a relatively short lived activation of the Na^{+}: Ca^{2+} exchange mechanism so that Na^{+} will leave the tissue in exchange for Ca^{2+} (Reuter, 1974). However the amount of Ca^{2+} which can enter in this way will be limited by at least two factors:-

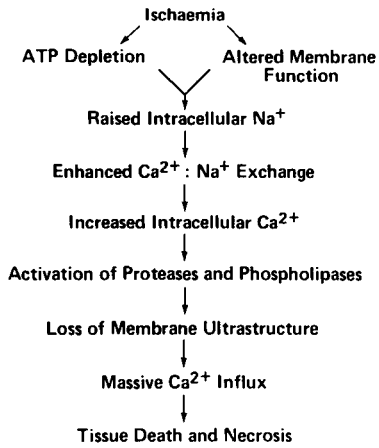

Fig. 1. Schematic representation of the events involved in the post-ischemic reperfusion-induced gain in tissue Ca^{2+}.

Table 1. Effect of H^+ accumulation on the Na^+-Ca^{2+}-exchange reaction in isolated cardiac sarcolemma.

pH	Ca^{2+} accumulated in exchange for Na^+ (nmoles/mg protein/min)
7.4	2.2
7.0	1.2
6.6	1.1

These results were obtained using isolated fragments of cardiac sarcolemma from normotensive Sprague Dawley rats.

(a) the amount of Na^+ available as an antiporter, and
(b) the inhibitory effect of the protons that accumulate under these conditions (Gevers, 1977, Table 1).

The early net gain in Ca^{2+} probably does not involve the entry of Ca^{2+} through sarcolemmal defects (Ashraf, White and Bloor, 1978) even although this may take place later (Jennings and Reimer, 1981). An excess entry of Ca^{2+} through the slow channels is also unlikely, because these channels require energy in the form of ATP for their maintenance (Sperelakis and Schneider, 1976).

Probably this early net gain in Ca^{2+} is accompanied, or even preceded, by another event - that is a redistribution of tissue Ca^{2+}. There are several reasons for believing that this may occur. Firstly energy (as ATP) is required for the correct functioning of the sarcoplasmic reticulum. Hence as the availability of ATP declines Ca^{2+} will remain in the cytosol where presumably it will remain free to activate the Ca^{2+}-dependent ATPases. Under these conditions, and because of the switch to anaerobic glycolysis, cytosol H^+ will rise. Tissue acidosis favours the process whereby Ca^{2+} induces a spontaneous release of Ca^{2+} from the sarcoplasmic reticulum (Fabiato and Fabiato. 1977; Dunnett and Nayler, 1978). Therefore, as the tissue becomes increasingly acidotic Ca^{2+} will tend to leave the sarcoplasmic reticulum, to remain in the cytosol. Some Ca^{2+} may also be displaced from the mitochondria, because a raised tissue Na^+ (Carafoli, Trozzo, Lugli, Grovetti and Kratzing, 1974) promoted a spontaneous release of Ca^{2+} from these organelles.

We may ask ourselves why this Ca^{2+} is not immediately extruded from the tissue. There are three possible explanations:-

(a) the prevailing concentration gradient will favour the entry and not the exit of Ca^{2+};

(b) the Na^+: Ca^{2+} exchange mechanism will be functioning in the opposite direction; and
(c) the Ca^{2+}-stimulated plasmalemmal-located Ca^{2+} pump will be failing because of ATP depletion.

In summary, therefore, early during the ischemic event there may be a modest gain in tissue Ca^{2+} and a large rise in cytosolic Ca^{2+} due to the intracellular redistribution of these ions. The progression of events does not stop here, however, because as the duration of the ischemia continues the cytosolic Ca^{2+} may reach the levels needed to activate phospholipase A_2 (Chien, Reeves, Buja, Bonte, Parkey and Willerson, 1981). Under these conditions membrane structure and function will be severely impaired, with large membrane defects appearing (Jennings and Reimer, 1981). Consequently Ca^{2+} will be free to flow along its concentration gradient, causing a massive and explosive rise in tissue Ca^{2+} (Figure 1). Some of this Ca^{2+} is rapidly accumulated by the mitochondria (Nayler, Ferrari and Williams, 1980) which are then rendered incapable of rephosphorylating adenosine diphosphate. Moreover, the large membrane defects permit the loss of the precursors of adenosine triphosphate (adenosine and its breakdown products, including hypoxanthine and inosine) from the cell.

One of the problems encountered when trying to quantitate the degree of protection that a particular regime provides involves the problem of assessing the damage in a quantitative manner. Measurements of protein release (or intracellular enzymes, including creatine kinase) have often been used but recent data indicates that this technique may give misleading results. Thus agents which cause the sarcolemma of heart muscle cells to remain intact may not prevent Ca^{2+} ions from entering. Presumably a small membrane defect will allow these ions to enter whereas a large hole may be needed for the exit of these larger macromolecules. Possibly the recovery of mechanical function upon reperfusion provides a better assessment of the effectiveness of a proposed protective regime, or of the deleterious consequences of other interventions.

Using this approach we have begun to determine some of the factors that influence the sequence of events that are precipitated by an ischemic episode. Pre-existing hypertension (Table 2) has been found to cause an exacerbation of the gain in tissue Ca^{2+} that occurs during post ischemic reperfusion, and this is accompanied by an excessive gain in Ca^{2+}.

That there is a positive correlation between the gain in tissue Ca^{2+} that occurs upon reperfusion and the recovery of mechanical function is shown by the data summarized in Table 3. Hyperthyroidism, and hyperthermia cause an exacerbation of this gain in Ca^{2+}; hypothermia and the administration of the Ca^{2+} antagonists protect against it.

Another interesting correlation which emerged during these studies

Table 2. Effect of Hypertension on the Post-ischemic induced gain in tissue Ca^{2+}, and the recovery of mechanical function.

Rat Model	Mean BP (mm Hg)	% Recovery	Gain in Tissue Ca^{2+} (μmoles/gm dry wt)
Normotensive			
Sprague Dawley	112.3 ± 3.2	32.6 ± 3.2	34.3 ± 5.5
Wistar Kyoto	138.3 ± 6.12	29.8 ± 4.6	38.2 ± 4.8
Hypertensive			
S.H.R.	212.5 ± 6.1	15.8 ± 1.6	51.4 ± 3.6
Stroke prone	204.2 ± 5.4	12.9 ± 2.2	55.9 ± 6.8

Each result is mean ± S.E. of 6 experiments. Gain in tissue Ca^{2+} refers to the gain during 20 minute's reperfusion after 60 minutes normothermic global ischemia.

ated to the relationship between the recovery of mechanical function upon reperfusion and the increase in mitochondrial Ca^{2+}. Thus as Figure 2 shows there is a linear but inverse relationship between recovery of mechanical function and mitochondrial Ca^{2+} overload. If we remember, now, that cardiac mitochondria that are overloaded with Ca^{2+} fail to rephosphorylate ADP (Nayler, Ferrari and Williams, 1980) then we can begin to understand why, when we plot percentage recovery of mechanical function during post ischemic reperfusion and the ATP content of the heart after the required period of post ischemic reperfusion - in this case twenty minutes, a straight line relationship (Figure 3) is found.

In summary, therefore, these results show that conditions which favour the recovery of mammalian hearts upon post-ischemic reperfusion prevent the heart from accumulating large amounts of Ca^{2+}. Since the initial gain in Ca^{2+}, or rise in cytosolic Ca^{2+}, may reflect an ATP-induced failure to maintain ionic homeostasis with respect to Ca^{2+}, protective procedures should, perhaps, be aimed at preserving the tissue levels of ATP. Alternatively we should begin to explore the possibility of protecting the heart against the consequences of a raised Ca^{2+}, perhaps by searching for drugs which will decrease the Ca^{2+}-binding activity of calmodulin, for the activation of phospholipase A_2 by Ca^{2+} is modulated by calmodulin.

Table 3. Correlation between gain in tissue Ca^{2+} and percentage recovery of mechanical function upon reperfusion.

% Recovery of mechanical function	Gain in tissue Ca^{2+} (μmoles/gm dry wt)
92 ± 3	1.8 ± 0.6
44 ± 4	5.2 ± 1.3
10 ± 3	9.6 ± 2.1

These results were obtained from rabbit hearts that were made globally ischemic for 60 minutes at 37°C and then reperfused for 20 minutes. Each result is mean ± S.E. of 6 experiments.

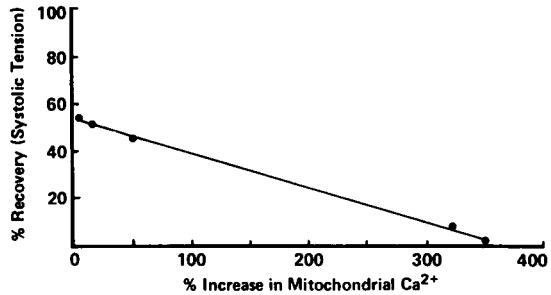

Fig. 2. Relationship between the recovery of mechanical function during post-ischemic reperfusion and the gain in mitochondrial Ca^{2+} expressed as a percentage change relative to the Ca^{2+} content of mitochondria from aerobically perfused hearts. Rabbit hearts: perfusion temp 37°C; perfusion buffer Krebs-Henseleit (Nayler, Ferrari and Williams, 1980).

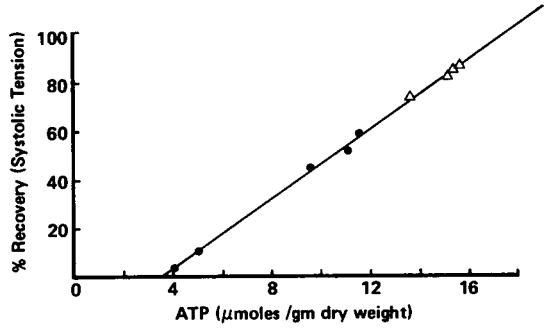

Fig. 3. Relationship between percentage recovery in mechanical function and ATP content of left ventricular heart muscle in rabbits made ischemic at 37°C for 60-90 minutes and then reperfused for 15-20 minutes. For perfusion details see Nayler, Ferrari and Williams, 1980.

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THEORETICAL IMPLICATIONS OF THE USE OF ANTIOXIDANTS FOR HEART PROTECTION AGAINST STRESS-INDUCED AND ISCHEMIC DAMAGES

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Ischemia, stress-induced catecholamine excess and, most commonly, a combination of these two factors are the key links in the pathogenesis of the coronary disease and other damages of the heart muscle. The experimental data obtained independently on models of acute ischemia (Meerson, Kagan et al., 1982; Vasdev et al., 1979; Chien et al., 1979) and emotional-painful stress (Meerson, Kagan et al., 1980; Meerson, Arkhipenko et al., 1981) suggest that the crucial role in the damaging action of these factors, in particular in transition of reversible damages into irreversible ones, belongs to the events occurring at the level of the membrane lipid bilayer of cardiomyocytes. The excessive activation of at least three physiologically significant factors is of primary importance here, namely the activation of lipases and phospholipases, the activation of lipid peroxidation and the detergent-like action of lysophospholipids and free fatty acids (Katz and Messineo, 1981).

These three processes are closely interrelated both in a healthy organism and under ischemic and stress damage of the heart; therefore they have been termed by us as a "lipid triad" of modification or of damage of biomembranes (Meerson, Arkhipenko et al., 1981).

In the present paper our attention will mainly be focused on the role of lipid peroxidation. It will be demonstrated that excessive activation of this process is an essential link in the pathogenesis of stress-induced and ischemic damages of the heart and can be prevented or limited by lipid peroxidation inhibitors - antioxidants.

The scheme on Figure 1 shows that there exist two pathways of oxygen utilization in the organism. The first of them, the well-known oxidase pathway, is related with oxidation of energy substrates;

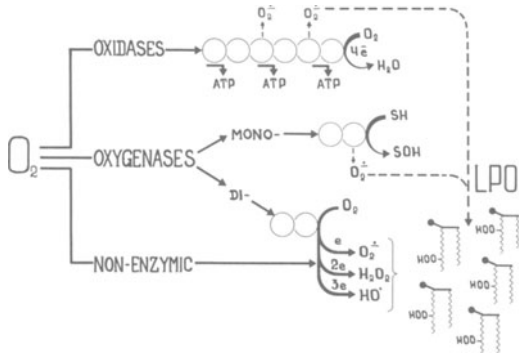

Fig. 1. Two pathways of oxygen utilization and lipid peroxidation. Explanation see in the text.

this pathway is realized by cytochrome oxidase, the terminal component of the respiratory chain. In the course of the oxidase reaction oxygen accepts four electrons; such a four-electron reduction of O_2 eventually results in a formation of a H_2O molecule. This pathway does not involve oxygen incorporation into the oxidized substrate molecule; under normal conditions it is coupled with a resynthesis of ATP, thus being the main energy source in living systems.

The other, oxygenase pathway involves incorporation of one or two oxygen atoms into the substrate molecule. The system of microsomal oxidation containing cytochrome P-450 can serve as a most typical and common example of this pathway. Such oxygenase reactions do not result in a complete reduction of O_2 to form water but give rise to a formation of activated oxygen species, i.e. superoxide anion radical, hydroperoxide, and hydroxyl radicals. It is essential that the formation of these activated oxygen species can also take place in the mitochondria by donating one electron to oxygen during electron transport to cytochrome oxidase. The activated O_2 species are capable of interacting with endogenous substrates, the structural components of the cell and, in the first place, with biomembrane phospholipids. The free radical oxidation of lipids gives rise to the so-called peroxide compounds; therefore the overall process has been termed lipid peroxidation, or LPO (Fridovich, 1974).

The results in Table 1 show that the heart muscle of intact animals always contains intermediate products of lipid peroxidation, such as lipid hydroperoxides, and LPO end products, namely Schiff's bases. Simultaneously we can observe a rather high activity of the antioxidant enzymatic systems, superoxide dismutase and catalase, which restrict the LPO process. It is essential that the activity of the antioxidant systems in the left ventricle myocardium is higher and the content of LPO products is lower than in the right ventricle myocardium.

Table 1. Activity of antioxidant systems and content of LPO products in heart ventricles.

	Right ventricle (10)	Left ventricle (10)
Superoxide dysmutase, conv. unit/mg.min	218±21	333±25
Catalase, nmol H_2O_2/mg.min	256±16	475±25
Fluorescence of Schiff's bases, rel. unit	1.64±0.12	1.00±0.15
Lipid hydroperoxides, opt. dens. unit/1 mg lipids in 1 ml.	1.49±0.2	1.03±0.13

Figures in parenthesis mean numbers of animals

Thus in normal heart muscle the lipid peroxidation process occurs continuously and its intensity is inversely dependent on the activity of natural antioxidant systems.

There are at least six factors known by now which can facilitate the switch-over of oxygen utilization from the oxidase pathway to the oxygenase one; they are the following:

1. Excess of catecholamines and products of their incomplete oxidation in stress (Kogan et al., 1976; Bors et al., 1978; Dilberto and Allen, 1981).

2. Excess of reduced pyridine nucleotides, the electrone donators in ischemia, hypoxia, and reoxygenation (Fridovich, 1974).

3. Increase in tension of oxygen, the electrone acceptor in hyperbaric oxygenation (Fridovich, 1974; Krachevskaya et al., 1980).

4. Inactivation of enzymic and non-enzymic antioxidant systems in avitaminosis E and other diseases (Arkhipenko et al., 1976).

5. Accumulation of unsaturated polyenic lipids attacked by activated oxygen species in obesity (Trostler et al., 1979).

6. Accumulation of metal-containing complexes with variable valency in hemolytic anemia and other diseases (Rachmilewitz et al., 1976).

Of primary interest here are the first four factors, namely the stress-induced catecholamine excess, ischemia and reoxygenation,

hyperbaric oxygenation and vitamin E deficiency, since in these situations the activation of lipid peroxidation in heart muscle does take place and the role of this activation in cardiomyocyte damage has been proved.

The scheme on Figure 2 shows two possibilities responsible for LPO activation by catecholamine excess and for the activation of its biosynthesis in stress (Bors et al., 1978; Dilberto, 1981). First, the activated oxygen species are formed at certain steps of catecholamine biosynthesis and second, epinephrine oxidation to adrenochrome gives rise to a semiquinone radical which can donate electrons to oxygen, thus generating the superoxide radical, an important LPO inducer.

In full agreement with these assumptions is the finding that the excitation of adrenergic regulation in emotional-painful stress reproduced as an "neurosis of anxiety" causes LPO activation in heart muscle and in other organs (Meerson, Kagan et al., 1980).

It can be seen from the Table 2 that in the myocardium of stress-exposed animals the content of intermediate and end products of LPO is increased more than 3-fold. It appears also that the stress-induced activation of the lipid peroxidation can largely be prevented by injecting ionol (2,6 ditertbutyl, 4 methyl phenol), a synthetic antioxidant of a phenolic type prior to stress exposure (Meerson, Golubeva et al., 1980). Further investigations have demonstrated that beside LPO activation prevention ionol and other LPO inhibitors prevent the stress damages of the heart and other organs (Meerson, 1981). The results obtained in our laboratory evidence that the stress-induced disturbances of metabolism such as a decreased activity of sarcoplasmic reticulum Ca-pump (Meerson, Arkhipenko et al., 1981) a lowered content of glycogen (Meerson et al., 1978), uncoupling between oxidation and phosphorylation in mitochondria (Meerson, Malyshev et

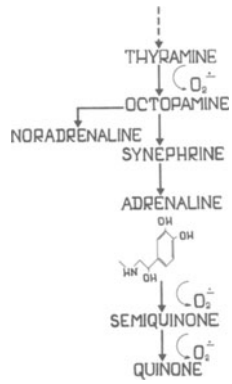

Fig. 2. Scheme of activation of lipid peroxidation by catecholamines. Explanation see in the text.

Table 2. Effect of ionol on LPO in myocardium in stress.

Series	Lipid hydroperoxides, opt. dens. unit/1 mg lipids in 1 ml.	Fluorescence of Schiff's bases, rel. units
1. Control (11)	1.08±0.13	1.00±0.33
2. Stress (10)	2.53±0.14	2.64±0.40
3. Ionol (10)	1.08±0.10	1.01±0.20
4. Ionol-stress (8)	1.71±0.14	1.26±0.17
Significance of differences, p.	p_{1-2} < 0.001	< 0.001
	p_{1-3} non-significant	non-significant
	p_{1-4} > 0.05	> 0.05

Figures in parenthesis mean number of animals

al., 1982), labilization of lysosomes (Meerson, 1981), and reversible damages of DNA (Meerson and Vasiliev, 1982) are partly or completely prevented by the LPO inhibitor ionol. In a similar way this antioxidant prevents other damaging effects caused by stress, namely the loss of enzymes by the myocardium (Meerson, Golubeva et al., 1980), an appearance of small-focal contractural and necrotic lesions (Meerson, 1981), a depression of the contractile function (Meerson and Trikhpoeva, 1980), and, finally the post-stress decrease of heart resistance to Ca^{2+} excess and hypoxia (Meerson, Golubeva et al., 1980).

Figure 3 shows the dynamics of the well-known event that is, hypoxic contracture of the myocardium. It can be seen that in the myocardium of rats exposed to stress the hypoxia-induced contracture is at least two times as high as in the control. A preliminary injection of ionol completely prevents the post-stress decrease of myocardium resistance to hypoxia.

It may thus be concluded that LPO activation which inevitably occurs in heart muscle upon stress is a key link in the stress damage of the heart and can therefore be prevented by antioxidants.

In terms of clinical cardiology it is essential that the observed changes in the metabolism and in the structure and function of the heart are not eliminated after cessation of the stress reaction. These relatively stable changes may be accumulated from one stress episode to another and become involved in a gradual development of cardiosclerosis and chronic heart insufficiency, which take place in aged individuals who have not suffered from the circulatory diseases before. The antioxidants prevent stress damages of the heart and, consequently, are cardioprotectors, the use of which can be a promising tool for prevention of cardiosclerosis and chronic heart insufficiency.

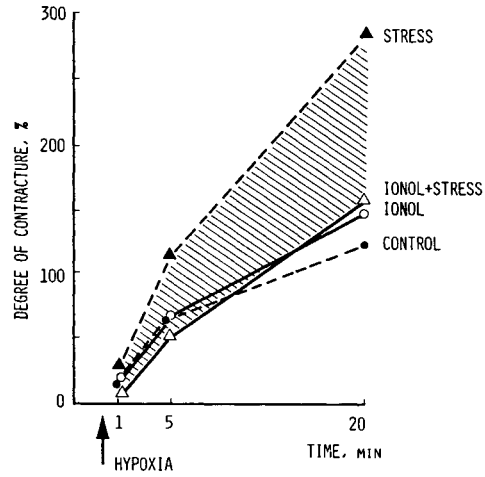

Fig. 3. Post-stress potentiation of the myocardium hypoxic contractur and prevention of this phenomenon by the antioxidant ionol.

When studying the role of lipid peroxidation in the pathogenesis of ischemic heart damage and the possibility of antioxidant protection of the heart against such damage, the investigator is immediately face with logical handicaps, since ischemic hypoxia is known to decrease oxygen tension necessary for LPO activation. However, a more detailed analysis shows that under "real organisms" conditions hypoxia and, in a greater degree, reoxygenation may induce LPO activation.

The scheme on Figure 4 shows that the four-electron reduction of oxygen on cytochrome oxidase can occur only upon continuous and well-coordinated functioning of the respiratory chain, that is, under conditions of normal oxygen tension. When the respiratory chain is blocked at its terminal component, i.e. in ischemic hypoxia, an inevitable reduction of NAD to NADH and a reduction of the respiratory chain carriers may cause a reduction of molecular oxygen dissolved in the membrane lipid matrix. However, this will not be a four-electron reduction to an end product, H_2O, but an incomplete reduction coupled with generation of activated oxygen species. Naturally under oxygen deficiency the accumulation of its activated species and LPO activatic are considerably limited and occur at a slow rate. A far more intensive production of activated oxygen species and LPO products occurs when the accumulation of reduced respiratory chain carriers, electron donors, is accompanied by a sufficiently high oxygen tension. This situation is observed during reoxygenation after hypoxia and ischemia, that is, in a situation apparently accompanying each coronary attack under conditions of natural reperfusion of primarily ischemic zones of the myocardium. A similar situation can be reproduced experimenta by restoration of oxygen transport to anoxic heart.

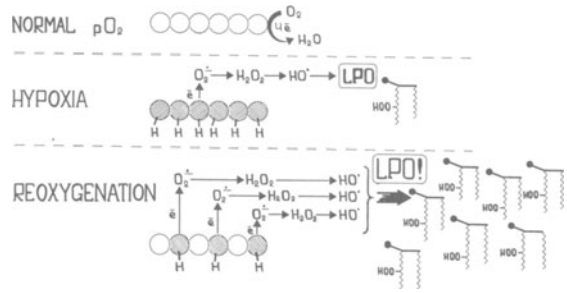

Fig. 4. Activation of lipid peroxidation in hypoxia and reoxygenation. Explanation see in the text.

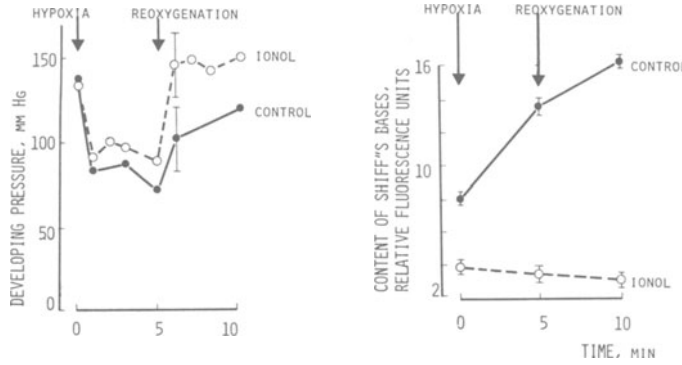

Fig. 5. Effect of preliminary ionol administration on the contractile function of the heart muscle and content of Schiff's bases in hypoxia and reoxygenation.

The left part of Figure 5 shows the pressure developed by left ventricle in the whole organism under hypoxia induced by a switch-off of the respiratory apparatus and a subsequent reoxygenation. The right half shows the concentration of end products of lipid peroxidation in the ventricle myocardium under the same conditions. It can be seen that a preliminary injection of the LPO inhibitor ionol prevents the rise in the concentration of Schiff's bases in hypoxia and, even in a greater degree, during reoxygenation. Ionol has no effect on the hypoxic depression of the contractile function and effectively prevents reoxygenational disturbances of this function. Thus, LPO activation in the myocardium can be due to the effects of at least three factors really existing in pathology namely to stress-induced

catecholamine excess, hypoxia and reoxygenation. All these factors are inevitably associated with myocardial infarction; therefore a considerable activation of LPO may be expected in this case.

It can be seen from the Table 3 that 24 hours after ligation of the left coronary artery descending branch and the development of experimental myocardial infarction in the rat the inactivation of antioxidant enzymatic systems and the significant increase of the intermediate and end products of LPO take place in the ischemic and, which is most essential, in the non-ischemic zone of the myocardium. This suggests a possibility of antioxidant use for protection of the heart against ischemic damage. However, our preliminary results were far from being encouraging.

Table 3. LPO activation and inactivation of antioxidant systems of myocardium in experimental myocardial infarction.

	Control (10)	Infarction	
		Ischemic zone (8)	Outside the ischemic zone (8)
Activity of enzymes:			
Superoxide dysmutase, conv. unit/1 g protein	344±25	283±18 $p < 0.05$	247±8 $p < 0.005$
Glutathion peroxidase I, nmol NADPH/1 mg protein in 1 min	315±18	134±22 $p < 0.01$	116±8 $p < 0.01$
Glutathion peroxidase II, nmol NADPH/1 mg protein in 1 min	218±11	74±13 $p < 0.01$	42±6 $p < 0.001$
Catalase, nmol H_2O_2/1 mg protein in 1 min	445±43	285±28 $p < 0.001$	178±8 $p < 0.005$
Lipid hydroperoxides, opt. dens. unit/1 mg lipids in 1 ml	1.08±0.13	3.71±0.42 $p < 0.005$	2.00±0.18 $p < 0.01$
Fluorescence of Schiff's bases, rel. unit	1.00±0.22	13.31±1.39 $p < 0.005$	5.11±0.60 $p < 0.005$

Figures in parenthesis mean number of animals

Data represented in Table 4 show that preliminary injection of the antioxidant did not significantly affect the size of the infarction zone 48 hours after ligation of the left coronary artery descending branch. Ionol only decreased the degree of fermentemia

Table 4. Effect of preliminary ionol administration on the necrosis size and fermentemia in experimental myocardial infarction

	Infarction (11)	Ionol-infarction (9)
Infarction area under epicardium, % of the total area of left ventricle	63.3±20	59.5±3.1 $p < 0.05$
Infarction area under endocardium, % of the total area of left ventricle	50.9±1.9	43.7±3.2 $p > 0.5$
Activity of aspartate transaminase, u/l	130.4±12.2	97.2±7.1 $p < 0.05$

Figures in parenthesis mean number of animals

associated with the infarction; the same effect of ionol was, however, observed in the case of emotional-painful stress. It may therefore be assumed that the fermentemia observed 48 hours after the onset of myocardial infarction is mainly due to stress damage of various tissues and its decrease is a result of the antistress effect of ionol.

The curves shown in Figure 6 reflect the dynamics of the pressure developed in the left ventricle during relative rest and at maximal load caused by tension after 25 sec clamping of this pressure and the velocity of its development are significantly decreased as compared to the control. This depression caused by infarction is maximal at the 25th second after aorta clamping when a fatigue of the safe zones of the myocardium is developed. The most essential experimental finding consists of the fact that a preliminary injection of the antioxidant considerably prevents the development of this contractile function damage. The hatched area in the slide designates the protective effect.

Since the size of the ischemic necrotic zone is decreased by the antioxidant only insignificantly, we have supposed that this effect may be due to the antioxidant protection of the safe, non-ischemic zones of the myocardium against the stress-induced damages concomitant with myocardial infarction. In other words, it seems probable that in this case we deal with a protection of the intact divisions of the myocardium against stress damage.

To test this assumption we studied the contractility of the experimentally non-ischemic zones of the heart, namely the right auricle of animals with experimental infarction of the left ventricle.

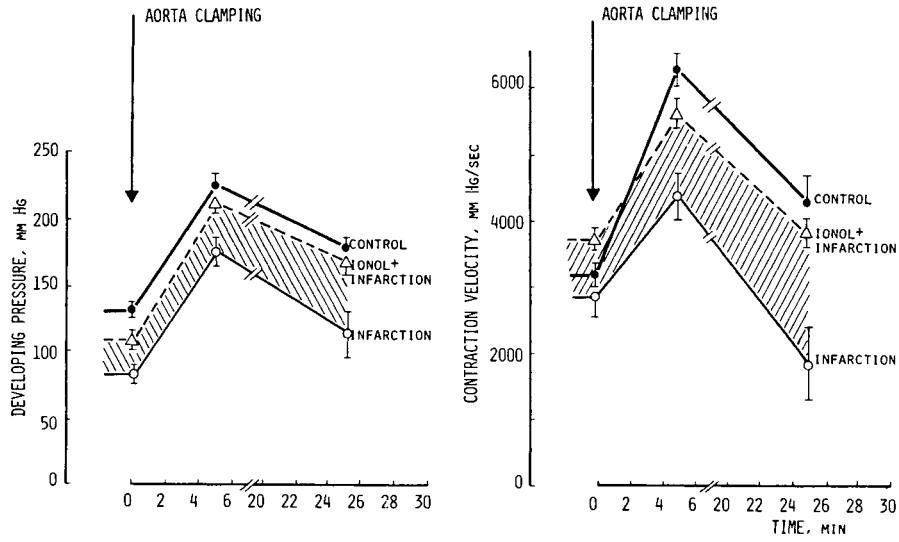

Fig. 6. Effect of preliminary ionol administration on the contractile function of the heart muscle at maximal load in control animals and in those sustained experimental infarction.

The Table 5 shows that 24 hours after the onset of experimental infarction in rat left ventricle the contractile function of the right auricle is strongly disturbed, showing in an almost 2-fold decrease of the maximal pressure developed by the isolated auricle during isometric contraction and of the intensity of its structure functioning which was determined as auricle tension/mass ratio multiplied by the contraction frequency. A preliminary injection of the β-blocker inderal prevents this defect of the contractile function. Consequently the auricular damage is due to catecholamine excess, that is, to adrenergic stress effect associated with infarction. The table also demonstrates that ionol prevents the disturbed contractile function of the auricle in the same degree as inderal. In this way the antioxidant prevents the depression of contractile function of the non-ischemic part of the heart under acute period of myocardial infarction and the antioxidant protection of the heart is in this case an antistressory one. Based on the facts given here it is purposeful to come back to the very beginning of this paper to remind you that the damages of the membrane lipid bilayer upon stress and ischemia are due not to isolated activation of LPO but to the "lipid triad" involving the activation of LPO and phospholipases and the detergent-like action of excessive amounts of fatty acids and lysophosphatides.

Table 5. Effect of preliminary inderal and ional administration on the right auricle contractility in the infarction of left ventricle.

Series	Developed tension, mg	Intensity of structure functioning, g/mg.min
1. Control (10)	382±12.5	3.8±0.2
2. Infarction (15)	198.6±4.3	1.8±0.1
3. Inderal-infarction (11)	331±15	2.9±0.2
4. Ionol-infarction (10)	315.2±15	2.9±0.2
P_{1-2}	< 0.001	< 0.001
P_{1-3}	> 0.5	> 0.5
P_{1-4}	> 0.5	> 0.5

Figures in parenthesis mean number of animals

Based on this assumption we used, in addition to the antioxidants, the phospholipase inhibitor chloroquene and the lipase inhibitor nicotinamide, for protection of the non-ischemic divisions of the myocardium.

Figure 7 shows that 24 hours after the producing of infarction the contractile function of the non-ischemic division of the myocardium, that is, of the right auricle, is characterized by a marked depression of the Sterling's curve, which is shifted downwards and to the right. A preliminary injection of the above-mentioned membranoprotectors prior to the infarction significantly prevents the disturbance of contractile function. The hatched area in the figure designates the protective effects of ionol, nicotinamide and chloroquene.

When evaluating the fact that inhibitors of different components of the lipid triade indirectly protect the safe divisions of myocardium one should keep in mind that in the whole organism conditions the lipid triade components are inseparably linked with each other. Really LPO activation results in labilization of lysosomes in which a considerable part of cell phospholipases is concentrated. The phospholipidases which are released from lysosomes and also the activated membrane-bound ones can play an important role in the destruction of the membranous lipid bilayer and in formation of lysophosphatides and free fatty acids (Katz and Messineo, 1981). Lysophosphatides and high concentrations of fatty acids which are observed in blood during stress and infarction, due to their detergent-like action, disturb the regulary arrangement of phospholipids in membrane which, in turn, may result in an additional LPO activation. This chain

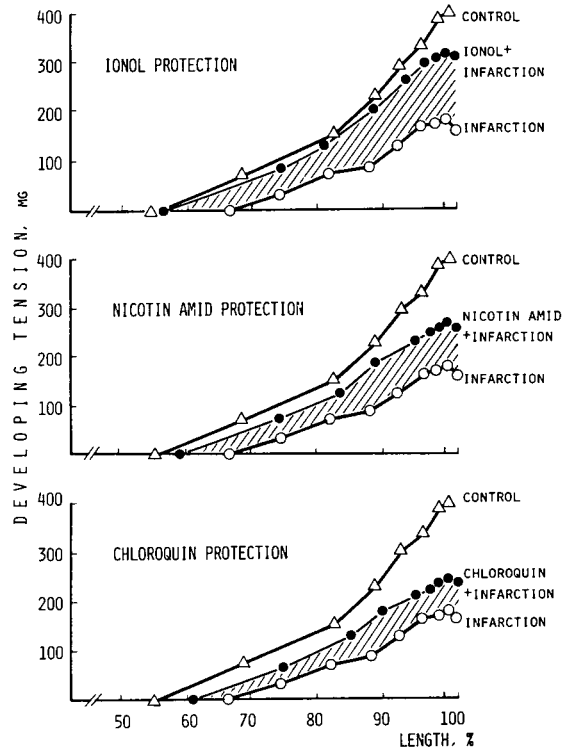

Fig. 7. Prevention of the Starling's curve depression in the non-ischemized zone by membrane protectors ionol, nicotinamide, and chloroquene in the experimental myocardial infarction.

reaction enables an increase in the membrane permeability for Ca^{2+} and the resultant excess of this cation, in turn, activates phospholipases (Vladimirov and Archakov, 1972). In the end the vicious circle closes which plays an important role in cell membrane damage, in transition of those damages to irreversible ones, and in myocyte death.

In this way one can imagine the process occurring in the ischemic zone in infarction and foci of necrotic damages. Outside the ischemic zone in infarction and in the most part of myocardium in stress the same cycle of events is less pronounced; it may be terminated at one of its links by inhibiting any component of lipid triads. This is precisely the point of view to understand the above-examined protective effect of the inhibitors of LPO, phospholipases, and lipases.

In conclusion we would like to emphasize that the contractile function of the safe parts of the heart muscle largely predetermines

the fate of a myocardial infarction patient. These divisions are damaged by stress which is always concomitant with infarction; therefore their protection by antioxidants and other membrane protectors seems to be a promising one.

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NUCLEAR MAGNETIC RESONANCE STUDIES OF INTRACELLULAR pH AND MYOCARDIAL CONTRACTILITY DURING ISCHEMIA

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INTRODUCTION

Most investigators in the field of molecular cardiology define myocardial ischemia in terms of a metabolic supply demand imbalance. Normal cell function is observed when supply is equal to or greater than demand. However, if coronary flow is reduced such that oxygen supply cannot meet the metabolic demands for the aerobic production of ATP (and phosphocreatine), then there must be a reduction in cell function[1]. In other words, cell function is in a delicate balance between metabolic (energetic) supply and its physiological demands, as illustrated by Equation 1.

$$\text{Cell Function} = \text{Supply/Demand} \qquad \text{Eq. 1}$$

By this definition, ischemic dysfunction of the heart may occur from either a reduction in blood supply at a constant physiological work demand, or from an increase in demand at a constant but limited rate of supply (flow). In cardiology, the latter condition is the cause of the clinical syndrome of angina pectoris, whereas the former initiates acute myocardial infarction, the leading cause of death in the Western world. While almost universally accepted, this fundamental metabolic definition of myocardial ischemia raises an important basic question. Is the ischemia, or flow induced depression of ventricular function always associated with observable alterations in the cell metabolic supply/demand balance? If the above equation is valid under all cases, then the answer should be yes.

The experimental observations leading to this question are well known. In an isolated, perfused isovolumic heart model, when the perfusion line is clamped, coronary flow is abruptly halted. As a result, we see an almost instantaneous and progressive fall in performance. During this time several key metabolic events are occurring. These include the hydrolysis of ATP and phosphocreatine, the cessation of aerobic metabolism, and the onset of anaerobic glycolysis leading to the accumulation of lactate and ultimately to intracellular acidosis[2,3]. Tissue oxygen is severely reduced, and as a consequence of bicarbonate buffering and diminished wash-out, tissue carbon dioxide rises[4]. These well documented observations have become the metabolic hallmarks of ischemic tissue. They have also led to the following hypothesis: the fall in contractility associated with myocardial ischemia may result from a decline in the high energy phosphates, the development of intracellular acidosis, the lack of tissue oxygen, or some combination of all three. A more critical examination of this hypothesis using 31-P nuclear magnetic resonance and mass spectrometry methods forms the subject of this report.

EXPERIMENTAL METHODS

Nuclear Magnetic Resonance Methods

Heart perfusion techniques. Hearts (5-6g) from young female New Zealand white rabbits weighing less than 2.0 kg are routinely used in our experiments. The hearts are perfused retrograde in a modified Langendorff mode previously described in detail[5]. The perfusion canula is positioned well above the aortic valve, which is then competent. The Krebs Ringer bicarbonate buffer is phosphate-free and contains 117 mM NaCl, 6.0 mM KCl, 3.0 mM $CaCl_2$, 1.0 mM $MgSO_4$, 0.6 mM EDTA, 16.7 mM glucose, and 24 mM Na bicarbonate, and is vigorously bubbled with 95% O_2/5% CO_2 for a final buffer pH of 7.40. The perfusate temperature is 40°C in the reservoir. The temperature of the perfusate overflow from the heart is 35-37°C. Since the perfusate is phosphate-free, all 31-P NMR signals arise from endogenous tissue metabolites. The hearts are paced at a rate of 150-170 beats per minute. Isovolumic ventricular pressure is measured using a latex balloon positioned through the mitral valve into the left ventricular cavity and connected to a Statham P 23 Db transducer, which is calibrated with a mercury manometer. The control end diastolic pressure is set at 10 mm Hg. Care is taken to prevent balloon herniation into the left atrium. Figure 1 illustrates a heart positioned in the NMR sample tube. Perfusate overflow is removed by vacuum aspiration. The rest of the accessory apparatus required for our studies is shown in Figure 2.

Instrumental methods. 31-P NMR spectra are obtained from a wide-bore superconducting magnet (4.23 T) at 72.89 MHz, using a Bruker WH-180 spectrometer. The stability of the magnetic field is such that

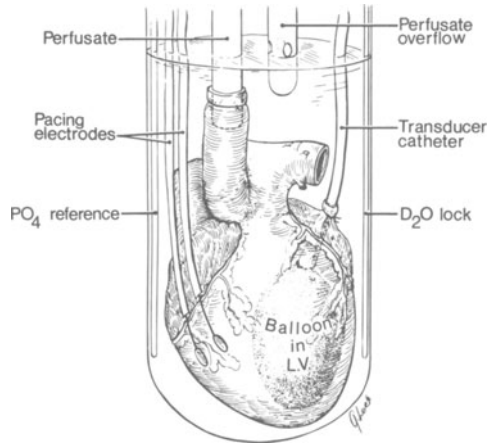

Fig. 1. A view of a perfused rabbit heart in the NMR sample tube. Tube diameter is 25 mm. Perfusate flows retrograde into the coronary arteries from the aortic perfusion cannula. The latex balloon in the left ventricle and the transducer catheter used for the measurement of isovolumic pressures are shown. Also illustrated are the pacing electrodes and the overflow line for removal of coronary flow. Reprinted with permission[5].

field/frequency D20 lock is not required; the probe diameter is 25 mm. The instrument is operated in the pulsed, Fourier transform mode, and is interfaced to a Bruker 1080 computer. Proton-decoupled spectra are obtained from transients following 25 μsec (45°) pulses delivered at 2-second intervals, conditions resulting in minimal spectral saturation. The data are collected at a 3,000 Hz width with a 2K data table, or at a 5,000 Hz spectral width with a 4K table. A heart spectrum with each peak labeled is shown in Figure 3. Spectra required 200-400 transients. Estimates of intracellular pH (pHi) are determined from the chemical shift (δo) of the Pi peak according to Equation 2.

$$pH = pK - \log (\delta_o - \delta_b/\delta_a - \delta_o) \qquad \text{Eq. 2}$$

To minimize the effects of tissue inhomogenity, all chemical shift values are determined relative to the phosphocreatine resonance, which because of its low pK (4.6) is rather insensitive to pH changes above 6.0. Our best estimates of the constants for Equation 2 are as follows: pK = 6.90; δa = 3.290; δb = 5.805. The validity of these values has been extensively discussed by us and others[5-7]. Using these constants, our current estimate of the heart's intracellular pH under various conditions is presented in Table 1.

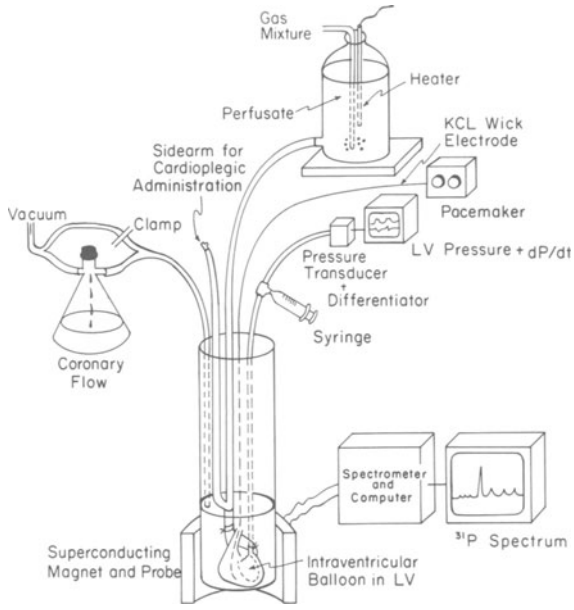

Fig. 2. A drawing of the accessory equipment required for physiological studies using NMR. LV is left ventricle; dP/dt is the first derivative of pressure. Reprinted with permission[5].

Table 1. Intracellular pH of perfused Rabbit Hearts.

Condition	Intracellular pH
Working (N = 15)	7.18 ± 0.02
KCL arrest (N = 4)	7.22 ± 0.02
Hypoxia (N = 7)	7.19 ± 0.01

Hearts were perfused with phosphate-free Krebs buffer. The working heart was the normal isovolumic model. Hearts were arrested with 37 mM KCl added to the perfusate. Partial hypoxia was induced by bubbling the perfusate with 65% O2/30% N2/5% CO_2. In all conditions, buffer pH was 7.45 - 7.48. Intracellular pH values are given as mean ± S.E.

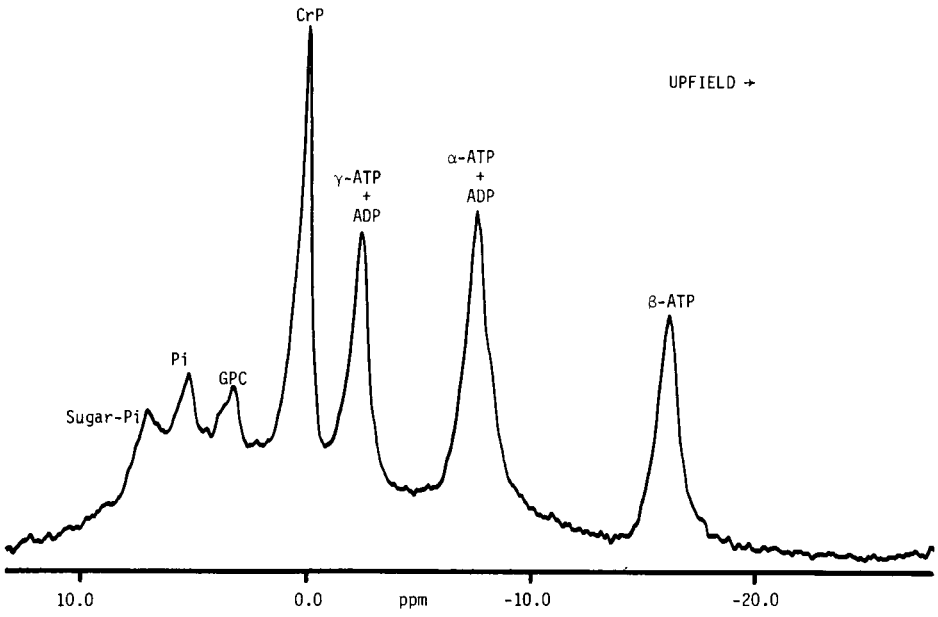

Fig. 3. A 31-P NMR spectrum of a perfused rabbit heart. This is a 9,000-pulse spectrum acquired at a 3,000-Hz spectral width. Peaks are observed for the glycolytic sugar phosphates (Sugar-Pi), inorganic phosphate (Pi), glycerol-3-phosphorylcholine (GPC), phosphocreatine (CrP), and ATP and ADP. The B-ATP peak is used to quantitate ATP. The abscissa scale is chemical shift in parts per million (ppm) with 0.0 at the peak of the phosphocreatine resonance. Reprinted with permission[5].

Mass Spectrometry Methods

The measurements of tissue gases were done by vacuum mass spectrometry methods standard in this laboratory. Briefly, a 22-gauge Teflon-coated probe is inserted tangentially into the free wall of the left ventricle and tightly sutured at the epicardial surface to prevent contamination by room air. The tissue gas mixture is withdrawn across the Teflon membrane, through stainless steel tubing, and into the mass spectrometer at a rate of 5×10^{-6} ml/sec. The 1/e response time of this system is 1.5 minutes for oxygen and 3 minutes for carbon dioxide, and the instrument is calibrated daily using gases of known composition. Further details of the mass spectrometry method have been reported[8].

Protocols

Since ferrous metals cannot be placed in the NMR magnet, separate protocols were required for our NMR and mass spectrometry experiments. In our NMR studies perfusion pressure was stepwise lowered from control of 110 cm H_2O (80 mm Hg) to 80%, then to 60%, and then to 40% of control. At each level of perfusion pressure, functional stabilization required 5 minutes, and NMR data acquisition required an additional 15 minutes. At the end of the protocol function recovered to 90% of the initial control performance when perfusion pressure was restored to 100%. A slightly different protocol was employed in our parallel mass spectrometry experiments. Perfusion pressure was lowered stepwise from a control value of 80 mm Hg to a final value of 20 mm Hg, in 10 mm Hg increments. Myocardial gas tensions were recorded 10 min. after each change in perfusion pressure, allowing time for instrument stabilization. Rates of coronary blood flow (CBF), oxygen utilization (MVO_2) and left ventricular developed pressure (DP) were also measured.

RESULTS AND DISCUSSION

From the perspective of heart energy metabolism, oxygen is by far both the most necessary and also the most limited substrate in the myocardium, there being no major cytoplasmic reserve. It has been estimated that within less than 5 seconds after coronary artery ligation, the tissue concentration of oxygen falls below the Km for cytochrome oxidase, and the mitochondrial electron transport chain becomes reduced. Oxidative phosphorylation, the source of 90% of contractile ATP[9], stops under these conditions. Therefore, in addition to the considerations of high energy phosphate metabolism expressed by Equation 1, tissue oxygen content could also be an important cellular regulatory signal for the heart. Physiologically important oxygen chemoreceptors exist in the carotid bodies of the carotid arteries. These receptors monitor blood oxygen content and help ensure adequate oxygen delivery to the brain. In a similar manner, oxygen per se could directly exert a regulatory effect over myocardial contractility If such were the case, it is possible that the changes in contractility observed at the onset of ischemia might parallel the decline in tissue oxygen tension, or PmO_2. To examine the dynamics of these potential regulatory parameters (ATP, phosphocreatine, pHi, and oxygen during conditions of reduced coronary flow, we used the techniques of NMR and mass spectrometry to monitor changes in these metabolites (Figures 4 and 5). Hearts were initially perfused at a pressure of 80 mm Hg. In a stepwise manner, perfusion pressure was reduced in 10 mm Hg increments from 80 to 20 mm Hg, or expressed as a percentage reduction, to 25% of control pressure. In both Figures 4 and 5, the line of identity is indicated by the dashed line. In Figure 4 we see that three parameters closely parallel the line of identity. These are the rates of coronary blood flow (CBF), left ventricular developed pressure (DP), and the rates of myocardial oxygen utilization (MVO_2).

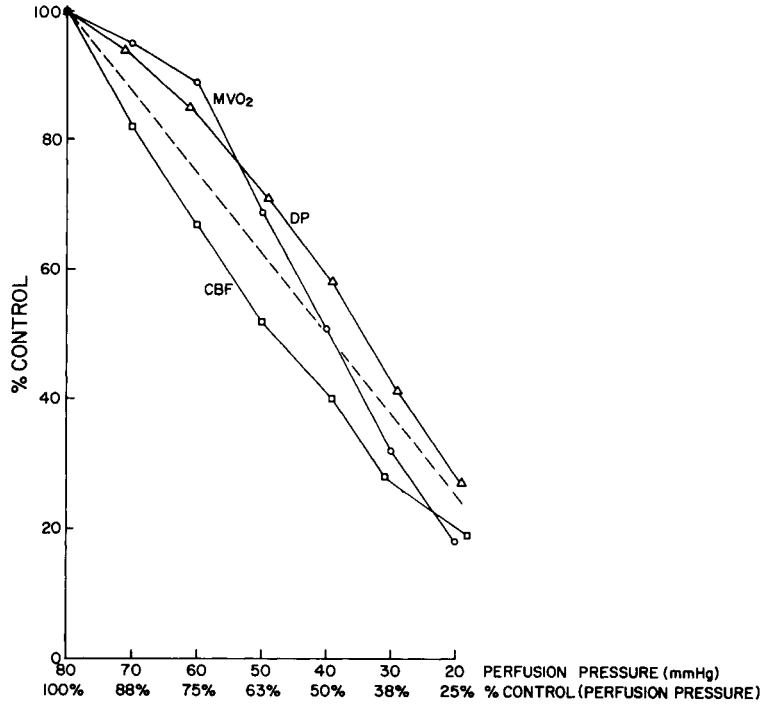

Fig. 4. Changes in coronary blood flow (CBF), developed pressure (DP), and the rate of oxygen utilization (MVO_2) as a function of decreased perfusion pressure in isolated, perfused rabbit hearts. Data were collected during a steady-state reduction in perfusion pressure, which was held constant at each point for at least 15 minutes. The dashed line is the line of identity. Reprinted with permission[5].

In other words, as perfusion pressure is diminished there is an almost linear decline in coronary blood flow, indicating a lack of coronary artery autoregulation. In conjunction, there is a similar decline in developed pressure, which, because of the reduction in ventricular work (ATP demand), leads to a parallel decline in oxygen utilization. These results were neither new or surprising.

In contrast, a rather striking and unanticipated set of results are seen in the NMR and mass spectrometry data (Figure 5). If our oxygen feed-back hypothesis had been correct, we would have predicted an immediate fall in tissue PmO_2, a sharp rise in tissue $PmCO_2$, with perhaps moderate changes in pHi and the high energy phosphate compounds. This was not observed (Figure 5). Initially, as perfusion pressure was decreased, PmO_2 increased while tissue $PmCO_2$ fell and pHi remained

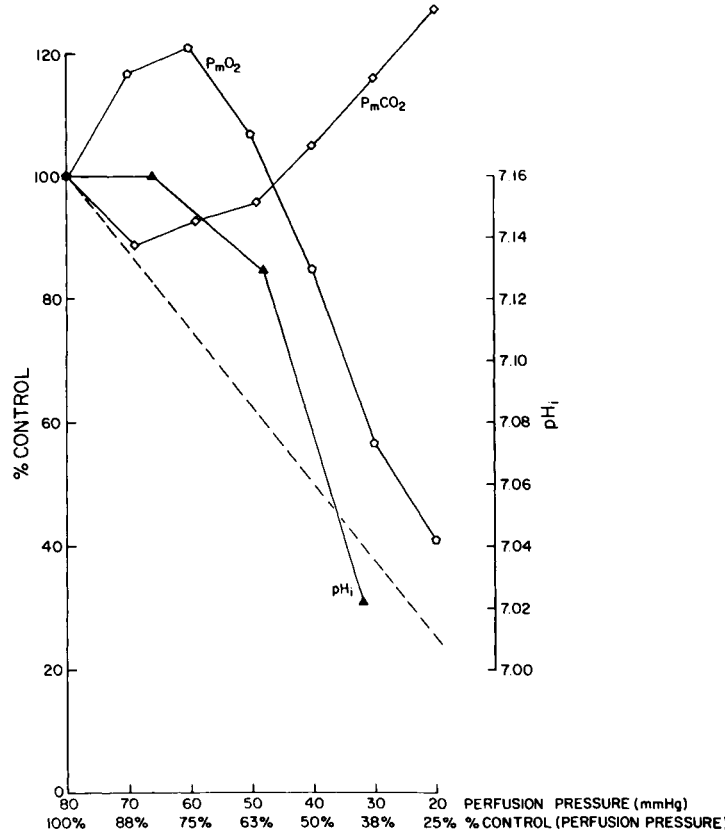

Fig. 5. Changes in tissue oxygen content (PmO_2), carbon dioxide content ($PmCO_2$), and intracellular pH (pHi) as a function of decreased perfusion pressure. Conditions were as described in Figure 4. Tissue gases were measured directly by mass spectrometry methods, and pH, was determined by NMR (see Experimental Methods). Reprinted with permission[5].

constant. As perfusion pressure was further decreased to about 75% of control, PmO_2 then started to decline and $PmCO_2$ began to rise, still at a rather constant pHi. The tissue gases continued to change until a crossover point was reached, which occurred at about a 44% reduction in perfusion pressure or a 52% reduction in flow (Table 2). Furthermore, at this point there was no change in the high energy phosphate content of the heart. ATP and phosphocreatine were present in normal amounts, up to the crossover point. Therefore, these hearts appear to be metabolically in balance (Eq. 1) even though performance is decreased by 32% (Table 2). This condition appears to be a direct violation of Equation 1. These data clearly show that tissue oxygen tension is not a primary parameter regulating myocardial function.

Table 2. Magnitude of the Changes Observed at the Crossover Point for PmO_2 and $PmCO_2$.

Perfusion pressure	44% reduction
Coronary blood flow	52% reduction
Left ventricular developed pressure	32% reduction
Rate of O_2 utilization	38% reduction
Intracellular pH	0.04 pH unit acidification
High energy phosphate	No detectable changes
Myocardial PmO_2 and $PmCO_2$	Normal

Data were derived from Figures 5 and 6.

These data do provide a new insight into the flow dependent control of performance. They show that up to 50% reduction in flow, some as yet undefined autoregulatory mechanism efficiently down-regulates myocardial contractility and thereby reduces oxygen utilization. Despite decreasing coronary flow, oxygen supply apparently exceeds oxygen demand as evidenced by the PmO_2 being higher than control. This level of tissue PmO_2 presumably allows aerobic metabolism to continue and intracellular pH to remain in balance (Table 2). Beyond a 50% reduction in flow, this mechanism apparently fails. Contractile demands then exceed supply and the classic metabolic indices of ischemia are expressed, including an elevated $PmCO_2$, and intracellular acidosis. In other words, moderate reductions in coronary flow did not result in a supply/demand imbalance (Equation 1), even though left ventricular function was markedly diminished.

Since this mechanism also prevents the "ischemic" accumulation of H^+ and Pi, agents known by themselves to induce cellular damage,[10,11] it would appear that this functional autoregulation might well act to protect the heart from "irreversible" damage during transient and moderate reductions in flow. The idea that functional "down-regulation" might reduce myocardial oxygen demands and thereby prevent irreversible ischemic damage could be applied to several clinical settings. In the case of myocardial infarction associated with thrombotic coronary occlusion, functional down-regulation could reduce metabolic demands such that in the presence of a sufficient degree of coronary collateral flow, myocardium could remain viable until an intervention such as thrombolysis could be employed to improve perfusion. In the case of angina pectoris due to either reduced supply (e.g. coronary spasm) or increased demand (e.g. exercise or atrial pacing) in the presence of fixed coronary obstructive disease, left ventricular angiography has demonstrated reversible segmental dyskinesis, akinesis, or hypokinesis. Functional down regulation of segmental contractility would be protective in any of these circumstances against irreversible

ischemic damage to cell membranes or enzyme systems. Segmental wall motion abnormalities on preoperative catheterization in the absence of symptoms suggesting angina pectoris have reversed or improved following coronary artery bypass surgery. This observation could be interpreted as suggesting the occurrence of functional down-regulation of segmental function distal to significant coronary narrowing in the absence of clinical or electrocardiographic signs of ischemia.

The results of this work leave us with a rather significant new question: how do moderate changes in coronary flow induce a depression in ventricular performance if there is no measurable decrease in tissue oxygen, intracellular pH, ATP or phosphocreatine? Certainly one can evoke the concept of "microcompartmentation" of metabolites. There is no current experimental methodology available to verify or refute this notion. On the other hand, it is possible that these effects are mediated by changes in wall tension[12]. The phenomenon relating coronary perfusion pressure to the internal stretching of the myocardial muscle fibers has been called the "garden hose" effect[13,15]. In brief, reductions in flow and perfusion pressure result in a decrease in the distending pressure in the coronary arteries, leading to a decrease in wall tension. By some intramural "internal Starling Mechanism", reduced wall tension appears to decrease contractility.

When a viscous nonoxygenated, nonnutrient containing substance was injected into the coronary arteries early after aortic occlusion at a time when contractility was depressed, function transiently returned toward normal[12]. These observations suggest that mechanical distention of the coronaries was sufficient to improve function, which supports the proposed "garden hose" effect. Although these experiments and the so called "garden hose" effect are consistent with our findings[12,16], whether or not this is the only major mechanism regulating myocardial function, during moderate ischemia, remains unclear.

The importance of our observed "crossover point" remains uncertain. One could speculate that if coronary blood flow is reduced by less than 50% that metabolic imbalance does not exist and therefore myocardial cells should remain viable indefinitely, albeit with depressed function. In contrast, if coronary flow is reduced more than 50% (i.e., to the right of the crossover point) then metabolic evidence of ischemia is present. Thus, if the duration of exposure to these conditions were sufficiently long, then myocardial necrosis would result. Jugdutt et al[17] have suggested that myocardial preservation within an anatomic vascular risk region is seen when collateral flow distal to a coronary ligation exceeds 50% of control. Regions receiving less than 50% of control flow will ultimately undergo necrosis. Whether the crossover phenomenon seen in the isolated perfused heart and this observation made in a conscious previously instrumented canine model are casually related or, more importantly, are applicable in the clinical setting remains open to further investigation.

SUMMARY

1. As coronary perfusion pressure was progressively reduced, coronary flow, left ventricular function, and oxygen utilization fell proportionately.
2. Under conditions of greater than 50% reduction in flow, energetic demands exceed supply, and the classic metabolic indices of is chemia were expressed, including decreased PmO2,: and intracellular acidosis.
3. However, with flow reductions of less than 50%, metabolic supply met or exceeded demands, and metabolic indices of ischemia were not observed. Under these conditions the heart was not in a metabolic supply/demand imbalance, even though function was reduced by 32%. These data show that the classic metabolic definition of ischemic cell dysfunction is not valid under all conditions of coronary flow reductions.
4. Therefore, some non-metabolic signal must be down-regulating contractility under the conditions of mild to moderate flow reduction. Although mechanical factors may play a role, the magnitude of its contribution along with other mechanisms responsible for this potentially protective, auto-regulation remain elusively undefined.

ACKNOWLEDGEMENT

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INTERVENTION TRIALS - PROBLEMS OF INTERNATIONAL COORDINATION AND EVALUATION

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THE SEARCH FOR PROOF

Does prevention really work? Until that question can be answered, inertia will persist; for it will continue to be said, "There is no proof".

How can the question be answered? If the demand is for proof (that is, for certainty) then that is and will remain beyond our reach. In clinical practice patients are not diagnosed and treated on the basis of certainties, but on a best clinical judgement of all the available evidence; and that is also how we have to manage populations.

Historically the great advances in public health have not been based on prior proof. The decision that sewage should be kept out of the water supply was taken before Pasteur had discovered bacteria. Recommendations on a healthy balanced diet preceded randomised controlled trials, and are still based on judgement rather than proof. Public health policy on control of air pollution and of cigarette smoking is based on an overall assessment of the epidemiological, laboratory and clinical evidence; where that evidence has been accepted, chronic bronchitis and lung cancer have declined - but that could not have been known until after the decisions had been taken.

Unfortunately a judgement based on indirect and circumstantial evidence may sometimes go wrong. Until recently it was thought that foods with high residue and high fibre content made a poor diet, especially in diverticular disease; and in my country at the end of the last century an unfortunate physician was banned from medical practice for life because he told the public that brown bread was

healthier than white. A controlled trial might have saved him! Views on that issue have now changed. No doubt there are many items of current medical practice and belief which are in fact mistaken. They ought to work, but for some reason they may not: why continue to guess, if you can perform the experiment?

WHAT DO CONTROLLED TRIALS OFFER?

Thus it became widely believed that a properly controlled intervention trial might at last offer certainty in public health decisions That view is now giving way to a more moderate assessment: the controlled intervention trial provides one more piece of evidence - uniquely important, because it is unbiased, quantitative and experimental - but yet providing neither unambiguous nor universal conclusions; and to be considered, not as the ultimate court of appeal, but as contributing one part of the totality of evidence.

There have been two reasons for this shift of opinion. The first is the realisation that we must be cautious about the generalisation of findings. Different trials may give different answers, and local results can depend on local circumstances. The acceptance of health advice varies. This has been the experience of the WHO European Multi factorial Trial of CHD Prevention (whose results are to be reported later in this Congress). In Britain we were less successful in gettin men to change their diet than were our Belgian and Italian colleagues; but we did rather better with cigarette smoking. Also, biological response may vary. Thus results of hypertension control may be different in men and women, or in blacks and whites; or the effect of reducing dietary fat in our trial could be different in well-fed Belgians from what it is in Poland, where alternatives are less readily available.

There is another reason why today we are more moderate in our expectations from controlled trials. This arises from the gap betweer the size of effect which a trial can hope to detect and the size of effect which would be of public health importance.

In 1971, when we started our multifactorial CHD prevention trial in Britain, our human and financial resources did not permit us to recruit and to follow for 5 years more than about 20,000 men. Statistical calculations suggested that such a trial might fail to detect any reduction in CHD incidence which was less than 25%. That was a serious limitation. We were evaluating a relatively cheap preventive program, operating largely through existing medical services. From a public health point of view, the costs would have been justified by a much smaller benefit: after all, a reduction of only 5% in CHD deaths would be more than equivalent to preventing all our deaths fror road accidents.

THE DEVELOPMENT OF INTERNATIONAL TRIALS

These same two concerns have prompted the important trend towards international collaborative trials. By involving a number of countries within a common protocol, they test the consistency of the findings in a variety of populations; and by combining the resources of several groups they increase the trial's size and statistical power.

The development of an international study is illustrated by the experience of our WHO European Collaborative Trial in the Multifactorial Prevention of CHD. The original UK centre was both too small, and also too narrow a base for generalizing the findings into international recommendations. Enlargement and diversification were clearly desirable, and the possibility of international extension was discussed among a small group of cardiovascular epidemiologists from Belgium, Italy, Poland and Spain. We already knew each other, and we welcomed the chance of working together. As a result the number of subjects grew to about 64,000.

I believe that these human relationships, which take time to develop, were vital; and they led directly to our efficient and pleasant working relationships. International studies cannot be initiated centrally according to some tidy plan: they have to grow out of existing scientific communications and dispositions. Thus their location cannot be freely determined.

In our trial, as in the WHO Clofibrate Trial and other collaborative studies, one center had already made a start before international collaboration was sought. This meant that our collaborators were faced with a predetermined protocol: they were asked to operate a study which they had not designed, and for highly experienced investigators this was not easy. To some extent the problem was reduced by our decision to standardize only a minimum core of protocol items that were essential to the pooling operation: each national center was free to extend the core study according to local interest. For example, the core protocol required electrocardiograms at entry to the control group in only a random 10% of subjects; in the Belgian center it was decided to extend this to 100% of control subjects, and as a result they are now able to test the effects of intervention according to whether or not there was early ischaemic injury.

The scientific status of individual centers was further strengthened by our agreement that each study was to be justified in its own right, with the pooled results of the whole collaborative group coming as a bonus. In each of the five countries it was necessary to know how much the intervention could change risk factors; and each study has been able to produce this important local information, together with estimates from the control group of naturally occurring changes in risk factors. There was also the possibility that the larger centers might (with some luck) see a significant change in incidence

of intervention. It is unlikely that individual centers would have been funded if they had not been seen as justified in their own right.

PROBLEMS OF DATA QUALITY

International collaboration involves major problems of standardizing the methods of data acquisition. This issue was crucial in a number of WHO studies which set out to compare in different countries the absolute levels of disease (for example, the incidence of heart attacks, or the severity of atherosclerosis). It is not nearly so serious in trials, whose purpose is to study the differences in incidence observed between intervention and control groups within individual countries: inter-center differences in the level of ascertainment should affect both the intervention and the control groups equally.

This does not mean, of course, that external quality control of data can be ignored in a collaborative trial. Inter-center difference may not bias the overall estimate of intervention effect, but they do influence its interpretation. To understand the results one needs to have used a common language for critical events; and incidence finding need to be related to absolute levels and trends of major risk factors In our collaborative trial we therefore followed the WHO registry procedures for defining coronary events, and we set up external quality control systems for cholesterol (through the WHO reference laboratory in Prague) and blood pressure. Electrocardiograms were reported according to Minnesota Code criteria, and each center submitted samples for central checking every 6 months. Inter-center differences were much smaller than is usual in multicenter studies, reflecting the fact that all the principal investigators had already worked together in using the Minnesota Code. Here, as at so many points, successful collaboration requires more than a common protocol: it depends on an accumulated mutual understanding between investigators who know each other and who are used to working together.

THE WHO CONTRIBUTION

The WHO contribution in our trial was important in two ways. First of all, the name of WHO sets a scientific and ethical standard which influences the investigators, the subjects and the sponsors - and all this at no financial cost to WHO! The job is done better because it is done in part for a supranational purpose.

At a more practical level, it is difficult for national studies to get money for those regular meetings with other investigators which are vital to the successful pooling of international data. A further expense arises for the data processing of pooled results. The amount of money involved may not be large; but without help from WHO I do not see how our collaboration could have succeeded.

HYPERTENSION CONTROL IN POPULATIONS

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Community Control Study Group*

In a masterly review of major developments and trends in health care in the twentieth century Milton Roemer quotes 16 points, including health manpower growth, specialization, organization for team work, population control, geriatrics and rehabilitation, health care planning, medical humanization, internationalism in health and health as a human right[1]. To this World Perspective on Health Care in the Twentieth Century one more important point should be added: the development of the concept of community control of disease. According to this concept, disease control rests on a defined community as a socio-biological entity. All its strata and components are taken into account and the community itself is being used to control and combat its own disease or diseases. There is an important difference between this participatory concept of community disease control and the earlier, classical public health approaches to disease prevention such as the draining of swamps as part of malaria prevention, or the provision of safe community water supply, or vaccination. In the classical situation, the community is rather the scene on which a medical or public health action is taking place. On the contrary, the community disease control programme of our days is like a happening in which all those present are required to participate.

Hypertension control provides an important example of the illustration of modern disease control programmes, if for no other reason than because of its very high prevalence a considerable part of the population has to be involved. With a prevalence of 15% in adult populations, hypertension may occur in one out of 3 to 4 families.

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In such a situation the only feasible solution is control through primary health care, backed by specialized services. Thus, blood pressure detection and treatment (in most cases) should be part of routine primary health care. For the diagnostic work-up of cases not responding to simple therapy, appropriate referral clinics should reinforce the primary health care services. In addition, for statistical (and clinical) surveillance of the great number of hypertensive subjects certain information systems are needed. These may be established in the form of various types of registers which, however, should be adapted to local circumstances. Such registers may range from simple manual card files in the offices of general practitioners to sophisticated and automated computer-based information systems, covering communities of several hundreds of thousands of people.

The real bases of hypertension community control programs consist, however, of a systematic education of both the population and the health workers. The two educational approaches should be initiated simultaneously, since they are complementary. A population aware of its hypertension problem should be backed up by its health corps motivated to spend time and effort on the control of this disease. Conversely, the health workers of a community should act in a population responsive to their efforts, or the community programs will remain barren. Such "formes frustes" of community programs have been observed. In fact, a community program triggers off an interplay between the population and its health workers: by creating, through health education, greater awareness and thus greater demand in the population, the health workers themselves will be stimulated; and conversely increasing offer from the part of the health services will in turn stimulate greater demand in the population. While in a number of health fields such a positive feedback mechanism may lead to an unwanted spiral of rising health consumption, in the field of hypertension it may result in adequate community control of hypertension, which was the original aim of the exercise. Whether this is invariably the best solution is, however, another question to which we shall come back somewhat later.

In 1972, the World Health Organization has started a multicenter international cooperative hypertension community control project with the aim of assessing whether such community programs are feasible and effective in various circumstances, such as socialized or private medical care, or developed and developing countries. Despite a number of difficulties, the project was carried to its end, and its final results are now being analyzed. It has been shown by the project that such community programs are, indeed, both feasible and effective.

In the following some findings from this study will be demonstrated on three simple graphs.

In a hypertension control program, blood pressure changes should be the most elementary indicator. Figure 1 shows the changes in blood

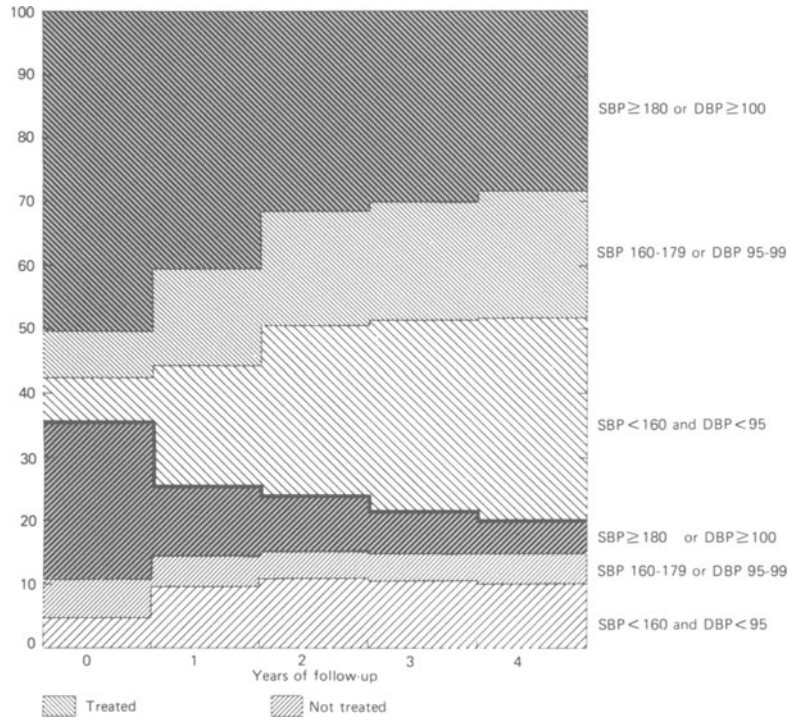

Fig. 1. Percentage of persons in various blood pressure categories in a cohort of 4,475 hypertensive subjects during a 5-year observation period; subjects without and with drug treatment.

pressure in a cohort of 4,475 hypertensive subjects, identified at the outset of the study and followed up throughout the first four years of the project. The graph shows that the proportion of non-treated hypertensive subjects was constantly decreasing, but that the most appreciable decrease occurred at the beginning of the project. Conversely, the proportion of treated subjects increased particularly at the beginning of the study, while in later years there was little further increase. The conclusion is that the effect of diagnosing hypertension is being immediately exploited for the starting of drug treatment. The graph also shows that the proportion of hypertensive subjects whose blood pressure had been brought under control (had become normal) increased considerably during the project, accompanied by an appropriate decrease in the group of severe blood pressure elevations. The proportion of mild hypertension, within the cohort of diagnosed hypertensives, increased due to the fact that a number of subjects with higher elevations of blood pressure moved down, due to treatment, to the mild hypertension zone.

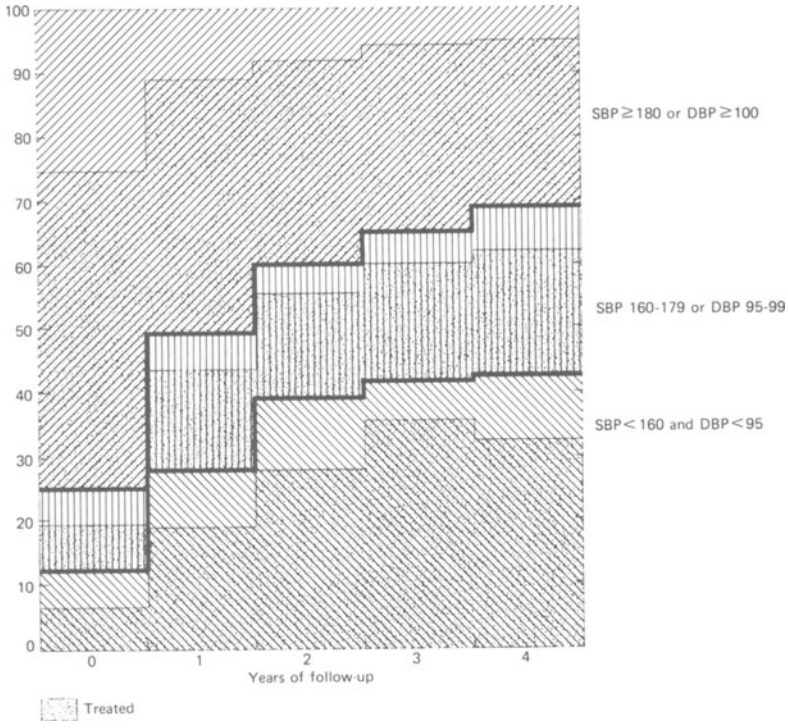

Fig. 2. Percentage of persons in various blood pressure categories in a cohort of 4,475 hypertensive subjects during a 5-year observation period; decreasing proportion of blood pressure values and increasing proportion of normotensive subjects.

Figure 2 presents the same information in a slightly different way. It shows more clearly the decreasing proportion of high values, the increasing proportion of normotensive subjects within the hypertensive cohort, and the moderate increase of the proportion of subjects in the range of mild hypertension. It should be noted that lowering of blood pressure to normal levels occurred also among the non-treated hypertensives. This finding is in agreement with reports from other studies, i.e. the Australian National Blood pressure Study[2].

It should be emphatically pointed out that the WHO Study was not a controlled therapeutic trial. Therefore, treated and untreated subjects may not be comparable. The WHO project was a community study where the option for treatment or non-treatment was left both at the discretion of the physicians and the patients. Figures 1 and 2 thus show what happens in a cohort of identified hypertensives in the community under "natural" circumstances. Part of the changes is, no doubt, due to the effect of regression to the mean. It is difficult, however, to assess what proportion of the observed changes should be interpreted in this way.

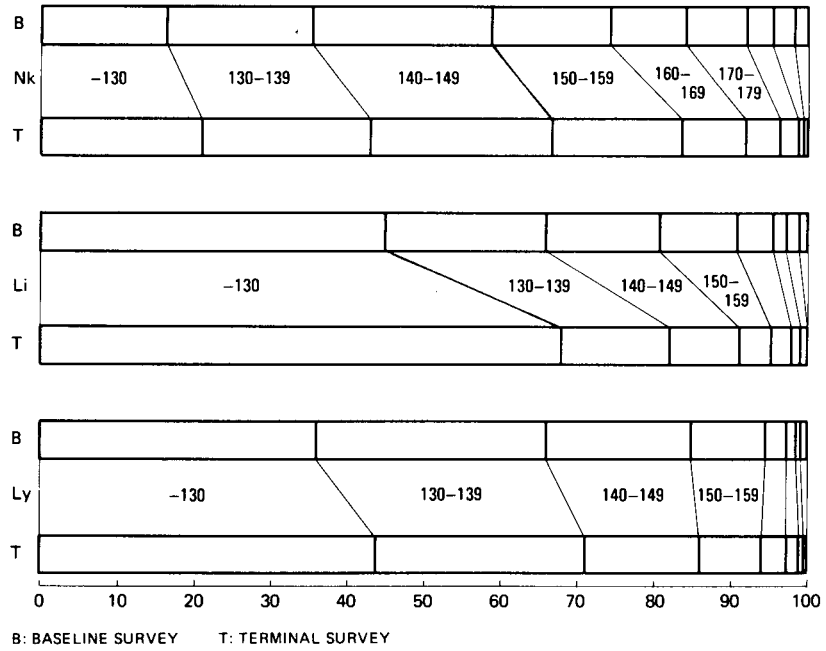

Fig. 3. Percentage distribution of systolic blood pressure in the population, age 30-59.
Blood pressure changes in all communities during a 5-year period, age group 30-59. Nk, Ly, Li: 3 communities where a blood pressure control program took place. The proportion of lower blood pressure classes is increasing; that of the higher classes diminishing (B: Baseline survey, T: Terminal survey).

Unlike the previous graphs which presented the changes in a cohort of hypertensive subjects, figure 3 demonstrates the blood pressure changes in whole communities based on a comparison of representative samples surveyed at the beginning and at the end of the project. In the three communities, selected as examples, the lower blood pressure classes were clearly expanding with an appropriate shrinking of the upper blood pressure classes. Such a shift in the distribution of blood pressure values in the entire community is, in fact, the ideal goal at which any community program should be striving. The figures on which graph 3 is based stem for a preliminary analysis of the study. It is to be hoped that this finding will be confirmed by the forthcoming final analysis of the data.

If, indeed, such shifts in blood pressure of the whole community can be achieved, hypertension community programs acquire a different meaning and become much more important than if the treatment of great

numbers of hypertensive subjects were their only result. In fact, although the achievement of high treatment rates is the aim of present hypertension community control programs, the attainment of this aim should be considered only as an intermediate step. Life-long drug treatment of some 15% of the population should not be the definitive goal. High drug usage in the community is close to abusage. Eventually, the whole community as a socio-biological entity should shift towards the lower end of the blood pressure distribution. This goal is hopefully achievable without the administration of potent chemicals.

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COMPREHENSIVE CARDIOVASCULAR COMMUNITY CONTROL PROGRAM IN THE PHILIPPINES

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In collaboration with the Ministry of Health (MOH), the Philippine Heart Center for Asia (PHCA) initiated in 1977 feasibility studies on community control of rheumatic fever-rheumatic heart disease[1] and, a year later, on hypertension and stroke[2], utilizing primary health personnel of MOH. The encouraging results obtained and the experiences gained from these studies logically lead us to embark on feasibility studies in comprehensive cardiovascular community control program (CCCCP).

THE RHEUMATIC FEVER - RHEUMATIC HEART DISEASE (RF-RHD) COMMUNITY CONTROL PROGRAM IN A RURAL AREA (PANGASINAN)

The general objectives of the control program are:

a) to promote early diagnosis and treatment of rheumatic fever and rheumatic heart disease occurring in a defined area of the province of Pangasinan, utilizing the primary health care personnel of the Ministry;
b) to establish an RF-RHD registry in the area; and
c) to promote secondary prevention of rheumatic fever.

The primary health workers including nurses and physicians were initially given training on the epidemiology, etiopathogenesis, clinical manifestations and on prophylaxis of RF-RHD before the program started. Posters, strategically placed in health centers, churches and market places were used for the education of the public.

After two years of operation, we demonstrated that the utilization of the primary health care personnel in the early recognition of rheumatic fever-rheumatic heart disease was very gratifying as judged by the number of referrals made by them and their ability to detect murmurs. Midwives alone referred nearly half of 472 referrals made during the period, and their specificity in the recognition of murmurs was 100%, although they missed the presence of a murmur in 8% of cases.

In addition, these midwives carried out the administration of the prophylaxis in 70.8% of the cases. The adherence of patients to a long-term program of subsidized chemoprophylaxis in a non-subsidized program.

HYPERTENSION AND STROKE COMMUNITY CONTROL PROGRAM

The pilot project has the following objectives:

a) to find an applicable way of control of hypertension and stroke within the existing delivery system of medical care;
b) to determine the feasibility of community-based hypertension and stroke control in rural areas of the Philippines; and
c) to evaluate the effect of long-term intervention of the knowledge, attitude and behavior on the cardiovascular health of the population.

The communities involved are part of the area covered in the RF-RHD community control program already described, involving 25,000 individuals in each of the study areas and a reference community.

As in the RF-RHD program, the existing primary health care personnel is being utilized. Baseline screening gave a prevalence rate for hypertension (i.e. 160/95 mm Hg or above) of 7% in both communities. Patients identified to be hypertensive in the study area were given antihypertensive therapy through their service of choice (i.e. through the hypertension clinic established by the program, or through their own physicians). Intensive public education, as well as education of practitioners in the area is being done. A stroke surveillance and registry is also being initiated. The control area will participate in all evaluative activities but will not have an active intervention program.

Evaluation of the project will be based on mortality and morbidity statistics, and for this purpose, periodic screening of the target populations will be carried out after three, five, and ten years.

COMPREHENSIVE CARDIOVASCULAR COMMUNITY CONTROL PROGRAM

The results in the community control program of RF-RHD and hypertension and stroke utilizing the existing primary health workers is very encouraging. The logical step is to unify these single disease projects into a comprehensive cardiovascular community control program. Such a program is now being conducted jointly by the PHCA and MOH in two rural areas north of Manila. Basically, it entails the study of two sets of communities; one representing the intervention area and the other as the reference. Baseline epidemiological survey as well as survey on the knowledge, attitude and practices (KAP) of the people are being done in both sets of communities.

Active community intervention will be carried out mainly through an educational approach. Hypertension being the most prevalent, will be the main target in the prevention and control program although the other CVD will be taken up also. The major risk factors to be controlled are cigarette smoking and salt intake. Morbidity and mortality registers will be set up in both communities.

After a period of five years, KAP, prevalence and risk factors surveys will be done for comparison with the results of the baseline survey.

The Ministry of Health is now developing pilot studies to make primary health care as the foundation of the national health service system. It is envisioned that CCCCP will be integrated into PHC.

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PREVENTION OF CARDIOVASCULAR DISEASES

WHO PROGRAMME

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The World Health Organization has concentrated in its cardiovascular disease activities specifically on those areas where international collaboration and coordination was essential. For that reason the area of epidemiology and prevention and control of different cardiovascular diseases became very soon much preferred field in all WHO activities. Starting with prevalence and incidence population surveys, preparation of teaching materials (among which "Cardiovascular Survey Methods: Methodology" by G. Rose and H. Blackburn (1968) was the most important publication) and intensive training program in epidemiology and prevention of cardiovascular diseases were essential. Later, WHO has initiated in collaboration with many centers, aided by its trainees, more systematic epidemiological research to assess the extent of the program of cardiovascular diseases in different countries, both industrial and developing, as well as entered the field of intervention trials. The project on myocardial infarction as well as stroke community registers carried out in the early 70's became the basis of presently formulated project on "The monitoring of the incidence and determinants of cardiovascular diseases in populations". It will help to provide the reasons for the different trends in the incidence of cardiovascular diseases and specifically coronary heart disease and cerebrovascular stroke in different populations. The clofibrate trial has provided beside its scientific results, new stimulants for and considerations about the ethical aspects connected with the application of long term drug therapies.

The wide network of collaborating institutions with epidemiological skills in the field of cardiovascular diseases provided the possibility of developing and initiating community control programs of different cardiovascular diseases.

From this background the symposium aimed at highlighting the successes as well as the mistakes and problems encountered while carrying different projects.

Professor G. Rose, London, the chief coordinator of the "WHO European trial in the multi-factorial prevention of coronary heart disease" discussed general problems of international trials and their possibility to give the answer to the major question "does prevention really work?". He stressed that the controlled intervention trial provides more scientifically important evidence because of its unbiased quantitative and experimental set-up. However, it still does not provide unambiguous nor universal conclusions. Different trials produced often different answers and results can depend on local circumstances. The advantage of international collaboration is the increase in trials size and its statistical power. The important lesson was that beside adhering to the standardized protocol to achieve the comparability of the results and standardization of the methods of data acquisition, provision for external quality control of data has to be made. The successful collaboration however, also depends on an accumulated mutual understanding between investigators who know each other and who are used to working together. The role of WHO set a scientific and ethical standard which influenced the investigators, the subjects and the sponsors.

The hypertension control in populations was discussed by Dr T. Strasser, WHO, Geneva. The concept of community control of disease rests on a defined community as a social, biological entity. The modern community disease control programs require full participation of the individuals, family, different sections of community including health services. The WHO hypertension control program initiated by the Organization in 1972 aimed at assessing whether community control of hypertension is feasible and effective in various circumstances, such as socialized or private medical care and in individual and developing countries. The control of hypertension in the population is feasible and effective. It leads to diminution of non-treated hypertensive subjects. The major changes in the percentage of treated population were registered specifically in the very beginning of the project. The proportion of hypertensive subjects whose blood pressure has been brought under control - had been normalized - increased considerably during the project accompanied by the decrease in the group of severe blood pressure elevations. The proportion of mild hypertension within the cohort increased due to the fact that the number of subjects with higher elevations of blood pressure moved down due to treatment.

The systematic hypertension control program effects the blood pressure of the whole community. That means that the mean blood pressure of the population studied is shifted to the biological normality. In such a way hypertension community control programs acquire a different meaning and become much more important from public health

point of view and health of the community as such. However, the study has also shown that life-long drug treatment of some 15% of the population should not be the definitive goal. Questions have been raised if high drug usage in the community is not an abuse.

Collaboration between the town of Kaunas, USSR, and WHO is of long-lasting tradition. Originally, a Kaunas-Rotterdam multi-factor preventive trial has been initiated coordinated by Dr I. Glasunov (WHO, Geneva), who was the speaker in the symposium. The population of Kaunas participated in other WHO activities such as myocardial infarction and the stroke registers, rehabilitation and secondary prevention of myocardial infarction patients, and recently in the project on comprehensive control of non-communicable diseases as well as in the project on the monitoring of the incidence and determinants of cardiovascular diseases (MONICA Project). This collaboration provided great experience for the Kaunas medical community as well as for WHO. Kaunas is one of the population areas where, for example, the myocardial infarction community register is being continued since 1971 until now. Therefore, its experience is of great value for the planning of the MONICA Project. Kaunas on the other hand serves to the public health service in the USSR as a public health laboratory specifically in studies in prevention of cardiovascular diseases and other non-communicable diseases.

Dr S. Guzman and his collaborators, Manila, Philippines, have discussed the problems of the establishment of comprehensive cardiovascular control program in a developing country. Feasibility studies on community control of rheumatic fever and rheumatic heart disease demonstrated that existing primary health care workers were able to recognise the disease with reasonable accuracy and were able to institute secondary prophylaxis in rheumatic heart disease programs on the level of peripheral health units. Later, encouraged by these results the community study was extended to control of hypertension and stroke. Screening for elevated blood pressure was done by primary health care workers and hypertensives discovered were registered. Medications were given under physicians' supervision and regular follow up and surveys were done again by primary health care workers. This project showed the feasibility of the community control programs to be carried out in developing countries when the role of the health personnel at different levels is appropriately identified and proper training is introduced. These programs increase the health education of the public and improve the surveillance in the community for cardiovascular diseases. The community control programs in cardiovascular diseases are now being integrated into the primary health care programs of the Ministry of Health. In such a way it is expected that the prevention and control of cardiovascular diseases will become an integral part of the health care programs of the public health service.

A new concept of primary prevention of cardiovascular disease in developing countries was presented by Dr S. Dodu, WHO, Geneva. "Primordial prevention" expressed the need to take appropriate action at an early stage aiming to prevent the emergence or entrenchment of

those social, economic and cultural backgrounds that have been shown to contribute to the high incidence of certain diseases in industrial populations. Some preliminary approaches have already been done and analysis of the country's situation have shown that in many developing countries realistic priorities for the primordial prevention of cardiovascular diseases will at best be restricted at the present time to smoking and hypertension control. The challenge of a broader based approach to primordial prevention is more likely to be accepted in the middle income developing countries where the cardiovascular and other diseases of "affluence" are emerging as a public health problem and where major risk factors are beginning to be established in the community at large and not merrily in the urban elite. Definite plans have been drafted by the World Health Organization to promote this approach and show by example of a few countries that it is feasible.

In its concluding remarks, the co-chairman, Dr Z. Pisa, WHO, Geneva, has stressed the importance of the work done by WHO and by other investigators in promoting and testing different methods to make the prevention and control of cardiovascular diseases a part of daily medical practice in countries with different social and economic structures. The problem of cardiovascular diseases is too extensive and emerging quite strongly now in developing countries that without prevention the problem cannot be tackled and controlled effectively. The lack of present acceptance of preventive programs is very often more in the resistance and traditions of the medical profession and its identification of prevention in populations with control of infectious diseases. Prevention of cardiovascular diseases is nevertheless more than medicine requiring the collaboration of different sections in the community involving also nutrition, city planning, agriculture, finances and others.

The present WHO strategy to prevention of cardiovascular diseases is based on three approaches: the first one and most important is the "population approach". It aims at altering the mass characteristics of life-style and environment which are underlying causes of cardiovascular diseases (for example coronary heart disease), or in low incidence populations preventing the developing of such precuros (primary prevention). The second approach is the identification of individuals with elevated risk factor levels and helping to modify these risk factors. This is a traditional approach in preventive efforts in the field of cardiovascular diseases as is the third approach, namely preventing the recurrence and progression of the disease - secondary prevention.

In the "population approach" the attempt is to shift the distribution curves of precursors and determinants of cardiovascular diseases in the whole population to the biological normality. Its importance is specifically stressed by the fact that the highest number of cases for instance of myocardial infarction is around the mean values of the population's risk factor levels, in spite of the fact that the risk of an individual itself increases with the increase in the level

of these factors. Further consideration for population strategy is based on the fact that artherosclerotic changes in the vessels start early in life. The population approach is specifically important in smoking control as well as in the approach to nutrition.

This strategy and its implementation through community control programs is of great significance and exceeds the importance and implications of cardiovascular diseases only. Examples of the cardiovascular control programs and preventive strategies are being now considered and adapted to the programs of other non-communicable diseases as well.

The work WHO could not be done without the enthusiasm and support of individual investigators, centers and Governments. WHO is most thankful to all those who are collaborating with it in their support.

INTEGRATED PREVENTION WITHIN PRIMARY HEALTH CARE

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After the "World Health Day 1972" with the slogan "Your Heart is Your Health", announced by WHO and ISC, considerable effort has been undertaken world-wide to control the major cardiovascular diseases, especially by preventive measures. Taking increasingly into consideration that cardiovascular disease is a social epidemic spreading from man to man and to women, from parent to child, and from culture to culture it became obvious: there are life habits that have to be changed. On this way, it might be possible to keep those diseases from appearing, stopping them when already going on, and even reversing their progression. But this is not a simple vaccination like in some communicable diseases!

Health starts in families. Home doctors, therefore, play a very important role joined by their nurses. They are able to detect easily people with elevated risk, provide them with the necessary preventive information, and make regular check-ups of what is going on, individually and on a community level. Being aware of that and of the fact that cardiovascular disease is a general public health problem in the German Democratic Republic as well as in other highly industrialized countries a model of prevention[1,2] has been tested integratable into the primary health care system of the country.

MATERIAL AND METHODS

The study was performed in the County of Cottbus, an energy production area based on brown coal mines. The study population incidentally screened within the daily practice of 24 General Practitioners comprised 6465 subjects, 2930 men and 3535 women,

within the age limits of 30 and 50 years, i.e. working people. They came to their doctors for different reasons and were checked for the main risk factors of coronary heart disease, additionally.

Before starting the study, doctors and nurses were trained in measuring blood pressure. Cholesterol determination has also been standardized from taking the blood samples until the laboratory which is working in coordination with the WHO Reference Lab. in Prague. The number of cigarettes per day has been registered by a short questionnaire for self-administration in which the Rose Questionnaire was partly included. In addition, some social factors were obtained on the basis of a national standard; some of those results have been reported already elsewhere[3].

Screening and prevention procedures were connected closely to get a high patients' compliance. The first step of treatment was without any (additional) drug, only giving information and advice to each individual, using printed material on smoking, hypertension, hypercholesterolemia, (physical inactivity and overweight). Intervention was started from 1 cigarette/day, blood pressure 140/90 mm Hg or 220 mg% cholesterol, i.e. borderline-ranges included. The patients were asked for a second visit to tell him/her the results at least. If there was a smoker consultation has been provided during the first visit. People with elevation of cholesterol or blood pressure, being classified during the second visit, then got preventive advice. (People with overweight or physical inactivity were only advised if one of the main risk factors was found, supporting non-medical treatment.)

RESULTS

Within the first, "hygienic" period a considerable shift took place in hypertensives. Three months after screening the prevalences had been diminished roughly by one third in both age and sex groups (Table 1). Simultaneously, a similar reduction could be seen with the prevalences of hypercholesterolemia, more pronounced in the age group 40-49 years concerning both sexes. In contrast, borderline hypertension prevalences did not change very much obviously due to the fact that a lot of hypertensives entered this group and only few (borderline) hypertensives reached the normal range of blood pressure. If looking at overweight nearly nothing changed, thus, indicating that losing weight cannot be a main reason for diminution of hypertension and hypercholesterolemia in the first period of intervention.

A second check-up, after 18 months, unfortunately showed that the gain was mostly lost with the exception of the severest hypertensive (Table 2 a/b) who got profit from the additional therapy provided during the second period by drugs. Their prevalence diminished already during the "hygienic period" and further decreased

Table 1. Prevalence reduction by hygienic approach only within 3 months.

Age groups	30 - 39		40 - 49		
Sex	males	females	males	females	
Hypertension[1]	21.9	18.9	31.3	32.7	
	14.8	11.6	23.4	23.0	per cent
Hypercholesterol[2]	21.1	12.8	27.2	20.8	
	17.4	9.2	21.5	14.0	

1) Definition acc. to WHO: 160/95 mm Hg or more
2) Serum cholesterol: 260 mg per cent or more

Table 2 a/b. Two step hypertension control within primary health care.

Prevalence changes:	Normotension	Borderline-H	Hypertension[1]
Males (n = 2340)[2]			per cent
Incidental screening	40.7	32.3	12.3 + 14.7
After hygienic approach	49.9	30.8	9.2 + 10.1
After drugs additionally	41.1	33.4	21.2 + 4.3
Females (n = 3037)[3]			
Incidental screening	43.1	30.5	10.3 + 16.1
After hygienic approach	54.6	27.7	7.2 + 10.5
After drugs additionally	46.8	32.5	17.8 + 2.9

1) Definitions acc. to WHO, but the hypertension group has been divided into those with blood pressures of 160/95-199/109 and those with 200/110 mm Hg or more.
2) Dropout 590 cases (20 per cent) after 18 ms.
3) Dropout 498 cases (14 per cent) after 18 ms.

considerably during the "medical period". They again shifted, however, only to blood pressures being still hypertensive since borderline hypertension increased not very much at long-term.

DISCUSSION AND SUMMARY

Prevention doesn't need special diagnostics. Home doctors, therefore, can perform the necessary basic examinations standardized within the daily practice. This allows a definition of individual

risk and provides the basis for interventional measures which should be first non-medical.

It is of strategical interest from the primary care point of view to know that prevalence of hypertension and hypercholesterolemia may remarkably be reducing within a few weeks, not being decidable whether this is due to a large regression to the mean or, at least partly, induced by the preventive information and check-up. That leads the GP to a much lesser number of risk people to be treated at lengthand serves to avoid a misuse of drugs which is, on the other hand, an essential condition for a good patients' compliance.

The study shows, however, that doctor's compliance is not a constant. GPs being primarily prone to use non-medical treatment may lose their interest with the guidance by the District Cardiologist is getting weaker. Then they return to their traditional, i.e. therapeutical way of practice, and the same may occur with the population.

In future it will be of high interest to get a) in general reliable basic prevalence data being comparable on a national and even international level, b) more implementable hygienic approaches, at least as first step of control of the main risk factors, c) systemic medical and public education in prevention and, last but not least, d) improved doctors' and patients' compliance.

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MULTIPLE RISK FACTOR INTERVENTION TRIAL:

6 YEAR RISK FACTOR CHANGES

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The Multiple Risk Factor Intervention Trial completed the follow up of the participants on February 28, 1982. This report describes the results of the changes in risk factors. Some of the data to be presented is still preliminary as final tabulations of the risk factor changes are being completed.

The design of the Multiple Risk Factor Intervention Trial has been published. A monograph describing basic intervention methods and preliminary risk factor changes has also recently been published. The methods will be briefly reviewed. The basic goal was to determine whether death rates for coronary heart disease could be reduced by a special intervention program aimed at reducing the serum cholesterol, blood pressure and cigarette smoking. Men were selected on the basis of a risk score distribution determined from the Framingham study. The risk score included the men's serum cholesterol, blood pressure and cigarette smoking. Initially men in the upper 15% of risk were eligible. After about one third of the screening had been completed, this was changed to the upper 10% of risk. The men were between the ages of 30 to 57 at entry to the trial and were free of clinical heart disease based on a history of myocardial infarction, positive Rose questionnaire or EKG evidence of myocardial infarction. The men were also to be free of other life threatening disorders. They had to be willing to participate in a six year intervention trial and at least attempt to make the risk factor changes which were proposed for the special intervention group.

The men were recruited in the community from available resources. No attempt was made to select a random sample of any defined population. Twenty-two clinical centers in the United States participated in the trial.

Table 1. Risk Factor Levels at each Screening Visit for Men Randomized to the Trial and Compared with All Men at First Screen.

Eligible Randomized	DBP	Cholesterol[a]	Percent Smokers
Screen 1	99.9	253.7	63.7
Screen 2	91.2	240.3	61.7
Screen 3	90.7	N.A.	59.2
All Screenees	84.0	214.6	36.8

[a]Screen 1 is serum and Screen 2 is plasma

The men had three preliminary examinations prior to randomization. The first screening examination included the measurement of blood pressure, serum cholesterol, and cigarette smoking behavior. These measurements were used to determine a risk score for eligibility for a second screen which included a physical examination and further evaluation of the individuals' health status. Finally, a third screening examination was done for those men still eligible which included a Bruce type submaximal exercise test as well as a behavioral evaluation prior to randomization. At the initial screen, 361,662 were seen. From the initial screenees, 7.1% were eligible for second screen and 3.6% or 12,866 men were ultimately randomized into special intervention (6,428) and usual care (6,438). The risk factor levels of the men randomized to the trial as compared to all men screened is shown in Table 1.

By the time the men were randomized to the trial at third screen, there had already been a substantial change in the risk factor levels especially for blood pressure and cholesterol. The primary reason for this decrease was probably regression to the mean. Other factors included changes made by the men prior to randomization and differences between plasma about 4 mg lower than serum cholesterol levels. At the time of randomization, the special intervention and usual care men were very similar for nearly all risk factors.

The development of the intervention process has been published. Men randomized to usual care were referred back to their physicians, contacted every 4 months to determine their vital status, and evaluated annually including a physical examination and measurement of risk factors.

The initial plan of intervention for those men randomized to special intervention was an intensive group intervention program which usually lasted about 10 sessions. During these 10 sessions, there was a structured program aimed at smoking cessation, changes in dietary

intake of cholesterol and saturated fat, and various aspects of the diagnosis and management of hypertension. Following the initial 10 week sessions, the men were placed in a follow up program based on their initial risk factor levels and changes during the intensive intervention. All special intervention men were seen at least every 4 months for evaluation of their risk factor levels and continued counseling. Many were seen more often. The special intervention participants also had an annual examination identical to that of the usual care men.

The specific goals of the intervention program were established prior to the trial and have been previously published. An important point to note is that little change in risk factors was expected among the usual care men.

The study has been very successful in maintaining the participation of the men. Only 7.2% of the special intervention and 9.1% of the usual care participants were not seen for their sixth annual exam. The result of the risk factor changes and morbidity to be published subsequently are, therefore, based on a very high level of ascertainment unusual for trials of this length and complexity. Very few participants were lost to follow up.

The Hypertension Treatment Protocol for special intervention participants was based on the Stepped Care model used in the Hypertension Detection and Follow up Program. Men were classified as having hypertension at baseline if the average of their second and third screen blood pressure was equal to or greater than 90 or if they were being treated for hypertension at the time of randomization.

Based on these criteria, 2,757 (42.8%) of the special intervention participants had a diastolic blood pressure greater than 90 at baseline and 1,261 (19.6%) were on drug treatment for hypertension, a total of 4,018 (62.4%) hypertensives. A participant normotensive at baseline could later be defined as hypertensive if at one of the four month visits, he had a blood pressure level greater than 89 and at a repeat confirmation visit usually one to four weeks later, it was still above 89.

Every special intervention participant defined by the criteria above was eligible to be given a goal blood pressure for the trial. The goal was based on the blood pressure level at the time of a confirmation visit. It was a diastolic blood pressure of 89 mm Hg or 10 mm Hg less than the average diastolic blood pressure at that visit. Men who had been previously on drug treatment prior to randomization and had a diastolic blood pressure less than 90 were given a goal of 80 mm Hg.

At 72 months, approximately two thirds of the special intervention participants had a goal blood pressure established. Their

mean blood pressure at baseline was 94.2 mm and at 72 months, 81.2 mm. Approximately two thirds had a blood pressure less than goal and 88% had a diastolic blood pressure less than 90 mm.

The mean blood pressure for all of the special intervention participants at year 6 was 80.5 compared to 83.6 for usual care men, a difference of 3.1 mm. This difference includes both the normotensive (Table 2) and hypertensive participants. The differences between special intervention and usual care participants were directly related to the level of the baseline blood pressure, about 1 mm for those with screen 1 levels less than 95 mm Hg and 4.9 mm for those with a screen 1 diastolic greater than 105. The number of special intervention participants prescribed drug therapy rose from 44.5% at 12 months to 58.2% at year 6. For usual care men the percentage rose from 30.3% at 12 months to 47.0% at year 6. The use of drug therapy for hypertension was directly related to the baseline blood pressure level. However, even among those with a baseline diastolic blood pressure less than 90 mm, 26% of special intervention and 18% of usual care men were on drug therapy for hypertension by year 6.

The mean serum cholesterol level at first screen was 253 mg%, and the mean plasma cholesterol level at second screen was 240 mg% At entry the men repeated an average about 2,500 kilocalories, 14.0 from saturated fat and 450 mg of cholesterol in their diet. Approximately 7.4% of the calories were derived from alcoholic beverages.

Table 2. Risk Factor Changes Between Baseline and Year 6 Special Intervention and Usual Care

	SI		UC		SI-UC
	Baseline	Year 6	Baseline	Year 6	Diff.
Serum cholesterol	254	236	254	240	-4 mg
LDL cholesterol	160	149	162	153	-4 mg
HDL cholesterol	42	42	42	42	0 mg
Triglycerides	192	198	192	199	-1 mg
Diastolic blood pressure	91	81	91	84	-3 mm
Reported cigarette smokers (per cent)	64	32	64	45	-13

The Nutrition Program was aimed at a reduction of saturated fat and cholesterol and total calories for weight reduction for those men whose weight was greater than 1.15 above their ideal weight. The initial dietary plan included reduction of saturated fat to less than 10% of calories, dietary cholesterol to less than 300 mg, and an increase in polyunsaturated fats to 10% of calories. In 1976, the amount of saturated fat was further reduced to 8% and dietary cholesterol to 250 mg per day.

At the end of 6 years, the serum cholesterol had decreased to 235.5 mg/dl for the special intervention and 240.3 for the usual care, a 4.8 mg/dl difference (Table 2). This difference in cholesterol was greatest for men with high initial levels of serum cholesterol, 2.7 mg/dl for those with screen 1 cholesterol 220-239 mg/dl and a 10.6 mg/dl difference between special intervention and usual care for those with cholesterol greater than 300 mg%.

Lipoproteins were measured at second screen and every two years following randomization. The mean baseline LDL cholesterol was 160 mg%. It had dropped to 148.7 mg/dl for special intervention and 152.9 for usual care by year 6 (a 4.2 mg/dl difference). The HDL cholesterol averaged 42 mg% at second screen and was approximately the same at year 6. Preliminary analysis has determined that a decrease in saturated fat and weight loss contributed to the decline of LDL cholesterol while alcohol intake, weight change and cigarette smoking were primary determinants of the HDL cholesterol levels (Table 2).

The smoking cessation program urged all special intervention participants to quit smoking. Initially no effort was made to change the smoking habits of men who smoked only pipes and cigars. As noted, about 60% of the participants smoked cigarettes at baseline (Table A). The dose and change of cigarettes smoking was monitored by questionnaire including information about number of cigarettes, brand and inhalation characteristics and objective changes in serum thiocyanate levels and expired air carbon monoxide levels (Table 2).

At the end of 6 years, 49.6% of the special intervention and 29.1% of the usual care reported that they had quit smoking cigarettes. The thiocyanate adjusted quit percentage was 46.1% for special intervention and 28.5% for usual care. The quit for special intervention men was greatest within the first 4 months following randomization but was maintained throughout the trial. There was a substantial increase in the usual care quit percentage as the trial progressed from only 11% at year one to close to 30% at year six.

The quit percentage was inversely related to the number of cigarettes smoked at first screen, ranging for special intervention men from 68.6% for those smoking 1-19 cigarettes to 40% for those smoking 2 or more packs per day. The differences between special intervention and usual care were also inversely related to the number of cigarettes at first screen.

Many men quit smoking for a period of time and then started again. Of the 1,399 special intervention men who quit smoking between randomization and the four month visit in year one, 36.5% were smoking at the time of their sixth annual examination. Therefore, the reported quit percentage represents a balance between current quitters and men who started to smoke again. Interestingly, it appeared that men who quit smoking later in the trial were more likely to start smoking again.

Not all of the men in the trial had the same distribution of risk factors. Most had at least 2 of the 3 major risk factors present. The simultaneous intervention on different risk factors had important effects on risk factor changes especially the use of drug therapy for the treatment of hypertension. For special intervention men, hypertension drug treatment tended to blunt the cholesterol lowering effects of the qualitative dietary and weight changes. A combination of diuretic therapy with weight gain or even weight stability was associated with substantial increase in plasma triglyceride levels. Hypertensive cigarette smokers had the least favorable change in their serum cholesterol levels. Reduction of alcohol intake was urged in order to reduce both calories and blood pressure levels but this may also have resulted in lowering of the HDL cholesterol levels for some men. Weight loss enhanced the qualitative dietary changes for cholesterol lowering effect but some cigarette smokers who quit gained weight.

Overall, the special intervention men did reasonably well in terms of their risk factor modification. Practically all normalized their blood pressure using pharmacological therapy. Over 40% had quit smoking by year 6. The cholesterol reduction was not as large as expected but for the hypercholesterolemic group was similar to that noted in other studies. The changes in usual care participants, however, were far greater than expected. This was especially true for the treatment for hypertension, for the reduction of serum cholesterol and somewhat for the changes in cigarette smoking. The overall difference between special intervention and usual care especially early in the trial were, therefore, less than planned. However, the maintenance of the changes in risk factors among the special intervention participants and their continued participation in the trial was unexpectedly strong and so by the end of the trial, the observed differences in risk factor changes although notachieving the goals approached those initially expected.

MULTIFACTOR PREVENTION OF ISCHEMIC HEART DISEASE

(A COOPERATIVE SURVEY)

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Prevention of ischemic heart disease and control of arterial hypertension is a most important task of health care in the USSR, since the implementation of this task is sure to result in decreasing morbidity and mortality. A lot has been done to the effect for the past years in this country, but there still exists a number of theoretical and practical problems, which are presently being worked at.

Since 1977 the USSR Cardiology Research Center has been guiding a cooperative survey of multifactorial prevention of ischemic heart disease (IHD). It is aimed at: 1) elucidating a possibility of reducing IHD morbidity and mortality by primary prevention - affecting such risk factors as arterial hypertension (AH), hypercholesterolemia (HC), smoking (S), excess body weight (EBW), low physical activity (LPA), and by secondary prevention; 2) working out a schedule of primary and secondary IHD prevention measures which can be put into practice within the framework of the existing health service.

The survey is being carried out among 40-59 year old randomized male population. According to the schedule (Figure. I) it involves three groups of population: an active intervention group and two control ones. The first group was subjected to examination, which includes the use of questionnaires to reveal angina of effort, possible myocardial infarction, intermittent claudication, chronic pulmonary diseases as well as smoking habits and physical activity level; registration of a 12-lead ECG with subsequent evaluation according to Minnesota code; measurement of AP (twice); biochemical determination of blood cholesterol content; anthropometry (height, weight, skinfold thickness).

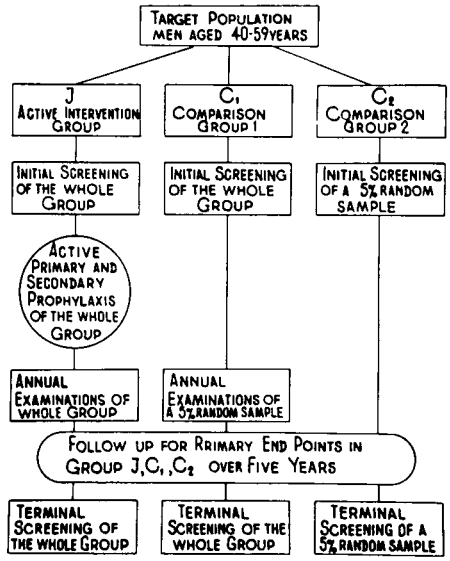

Fig. 1. General study design.

IHD and risk factors were established on the basis of the following criteria;

1) IHD - myocardial infarction (MI) in the anamnesis, I-I, I-2 category alterations in ECG and (or) documented MI. (The diagnosis in each case is decided upon by the consultative committee on the basis of criteria used in WHO "Myocardial Infarction Register"); angina of effort - the corresponding WHO questionnaire; painless form (in the absence of MI and angina of effort - ECG alterations (type 1-3; 4-1,2,3; 5-1,2,3; 6-1,2; 7-1; 8-3 of Minnesota code).

2) AH - diastolic arterial pressure (DP) of 95 mm Hg or higher; any DP on the background of hypotensive drug therapy for the last 2 weeks.

3) Smoking - regular smoking of at least one cigarette a day.

4) Hypercholesterolemia - blood cholesterol of 260 mg% or more.

5) EBW - $\frac{\text{weight (kg)}}{\text{height}^2\text{(m)}}$ index equal to or exceeding 30.

6) LPA - sedentary work for 5 and more hours a day and active leisure of less than 10 h a week.

After a screening and a classification of patients the first group (active intervention) is subjected to active primary and secondary IHD prevention measures. The latter are being carried out on the basis of maximal involvement of the existing health service personnel. The second group (I control) undergoes the same examination as the first one; the obtained data are handed over to practising physicians; preventive measures are taken according to the usual procedure. In the third group (II control) a list of the population is made and only a 5% random selection is examined and subsequently subjected to routine preventive measures. The whole of the first group and a 5 or 10% random selection from the second are seen annually while the third group is not examined at all.

The minimal follow-up period is 5 years. During the whole period we register mortality and new cases of MI and brain stroke in all three groups.

Besides, dynamics of risk factor levels, new cases of angina of effort, ischemic ECG alterations, temporary and permanent disability, and some other health indices are analyzed in the I and II group at annual examinations.

When the follow-up period is over the whole of the first two groups and a 5-10% random selection from the third are examined.

The results of the survey will be evaluated by comparing the number of the following dead points in three groups:

major points
- new cases of MI
- brain stroke
- IHD mortality
- total mortality;

secondary points
- risk factor levels
- new cases of stable disablement due to IHD and brain stroke
- new cases of angina of effort and ischemic ECG alterations.

The following centers collaborate in the survey: USSR Cardiology Research Center and Kaunas Medical Institute (since 1977), Belorussian Scientific Research Institute (SRI) of Cardiology in Minsk (since 1979), Kirgizian SRI of Cardiology in Frunze and Uzbek SRI of Cardiology in Tashkent (since 1981), Kharkov branch of Ukrainian Strashesko SRI of Cardiology (since 1982).

The population observed in each center in all three groups ranges from 9,000 to 16,000. In Frunze, Tashkent and Kharkov primary examination of the selected groups is not over yet: the total number of people observed is 71,000: 23,000 have been examined by now in all centers; 12,000 have been assigned to a program of special intervention.

The survey is conducted according to a common plan and protocol using unified examination techniques. For a better coordination of efforts a special committee incorporating the representatives of all centers was set up. All cases involving major final points of the survey are considered by an intercenter consultative body which makes it easier to unify the evaluation of results.

Before starting the program the members of the teams from all the centers had a special training course and all the examination techniques were standardized (questionnaires, AP measurement, ECG coding). In the course of the survey training and control standardization is annually repeated which makes it possible to compare the data obtained by various centers.

This report analyzes and compares the tentative data of male population screening in 5 cities (Moscow, Kaunas, Minsk, Tashkent and Frunze) and the tentative results of special intervention toward altering the risk factors in Moscow, Kaunas and Minsk.

The analysis and comparison of IHD and risk factors spread were made on the basis of random selections with a 65-69% response (Moscow - 6737, Kaunas - 5929, Minsk - 1806, Tashkent - 1284, Frunze - 4719).

According to the primary examination data (Figure. 2) only 17-25.3% of the subjects observed had neither IHD nor the risk factors. The lowest number was in Moscow (17%) and the highest - in Tashkent (25.3%) (Kaunas - 21.5%, Minsk - 22.5%, Frunze - 20.5%). From 64 to 68% of the subjects observed had one or more risk factors, and half of them had two or more. At the time of the examination from 8.6-15% had IHD and brain stroke record in the anamnesis. Moscow accounted for the largest number of such cases (15%), Tashkent - for the fewest (8.6%) (Kaunas - 11.5%, Minsk - 13.2%, Frunze - 11.5%).

Thus, in the 5 cities of the Soviet Union from 75% to 83% of 40-59 year old men observed needed primary and secondary preventive measures.

The distribution of IHD incidence was as follows: Moscow (14.5%) Minsk (12.6%), Kaunas (10.9%), Frunze (10.3%), Tashkent (8.4%). It is of interest that major differences are registered in the incidence of such forms of IHD as myocardial infarction (from 1.1% in Tashkent to 3.7% in Moscow: Kaunas - 2.7%, Minsk - 2.5%, Frunze - 1.5% and angina of effort (from 2.8% in Tashkent to 6.5% in Moscow; Kaunas - 2.5%; Minsk - 4%; Frunze - 3.4%). At the same time there is a lesser variety in the incidence of painless IHD form (from 4.4% in Moscow to 6.1% in Minsk). Moscow is distinguished both by a considerable spread of IHD and the domination of its painful forms (10.1% of MI and angina pectoris of 14.5%). In Kaunas, Tashkent, Frunze and Minsk painless IHD either prevails or insignificantly

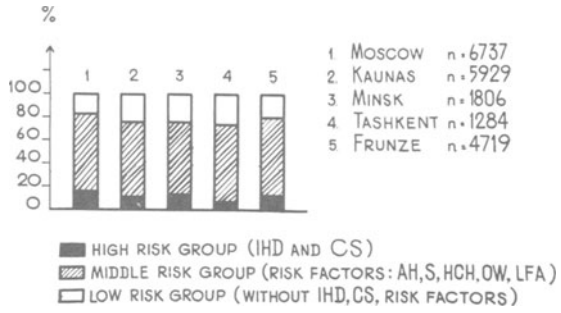

Fig. 2. Prevalence of the three risk groups.

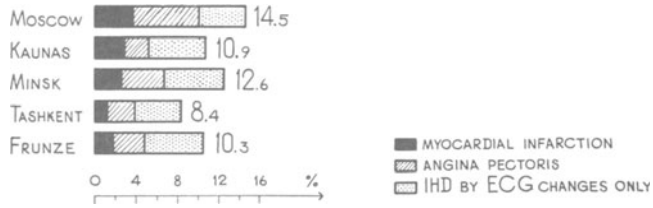

Fig. 3. IHD prevalence in examined populations.

differs from the incidence of painful forms. There are subjects in MI group who have only ECG signs (1-1, 1-2 according to Minnesota code) without MI in the anamnesis.

The distribution of the risk factors (Figure. 4) was as follows: the incidence of AH varied from 22.5% (Minsk) to 27.9% (Moscow) (Kaunas - 23.9%, Tashkent - 25.0%, Frunze - 25.2%); smoking - from 42.9% (Kaunas) to 52.3% (Tashkent) (Moscow - 46.2%, Minsk - 51.8%, Frunze - 48.0%); HH - from 20.0% (Frunze) to 26.8% (Tashkent) (Moscow - 21.1%, Kaunas - 24.2%, Minsk - 23.6%); EBW - from 8.4% (Frunze) to 21.2% (Kaunas), (Moscow - 9.8%, Minsk - 13.7%, Tashkent - 19.9%); LPA - from 11.5% (Kaunas) to 22% (Frunze) (Moscow - 21.0%, Minsk - 11.9%, Tashkent - 13.7%).

The above mentioned variety in the incidence of IHD and risk factors is quite natural since the people assigned to the program live in different geographical and climatic conditions, and have their own national peculiarities and traditions, eating habits, etc.

The incidence of IHD and risk factors turned out to be rather high in all the cities which proves the necessity of the most active primary and secondary preventive measures. For example, in Tashkent IHD incidence is the lowest, while the spread of risk factors is practically the same or even higher than in other cities. That

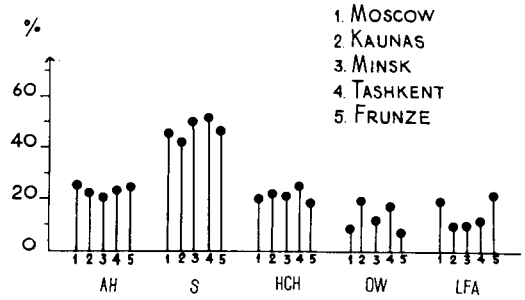

Fig. 4. Prevalence of risk factors in examined populations.

indicates a possibility of the rise in the incidence of IHD in Tashkent if timely intervention does not alter the risk factors.

Preventive measures in the active intervention group are carried out by specially trained health service personnel and scientific research centers.

The intervention program in each center may have its local characteristic features if it keeps to basic common principles. These are as follows: 1) maximally differentiated approach to all patients; 2) mostly non-medicamental preventive measures against smoking, hypercholesterolemia, excess body weight and low physical activity; 3) medicamental therapy conditioned by the state of each patient in the group of subjects with AH and IHD.

In Moscow, Kaunas and Minsk the annual examinations of random samples selected from the same group of people who passed 3 examinations (primary, after 1 and 2 years of follow-up) elucidated the dynamics of risk factor levels on the background of intervention.

During the first two years AH, smoking and hypercholesterolemia dropped in all three centers. AH decreased by 7.8% in Moscow, by 6.1% in Kaunas and by 9.8% in Minsk (Figure. 5). The decrease in the spread of AH is accompanied by significant lowering of mean levels of systolic and diastolic AP in populational groups. Even in the first year the number of people subjected to effective treatment considerably increased: Moscow (to 48%); Kaunas (to 51%); Minsk (to 37.7%).

Smoking (Figure. 6) in Moscow, Kaunas and Minsk dropped by 7.9%, 8.1%, 14.8%, respectively. In subjects who gave up smoking external breathing indices (spirometry data) and tolerance to physical load (cycloergometry data) significantly improved even during the first year.

In Moscow, Kaunas and Minsk hypercholesterolemia (Figure. 6) decreased by 4%, 4.9% and 8.4% respectively.

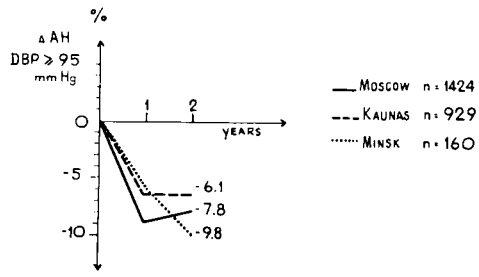

Fig. 5. Dynamics of the prevalence of risk factors in the active prophylaxis group (Figure 5,6,7).

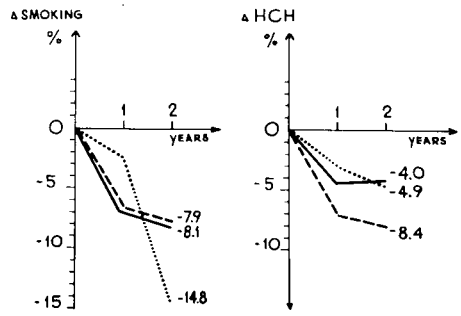

Fig. 6.

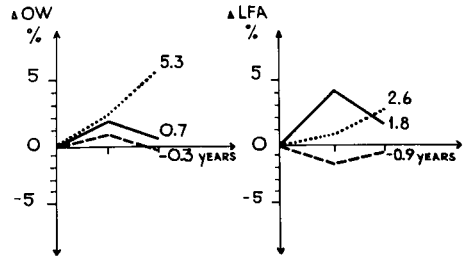

Fig. 7.

As far as excess body weight and low physical activity are concerned, different trends were observed in 3 centers. The first year in all three centers saw an increase in the number of people with EBW: in Moscow and Minsk - by 2.8%, in Kaunas - by 0.7%. During the second year in Moscow and Kaunas the trend to weight reduction appeared: in Kaunas EBW dropped below the baseline and in Moscow returned to it. At the same time EBW increased in Minsk.

No doubt, it is still too early to estimate the efficacy of preventive measures. It needs a longer observation of a larger

population and a thorough comparison with the control. This report constitutes an attempt to demonstrate the trends in the dynamics of major risk factors that were found in the first years of multifactor program inplementation.

The survey continues and its results will be evaluated and summed up in the future.

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EFFECT OF DIET AND SMOKING INTERVENTION

ON THE INCIDENCE OF CORONARY HEART DISEASE

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Improved hospital care has lowered hospital mortality from acute coronary heart disease (CHD). However, the goal of a sizeable reduction in the prevalence of CHD in young and middle age can only be achieved by postponing or preventing the disease. Intervention trials have been carried out in order to be able to demonstrate that such prevention is possible. These trials have not shown unequivocal results, except for the beneficial effect of antihypertensive treatment on prevention of stroke.

No intervention trials on healthy middle aged subjects at high risk of CHD have earlier been carried out. The purpose of our trial was to find out whether the lowering of high levels of blood lipids by dietary changes and the cessation of smoking, if maintained for many years, would reduce the incidence of first attacks of CHD in men aged 40-50 years.

All Oslo men aged 40-50 years were invited to a screening for coronary risk factors.[1] 65% (16202 men) attended the screening and from this cohort men with high risk were selected for a controlled trial[2] if they had serum cholesterol levels (mean of two measurements) of 7.5 - 9.8 mmol/l, coronary risk score (based on cholesterol levels, smoking habits and blood pressure) in the upper quartile of the distribution, and systolic blood pressure (mean of two measurements) below 150 mm Hg. A full clinical examination and exercise ECG was performed. Those with cardiovascular disease, diabetes, cancer, disabling disease, psychopathological disease and those already on lipidlowering diet were all excluded before randomization.

1232 men satisfied the inclusion criteria and were randomized to the intervention (n=604) and control group (n=628).

Table 1 shows that the groups were well comparable before start. The men in the intervention group were all subjected to dietary advice in order to lower their elevated blood lipids. Antismoking advice was given to all smokers. Dietary advice was given individually and based on each man's diet record, on body weight, serum cholesterol and triglyceride levels and his general background. For those with high serum cholesterol, without elevated triglyceride levels, the diet change mainly consisted of a reduction in saturated fat. In addition total energy intake was reduced in those who also had elevated triglycerides. The wives of the subjects in the intervention group were invited in groups of 30-40 together with their husbands for diet and smoking information. Other risk factors were not subjected to intervention.

Follow-up examinations were made every 6 months in the intervention group and every 12 months in the controls. A short clinical examination was made, 12 had ECG recorded, and at each follow-up the men in the intervention group were asked about their eating and smoking habits. A cholesterol curve was made for each man and shown to him.

Follow-up compliance was good. Only 1 control subject and 9 subjects from the intervention group refused to attend at the final exercise - ECG examination. For only 5 subjects of those still alive at the end of the observation period, information was lacking.

An attempt was made to separate the effects on CHD incidence of reducing cigarette consumption and of reducing serum cholesterol. The analysis included a Cox proportional hazard model with the following explanatory variables: change in serum cholesterol, change in cigarette consumption, initial serum cholesterol, initial cigarette consumption and initial age. An extensive statistical analysis has been made elsewhere.[3]

The cardiovascular events counted were fatal and non-fatal myocardial infarction, sudden death and stroke. All events were diagnosed by a diagnostic board not involved in the study and ignorant of the group to which the men belonged. Detailed diagnostic criteria had been listed before the start of the trial.

Results. The effects on serum cholesterol and smoking are shown in Figures 1 and 2. Before randomization, 3 determinations of serum cholesterol were made. As can be seen from the figures, the pre-randomization level of these two risk factors were similar. Immediately after randomization, the efforts started to lower serum cholesterol and cigarette consumption in the intervention group.

In the intervention group there was a reduction of 17% of mean serum cholesterol from screening to first follow-up. The mean difference in serum cholesterol between the groups during the study was

Table 1. Comparability of Study Groups before Trial

	Intervention group (n=604)	Control group (n=628)
Sex	Male	Male
Age (mean and range;yr)	45.2(40-49)	45.2(40-49)
History/symptoms of CHD	None	None
Mean daily cigarette consumption	13.0	12.5
Smokers (%)	79.1	79.6
Body weight*(kg)	77.3±10.3	78.2±9.8
Height (cm)	177.4±6.0	176.9±6.3
Serum cholesterol*(mg/dl)		
Screening examination	328.2±26.9	329.2±27.5
1st re-examination	322.7±27.6	322.5±28.9
Range (mean of these 2 examinations)	290-379	290-379
Serum triglycerides*(mmol/l)		
Screening examination	2.80±1.5	2.84±1.5
1st re-examination	2.21±0.9	2.25±1.1
SBP (mm Hg)	<150	<150
% with SBP≥150 mm Hg and/or DBP≥98 mm Hg at screening	23	20
Sedentary workers (%)	50	48
Diet score*	14.8±6.1	14.1±6.1

*Value±1 SD. Subjects were non-fasting at the screening examination and fasting at the 1st re-examination. SBP=systolic blood pressure; DBP=diastolic blood pressure.

13%. Tobacco consumption (expressed as number of cigarettes per man per day) fell about 45% more in the intervention group than in the controls. However, only 25% of the smokers in the intervention group completely stopped smoking as compared with 17% in the control group.

Mean of fasting and non-fasting serum triglycerides levels was 20% and 25% lower in the intervention group than in the controls during the trial. In a subgroup of good diet responders it was shown[4] that the ratio of high density lipoprotein (HDL) cholesterol to the other serum cholesterol fractions after 4 years of intervention was 66% higher than in controls matched with respect to initial HDL cholesterol, triglycerides, cholesterol, body weight, cigarette smoking, level of physical activity and diet score, all recorded before randomization.

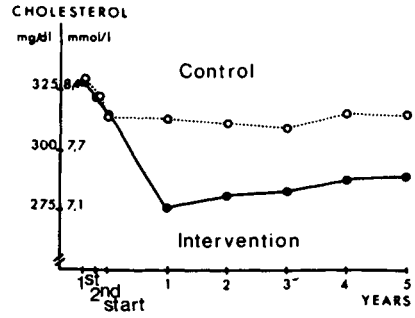

Fig. 1. Intervention effect on serum cholesterol. Oslo Study.

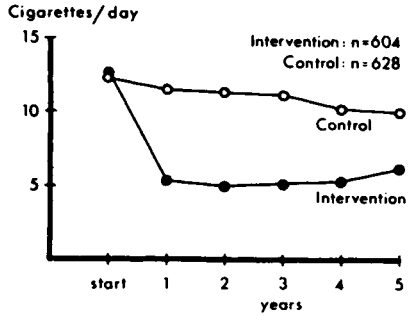

Fig. 2. Intervention effect on cigarette smoking. Pipe smoking is included: 50g pipe tobacco/week equals 7 cigarettes/day. Oslo Study.

The incidence of fatal and non-fatal MI and sudden death is shown in Figure 3 as a life table. The difference between the intervention and control group is significant, $p = 0.03$, two-tailed test, (one-tailed test should also be accepted). In Table 2 the different cardiovascular events in the two groups are listed. The endpoints sudden death, sudden coronary death, major CHD (sudden coronary death + MI) and total cardiovascular events show significant differences between the two groups. Table 3 shows no significant difference for total mortality but a trend is present in the same direction as for the cardiovascular events. It should be emphasized that the trial was not dimensioned for showing differences in mortality. When evaluating the trial, it is important to note that the trend for mortality is in the same direction as the difference in CHD-incidence. And if a one-tailed test is used (which should be acceptable for this trial), the difference between the groups for total coronary death is also significant.

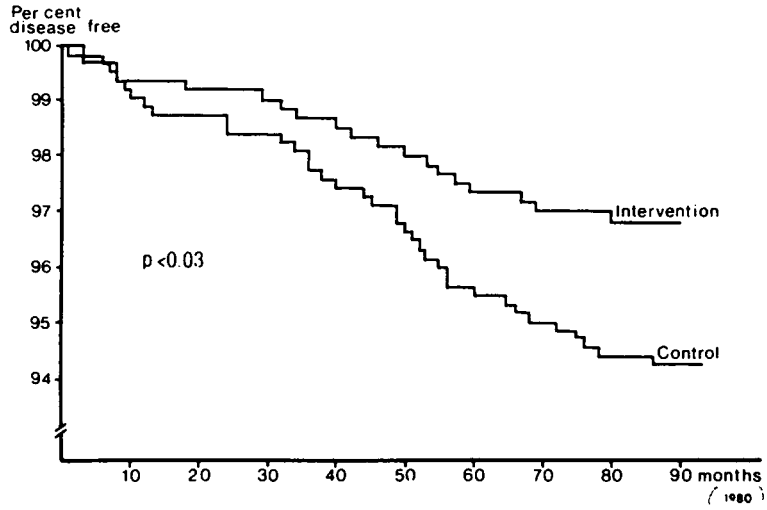

Fig. 3. Life table analysis of CHD (fatal and non-fatal myocardial infarction and sudden death) in intervention and control groups. Oslo Study.

Table 2. Cardiovascular Events

	Intervention group (n=604)		Control group (n=628)		
	No events	Rate (per thousand)	No events	Rate (per thousand)	p*
Sudden coronary death	3	5	11	18	..
Sudden unexplained death	0	..	1	2	..
Sudden coronary + unexplained death	3	5	12	19	0.024
Fatal MI	3	5	2	3	..
Fatal MI + sudden death	6	10	14	22	0.086
Non-fatal MI	13	22	22	35	0.153
Total coronary events	19	31	36	57	0.028
Fatal stroke	2	3	1	2	..
Non-fatal stroke	1	2	2	3	..
Total cardiovascular events	22	36	39	62	0.038
Bypass surgery	1	2	3	5	..

*By two-tailed test.

Table 3. Mortal Events

	Inter-vention group (n=604)	Control group (n=628)	p
Sudden coronary death	3	11	0.024
Sudden unexplained death	0	1	..
Fatal MI	3	2	..
Fatal MI + sudden death	6	14	0.086
Fatal stroke	2	1	..
Total cardiovascular events	8	15	0.168
Cancer deaths	5	8	..
Suicide/accidental death	3	1	
Total mortality	16	24	0.246

The two risk factors influenced in this study was smoking and eating habits. Physical activity not influenced, as assessed by a questionnaire.

The men in the intervention group were examined and subjected to intervention every 6 months, but because of capacity problems the controls were examined only every 12 months. This difference between the groups in frequency of medical contact might have favored the intervention group, if a feeling general attention had a reassuring effect. On the other hand, the more frequent medical contact in the intervention group might have led to increased reporting of symptoms and more events may have been diagnosed.

It is important to find out to what extent each of the two factors influenced in this trial was responsible for the observed difference in incidence. As mentioned, a multivariate statistical model was used in an attempt to separate the two factors. Changes in serum cholesterol appeared to correlate more closely with CHD incidence than did the change in cigarette consumption. The statistical model predicted that the changes in serum cholesterol accounted for about 60% of the reduction in CHD incidence, whereas, most optimistically, the cigarette factor accounted for 25%.

Thus, although a substantial reduction in cigarette consumption was achieved during the trial, it was not large enough to create a significant reduction in risk. The statistical power of the trial was, in this respect, too low. The effect of change in smoking was also less than predicted from earlier observational studies in Oslo. It is possible that observational studies have over-estimated the decline in risk of CHD brought about by reducing (not quitting)

cigarette smoking. This is in accordance with an earlier monofactorial, controlled trial on smoking reduction.[5]

It is concluded that in healthy men aged 40-49, at high risk of CHD advice to change eating habits and to stop smoking significantly reduced the incidence of first event of major CHD. Statistical multivariate analysis indicate that the reduction in incidence in the intervention group is correlated with the reduction in serum cholesterol and to a lesser extent with smoking reduction.

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THE PRESENT STATUS OF CARDIOVASCULAR MEDICINE IN AFRICA

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The three dominant groups of cardiovascular conditions in Africa today are: Hypertension, Rheumatic Heart Disease and the Cardiomyopathies (including Endomyocardial Fibrosis). Less common are congenital heart disease, cor pulmonale, pericarditis, the arrhythmias, anaemia or beriberi heart disease, ventricular aneurysms and idiopathic aortitis. It is worthy of note that Ischaemic Heart Disease remains distinctly uncommon among indigenous African communities, although sporadic cases are now beginning to emerge in the higher socio-economic strata of certain urban populations[1-3].

Hypertension in Africa forms part of a world-wide aetiopathogenic problem. In the majority of African populations (rural and urban) mean blood pressure rises with age and the prevalence rate of arterial pressure differs little from that in black and white communities in other parts of the world[4-10]. However, pockets of population exist in Africa and elsewhere, notably in the rural setting, in whom the blood pressure does not appear to rise with age[11-14]. Most reported studies relate this phenomenon to diet, life-style and the environment. Much has been made of the preliminary observations that when Masai tribesmen in the rural north of Kenya were recruited into the army and migrated to urban Nairobi, their gain in weight was followed some months later by a gradual rise in mean arterial pressures, and that these pressures fell in those who subsequently returned to their former habitat on demobilization[15].

The lifestyle in most African cities is undergoing remarkable transformation. Although the role of such acknowledged risk factors as atherosclerosis, tobacco, alcohol, lack of exercise, diet and salt in the genesis of hypertension is still to be critically assessed in the context of Africa, there is little doubt that the natural history

of hypertension continues to be affected by the changing socio-economic circumstances of those communities.

Cardiac ischaemic complications are as rare as cerebrovascular are common[16,17]. The low incidence of certain atherosclerotic risk factors may be partly responsible for this apparent immunity of black communities to heart attacks - relatively low levels of plasma triglycerides, cholesterol and high-density-lipoprotein cholesterol as compared with whites[18,19]. Haemorrhagic cerebrovascular complications occur more frequently in blacks than in whites; this is probably attributable to the height of the blood pressure itself[20]. The apparent rarity of severe retinopathy in the hypertensive African constitutes yet another conundrum[21].

Hormonal studies have been undertaken in several biracial population groups but the results are often difficult to interpret[22,23]. Renin suppression is a common finding in black hypertensive populations but sympathetic activity as determined by plasma levels of noradrenaline is inconsistent. Studies in blacks outside Africa have confirmed lower levels of dopamine β-hydroxylase than in whites, but its significance remains doubtful[24].

Another area worthy of more attention is the role of sodium and potassium[25-27]. It is clear that dietary salt excess alone cannot be held as a major contributory factor to hypertension in black Africa, nor can its lack be directly related to the low prevalence of hypertension in certain defined tribal societies on the continent[22]. Of more interest in recent times are observations on the cellular transport of sodium and potassium in normotensive and hypertensive black communities[28]. These communities have a significantly reduced ouabain-insensitive sodium/potassium pump activity compared to white normotensives, implying a diminished cellular capacity to eliminate sodium. Differences have also been observed in urinary sodium/potassium ratios as between blacks and whites[29]. All these suggest a genetic basis and much further work needs to be done to unravel any cause-effect relationships.

Rheumatic Heart Disease (RHD), an eminently preventable condition constitutes a major cause of cardiovascular morbidity and mortality in developing countries[30]. It occurs in a significantly higher proportion of children and adolescents in Africa than in industrialized societies. Refinements in the clinical diagnosis of complicating arrhythmias or established valvular disease (phonocardiography, echocardiography, catheterisation, coronary arteriography etc.) are still confined to very few cardiological centers in Africa - within or attached to Teaching Hospitals. So also are such specialized procedures as cardiac pacing, valve replacement, coronary bypass and open heart surgery. The major thrust in the control of RHD is obviously the integration, however modest, of preventive measures with national and primary health care programs[31]. Infective endocarditis complicating known RHD needs to be distinguished from the persistent fever

in RHD itself, coexistence of RHD with haemoglobinopathy, or fever following urological instrumentation. Staph. aureus is more commonly encountered than Strept. viridans, or there may be such unusual organisms as Strept. faecalis, Haemophilus parainfluenza, Pseudomonas, Klebsiella and Escherichia coli[32]. Primary sites of infection include skin and scalp sepsis, osteomyelitis or pyomyositis.

Classification of the Cardiomyopathies has only recently been tidied up, and the conflict of terminology that has always bedevilled the subject is now hopefully resolved[33]. Congestive (dilated) cardiomyopathy is easily the dominant type of myocardial disease of unknown cause on the African continent. Alcohol, malnutrition and hypokalaemia have been variously blamed as aetiological factors but evidence remains unconvincing[34-36]. Peripartal cardiomyopathy[37] is but a facet of this "congestive" variety, and its descriptive appellation merely signifies its occurrence in or around pregnancy, often in situations associated with volume overload or vitamin deficiency. Toxoplasma[38] or Coxsackie B viruses[39] are infections often associated with the presence of congestive cardiomyopathy, but the new classification rightly places these infections in the category of "Specific heart muscle disease of known cause". The relationship between cardiomyopathy and hypertension has often led some investigators to regard congestive cardiomyopathy as synonymous with hypertensive heart failure[40], but haemodynamic studies would seem to refute such hypothesis.

Restricted cardiomyopathy manifests in the tropical milieu essentially as Endomyocardial Fibrosis (EMF)[41]. Some would regard Loeffler's cardiomyopathy as an early phase of EMF, and have even attempted to rename EMF "Eosinophilic endomyocardial disease"[42].

Health care systems in Africa, beset as they are by the burden of infectious disease, are yet to be fully alive to the problems that cardiovascular diseases are destined to pose in the coming decades. The answers to many of the intricate problems in the aetiology and pathogenesis of hypertension, ischaemic heart disease and cardiomyopathy in Africa will come, not through superficial surveys of populations or half-hearted and poorly supported haemodynamic studies, but through well-ordered, in-depth clinical experimental and epidemiological application. Inter-regional scientific collaboration is thus vital as the ultimate realization of these goals.

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CHAGAS' CARDIOMYOPATHY

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Chagas' disease is restricted to the American continent and has been identified from the southern United States to Argentina[1]. Its epidemiology is related to poor housing in the rural communities and to trypanosoma cruzi infected triatominae. Less frequently the disease may be transmitted through blood transfusion, congenitally or by accident in the laboratory[2]. It is estimated that a total of 7 million people are infected[3].

Chagas' cardiomyopathy, however, is highly prevalent only in Argentina, Brazil and Venezuela. In Bahia, Brazil, at the University Hospital, it is responsible for 42.4% of all cardiac deaths. In a few cases, it may be preceded, 10 to 30 years ago, by an acute myocarditis with complete recovery in 90.0% of the cases and death in 10.0%. Forty six percent of such cases occur in the first decade of life[4].

Basic pathologic changes are related to a diffuse chronic myocarditis. The degree of involvement of the ventricular myocardium and/or the excitation-conduction system of the heart is a major determinant of the type of clinical presentation. Extensive myocardial damage leads to progressive dilatation of the heart with congestive heart failure (CHF). Lesions of the excitation-conduction system are common and give rise to the rhythm and conduction disturbances frequently seen in this condition[5].

The clinical presentation of chronic Chagas' cardiomyopathy comprehends asymptomatic patients with electrocardiographic changes and no cardiomegaly to patients with symptomatic cardiac arrhythmias and/or CHF[6]. These latter patients are more common (71.1%) between the 3rd and the 5th decades of life, with a 2.7 male to female ratio (Table 1). Complete right bundle branch block (CRBBB), isolated or

Table 1. Sex and age distribution in Chagas' cardiomyopathy[a]

Age yrs.	11-20	21-30	31-40	41-50	51-60	>60	Total
Male	13	16	12	20	9	3	73
Female	1	8	8	7	2	1	27
Total	14	24	20	27	11	4	100

[a]Data of 100 patients who died with advanced Chagas' cardiomyopathy at the University Hospital, Bahia, Brazil.

associated with left anterior hemiblock (LAHB), is the most frequent and characteristic ECG abnormality, occurring in 30 to 60% of the cases[7].

Ten percent or less of patients with Chagas' cardiomyopathy may present with total A-V block, requiring pacemaker implantation[8]. In those with a normal or mildly enlarged heart this procedure seems to improve survival in addition to symptomatic control. Improvement of survival, however, doesn't occur in some patients with a moderately to severely enlarged heart, whose CHF shows a progressive downhill course immediately after pacemaker implantation. In order to understand how the heart of these patients responds to ventricular pacing, 9 of them were studied in our laboratory. They were divided in 2 groups according to their left ventricular end-diastolic pressure (LVED): Group I - 5 patients, LVED from 10.5 to 14.9 mm Hg, mean 12.7 mm Hg; Group II - 4 patients, LVED from 20.8 to 26.6 mm Hg, mean 23.5 mm Hg (Table 2). During pacing, G.I. patients showed a uniform LVED decrease between idioventricular rate and 100 b/min pacing rate, ranging from 3.7 to 11.8 mm Hg, mean 9.0 mm Hg. In G.II, LVED decreased in 3 patients, reaching normal values in nos. 1 and 3 (10.5 and 13.2 mm Hg) and remaining elevated in no. 2 (22.3 mm Hg); in patient no. 4 it rose from 25.8 to 32.5 mm Hg. The lowest LVED was reached at pacing rates equal or above 80 b/min in all but patient no. 5 (G.I.), in whom it happens at 70 b/min. Although the still limited number of patients studied, this data suggests the following: 1) artificial pacing seems to improve left ventricular function of total A-V block chagasic patients with a normal, mildly or moderately elevated LVED; 2) this improvement seems to be maximal at pacing rates equal or above 70 b/min; 3) in patients with severely elevated LVED, left ventricular function doesn't improve or may even become worse. Based on still very limited data, it has been claimed that 50 b/min is the best pacing rate for these latter patients[9]. Our data doesn't confirm this assertion, but further studies using larger series of patients are necessary to clarify this important point.

Table 2. Hemodynamic data in chagasic total A-V block[a]

Groups	Patient No.	Basal HR (b/min)	Basal LVED (mmHg)	Pacing (b/min) LVED (mmHg) 60	70	80	90	100	LVED Gradient Basal-100 (b/min)
I	1	33	13.4	4.4	4.0	4.3	2.1	1.6	11.8
	2	20	11.2	7.2	7.2	6.2	5.2	3.3	7.9
	3	30	10.5	6.8	6.6	6.3	6.1	6.8	3.7
	4	32	13.5	9.8	5.1	6.2	3.3	1.8	11.7
	5	35	14.9	4.6	2.1	3.6	3.8	4.8	10.1
II	1	27	20.8	13.4	10.5	9.5	9.5	10.5	10.3
	2	25	26.6	20.7	20.7	19.2	19.2	22.3	4.3
	3	20	20.8	16.3	14.9	12.4	11.3	13.2	7.6
	4	32	25.8	22.5	23.2	21.7	24.1	32.5	-6.7

[a]Left ventricular end-diastolic pressure (LVED) response to pacing rates 60 to 100b/min of 9 patients divided in Groups I and II according LVED basal level

Ventricular arrhythmias are one of the major problems to deal with in patients with Chagas' cardiomyopathy. In endemic areas sudden death is a common cause of death among young and middle aged adults. The presence of these arrhythmias doesn't keep any relationship with heart size; patients with normal heart size may present with life threatening ventricular arrhythmias.

Although there are no definitive data for identification of high risk sudden death patients with this condition, the presence of couplets or salvos in the standard ECG has been considered as an indication for permanent antiarrhythmic treatment. In this regard the sensitivity of the 12 leads ECG with a 30" V_1 rhythm strip was compared with that of the 24:00 hs Holter monitoring[10], a high sensitive technique but too much expensive and time consuming to be used on a large scale basis, especially in rural communities. Three groups of chagasic patients were studied: Group I - normal ECG (N = 43); Group II - conduction defects but no ventricular premature beats (VPB's) (N = 29); Group III - one or more VPB's (N = 16). Age ranged from 18 to 55 years, mean 38.4 years; there were 27 females and 12 males. The number of couplets and salvos in the 24:00 hs. Holter increases sharply from Groups I and II to G.III (Table 3). In G.I., the number of couplets ranged from 1 to 7, mean 1.7, in G.II from 1 to 38; mean 9.4, and in G.III from 2 to 5.749, mean 563.1. Salvos occurred only once in G.I. and G.II patients, but in G.III ranged from 1 to 855, mean 76.1. Based in these findings, it was possible to evaluate the predictive value of the 12 leads ECG in relation to the 24:00 hs Holter (Table 4). So, in G.I. patients 11.6% will have chance to present with couplets or salvos in the 24:00 hs Holter, in G.II 24.1%, and in G.III 92.0%. Then, the existence of couplets and/or salvos should be strongly suspected in chagasic patients with one or more VPB's in the standard ECG.

Low adherence to treatment secondary to poor socio-economic conditions, and drug selection make antiarrhythmic treatment a difficult task. Any drug that decreases contractility or conduction should be avoided or used with utmost precaution in this condition, especially in patients with a dilated heart.

At the present moment, amiodarone is the most useful drug for the treatment of VPB's in Chagas' cardiomyopathy. Its efficiency and safety has been documented elsewhere[11] and in our laboratory in 25 patients[12], divided in Group I, 19 patients with at least couplets in the one hour sedentary ECG, and Group II, 6 patients with the same arrhythmia only on exercise. In G.I., after 10 days of 600mg daily of oral amiodarone, the mean total number of VPB's decreased from 393.4 to 52.5 ($p<0.01$), and remained stable until the end of the study, on the 22nd day. In G.II, this number decreased from 65.2 to 2.2 ($p<0.05$) during this latter period of time (Table 5). From the qualitative stand-point, amiodarone was able to suppress the more

Table 3. Ventricular premature beats (VPB's) in Chagas' cardiomyopathy

	GROUP[b]					
	I(N=43)		II(N=29)		III(N=25)	
	C	S	C	S	C	S
Range	1-3	1	1-38	1	2-5,740	1-855
Mean	1.7	1	9.4	1	563.1	76.1

[a]Frequency of couplets (C) and salvos (S) of VPB's in the 24:00 hs Holter monitoring of 97 patients.

[b]Groups selected according the 12 leads ECG with a 30 seconds V_1 rhythm strip: G.1 = normal ECG; G.II = conduction disturbance, no VPB's; G.III = VPB's

Table 4. Predictive value of the standard ECG in Chagas' cardiomyopathy[a]

VPB's[b]	VPB's in the 24:00 hs Holter		
	G.I.(%)	G.II.(%)	G.III(%)
At least 1	36(83.7)	26(90.0)	25(100.0)
Couplets (C)	3(7.0)	7(24.1)	23(92.0)
Salvos (S)	2(4.7)	1(3.4)	16(64.0)
C + S	5(11.6)	7(21.4)	23(92.0)

[a]Same groups as in Table 3.

[b]Number and type of VPB's in the 12 leads ECG with a 30 seconds V_1 rhythm strip.

Table 5. Antiarrhythmic action of oral amiodarone (600 mg daily) in Chagas' cardiomyopathy

Treatment Day	Mean total of VPB's[a]			Extrassystolic Index		
	Range	Mean	SD	Range	Mean	SD
			GROUP I (N = 19)			
Before	90-1,044	393.4	249.8	2.0-23.0	10.1	6.8
10th	0-278	52.5[b]	97.3	0-10.6	1.8[b]	3.6
22nd	0-603	69.9	160.3	0-21.6	2.5	0.7
			GROUP II (N = 6)			
Before	35-177	65.3	55.3	2.7-19.2	9.7	6.0
22nd	0-8	2.2[c]	3.5	0-1.2	0.3[c]	0.5

[a] VPB's were counted in one hour of sedentary ECG monitoring (paper speed of 10min/sec) in Group I and during bicyclergometer exercise test in G.II.

[b] $p < 0.01$

[c] $p < 0.05$

complex forms of VPB's (R/T, couplets and salvos), both at rest and on exercise, in all but one G.I. patient, who still showed couplets on exercise even after further 10 days of 800 mg of amiodarone per day.

Amiodarone also decreased significantly the heart rate ($p<0.01$) and increased significantly the QT interval ($p<0.05$), but didn't cause any change in the PR interval whatsoever. In one patient with recurrent ventricular tachycardia this amiodarone induced sinus bradycardia was so marked that required pacemaker implantation in order to continue drug administration. In general, drug tolerance has been good, except for a patient with severe partial loss of vision after 3 months on a 600 mg daily dosage, but who recovered completely after withdrawal of the drug.

The treatment of advanced CHF in Chagas' cardiomyopathy is another challenge for the clinician. These patients are very sensitive to digitalis, so, usually, only half to one third of the regular dosage can be employed. The use of vasodilators, specially prazosin, has been of great help in management of such patients. Nevertheless, diuretics continue to be the basis of anti-congestive treatment, with refractory patients showing a very good response to the association of furosemide, hydrochlorothiazide and spirolactone.

Another great problem concerns the natural history of seropositive individuals without or with ECG changes like bundle branch block or primary ST-T wave changes and a good functional heart. The potential risk of severe heart disease in these individuals has created serious problems regarding employment opportunities.

Finally, the greatest challenge will be the eradication of the disease through better socio-economic conditions for the rural inhabitants and through a triatominae extermination program in order to keep the houses free of the vectors of the diseases.

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CARDIOVASCULAR DISEASE IN DEVELOPING COUNTRIES

RHEUMATIC HEART DISEASE

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PREVALENCE

Since World War II, published literature points to a high prevalence of RHD in developing countries, which contribute to two thirds of the world population. Rheumatic heart disease is the commonest type in children and adolescents and one of the most frequent in adults. For many parts of Latin America (Brazil, Chile, Columbia, Guatemala, Mexico, Panama, Peru), the West Indies, in the Middle East (Algeria, Cyprus, Egypt, Morocco, Sudan), in Ethiopia, Nigeria, Senegal, Iran, Pakistan, India, Burma, Hongkong, Indonesia, Thailand, Sri Lanka and Mongolia this holds true[1].

The sources of data unfortunately for RHD are mostly from surveys in vulnerable groups such as school children and not from autopsy or death statistics, as the latter are unreliable or unobtainable in these countries. Hospital statistics point to RHD accounting for 22 to 50 percent of all cardiac cases. In school children the prevalence ranges from 6 to 22 per 1000. The position is similar to what is obtained in Western countries e.g. the UK and USA at the turn of the century[1].

The incidence of acute rheumatic fever declined in the West long before the antibiotic era. The incidence of acute RF in developing countries is not clearly known because such patients do not reach hospital except in the cities. The symptoms of acute RF are no different from that described in Western literature, in those that reach hospital[2].

It has been pointed out that RHD in the developing countries shows special clinical features, viz. accelerated course, multi-valvular lesions, congestive heart failure, severe tricuspid insufficiency and "juvenile mitral stenosis". The Jones' criteria appears to be as applicable in developing countries as in the West[2].

NATIONAL AND INTERNATIONAL PROGRAMS FOR RHEUMATIC FEVER CONTROL

In recent years developing countries, with and without the aid of WHO have made co-ordinated or individual efforts for the study and control of the problem of RF and RHD.

A collaborative study involving seven centres was conducted between 1972 and 1979 with WHO collaboration[3,4,5]. A similar program was followed in Latin-American countries during the years 1975-1976. A WHO meeting on the "Community control of RF and RHD" was held in New Delhi in 1979. An Indo-US Conference-Workshop on "Rheumatic Fever in the 1980's" was held in 1981 in New Delhi, India under the auspices of the Indian Council of Medical Research, All India Heart Foundation and the National Heart Lung and Blood Institute (USA)[6].

A national program for control of RF and RHD was started in Japan in 1969. A South-East Asia RF and RHD Prevention Conference was held in Japan in 1973. At the present time three countries - Japan, Indonesia and the Philippines are carrying out a cooperative study for isolation of streptococci, their grouping and typing, from the throats of school children during various seasons and for determination of prevalence of RF and RHD[7].

India has its own programs for RF control, some of the results of which will be discussed later in this paper. These programs started in 1966 are:

- A collaborative study on prevalence of RHD in school children.
- A secondary prophylaxis pilot program for RF and RHD.
- A RF criteria study.
- A longitudinal study on 2000 children at 6 monthly intervals for evidence of streptococcal infection, RF and RHD: factor analysis of prevalence, evaluation of current methods of surveillance and research into alternative approaches for control.

STUDIES IN INDIA

Prevalence

It is believed that there are some 6 to 8 million children in India with RF and RHD today. The prevalence from school surveys is 6 to 11 per 1000 with a national average of 6 per 1000. Delhi has

the highest figure of 11 per 1000 school children between 5 and 15 years. Hospital admissions account for 33 to 50 percent of all heart cases. Surveys in adult populations point to a figure of 123 to 200 per 1,000,000. The differences in clinical and epidemiological features between developed and developing countries has been brought out in recent publications[1,4].

Table 1. RHD in India , Age and Sex distribution of 885 cases.

Age Group	Male	Female	Total
Up to 5 yrs.	-	-	-
5-9 yrs.	97	47	144 (16.3%)
10-14 yrs.	323	215	538 (60.8%)
15-19 yrs.	86	84	170 (19.2%)
20 yrs. and above	19	14	33 (3.7%)
Total	525 (59.37%)	360 (40.7%)	885

Table 2. RHD in India, Socio-economic Status of 885 cases.

Income group	No. of Patients	% age
100 or less	6	81.1
101 - 300	293	
301 - 500	358	
501 - 700	61	
701 - 1000	112	12.7
1001 - 1500	38	4.3
1501 - 2000	15	1.7
2000 and above	2	.2
Total	885	100.0

Table 3. RHD in India, Rheumatic Manifestations and Rheumatic Recurrences.

Total No. of Cases	885	
	First Attacks	Recurrence
Arthritis	323 (36.5%)	169 (19.1%)
Acute Carditis	247 (27.9%)	131 (14.8%)
Chorea	40 (4.5%)	17 (1.9%)
Subcutaneous nodules	14 (1.6%)	9 (1.0%)
Polyarthralgia	406 (45.9%)	352 (39.8%)
Erythema marginatum	Nil	Nil

Table 4. RHD in India, Applicability of Jones' Criteria.

	No. of cases	% age
Total No. of cases	885	
I. Patients satisfying Jones' criteria.	514	58.1%
a. Two or more major criteria	96	10.9%
b. One major with two minor criteria.	418	47.2%
II. Patients not satisfying Jones' criteria.	371	41.9%

Epidemiological and Clinical Features

The facts given in the following paragraphs are from a group of 885 children being followed up at the present time in Delhi. These children were referrals from School Health Services, general practitioners or hospital discharges. The age distribution was highest in the 10 to 14 year group. The sexes showed a slight male preponderance. The socio-economic status was the lowest income group in 80 percent of patients. Rheumatic manifestation in first attacks and recurrences followed the same pattern with polyarthralgia in the majority followed by arthritis and acute carditis. Nodules and Chorea were infrequent and no case of erythema marginatum was seen.

Table 5. RHD in India, Break-down of 885 Cases by Severity of Disease*

	No. of cases	% age
Group I		
Arthritis & Chorea	121	13.7%
Group II		
Carditis alone or associated with arthritis, Chorea.	396	44.7%
Group III		
With no CHF or carditis	321	
With CHF alone	41	
With CHF & carditis	2	
With carditis alone	4	
Total	368	41.6%
Total	885	

*Ref. Circulation 32,457. 1965.

Table 6. RHD in India, Valvular Lesions in 885 Cases.

Valvular Lesion	Male	Female	Total
MS	79	50	129 (14.6%)
MR	211	120	331 (37.4%)
MS + MR	88	74	162 (18.3%)
AR	9	8	17 (1.9%)
Combined aortic & mitral valve lesion	75	50	125 (14.1%)
No valvular lesion	63	58	121 (13.7%)
Total	525	360	885 (100.0%)

The Jones' criteria were applicable in 58 percent of patients with 11 percent showing two major criteria and 47 one major and two minor criteria. Nearly 42 percent of patients did not satisfy the Jones' criteria on admission to the study but in view of the low economic status of most of these patients it is possible that minor illness was not taken note of.

The severity of disease on entrance into the study was Group III in 42 percent, Group II in 45 percent and Group I (with no carditis) in 13 percent according to the criteria used in the UK-USA combined trial[8].

Of the valvular lesions, the order of frequency was mitral insufficiency followed by MS and MI, pure MS and combined mitral and aortic valvular lesions. Lone A I was present only in a small number. 2 percent of patients had a familial pattern with two or more members of a family being affected.

LABORATORY DATA

Throat Cultures

Beta haemolytic streptococci was isolated in 211 out of 885 cases on entry into the study (23.9 percent). The largest number of cases were of Group A, 91 (43.13 percent), followed by Group G, 83 (39.34 percent).

Table 7. RHD in India, Beta Haemolytic Streptococci Isolations in 885 Cases.

Group	No. of Cases	Total isolations	Serological Groups			
			A	B	C	G
Regular	703	133 (18.92%)	57 (42.86%)	2	20	54 (40.60%)
Irregular	182	78 (42.89%)	34 (43.59%)	3	12	29 (37.19%)
Total	885	211 (23.89%)	91 (43.13%)	5	32	83 (39.34%)

The dominant T types were 3/13/B-3264 followed by 5/11/12/27/44 and 8/25/Imp.19.

M typing could only be done in 20 percent of Group A strains, most of whom were positive for serum opacity reaction. This last is believed to be an epidemiological marker. M-types isolated were 1, 3,11,12,22,31,60,52,56,63 and 28. No prevalent M types were found.

Table 8. RHD in India, ASO Titre/Beta Haemolytic Streptococci in 885 Cases

Clinical Groups	No. of Cases	ASO Levels			
		200	200-400	400-596	596 & above
I. RHD cases without isolations of Beta-haemolytic strepto-cocci	674	409	179	66	20
II. RHD cases with isolations of Beta haemolytic strepto-cocci.	211	133	56	15	7
Isolations:					
A	91	91	25	9	5
B	5	4	1	-	-
C	32	24	7	1	-
G	83	53	23	5	2

Serology

High ASO titres were obtained in 343 cases (38.8 percent). In 265 cases (29.9 percent) there was no positive throat culture. With the use of both ASO and ADnase-B estimations it was possible to detect 578 (65.3 percent) cases of streptococcal infection whereas by the use of ASO alone this was possible only in 38.8 percent and by the use of ADnase-B alone only in 235 (26.5 percent) cases. Antipolysaccharide antibodies showed high values in 60 percent of cases with valvular involvement.

IMMUNOLOGIC STUDIES

T & B Lymphocytes

Peripheral blood T & B lymphocytes were studied in cases of acute RF (36), chronic RHD (25), acute streptococcal pharyngitis (37) and in 24 normal controls. A distinct depression of T cells was observed in cases of acute RF and acute streptococcal pharyngitis ($P<.05$). Statistically significant elevation of B cells was noted in acute RF RHD and acute streptococcal pharyngitis cases.

A positive humoral response was seen in cases who had elevation of B cells in acute RF and RHD cases. No such correlation could be made out in cases of acute streptococcal pharyngitis[9].

HLA Mapping

HLA mapping was done in 22 cases of acute RF and RHD and 18 normal controls. DR typing showed 55 percent cases of RHD, RF to be positive for 885 sera-DR specificity undefined. In the normal population of the same age group this type of genetic marker was seen in 16.6 percent. The HLA types reported from other centers in India for cases of acute RF showed HLA-A 28 in 48.2 percent (14.4 percent in controls), HLA B 5 in 48.2 percent (28.8 percent in controls), HLA-B 18 in 14.8 percent (4.4 percent in controls). This work is being continued in a larger number of cases.

VALIDATION OF JONES' CRITERIA

In 162 cases with acute major manifestations (carditis, polyarthritis, nodules, chorea and arthralgia) studied at Delhi, ESR and ASO elevation were present in 98.6 percent to 92.6 percent of patients with carditis, polyarthritis and chorea and in 100 percent of patients with subcutaneous nodules. In the case of arthralgia, when accompanied by a rise of ASO and ESR in patients with previous RHD, it was considered a significant symptom of RF. Immunologic studies in these patients have not yielded any new facts.

Seasonal Throat Culture

In a study of 2000 children being carried out now, throat swabs in summer and winter months showed a significant difference in five consecutive surveys, being nearly double in winter months. ($P = <0.01$). This is an important pointer to control measures which may have to be taken for primary prophylaxis.

Table 9. RHD in India, Seasonal Incidence of B-Haemolytic Streptococci along with Group-wise Break-up.

Season	No. of Specimens	Isolation	Serological Groups			
			A	B	C	G
Winter'79	2034	357* (17.6%)	207 (10.2%)	8 (0.4%)	52 (2.6%)	90* (4.4%)
Summer'80	2034	217* (10.5%)	94 (4.6%)	9 (0.4%)	54 (2.7%)	60* (2.9%)
Winter'80	2034	369* (18.2%)	192 (9.4%)	10 (0.5%)	63 (3.1%)	104* (5.2%)
Summer'81	2034	224* (11.1%)	104 (5.1%)	1 (0.05%)	55 (2.7%)	64* (3.1%)
Winter'81	2034	338* (16.6%)	165 (8.1%)	- -	55 (2.7%)	118* (5.8%)

*p = 0.01

COLLABORATIVE STUDY IN ASIA

A similar study has been carried out in 3 countries of South-East Asia (Japan, Indonesia and the Philippines) under a cooperative study in school children between 6 and 8 years of age from 1977. The prevalence of Group A streptococci using identical methods was 10 to 40 percent and was similar among the participating countries. It was significantly higher during the rainy months in Indonesia and the Philippines and high in winter in Japan. A higher prevalence was observed also in the lower income groups in Indonesia and the Philippines during the rainy season. Group A accounted for the largest number of streptococci isolated. There was a variation in predominant T types in the three countries and during the different seasons.

The prevalence of RF was 0.8 to 1 per 1000 in Indonesia and the Philippines and well below 0.1 per 1000 in Japan.

The low prevalence of RF and RHD in Japan compared with the higher rates in Indonesia and the Philippines in spite of the same isolation rates of streptococci in the throat raised questions regarding host response and virulence of bacteria.

CONTROL OF RHEUMATIC FEVER AND RHEUMATIC HEART DISEASE

Primary Prevention

In the Indian experience primary prevention still remains the ideal goal but is hard of achievement, because of the large population involved and the lack of medical facilities over the greater part of the country, in rural and semi-rural areas.

One method of primary prophylaxis to be attempted in India is seasonal prophylaxis of all school children during the months of peak streptococcal infection which may vary in different parts of the country.

The difficulty of recognising streptococcal infection except by culture stresses the need for a quick method of recognising such infection.

SECONDARY PREVENTION

Pilot studies at Delhi and at Hyderabad have conclusively shown the feasibility of secondary prevention even in a developing country.

Prophylaxis has been given in the Delhi clinic regularly from 1966 onwards. In 1975 a change was made from 4-weekly to 3-weekly injections. The differences in the number of positive throat cultures

Table 10. RHD in India, Streptococcal Infection and Rheumatic Recurrence.

	1966-75		1976-81	
	No.	Rate/pt.year	No.	Rate/pt.year
No. of patient years	1656.16	-	3062.5*	-
Positive throat culture	263	0.15	133	0.04
Raised ASO	257	0.155	206	0.06
No. of cases with raised ASO & positive Group A throat culture	118	0.07	22	0.007
Rheumatic recurrences	21	0.012	3	0.0009

*No. of patient years with 3 weekly Penicillin injections.

Table 11. RHD in India, Cost of Chemoprophylaxis

Drug	Cost per child per year	
	US $	Rupees
1. Penicillin Benzathine (injection 3 weekly)	5	42.00
2. Penicillin G oral	18	142.00
3. Penicillin V oral	15	102.00
4. Sulpha oral	2.4	15.30
5. Erythromycin	65	520.00

raised ASO and of rheumatic recurrences showed very significant differences suggesting the efficacy of 3-weekly as opposed to 4-weekly injections even in children[5].

Lessons that have been learnt from our own experience have been:

a) the need for early start between 5 and 9 years, when the disease is relatively mild and which makes allowances for a longer follow up as many children drop out of school between 12 and 15 years of age.
b) 3-weekly instead of 4-weekly injections in children.
c) assiduous follow up with the help of social workers of all patients who do not come to clinic regularly.

The minimum staff needed for such a clinic and the cost involved of Penicillin injections and other chemoprophylactic agents have been worked out and do not appear formidable.

Injections are preferable to oral tablets because of poor adherence to the regimen in children of the poor social classes due to overcrowded living conditions.

Another encouraging factor has been the results of a collaborative study carried out under WHO auspices in seven centers. This showed that control projects as outlined in the WHO protocol are feasible in developing countries and that even with the follow up rate of around 50 percent and only half of these receiving regular prophylaxis the gains achieved are considerable.

"These benefits are related to the level of prophylaxis, so that while full prophylaxis is obviously desirable, imperfect prophylaxis is better than none at all. Thus, the difficulties associated with establishing a full rheumatic fever control program should not deter authorities from building up similar programs with lower co-efficients of efficiency, since they will also yield considerable benefits[5]."

The most important problem is the extension of the rheumatic fever prophylaxis program to cover the entire population. In India two methods are being tried, one through the school health services and the other through the Primary Health Care centers. The first method has also been tried in the Philippines and the second in Cyprus[6].

DISCUSSION

Considering the urgency and magnitude of the problem there is need for research in the following areas:

1. A quick method for identifying streptococcal infection without throat culture using perhaps monoclonal fluorescent antibodies.

2. A fool proof method for clinical diagnosis of acute RF.
3. Biologic markers indicating rheumatogenic potential or lack thereof, other than serotypes as in M 5 and M 4 strains.
4. The comparative efficiency of Penicillin injections given through school health services and primary health centers.
5. Efficacy in raising serum Penicillin levels of brands of benzathine Penicillin currently in use.
6. A cooperative study to document the incidence and nature of reactions to Penicillin in children.
7. Trial of seasonal primary prophylaxis.

There is need for education among all sections of the population including doctors and parents on the relationship between sore throat and heart disease.

There is also need for improvement in surgical facilities for RHD especially mitral stenosis by open and closed technics, for causes of restenosis and thrombogenicity of prosthetic valves. Better tissue or biological valves which do not need anticoagulation and suitable for children should be sought.

SUMMARY

1. There is a high prevalence of RF and RHD in children and young adults in the developing countries.
2. The special features in developing countries are severe disease, multi-valvular lesions, TR, CHF, juvenile MS, frequent hospital admissions and high morbidity and mortality.
3. Primary prevention is ideal but difficult of achievement. Seasonal primary prophylaxis may be feasible.
4. Secondary chemoprophylaxis remains the best weapon at the present time. Recent collaborative studies have shown encouraging results, and even submaximal prophylaxis is worthwile.
   Early start between 5 and 9 years of age, 3-weekly injections and close follow up yields excellent results. The cost of staff and drugs are not exorbitant. The scheme is eminently feasible in developing countries. Its incorporation into the health care delivery system either through primary health care centers or school health services, would ensure universal coverage.
5. Suggestions for future research include identification of biologic markers for rheumatogenic potential, diagnosis of streptococcal infection without culture and a test for definite clinical diagnosis of acute RF. The relative merits of the school health or primary health care systems for secondary and primary prophylaxis and improvement of surgical facilities and methods are other areas for research.

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LONG-TERM RESULTS OF CORONARY REHABILITATION:

RATIONALE, IMPLEMENTATION AND SIGNIFICANCE OF PROGRAMS

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INTRODUCTION

Rehabilitative programs are based on the concept that many patients with symptomatic coronary disease can and should return to a productive and active life. In the United States, particularly during the past decade, this rehabilitative approach has been progressively incorporated into traditional medical care. Because rehabilitation should begin at the onset of the acute illness and continue during the long-term care of the patient, the primary physician has a pivotal role in initiating and coordinating rehabilitative efforts.

A major component of rehabilitation involves activity: early ambulation during the hospitalization and subsequent prescriptive exercise training after discharge from the hospital. The second component is education of the patient and family, including the provision of a variety of counseling services as warranted.

EARLY AMBULATION

In recent years there has been unequivocal documentation of the safety of early ambulation for appropriately selected and supervised patients with acute myocardial infarction. There has been no increase in the complications of infarction; indeed, some studies suggest a more favorable outcome with this approach.

The major benefits of early ambulation are evident in the short-term. These include prevention of the deconditioning effects of protracted immobilization at bed rest, decrease in pulmonary atelectasis

and thromboembolic complications, and lessening of the anxiety and depression commonly associated with myocardial infarction; the reassurance offered by the performance of progressive physical activity improves the patient's self-confidence and self-image.

Early ambulation permits the patient's functional capacity to be reliably ascertained at pre-discharge exercise testing; it enables the current shorter hospital stay with its saving in medical care costs. Because physiologic and emotional deconditioning has been limited the functional status of the patient is improved at the time of discharge from the hospital; this is reported to facilitate an earlier and more complete return to work.

EXERCISE TRAINING AFTER MYOCARDIAL INFARCTION

Convalescent Physical Activity

Physical activity during convalescence is designed to increase endurance to a level which will enable a prompt return to work and/or to usual preinfarction activities. In the United States, over 85% of patients with an uncomplicated infarction, employed at the onset of illness, currently return to work within 2 to 3 months, typically resuming their former job. Walking is the major activity during convalescence, with patients monitoring their pulse rate response to exercise. Ideally, patients should enter a supervised (and often a hospital-based) progressive activity program within the first month after infarction. The pre-discharge exercise test is often used for the initial exercise prescription. When this is not available, the symptomatic, heart rate, and electrocardiographic response to an exercise regimen can guide the level of recommended activity. In addition to the safety features of a supervised and intermittently monitored exercise regimen, the group setting provides emotional support and facilitates educational efforts and counseling.

Individualized Prescriptive Exercise Training

After convalescence, individualized prescriptive physical activity is designed to enhance cardiovascular function. When the patient has recovered and typically has attained sufficient endurance to return to work (commonly within 4 to 8 weeks after infarction), more intensive exercise training can be undertaken; the exercise prescription is based on results of a sign-symptom limited exercise test. Patients typically exercise at least 2 or 3 times weekly, preferably on nonsuccessive days; exercise sessions are 30-45 minutes in duration, including warm-up and cool-down periods. The target heart rate intensity of 70-85% of the highest level safely achieved at exercise testing corresponds to 60-78% of the peak oxygen uptake, an effective yet safe range within which to stimulate aerobic metabolism.

An ideal regimen initially involves medically-supervised exercise to enable exercise guidance, increase motivation and reassurance, and insure care for cardiovascular emergencies; as exercise performance and fitness improve, medical supervision can be decreased. The ultimate goal is reasonable independence in exercising; this mandates a weaning, initially from the monitored setting and subsequently from the ritualization of formal exercise training, with progressive involvement in an exercise setting that is social, pleasurable, convenient, and appropriate.

The characteristic design of supervised exercise regimens entails a 5- to 10-minute warm-up period of stretching and a range of motion exercises, a 15- to 20-minute aerobic or endurance component which may consist of walk-run sequences, stationary bicycle exercise, arm exercises and calisthenics, and aerobic games. The final 5- to 10 minute cool-down segment involves a gradual decrease in exercise intensity. Exercise recommendations for unsupervised home exercise vary considerably, but in the United States are the pattern of care for about half of patients for whom an exercise regimen is recommended. Some initial home exercise instructions involve progressive walking and walk-jog sequences, others use a stationary bicycle and/or a variety of community-based aerobic activities and sports.

Serial assessment of exercise capacity is recommended at 3- to 6-month intervals to document performance changes, permit revision of the exercise prescription, and/or define the need for changes in the medical regimen. Patients who attain a 7- to 8-met level of performance are often progressed to an unsupervised or minimally supervised exercise setting; this typically occurs within 3 to 6 months after infarction.

An important benefit of exercise training is a decrease in myocardial oxygen requirement for any submaximal task, allowing the patient to function farther from the ischemic threshold in usual daily activities; this enables an increased intensity and duration of work and a reduction in chest pain symptoms by increasing the exercise threshold for angina. Because the energy requirement for any task is a lesser percent of this increased physical work capacity, patients perceive less exertion as they work, which they describe as improved "endurance". The hemodynamic determinants of this increase in functional capacity include an increase in maximal cardiac output and oxygen consumption, a decrease in resting heart rate, a lesser increase in heart rate and systolic blood pressure for any level of submaximal work, and more rapid return to normal of the exercise heart rate. Peripheral oxygen extraction by working muscle improves, as does the redistribution of cardiac output; both further decrease the demand for oxygen transport. There is currently little or no evidence that short-term, modest-intensity exercise improves intrinsic myocardial performance, especially in older individuals with significant coronary disease.

Neither is there evidence that exercise training alters the coronary collateral circulation in man, either as detected angiographically or by myocardial perfusion studies. It remains controversial whether exercise training affects the incidence or severity of cardiac arrhythmias. Exercise training has not been shown to alter the natural history of coronary atherosclerotic heart disease - the recurrence of myocardial infarction or the incidence of coronary death. However, recent studies suggest that fatal reinfarction is decreased.

Serum triglyceride levels decrease with exercise training, but the effect on total serum cholesterol is not predictable; high-density lipoprotein cholesterol increases. Long-term effects on fibrinolysis and platelet function are inconclusive. Exercise may exert a beneficial effect on coronary disease by modifying other more powerful coronary risk factors: discontinuation of cigarette smoking, weight reduction, favorable dietary alterations, blood pressure control etc. Psychosocial benefits are that patients who exercise often feel better, have improvement in self-confidence and self-esteem, show less depression and dependency on standard psychometric tests, and appear better able to tolerate life crises.

PATIENT AND FAMILY EDUCATION

Education of the patient and family is designed to provide the information about coronary disease and its management that enables patients to assume some responsibility for continuing health care. During the hospitalization, a variety of health professionals can readily institute an educational program. However, if the benefits are to be maintained, the recommendations for care and their implementation must be reinforced after return home.

The patient education curriculum should include a brief review of normal cardiac structure and function and of the atherosclerotic process causing coronary obstruction; this forms the basis for subsequent recommendations for care. Prevalent myths regarding the precipitation of myocardial infarction must be dispelled. Prescriptive components - dietary changes, cigarette smoking cessation, physical activity, etc. - are explained,defining the rationale for each and offering suggestions for effecting the changes; community resources that may be helpful should be identified. Discussions should include advice about resumption of sexual activity, response to new or recurrent symptoms, control of associated diseases, and return to work. All medications should be reviewed in detail. Additionally, in many hospitals cardiopulmonary resuscitation is taught to families of myocardial infarction survivors; family counseling addresses lifestyle adjustments necessary during convalescence, focusing on averting unnecessary invaliding of the patient.

Patients who understand their disease and the rationale for management have improved ability and motivation to cooperate in re-

ommendations for care; since many components of the care of coronary patients involve a lifetime change in habits, intensive education appears appropriate.

SUMMARY

Effective rehabilitation can enable survivors of myocardial infarction or patients after coronary bypass surgery to return rapidly to a relatively normal lifestyle; patients with angina pectoris may be kept at work or at desired leisure activities. A plan of care must be designed to help the patient achieve realistically optimal physiologic improvement, attain an acceptable level of self-care, and to resume a useful activity level in the home and/or work environment. The majority of patients who recover from myocardial infarction can achieve a functional capacity adequate to perform most moderate personal, occupational, and recreational activities. The rehabilitative approach should limit the economic impact of the illness on the patient, family, and community through a shortened hospital stay, deemphasis of invalidism, a decreased need for convalescent care, and an earlier and more complete return to work. The risk of recurrent coronary events and the incidence of late complications of myocardial infarction may be lessened by a secondary prevention program. Ideally, rehabilitation is incorporated into the traditional care during the hospitalization for myocardial infarction or coronary bypass surgery, involves the patient's family and social environment as a support system, and continues in the office of the patient's physician and/or in a variety of community facilities.

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LONG-TERM RESULTS OF CORONARY REHABILITATION:

FACTORS AND REASONS FOR SUCCESS AND FAILURE

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The short term benefits of rehabilitation after myocardial infarction are now well documented, such as shorter hospitalization, improvement of physical capacity by training, psychological effects and earlier return to work.

Unfortunately, the long term studies failed to demonstrate a significant benefit of a comprehensive rehabilitation program or of a supervised physical training, on morbidity and mortality in randomized studies.

A few studies reported in the past encouraging results in cases of exercise rehabilitation after myocardial infarction, but these studies were not randomized and the exercise protocol as well as the duration of follow up were generally not specified.

If we consider the effects of physical exercise training as a secondary prevention measure in patients who have survived a myocardial infarction, we find several prospective randomized trials, in the United States, in Canada and in Europe.

In the US the National Heart Disease Project was proceeded to evaluate the effects of regularly performed medically prescribed exercise in male survivors of MI, who were assigned by randomization to either an exercise group or a non treated group[1]. This trial was effective in inducing a physiological training effect, at least for a short time. A reduction in mortality of 37% was observed in the exercise group; there is no suggestion of a benefit in cardiovascular morbidity. The number of patients is low (651) and some of them enter the program 36 months after MI; the observed reduction in mortality was not significantly different.

In Canada, in the Ontario Exercise-Heart Collaborative study[2], a total number of 751 patients was divided in 2 groups submitted to high intensity exercise or to low intensity exercise; the drop out was very high in the 2 groups and the rate of recurrence was not significantly different between the groups.

In Europe, the studies of SANNE, KENTALA, PALATSI and KALLIO, with a number of patients around 300 to 400, demonstrate some benefit in the trained group but again the difference with the control group were not significant[3].

In Europe also, as a result of several years preparatory work, WHO coordinated study or exercise training and comprehensive secondary prevention was designed: this study was aimed at assessing the effectiveness of comprehensive rehabilitation in reducing recurrent MI and cardiovascular mortality[4,5]. Patients under the age of 65 with definite MI, treated in hospital, were admitted to the study. The intervention measures were to be applied according to the best knowledge in each individual center; the follow up period was 3 years; the main end points were death and morbidity, but physical working capacity, changes in the quality of life and reduction in risk factors were used as additional criteria. The total number of patients (3,118) coming from 19 centers were divided into two groups by randomization.

Considering the end points, the pooling of all data is quite impossible, related to local attitudes or other reasons. If we consider the 17 centèrs with a sufficient number of patients in the treated and control group: 13 have a mortality experience which favors the rehabilitation group, but the difference in reinfarction is not significant. There were big intercenter differences in return to work. In most of the centers the physical capacity is improved.

Finally, from all these studies it appears that none of the trials showed a statistically significant difference in total mortality between the exercise and control groups; some showed a trend in favor of the exercise group. There was also only a little effect on the incidence of reinfarction[2,3].

The same type of doubtful answer is observed when other secondary prevention methods are used, such as long term prescription of antiarrhythmias drugs, lipid lowering drugs and diet, anticoagulant drugs, platelet active drugs, beta-blocking drugs, and others. The only attitude for which a strongly positive result is observed is the suppression of smoking habits[3-7].

In conclusion, all these studies failed, till now, to provide a clear answer to the question whether a comprehensive program could reduce mortality or morbidity after MI in the long run.

Should such a conclusion appear as a condemnation of our rehabilitation programs: of course not, it is probably only a demonstration that our trials are not well conducted.

The reasons for this are numerous: low number of patients in most of the studies; insufficient definition of the patients, from the physiological and psycho-social point of view; compliance of the patients to the program; drop out; drop in related to a scattering of the knowledge; unadapted level of training; therapeutic local traditions and use of different drugs for secondary prevention; local organization; technical resources, cost of programs, economical resources, role of surgery etc.

A good conclusion is presented by R. Shepard: "Pessimists may conclude that a control study to determine the influence of exercise upon the rate of reinfarction is logistically impossible ... optimists might attempt to meet the requirement of long numbers by pooling data from various current controlled studies, ignoring obvious differences of philosophy and protocol".

But from a scientific point of view, the only satisfactory course is to regard existing experients as pilot trials[6]...

I could not conclude on the success or the failure of cardiac rehabilitation in the long run, but only stress the difficulties in an evaluation of the comprehensive post coronary care programs generally adapted.

Important changes in the therapeutic approach to MI have appeared during the last 2 decades. The current attitudes have modified the way of life and the perspectives of our patients: most of those with a good clinical condition reintegrate easily the family and the professional life and even if it is not ascertained that their life expectation is improved, the quality of their life is. But we have to accept that, in spite of excellent results at short time, the progressive changes in our attitude are based on empiricism and that most of the hypotheses on which our actual programs are based, are neither proved or disproved.

More research is needed, especially for what concern the implementation of an optimal comprehensive program. Such programs should include exercise, for the reason that, at least, it brings up a positive psychological and physiological improvement.

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THE ECONOMIC EFFECTS OF CARDIAC REHABILITATION:

PRINCIPLES, METHODS AND PROBLEMS OF EVALUATION

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Two major indicators of successful rehabilitation are social integration of the patient and return to meaningful and productive activities. The economic effects of rehabilitation - both costs and benefits - may often be difficult and even impossible to calculate with any precision. Improvement of quality of life, such an important goal for rehabilitation, is not easily transferable to monetary units. Nevertheless, attempts have and should be made to evaluate rehabilitation activities even from economical points of view. It will help to direct investments of limited resources available for development of rehabilitation services[1],

In order to give a general overview of changes in the cost of burden of both health and sickness attributable to various diseases, some data from the USA are presented in Table 1. It shows the percent distribution of costs attributable to the three leading diagnostic categories in 1930 and 1975[2]. It can be seen that the cost of sickness, in particular, has increased remarkably as regards cardiovascular diseases, while the cost of premature deaths has increased less in this diagnostic group compared to accidents, poisoning and violence, and neoplasms. It remains to be seen how the downward trend in cardiovascular mortality will affect these figures.

As regards the overall economic burden of diseases of the circulatory system an estimation has been presented indicating that 1.7% of the adjusted Gross National Product was claimed by these diseases in the USA in 1975.[2] There is good reason to believe that the situation is similar in many industrialized countries in Europe.

Together with the development of public health policy and measures to combat cardiovascular diseases the importance of rehabilitation has increased rapidly. This may be due to several factors,

Table 1. Cost burden of death and sickness by disease in 1930 and 1975 in the USA[2]. (Percent distribution)

	Cost of premature deaths		Cost of sickness	
	1930	1975	1930	1975
Diseases of the circulatory system	12.9	22.4	8.3	15.1
Accidents, poisoning and violence	14.6	30.2	14.2	9.8
Neoplasms	4.9	14.8	1.2	1.9

such as increased demand for these services and emphasis on high-quality after-care. Measures should be resorted to in order to develop rehabilitation services in the most cost-effective manner.

Evaluation of the effects of rehabilitation depends on whether the impact is analyzed from the point of view of an individual or of society. These two different view points, subjective and objective need, must be taken into account when evaluating the overall economic effect of rehabilitation.

As rehabilitation represents an important field of social policy, methods used in social policy research can be applied in the evaluation of the economic effects of rehabilitation measures. Cost-benefit analysis and cost effectiveness analysis have both been used for evaluation. Cost-benefit analysis was originally developed for evaluation of physical investments in order to provide information required for economically efficient decision making[1]. In the 1960s it was recognized that many social expenditures could reasonably be considered investments and along with this development cost-benefit analysis was being increasingly applied to social programs. Since its sole concern is economic efficiency, cost-benefit analysis cannot be used alone in decision making concerning complex programs such as rehabilitation. It is thus merely a method to provide information about the problems and not the method to solve them.

Cost-effectiveness analysis on the other hand can be applied when the economic effects cannot be easily measured or they cannot be made commensurable. Cost-effectiveness analysis is particularly useful in problem areas such as public health when the output cannot be measured in terms of market price. The effectiveness can be evaluated for instance by increase in years of life or decrease in days due to inability to work. A typical example of this type of approach

is as follows: it has been calculated that the average expected duration of life of a new-born boy in Finland would increase by 6.8 years if all cardiovascular diseases could be abolished[3].

Typical problems in cost-benefit analysis of rehabilitation are presented in the following:

- calculation of direct costs to persons and families such as loss in earnings, loss in production efficiency, added costs etc.
- calculation of costs to industry such as loss in product and work time
- calculation of secondary or indirect costs such as loss in taxable revenue, loss in productivity, continuing costs of medical and other services etc.
- calculation of the capital cost
- calculation in monetary value of the increase in social welfare and quality of life resulting from rehabilitation
- differences in patient groups with regard to life expectancy

In spite of apparent problems of evaluation cost-benefit analysis has been applied in some studies on cardiac patients published recently. The economic impact of coronary by-pass surgery and subsequent rehabilitation was analyzed by Liddle[4]. The patients provided information by completing a questionnaire concerning pre- and postoperative salaries, disability payments, costs of medical care, and return to work. Of 607 questionnaires submitted to patients 90% were returned. Among all reporting patients 75% returned to employment within the first 6 postoperative months. Cost-benefit analysis of the 153 patients not working before the operation but who returned to work after the operation is shown in Table 2. The patients are divided into 5 groups according to years worked after the operation by the time of follow up study. The average 1-year salary was in all groups at least somewhat higher than total cost of care. If these disabled patients had not undergone operation and had been rehabilitated and returned to work, the indirect costs caused by lost working years, disability payments etc. would have exceeded the costs of operation and rehabilitation by many times.

Table 3 shows the cost-production relationship of the 152 men who were working preoperatively and returned to work. In all subgroups formed according to years worked after operation, total cost of care was substantially less than their reported annual salary. The annual salary after operation compares well with that before the surgery.

Crosby et al.[5] have presented their series of 66 patients who underwent aneurysmectomy and revasularization and were encouraged to participate in individual physical rehabilitation postoperatively. The average follow-up was 20 months. Full employment increased from 33% preoperatively to 63% postoperatively, and total disability rate

Table 2. Cost-production relationship in 153 patients incapacitated for work before coronary by-pass operation but working after operation[4].

		Average income per year and cost in US dollars		
Number of patients	Years worked	Disability payment before operation	Salary after operation	Total cost of care
30	< 1	1492	16467	15436
70	1 - 5	830	16257	14256
39	5 - 8	790	16705	13076
13	8 - 10	783	19461	12443
1	> 10	0	25000	10000

Table 3. Cost-production relationship in 152 patients working both before and after coronary by-pass operation[4].

Average income per year and cost in US dollars.

N	Year worked after operation	Salary before operation	Salary after operation	Total cost of care
20	< 1	19400	19550	12785
67	1 - 5	18708	20410	13166
51	5 - 8	15892	19421	11785
14	8 - 10	17857	22500	11959

decreased from 60% preoperatively to 29% postoperatively. When the employment capability was used as an indication of improvement after surgery, the results were even more favorable.

When the economic impact of the procedure in the community in terms of improvement in income, decrease in disability payments and reduction in tax revenues was calculated, the positive balance was such as to allow the cost of the procedure to be paid back over 1.68 years.

The authors emphasize the need of aggressive physical and vocational rehabilitation programs for patients undergoing expensive cardiac operations in order to make them cost effective.

In some studies[6,7] the return to work after myocardial infarction has been reported to be relatively low or 42 or 44%, respectively. There is evidence from literature indicating that return to work can be improved by active rehabilitation programs[7,8] although essential differences between countries exist even in this respect.

We have analyzed the individual economic impact of return to work in a material of men under 65 years who were treated in hospital because of acute myocardial infarction. The patients were randomly allocated in two groups: one with a comprehensive rehabilitation program and the other with ordinary services. All patients provided information by filling in a questionnaire concerning return to work and the salary or disability payment before and one year after the myocardial infarction. Return to work was almost similar in both groups of 58 and 55% respectively. There were, however, other positive effects which we consider worthy of investment required for the rehabilitation program[9,10,11,12]. Furthermore, the patients of the rehabilitation group who returned to work paid back in taxes the costs of the comprehensive rehabilitation program for all patients in about 1 year.

The effect on income of return to work is presented in Table 4. The results refer to patients who were working before the myocardial infarction. The pre-infarct salary was given an index value of 100 and the subsequent salries and disability payments were calculated as percentages of the pre-infarct salary. The average salary one year after the myocardial infarction was almost on the pre-infarct level, while the patients who were not working experienced a considerable economic loss which was bigger than expected. From the community point of view, the economic impact of not working after the myocardial infarction is substantial due to disability payments, lost taxes, inability to pay for medical treatments etc.

In our experience about 20% of men with acute myocardial infarction below the age of 65 receive disability payments at the time of the acute event. It is extremely rare for these patients to return

Table 4. Effect of return to work, disability or old age pension on income 1 year after myocardial infarction. (Kallio et al., unpublished data)*.

	Number	Mean age	Income index
Patients employed before the acute myocardial infarction	226	51.9	100
Work status one year after infarction			
- Patients returned to work	108	49.0	97
- Patients on disability pension	83	53.7	35
- Patients on old age pension	8	63.7	52
- Deaths or no income data	27	-	-

*Index figures refer to salary before the new infarction

to work in spite of an active rehabilitation program. Vocational rehabilitation seems, therefore, best indicated in patients who are employed when they get their myocardial infarction.

It can be concluded that rehabilitation measures which improve the working capacity and especially return to work are cost-effective particularly if all costs incurred by permanent disability are taken into account. New studies are, however, needed especially on patients with coronary heart disease in order to develop the most cost-effective organization and program of rehabilitation. Finally, many effects provided by cardiac rehabilitation cannot be measured in terms of economic benefit. These effects are at least as important and worth striving for as the economic gain.

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