THE YEAR IN DYSLIPIDAEMIA

2002

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Contents

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Part I
Lipid disorders

1
Familial hypercholesterolaemia and related disorders

Introduction

Familial hypercholesterolaemia (FH) has been known to be associated with premature coronary heart disease (CHD) since the work of Muller |**1**| and others almost 70 years ago. In the 1970s Brown and Goldstein showed that the marked increases in serum low-density lipoprotein (LDL) concentration were the result of impaired catabolism due to defective LDL-receptor function |**2,3**|. Homozygotes are rare, perhaps one in a million in many societies, but have total serum cholesterol levels that are often in the range 15–30 mmol/l. They may exhibit clinical CHD and even die in childhood.

The heterozygotes are much more common with a frequency usually estimated at about 1 in 500, although they are commoner still in some groups where there is a strong founder effect, including Lebanese Christians, French Canadians, Afrikaners, Finns and Icelanders. They usually have serum cholesterol concentrations in the range 8–15 mmol/l. The heterozygotes are also at particularly high risk of premature CHD and it is believed that this relates to the fact that FH subjects are hypercholesterolaemic from birth, whereas hyperlipidaemia is only expressed in adolescence or early adult life in some other genetic disorders, such as familial combined hyperlipidaemia (FCH) and type III hyperlipoproteinaemia (familial dysbetalipoproteinaemia).

It has been estimated that 50% of men and 15% of women with heterozygous FH will have died of CHD by the age of 60 years, with 75% and 45%, respectively, having clinical manifestations of CHD. There is, however, evidence that the ability to reduce serum, and in particular LDL, cholesterol to a much greater extent over the last decade since the advent of the statins is improving the prognosis.

Detection and diagnosis of familial hypercholesterolaemia

FH is probably greatly underdiagnosed. In the absence of a strong family history or gross tendon xanthomas the disorder will not normally be identified by the usual high-risk screening strategies. Tendon xanthomas are not always present especially in young subjects and when they are small they are commonly overlooked by patients and clinicians. FH subjects are frequently not overweight and hypertension and diabetes are not over-represented. There is not always an adverse family history, especially when inheritance is from the maternal side. It is perfectly possible for an affected man in his thirties or forties to suffer a myocardial infarction while his affected mother remains undiagnosed and apparently healthy. For an extensive review of the merits of case finding

in FH see Marks *et al.* |**4**| (also at www.ncchta.org). To what extent is FH underdiagnosed?

Extent of underdiagnosis of familial hypercholesterolaemia in routine practice: prospective registry study.
H A Neil, T Hammond, R Huxley, D R Matthews, S E Humphries. *BMJ* 2000; **321**:148.

B**ACKGROUND. To determine the prevalence of diagnosed FH and to estimate the proportion of expected cases that are identified in routine clinical practice, diagnosed patients living in Oxfordshire were identified from lipid clinic computerized diagnostic and other registers, and from general practice records.**

I**NTERPRETATION.** In all, 38 families with 88 affected members were identified. The overall prevalence of FH was 0.54/1000 and was highest in men aged 50–59 years and in women aged 60–69 years. Underdiagnosis was greatest in children and young adults: only two children under 10 years and 12 aged 10–19 years had been identified. About a quarter of the predicted cases of FH were diagnosed routinely; most remained undiagnosed until middle age. Underdiagnosis implies that patients are denied early treatment to reduce the risk of coronary events.

Comment

This study based on FH cases resident in Oxfordshire found a prevalence of 0.54 cases per 1000 overall—about 25% of those predicted. A higher proportion of middle-aged subjects is diagnosed and underdiagnosis is correspondingly greater in the young who have the potential to be spared premature CHD by treatment with statins.

Cardiovascular risk factors and testing of relatives amongst patients with familial hyperlipidaemia one decade after a clinical trial.
S Tonstad, I Hjermann. *J Intern Med* 2000; **248**:111–18.

B**ACKGROUND. Cardiovascular disease risk factor status of men and women with familial hyperlipidaemia 10–11 years after a clinical trial was studied. Participants started lipid-lowering drugs in 1987–88. Of 60 participants, 12 had died, one**

emigrated and 35 men and 12 women took part in a follow-up clinical examination in 1998.

I**NTERPRETATION.** Compared with baseline (diet alone), total cholesterol was reduced by 41% and high-density lipoprotein (HDL) cholesterol was increased by 13%. LDL cholesterol was lower at the end of the trial than at follow-up and was higher in those taking a low dose of a statin alone compared with other drug groups. To reach target lipid levels two-thirds of participants needed adjustment of lipid-lowering drugs. Most participants were undertreated. Increases in body mass index, blood pressure and glucose levels, and the diet posed challenges to risk reduction. Plasma homocysteine levels should be considered in this group. Testing of all first-degree relatives may not be achievable because of psychological barriers.

Comment

Forty-seven patients with a familial hyperlipidaemia were investigated in this study, ten years after they had participated in a clinical trial of lovastatin with or without cholestyramine. They included 37 with familial hypercholesterolaemia and seven with probable familial combined hyperlipidaemia (FCH). Half had CHD. Twenty per cent of the original cohort had died in the decade since the original study. The median reduction in total cholesterol of 41% and increase in HDL cholesterol of 13% is impressive, but the authors state that most were still undertreated. Those followed up in hospital had lower cholesterol levels. A study of this sort provides the opportunity to identify a large number of new subjects with genetic hyperlipidaemia. Of 136 identified siblings and offspring, 60 were deemed to be unaffected and 45 were affected and treated, although we are not told how successfully. Three affected children refused treatment and 28 first-degree relatives were uncontactable or refused participation. The results presented here are probably much better than in routine clinical practice, but they emphasize that many subjects likely to have a genetic hyperlipidaemia still do not get the opportunity for diagnosis and treatment.

Outcome of case finding among relatives of patients with known heterozygous familial hypercholesterolaemia.

D Bhatnagar, J Morgan, S Siddiq, *et al. BMJ* 2000; **321**:1497–500.

B**ACKGROUND. The feasibility of detecting new cases of heterozygous FH by case finding among relatives of patients with FH was assessed using a nurse-led genetic register. The 259 probands had 285 first-degree relatives.**

I**NTERPRETATION.** Of the 200 first-degree relatives tested, 121 (60%)

had inherited FH. The newly diagnosed patients were generally detected before they had clinically overt atherosclerosis. Screening for risk factors, as recommended in recent guidelines for CHD prevention, would not have identified most of the affected relatives. Because 1 in 500 people in the UK are affected by this condition, 60 000 tests would be needed to detect a similar number by population screening, and only a few of these patients would

have been detected had cholesterol testing been restricted to those with other risk factors. A genetic register linking lipid clinics nationally may be needed.

Comment

This paper, based on two lipid clinics in Manchester, emphasizes the power of the genetic register approach to the detection of FH. Screening 200 first-degree relatives of known FH cases yielded 121 new cases (one would have predicted 100). Based on a frequency of 1 in 500 it would have been necessary to take blood from more than 60000 people in a random population screening exercise to identify the same number of new cases. A large screening programme of this sort would be not only costly but would identify a large number of hyperlipidaemic individuals of uncertain risk. Identifying those with FH would still often be difficult without information from relatives. Finding a hypercholesterolaemic subject who in fact has FH in a general screening exercise but failing to recognize them as having FH may deny them much needed treatment. Risk calculation charts based on the Framingham equation, such as those in the Joint British Guidelines and the Sheffield Tables, underestimate risk in FH and are not suitable for use in such subjects.

In the absence of cholesterol screening of the whole adult population, which is not currently regarded as cost-effective, rigorous pursuit of close relatives of FH cases seems the best approach. Addition of DNA testing would improve the sensitivity and specificity of the process |**5**|.

Does treatment with statins modify outcome in familial hypercholesterolaemia?

Statins lower LDL cholesterol very effectively in most cases of heterozygous FH by upregulating the activity of the functional LDL receptors. There is no reason to believe that they would be ineffective in reducing CHD events. It would not now be ethical to perform placebo-controlled end-point trials in FH subjects and they were probably represented by small numbers in the original statin trials and not usually specifically identified. An angiographic study |6| suggests that drug treatment is associated with benefit. Angiographic and clinical benefit are often but not necessarily synonymous, however.

Mortality in treated heterozygous familial hypercholesterolaemia: implications for clinical management.
Scientific Steering Committee on behalf of the Simon Broome Register Group. *Atherosclerosis* 1999; **142**:105–12.

B**ACKGROUND. Men (*n*=605) and women (*n*=580) aged 20–79 years with heterozygous FH, were followed prospectively for 15 years (8770 person-years).**

INTERPRETATION. Forty-six of the 73 deaths were due to CHD. Despite treatment, the relative risk of a fatal coronary event in those aged 20–39 was increased 125-fold in women and 48-fold in men. The relative risk decreased with age but the absolute risk increased. For men and women aged 60–79, the annual coronary mortality was 1.1%, a significant excess mortality for women but not for men. Non-coronary mortality was not increased at any age. The relative risk for coronary mortality in patients aged 20–59 fell from an eightfold increased risk before 1992 to 3.7-fold thereafter, suggesting that the prognosis for patients with heterozygous FH has improved with the introduction of more effective treatment, and that lipid-lowering therapy does not increase non-coronary mortality. The findings suggest that all affected adult men and post-menopausal women should be treated with statins.

Comment

The Simon Broome Register was founded in Oxford in 1980 supported by a bequest from the estate of a patient with heterozygous FH. Information on FH patients has now been collected from an increasing number of UK lipid clinics for more than 20 years. The observation that relative risk of CHD death was approximately halved in 1992–95 compared with the previous 12 years fits well with the introduction of the statins at the beginning of the nineties. The potential benefit is probably greatly underestimated as with the passage of time growing confidence in the safety and efficacy of the statins has led to their much more aggressive use with more stringent targets for total and LDL cholesterol.

Why does risk vary so much in familial hypercholesterolaemia?

LDL cholesterol is clearly a major determinant of risk in FH. Several hundred different mutations in the LDL receptor gene have been described. Receptor-defective homozygotes, who have some residual receptor function, have a less severe form of the disease, in terms of LDL concentration and CHD risk, than those without detectable receptor function (receptor negative) |**2,7**|. One would predict that those with more

receptor function might also be more responsive to treatment with statins. However, the age at which FH subjects develop clinical CHD varies widely and is not always readily explicable on the basis of LDL cholesterol concentration. The age at first atherosclerotic event tends, however, to be fairly consistent within families |**8**| and this has prompted a search for other genetic factors. Other candidates to explain this variability in risk include conventional risk factors such as smoking and HDL cholesterol, but also apolipoprotein (apo) E genotype, lipoprotein(a) (Lp(a)) and homocysteine concentrations and recently a polymorphism of the renin-angiotensin system.

Additional risk factors influence excess mortality in heterozygous familial hypercholesterolaemia.

E J Sijbrands, R G Westendorp, M Paola, *et al. Atherosclerosis* 2000; **149**: 421–5.

B**ACKGROUND. All cause mortality in all 855 first-degree relatives of 113 unrelated FH patients was compared with that of the Dutch population, after standardization for age, gender and calendar period. All first-degree relatives were analysed (whether affected by FH or not) and as a result the standardized mortality ratios (SMRs) show only 50% of the excess mortality from FH.**

I**NTERPRETATION.** The 190 deaths in 32 048 person-years led to an overall SMR of 1.34. A high excess mortality occurred in men aged 40–54 years but decreased in older men indicating that additional risk factors modulate mortality. The SMR was 1.6 in 62 families referred with premature coronary artery disease (CAD) and 1.1 in 51 families without premature CAD. The mortality risk of relatives with null alleles was similar to that of relatives with other mutations. Additional risk factors increased excess mortality significantly and are highlighted by the presence of premature CAD among first-degree relatives. All hypercholesterolaemic relatives of such patients should be actively Identified.

Comment

Analysis of the patients in this study stopped in 1989 at about the time that statin treatment was introduced. Nevertheless there was a reduction in the SMR for all first-degree relatives of probands with FH (half of whom are likely to have had FH) in 1965–89 compared with 1935–64. This secular trend may have contributed to the reduction in mortality seen after the introduction of statins in the Simon Broome Register study (see above). Nonetheless, the statins are likely to have played the dominant part in what was a marked reduction in mortality over a very short time in the latter study.

In the present study, which contrasts with others in this respect (see Bertolini *et al.* below) no difference in mortality was observed between relatives of probands with null

alleles compared with relatives of those with other mutations. Relatives of probands who had presented with clinical CHD had a higher mortality than the relatives of those who were diagnosed as hyperlipidaemic by screening in the absence of clinical vascular disease. This is not surprising but it is unclear what precisely it is that confers the additional risk. This study once again emphasizes the variability in prognosis in FH even though it can tell us little about the underlying mechanisms.

Clinical expression of familial hypercholesterolemia in clusters of mutations of the LDL receptor gene that cause a receptor-defective or receptor-negative phenotype.

S Bertolini, A Cantafora, M Averna, *et al. Arterioscler Thromb Vasc Biol* 2000; **20**: E41–52.

B**ACKGROUND. Seventy-one mutations of the LDL receptor gene were identified in 282 unrelated Italian FH heterozygotes. Extending genotype analysis to families of index cases, we identified 12 mutation clusters localized in specific areas of Italy. The clusters were separated into receptor-defective and receptor-negative groups, according to the LDL receptor defect caused by each mutation.**

I**NTERPRETATION.** Receptor-negative subjects had higher LDL cholesterol, and lower HDL cholesterol and were twice as likely to have tendon xanthomas and CAD compared with receptor-defective subjects. In patients>30 years of age in both groups, the presence of CAD was related to age, arterial hypertension, previous smoking and LDL cholesterol level. Independent contributors to CAD in receptor-defective subjects were male sex, arterial hypertension and LDL cholesterol level. Overall, in receptor-negative subjects, the risk of CAD was 2.6-fold that of receptor-defective subjects. ApoE genotype accounted for only 4% of the variation in LDL cholesterol. All families of the major clusters shared the same intragenic haplotype cosegregating with the mutation, suggesting the presence of common ancestors.

Comment

This study confirms that the nature of the LDL-receptor defect is an important factor in determining phenotype and prognosis in heterozygous FH. The receptor-defective cases had lower LDL cholesterol than the receptor-negative cases and a lower prevalence of tendon xanthomas and clinical CHD. ApoE genotype made only a small contribution to variability in LDL cholesterol.

Renin-angiotensin system polymorphisms and coronary events in familial hypercholesterolemia.

A S Wierzbicki, M Lambert-Hammill, P J Lumb, M A Crook. *Hypertension* 2000; **36**:808–12.

B**ACKGROUND. The role of renin-angiotensin system polymorphisms as risk factors for CHD is controversial. This study investigated their role in patients with heterozygous FH.**

I**NTERPRETATION.** Polymorphism frequencies for angiotensin-l-converting enzyme insertion/deletion, angiotensinogen M235T and angiotensin-II type I receptor (AG2R) A1166C were determined in 112 patients with FH and 72 with polygenic hypercholesterolaemia, of whom 26.7% and 41.6%, respectively, had established CHD. None of the polymorphisms were associated with CHD risk in patients with polygenic hypercholesterolaemia. Male sex, smoking, diastolic blood pressure, plasma glucose and AG2R A1166C polymorphism were risk factors for CHD in patients with FH. AG2R A1166C polymorphism may interact with severe hypercholesterolaemia and other risk factors to increase the risk of CHD in FH patients.

Comment

The significance, for CHD risk in the general population, of polymorphisms in the genes for key proteins in the renin-angiotensin system is contentious |**9**|. There are few studies in FH. In this study a polymorphism (A1166C) in the gene for the AG2R confers a threefold increase in risk, after adjustment for other risk factors, of an FH patient having clinical CHD, although it was not predictive in subjects with polygenic hyperlipidaemia. The ACE DD genotype, in contrast with previous work in patients with FH and familial defective apoB (FDB) |**10**|, was not a risk factor. Further studies are needed.

Lipoprotein(a) in homozygous familial hypercholesterolemia.

H G Kraft, A Lingenhel, F J Raal, M Hohenegger, G Utermann. *Arterioscler Thromb Vasc Biol* 2000; **20**:522–8.

B**ACKGROUND. Lp(a) is a quantitative genetic trait that in the**

general population is largely controlled by the locus for the apo(a) gene. Mutations in apolipoprotein B and in the LDL receptor (LDL-R) gene may also affect Lp(a) plasma concentrations, but this is controversial.

I**NTERPRETATION.** When 69 members of 22 families with FH were analysed for mutations in the LDL-R as well as for apo(a) genotypes, apo(a) isoforms, and Lp(a) plasma levels, 26 were homozygous and 43 were heterozygous for FH. Heterozygotes had significantly higher Lp(a) than did non-FH individuals from the same population. FH homozygotes with nonfunctional LDL-R alleles had almost twofold higher Lp(a) levels than did FH heterozygotes. This increase was not explained by differences in apo(a) allele frequencies. Phenotyping of apo(a) and quantitative analysis of isoforms in family members allowed the assignment of Lp(a) levels to both isoforms in apo(a) heterozygous individuals. Thus, Lp(a) levels associated with apo(a) alleles that were identical by descent could be compared. In the resulting 40 allele pairs, significantly higher Lp(a) levels were detected in association with apo(a) alleles from individuals with two defective LDL-R alleles compared with those with only one defective allele. Mutations in the LDL-R showed a clear gene-dosage effect on Lp(a) plasma concentrations.

Comment

Lp(a) is a particle predominantly in the LDL density range that has as its protein component a molecule of apoB100 covalently linked to a molecule of apo(a). Apo(a) is a unique apolipoprotein that has sequence homology with plasminogen and a variable number of repeats of kringle IV leading to great variability in its size. The protein composition of Lp(a) led to the proposition that it might be involved in both atherogenesis and thrombogenesis and indeed a substantial number of cross-sectional studies have identified it as a risk factor |**11**|.

Little is known of the metabolism of Lp(a) and the presence of apoB100 suggested that it might be cleared by the LDL receptor. If so one would predict that Lp(a) concentrations would be increased in FH and that Lp(a) might contribute to and perhaps explain some of the variability in risk in this condition. In the event studies of Lp(a) concentrations in FH have yielded conflicting results. Control of Lp(a) concentration is complex and is determined by the size of the molecule (the number of repeats being inversely related to Lp(a) concentration) and sequence variability in the region of the apo(a) gene locus. Inability to control adequately for these factors may have confounded some previous studies of Lp(a) concentrations in FH heterozygotes.

Recently, Lingenhel *et al.* |**12**| have shown in sibling pairs, identical by descent for their apo(a) alleles, that FH was associated with higher apo(a) concentrations. Here they confirm this observation and demonstrate a gene dosage effect by studying FH

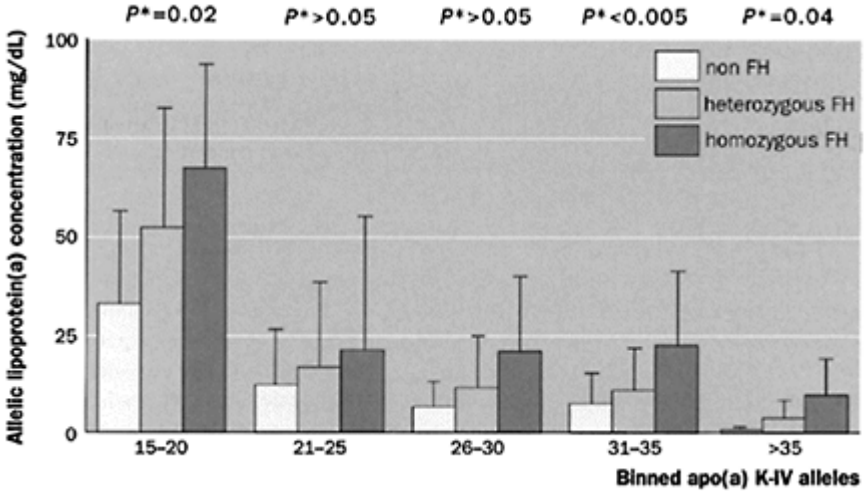


Fig. 1.1 Bar graph showing average Lp(a) concentrations (and SD) associated with five groups of binned apo(a) alleles by size (designated by the number of K-IV repeats). In every group, Lp(a) levels increase with the number of LDL-R mutations, demonstrating a positive gene-dosage effect. Values of *P* indicate statistical significance calculated (or the three genotypic groups by Kruskal-Wallis test and adjusted for multiple comparisons by the Bonferroni method. Source: Kraft *et al.* (2000).

homozygotes. It appears to be firmly established that defects in the LDL receptor are associated with an increase in Lp(a) concentrations, but the mechanism remains uncertain. The effect on LDL concentration exceeds that on Lp(a). This study does not address the contribution of Lp(a) to the variability in risk in FH.

Influence of beta^0-thalassemia on the phenotypic expression of heterozygous familial hypercholesterolemia: a study of patients with familial hypercholesterolemia from Sardinia.
L Deiana, R Garuti, G M Pes, *et al. Arterioscler Thromb Vasc Biol* 2000; **20**: 236–43.

B**ACKGROUND. Previous studies have shown that Sardinian β-thalassaemia carriers have lower total and LDL cholesterol than non-carriers. This study investigated whether the LDL-lowering effect of the β-thalassaemia trait was also present in subjects with FH.**

I**NTERPRETATION.** In 63 Sardinian patients with a clinical diagnosis of

FH, the plasma LDL cholesterol level was found to be lower in subjects with the β^o-thalassaemia trait than in subjects without this trait. The LDL-lowering effect of β^o-thalassaemia may be related to the mild erythroid hyperplasia, which would increase LDL removal by the bone marrow, and to chronic activation of the monocyte-macrophage system, causing increased secretion of some cytokines known to affect the hepatic secretion and receptor-mediated removal of lipoproteins containing apoB. The lifelong LDL-lowering effect of β^o-thalassaemia trait might slow the development and progression of coronary atherosclerosis in FH.

Comment

In Sardinia a single mutation of the β-globin gene accounts for 95% of β-thalassaemia cases, and 6–17% of the population are β-thalassaemia carriers. It has been known for about 20 years that subjects with an allele for β-thalassaemia have lower serum lipids than controls and more recently that this may contribute to a reduced risk of CHD in such subjects. It is now confirmed that this phenomenon is also observed in subjects with heterozygous FH, when matched for a given LDL-receptor mutation. The mechanism for the relative hypolipidaemia is speculative. It is postulated that the mild anaemia seen in β-thalassaemia carriers, which leads to increased bone marrow activity, increases the requirement for cholesterol, which might be met by upregulation of LDL receptors. Alternatively or in addition it is suggested that the tendency to chronic haemolysis leads to mild chronic activation of the macrophage system with cytokine production as in chronic inflammatory states. It has been shown that interleukin-1 and -6 and tumour necrosis factor-α can influence lipoprotein metabolism. In HepG2 cells they inhibit secretion of apoB-containing lipoproteins as well as upregulating the LDL receptor. In addition

Table 1.1 Plasma lipid values in heterozygous FH subjects with Fs572 mutation of LDL-R gene with and without Q39X mutation in β-globin gene

	Non-carriers (*n*=27)	**Q39X Heterozygotes (*n*=12)**	***P***	**Q39X Homozygote (*n*=1)**
BMI, kg/m^2	22.7±2.5	22.3±2.3	NS	18.7
TC, mmol/L	9.96±1.71	7.25±0.99	<0.001	3.67
LDL-C, mmol/L	8.25±1.66	5.76±1.08	<0.001	2.61
HDL-C, mmol/L	1.19±0.21	1.10±0.23	NS	0.67
TG, mmol/L	1.20±0.51	0.86±0.61	NS	0.87

BMI values (mean±SD) are adjusted for sex and age; lipid values (mean±SD) are adjusted for

age, sex, BMI, and apoE genotype. NS indicates not significant.
Source: Deiana *et al.* (2000).

to the more obvious candidate genes there are likely to be many other genetic and environmental factors that contribute to the phenotypic variability in FH.

A functional polymorphism in the promoter region of the microsomal triglyceride transfer protein (MTP −493G/T) influences lipoprotein phenotype in familial hypercholesterolemia.
B Lundahl, T P Leren, L Ose, A Hamsten, F Karpe. *Arterioscler Thromb Vasc Biol* 2000; **20**:1784–8.

B**ACKGROUND. The microsomal triglyceride transfer protein (MTP) has a key function in intracellular apoB lipidation and secretion of very low-density lipoprotein (VLDL). Functional polymorphism in the promoter of the MTP gene (−493G/T) affects the plasma concentration of LDL cholesterol and the VLDL distribution between large and small particle species in healthy men. We studied the effect of the MTP−493G/T polymorphism in 217 men and 211 women with heterozygous FH.**

I**NTERPRETATION.** The LDL cholesterol-lowering effect of the rare MTP gene promoter variant (MTP −493T) present in healthy subjects is shifted to a triglyceride (TG)-lowering effect in FH. These data suggest that the MTP gene has a role in modulating the clinical phenotype of FH.

Comment

MTP is found on the luminal side of the endoplasmic reticulum of tissues that secrete apoB-containing lipoproteins. These include not only the liver and small intestine, but also the heart, which was recently shown to secrete particles containing apoB-100 |**13**|. MTP transfers lipids, predominantly TGs and cholesteryl esters to nascent apoB-containing lipoproteins |**14**|. Absence of functional MTP causes abetalipoproteinaemia. The gene for MTP is another candidate to modify phenotype and possibly prognosis in FH. Here a polymorphism in the promoter region of the MTP gene is shown to lower serum TG levels in heterozygous FH (whereas it leads to lower LDL cholesterol in the general population). It remains to be seen if, as might be expected, it is associated with a lower incidence of CHD.

How can we assess risk better in familial hypercholesterolaemia?

Given that risk of CHD and the age of clinical onset are so variable in heterozygous FH ways of refining risk assessment over and above the family history and standard laboratory variables are sorely needed. Over the last few years there has been considerable interest in the use of coronary calcification, measured by electron beam or spiral computed tomography, as an indicator of coronary atherosclerosis.

Association of coronary heart disease with age-adjusted aortocoronary calcification in patients with familial hypercholesterolaemia.

J M Jensen, L U Gerdes, H K Jensen, *et al. J Intern Med* 2000; **247**: 479–84.

B**ACKGROUND. Existing diagnostic models of risk of CHD do not pertain to patients with FH, whose arteries have been exposed to hypercholesterolaemia since birth. Four diagnostic models of CHD (traditional risk factors of CHD—age, sex, cholesterol, hypertension, smoking and body mass index), cholesterol year score, and aortic as well as coronary calcium were compared in 80 individuals with molecularly defined FH.**

I**NTERPRETATION.** The age-adjusted coronary calcium score shows promise as an indicator of CHD in FH patients; it was significantly more strongly associated with clinical manifestations of CHD than were traditional risk factors, cholesterol year score and the age-adjusted aortic calcium score.

Comment

This is a cross-sectional study that investigates a variety of methods for predicting prevalent clinical CHD among patients with heterozygous FH. Coronary calcium score did best of the measures assessed. It was better than both conventional risk factors (though family history was not included) and aortic calcium score, even though in homozygous FH calcium is deposited in the ascending aorta ahead of the coronary arteries. Prediction is far from perfect but probably represents an improvement on conventional methods. It would be relevant, but more difficult, to determine whether measurement of coronary calcium can predict development of new clinical CHD in asymptomatic patients.

The vessel wall, homocysteine and familial hypercholesterolaemia

Impaired endothelium-dependent vasodilatation has been demonstrated previously in subjects with atherosclerosis and its conventional risk factors, including adults and children with FH |**15,16|.** Over the last decade there have been numerous studies looking at the relationship between dyslipidaemia and a number of variables related to vessel wall

mechanics and function, including endothelium-dependent and independent relaxation, intima-media thickness and wall compliance. There are inconsistencies in the data and considerable variation in the techniques used and the vascular territories and patients studied. Overall, however, it is impressive that there tends to be a rapid improvement in response to lipid-lowering treatment in at least some of the variables measured in most studies.

The effect of cholesterol lowering on carotid and femoral artery wall stiffness and thickness in patients with familial hypercholesterolaemia.

T J Smilde, F W van den Berkmortel, H Wollersheim, *et al. Eur J Clin Invest* 2000; **30**:473–80.

B**ACKGROUND. The effects of cholesterol reduction on artery wall stiffness and intima media thickness were investigated in 45 patients with FH, with untreated LDL cholesterol concentrations>9 mmol/l, before and after 1 year of cholesterol lowering therapy with statins (simvastatin, atorvastatin 40–80 mg daily).**

I**NTERPRETATION.** In FH, treatment decreased wall stiffness in the common femoral artery and wall thickness in the common carotid artery. Mean total cholesterol, LDL cholesterol and TGs were reduced significantly by 43%, 51% and 25%, respectively, whereas HDL cholesterol increased by 13%. Distensibility and compliance increased significantly in the common femoral artery but not in the common carotid artery. Intima medial thickness decreased significantly in the common carotid artery of both sexes, and in the bulb and internal carotid artery in premenopausal women. LDL cholesterol reductions of about 45% were needed to induce significant decreases in intima medial thickness and increases in distensibility and compliance.

Comment

Reducing LDL cholesterol for a year by about 45% produced significant improvements in the distensibility and compliance of the common femoral but not in the common carotid artery of this group of FH patients. In the carotid, however, intima-media thickness was reduced. The reasons for the different effects in different territories are unclear and may be partly methodological and partly related to different effects in elastic (carotid) as opposed to muscular (femoral) arteries.

Calf and forearm blood flow in hypercholesterolemic patients. A Cortella, S Zambon, G Sartore, *et al. Angiology* 2000; **51**:309–18.

B**ACKGROUND. Patients with hypercholesterolaemia without vascular disease have impaired endothelium-dependent (nitric oxide-mediated) vasodilation in coronary and peripheral vascular beds. To establish whether hypercholesterolaemia (and its reduction) also affects the microcirculation, vasomotion during postischaemic hyperaemia in both calf and forearm in 13 male patients with heterozygous FH and 10 male controls, free from vascular lesions, were studied. Plasma lipids, haematological parameters and limb vasoreactivity were evaluated while the patients were treated with diet alone, and during therapy with simvastatin.**

I**NTERPRETATION.** Hypercholesterolaemia does not affect vasodilation in the forearm as determined by postocclusive reactive hypeaemia, while in the calf hyper-cholesterolaemia is associated with higher resting vascular resistance, lower peak flow during reactive hyperaemia, and lower flow debt repayment. These abnormalities are corrected by the hypocholesterolaemic treatment.

Comment

In the 13 patients with FH in this study no abnormalities were seen in the haemo-dynamic variables studied in the forearm during reactive hyperaemia or after isosorbide dinitrate, with or without treatment with simvastatin. In the calf, however, resting vascular resistance was higher and peak flow during reactive hyperaemia was lower in the FH patients than in controls and tended to improve after treatment with simvastatin for 3 months. Peak blood flow during reactive hyperaemia is not nitric oxide dependent. This study contrasts with some previous work therefore in that no defect of endothelium-dependent vasomotion was found in the FH subjects. Neither were any abnormalities found after isosorbide dinitrate, which operates by endothelium-independent mechanisms. Nevertheless, a defect of lower limb blood flow that tended to improve after simvastatin treatment was identified in the FH subjects, although the precise mechanisms were unclear.

Effect of common methylenetetrahydrofolate reductase gene

mutation on coronary artery disease in familial hypercholesterolemia.
M Kawashiri, K Kajinami, A Nohara, *et al. Am J Cardiol* 2000; **86**:840–5.

B**ACKGROUND. CAD development in FH shows considerable interindividual variations. Elevated levels of plasma homocysteine have been recognized as independent risk factors for CAD. A 5,10-methylenetetrahydrofolate reductase (MTHFR) gene mutation (valine [V] substituted for alanine [A]) has been reported to be associated with elevated levels of plasma homocysteine in mutant homozygotes (i.e. W). Male heterozygous FH patients, 99 with and 100 without CAD, were studied.**

I**NTERPRETATION.** The MTHFR mutation appears to accelerate the onset of CAD through elevation of plasma homocysteine levels in male heterozygous patients with FH. In the CAD group, genotype VV and V alleles were significantly more frequent than in the non-CAD group. CAD development was significantly accelerated by MTHFR mutation. Mean plasma homocysteine levels of genotype VV were significantly higher than those of the other two genotypes.

Comment

Elevated plasma homocysteine levels are an independent risk factor for cardiovascular disease. They can usually be reduced by taking folic acid supplements. Possible further reductions may be achieved with vitamins B_{12} and B_6. A number of randomized controlled clinical trials are in progress and it remains to be seen if intervention to lower homocysteine levels will reduce clinical events. In the

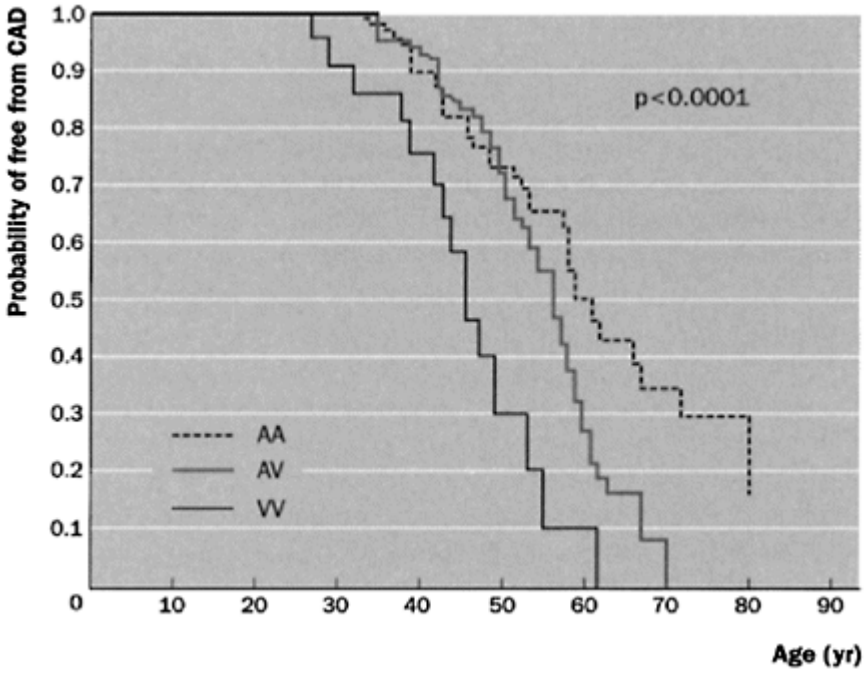


Fig. 1.2 Kaplan-Meier survival curves showing probability of freedom from CAD. Onset ages in the CAD group were considered as the time of events. The ages of patients in the non-CAD group, which indicated potential lifetime free from CAD, were considered as the time of the censored data. By log-rank test, a significant difference in survival curves was observed among these three genotypes (P<0.0001), and also was observed between all three pairs of genotypes (P=0.022 in AA versus AV, P=0.0002 in AV vs VV and P<0.0001 in AA versus VV). Source: Kawashiri et al. (2000).

meantime it is suggested that folic acid supplementation (400 μg daily may be sufficient for most people) is likely to be harmless and potentially beneficial for patients at high risk of cardiovascular disease |**17**|.

If homocysteine is a predictor of cardiovascular risk in the general population, is it also a factor in the determination of risk in FH? This study of male Japanese FH heterozygotes suggests that it may be. HDL cholesterol was significantly lower in the FH subjects with CHD and impaired glucose tolerance was more common. In a multiple regression analysis, however, only the VV genotype of methylenetetra-hydrofolate reductase, which was associated with the highest homocysteine levels, was an independent predictor for the age of onset of CHD in these subjects. Folate supplementation merits consideration in high-risk FH subjects.

If homocysteine is a risk factor in FH, does supplementation with folic acid reduce risk in these patients?

Effects of oral folic acid supplementation on endothelial function in familial hypercholesterolemia. A randomized placebo-controlled trial.

M C Verhaar, R M Wever, J J Kastelein, *et al. Circulation* 1999; **100**: 335–8.

B**ACKGROUND. Folates may help to reduce cardiovascular risk. This prospective, randomized, double-blind, placebo-controlled cross-over study examined whether oral folic acid supplementation (5 mg orally for 4 weeks) could improve endothelial function, as an intermediate end-point for cardiovascular risk, in 20 patients with FH.**

I**NTERPRETATION.** Oral supplementation with folic acid can improve endothelial function in patients with increased risk of atherosclerotic disease due to hypercholesterolaemia, without changes in plasma lipids. In FH patients, folic acid supplementation restored the impaired endothelium-dependent vasodilation, but did not significantly influence endothelium-independent vasodilation or basal forearm vasomotion.

Comment

Folic acid in a dose of 5 mg/day improves impaired endothelial function as judged by forearm blood flow in these patients with FH. The study used a cross-over design without an intervening washout period. This may have led to some carry-over effect from the folate supplementation, although presumably this would tend to minimize the differences between the folate and placebo treatments.

As the authors point out the effects of folate may be mediated by a reduction in homocysteine levels but other mechanisms are possible, given that these subjects had normal homocysteine levels to start with and that the reduction on treatment was modest. Other work suggests that 400 µg of folic acid daily may be sufficient to produce the maximum folate-dependent reduction in homocysteine concentra-tion. Here a much higher dose was used over a short period (4 weeks) and if other mechanisms are involved it remains to be established what the optimum dosage is over the long term. It also remains to be seen whether improved endothelial function secondary to folate supplementation translates into improved prognosis.

Pharmacological treatment of familial hypercholesterolaemia

There are currently five statins available in the UK and all are valuable in the treatment of hypercholesterolaemia generally as well as in FH |**18**|. Lovastatin, which is widely used in the USA, is not available in the UK.

In the UK recent evidence suggests that drug treatment to lower cholesterol generally

is used in only a relatively small proportion of individuals who meet the recommended criteria.

Lipid concentrations and the use of lipid-lowering drugs: evidence from a national cross sectional survey.
P Primatesta, N R Poulter. *BMJ* 2000; **321**:1322–5.

B**ACKGROUND. The prevalence of the use of lipid-lowering agents and its relation to blood lipid concentrations in English adults was determined in a cross-sectional survey of 13 586 adults (aged ≥16 years) living in non-institutional households.**

I**NTERPRETATION.** Only 2.2% of adults were taking lipid-lowering drugs in 1998 despite the high prevalence of dyslipidaemia in English adults. Among high-risk patients who were eligible for primary prevention with lipid-lowering drugs, rates of treatment were low (3%), and less than a third of patients with established cardiovascular disease received such treatment.

Comment

Recent guidelines produced in the UK by the relevant specialist societies |**19**| and endorsed by the National Service Framework on Coronary Heart Disease produced by the National Institute for Clinical Excellence (NICE) recommend that in subjects with overt vascular disease serum cholesterol should be reduced to 5.0 mmol/l or less and LDL cholesterol to 3.0 mmol/l or less. In the context of primary prevention subjects whose 10-year risk of a CHD event exceeds 30%, as assessed by the Framingham risk equation, are recommended for lipid-lowering treatment with the same targets as in secondary prevention.

This study, based on information from the Health Survey for England for 1998 suggests that only about 30% of patients with established CHD were taking lipid-lowering treatment and only some 3% of those with a greater than 30% risk of a CHD event over 10 years. Overall only 2.2% of adults under the age of 75 years were on lipid-lowering medication—hardly impressive for a nation with such a high incidence of CHD. The situation is, however, gradually improving. Only 0.4% of adults were being treated with lipid-lowering drugs in 1994.

The statins differ in potency and cost and possibly in a variety of LDL-independent effects, which may be important for their antiatherogenic properties |20|. Atorvastatin is particularly potent at lowering LDL |**21**|. Recently however, there has been some interest in possible differential effects of high doses of statins on HDL cholesterol |**22**|. These authors draw attention to a positive dose-response relationship between simvastatin and increase in HDL cholesterol and a negative relationship for atorvastatin. This does not seem invariably to be the case, however, and a very recent study of atorvastatin 80 mg daily in patients with FH was associated with a substantial increase in HDL cholesterol of

13.2%, which was indistinguishable from that obtained with simvastatin 40 mg |**23**|.

Comparison of therapy with simvastatin 80 mg and atorvastatin 80 mg in patients with familial hypercholesterolaemia.
A S Wierzbicki, P J Lumb, G Chik, M A Crook. *Int J Clin Pract* 1999; **53**: 609–11.

B**ACKGROUND. This study compared the efficacy of simvastatin 80 mg and atorvastatin 80 mg daily in the treatment of 26 patients with FH over 12 weeks using an open cross-over design.**

I**NTERPRETATION.** Both drugs appear to be equally effective in LDL reduction but simvastatin is superior in raising HDL and causes fewer side-effects. Both drugs similarly reduced LDL and median TGs but atorvastatin reduced HDL while simvastatin increased it affecting the LDL: HDL ratio (4.5 versus 3.7). Atorvastatin raised median fibrinogen by 15% compared with a non-significant 5% increase with simvastatin. Simvastatin reduced Lp (a) by a median 20% compared with baseline compared with 5% for atorvastatin. Side-effects, mostly gastrointestinal, were seen in four patients (16%) with atorvastatin compared with one case of myalgia with simvastatin (4%).

Comment

Although at lower doses atorvastatin has generally been reported to lower LDL cholesterol more than simvastatin, in this group of 26 heterozygous FH patients 80 mg of the two agents produced similar reductions in LDL cholesterol. The mechanism for the significantly different effects on HDL cholesterol (atorvastatin −2%, simvastatin+8%) is unknown, as indeed is its clinical significance. The inverse relationship in epidemiological studies between HDL cholesterol and CHD events is well established. It is not known if changes in HDL cholesterol induced by diet or pharmacological agents have the same clinical significance, although they may. The Veterans Affairs HDL Intervention Trial (VAHIT) |**24**| seems at first sight to suggest that increasing HDL cholesterol with a fibrate (gemfibrozil) does reduce CHD events as there was no effect on mean LDL cholesterol. This remains uncertain, however, as there were significant reductions in serum TGs and fibrates are known to exert favourable effects on LDL composition by reducing small dense LDL while preserving the more buoyant lipid-rich particles that are believed to be less atherogenic. The issue could only be resolved by a large head-to-head endpoint study, which is perhaps unlikely to be forthcoming. The outcome is likely to depend on the mechanisms by which HDL exerts its beneficial effects. These are incompletely understood but may involve promotion of reverse cholesterol transport and inhibition of oxidation of atherogenic particles such as LDL. These effects are probably dependent on specific HDL subfractions and components and

it remains to be established to what extent these are differentially modified by different statins.

Stanol ester margarine alone and with simvastatin lowers serum cholesterol in families with familial hypercholesterolemia caused by the FH-North Karelia mutation.

A F Vuorio, H Gylling, H Turtola, *et al. Arterioscler Thromb Vasc Biol* 2000; **20**:500–6.

B**ACKGROUND. To retard development of atherosclerosis, cholesterol-lowering treatment should start in childhood in FH. Nineteen FH families consumed stanol ester margarine, which lowers serum cholesterol by inhibiting cholesterol absorption, for 12 weeks. Each individual replaced part of their dietary fat with 80% rapeseed oil margarine containing stanol esters. The families included 24 children with the North Karelia variant of FH (FH-NK), four FH-NK parents, and 16 healthy family members, and a separate group of 12 FH-NK adults who consumed the margarine for 6 weeks and who were taking simvastatin (20 or 40 mg daily).**

I**NTERPRETATION.** In a genetically defined population of FH patients, a diet including stanol ester margarine was a safe and effective hypolipidaemic treatment for children and adults. In FH-NK adults on simvastatin therapy, including a stanol ester margarine in the regimen, could reduce serum LDL cholesterol levels even further.

Comment

There has been considerable interest recently in the use of poorly absorbable esters of plant origin taken orally in the form of margarine to interfere with cholesterol absorption and lower cholesterol. Of two such spreads that have been marketed one contains stanol esters (predominantly sitostanol), the other phytosterol esters. They commonly reduce LDL cholesterol by an average of 0.3–0.5 mmol/l |**25**| and the effect is dose related, so some patients who have become used to minimal amounts of margarine on bread need to take more to get the optimal effect with an associated increase in calories.

The question arises as to the usefulness of such products as an adjunct to statin treatment in patients with severe genetic hyperlipidaemias such as FH. In this open study of FH heterozygotes with FH-NK mutation the stanol ester spread reduced LDL cholesterol by 18% in children and by 11% in adults. In adults who were also taking simvastatin the LDL cholesterol reduction mediated by the spread was 20%. TGs and HDL cholesterol were unchanged. These results look encouraging but it remains to be seen if such impressive results can be obtained with other FH mutations and in longer-term placebo-controlled studies.

Inhibition of cholesterol synthesis by atorvastatin in homozygous familial hypercholesterolaemia.
F J Raal, A S Pappu, D R Illingworth, *et al. Atherosclerosis* 2000; **150**: 421–8.

B**ACKGROUND. Patients with homozygous FH have markedly raised LDL cholesterol levels that are refractory to standard doses of lipid-lowering drug therapy. The effects of atorvastatin were evaluated in 35 patients with well-characterized homozygous FH. Patients received atorvastatin first 40 mg/day and then 80 mg/day with possible further dose increases as needed.**

I**NTERPRETATION.** LDL cholesterol levels were reduced by 17% at the 40 mg/day dose and by 28% at the 80 mg/day dose. Reductions in receptor-negative patients were similar to those in patients with residual LDL receptor activity. Plasma mevalonic acid (MVA) and 24-h urinary excretion of MVA, as markers of *in vivo* cholesterol synthesis, were raised at baseline and decreased markedly with treatment. The reduction in LDL cholesterol was correlated with the reduction in urinary MVA excretion. Increasing the dose of atorvastatin to 120 and 160 mg/day did not further reduce LDL cholesterol or urinary MVA excretion suggesting a plateau effect.

Comment

When statins were first introduced, knowing that their primary action was to inhibit cholesterol synthesis, many intuitively assumed that they would lower serum cholesterol by inhibiting lipoprotein secretion from the liver. This mode of action has been rather neglected since it became apparent that the statins worked principally by depleting hepatocyte cholesterol leading to upregulation of LDL receptors and thus promoting LDL catabolism. They were not effective, and would not be expected to be, in homozygous FH where there was little or no LDL-receptor function to be modulated.

Recently it has become apparent that high-dose simvastatin or atorvastatin can be helpful in the treatment of homzygous FH |**26,27**|. The present study confirms that LDL cholesterol can be reduced in homozygous FH by high-dose atorvastatin and that the reduction was similar in receptor-negative and receptor-defective patients. The average reduction of 28% in LDL cholesterol on 80 mg/day of atorvastatin is not a panacea in these patients with very high cholesterol levels but it is worth having and a useful adjunct to extracorporeal methods of LDL removal |**27**|. Increasing the dose of atorvastatin to 120 or 160 mg/day did not yield additional benefit. In the receptor-defective cases some of the effect may have resulted from a degree of increased LDL-receptor activity. The efficacy in the receptor-negative cases and the correlation between the decrease in LDL cholesterol and urinary mevalonate excretion (see Fig. 1.3) support the notion that a

major part of the effect relates to decreased lipoprotein production.

Management of familial hypercholesterolaemia in pregnancy

The statins are not known to be safe for the fetus and are generally regarded as contraindicated in pregnancy. Women with FH have a better prognosis than men and in young women not thought to be at particularly high risk it may be reasonable simply to ask them to discontinue their statin treatment, or perhaps use a resin with folic acid supplementation, before trying to conceive and during gestation. There will be those, however, where this approach is unacceptable and this is likely to be the case in severe heterozygotes with evidence of vascular disease and in homozygotes. Extracorporeal removal of LDL, by LDL apheresis or plasma exchange, is a

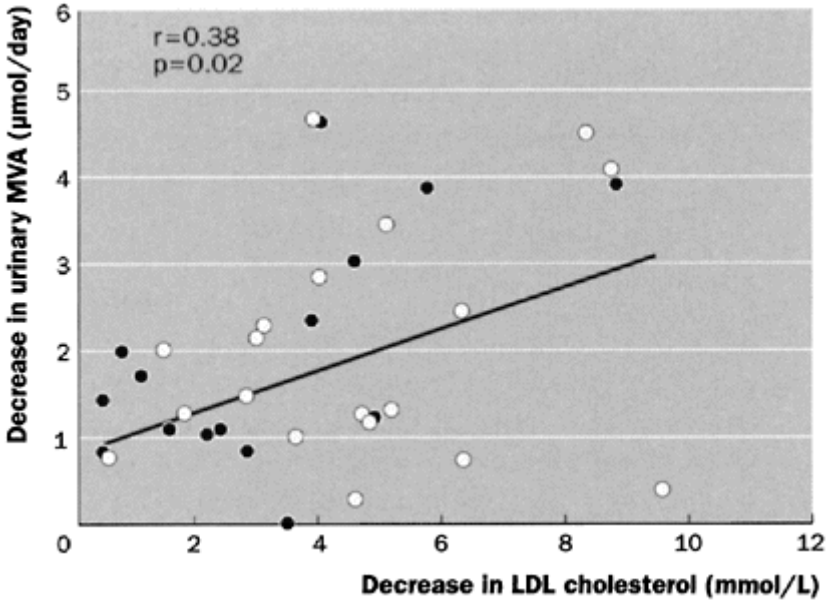


Fig. 1.3 Correlation between changes in LDL cholesterol and 24-h urinary excretion of mevalonic acid (MVA) with atorvastatin 40 mg/day (•) and 80 mg/day (O) in patients with homozygous FH. Source: Raal *et al.* (2000).

standard treatment for homozygous FH |**28**|, which in principle could also be used for homozygotes and high-risk heterozygotes during pregnancy.

Low-density lipoprotein apheresis therapy during pregnancy.
L Cashin-Hemphill, M Noone, J F Abbott, C A Waksmonski, R S Lees. *Am J Cardiol* 2000; **86**:1160 (A10).

B**ACKGROUND. Pregnancy in patients with severe hypercholesterolaemia and CAD is problematic both for mother and fetus; the most potent agents for LDL cholesterol reduction, the HMG-CoA reductase inhibitors (statins) cannot be used during pregnancy.**

I**NTERPRETATION.** In the case presented LDL apheresis via heparin-induced extracorporeal LDL precipitation was used safely and efficaciously during pregnancy in a woman with heterozygous FH and stable CAD.

Comment

This brief report describes the use of extracorporeal LDL precipitation in a 41-year-old FH heterozygote who had had a previous cardiac arrest with subsequent coronary artery bypass grafting. After successful vaginal delivery apheresis was continued for a further 2 months during breast-feeding.

Unstable angina during pregnancy in two patients with premature coronary atherosclerosis and aortic stenosis in association with familial hypercholesterolemia.
A B Hameed, P P Tummala, T M Goodwin, *et al. Am J Obstet Gynecol* 2000; **182**:1152–5.

B**ACKGROUND. Obstructive atherosclerotic CAD is rare in women of child-bearing age. Two young patients with premature CAD in association with FH had unstable angina in the second trimester of pregnancy, and were found to have CAD and aortic stenosis.**

I**NTERPRETATION.** The management of these conditions during pregnancy is influenced by the effects of available treatments on both maternal and fetal outcome. One patient opted for abortion at the twentieth week of gestation, and the other decided to continue pregnancy, and was delivered by caesarean at 28 weeks gestation. After pregnancy both patients had coronary artery bypass grafts; one also underwent aortic valve replacement, and the other had the narrowed ascending aorta replaced with an uneventful recovery.

Comment

Lipid deposits in the aortic root can give rise to an ejection systolic murmur and to supravalvar stenosis with a haemodynamically significant gradient in FH |**2**|. This is a frequent problem in homozygotes but can also occur in severe heterozygotes. This report describes two severely hypercholesterolaemic women presenting with unstable angina in pregnancy. Their serum cholesterol levels were 18.4 and 14.0 and from the description of their xanthomas it is possible that they were homozygous for FH, or at least very severe heterozygotes. Both required surgical widening of the aortic root and one also needed aortic valve replacement. Patients with severe FH can develop problems other than CAD and should be checked periodically for cardiac signs. When an aortic systolic murmur is present echocardiography should be performed and cardiological advice sought if appropriate.

Liver transplantation and low-density lipoprotein apheresis for homozygous familial hypercholesterolaemia

Liver transplantation in patients with homozygotic familial hypercholesterolemia previously treated by end-to-side portocaval shunt and ileal bypass.

M Lopez-Santamaria, L Migliazza, M Gamez, *et al. J Pediatr Surg* 2000; **35**: 630–3.

B**ACKGROUND. FH results from mutations in the gene that encodes the synthesis of the cellular receptor for LDL. Homozygous FH results in severe premature atherosclerosis and death from CAD usually before the age of 20. The only effective treatment is liver transplantation, which, alone or with medications, normalizes plasma cholesterol levels. The cases of two siblings with homozygous FH are reported.**

I**NTERPRETATION.** Portocaval shunt and ileal bypass are not indicated in homozygous FH, not even for the purpose of delaying liver transplantation. The patients underwent portocaval shunt at the ages of 2.5 and 1.5 years, respectively, resulting in immediate, but insufficient, reductions in cholesterol. Ileal bypass a year later was ineffective. Liver transplantation at the ages of 18 and 16 years, reduced plasma cholesterol to 129 and 225 mg/dl, respectively. The earlier operations seriously increased the technical difficulty of liver transplantation and did not have a favourable effect on the natural course of the disease.

Comment

Subjects with untreated homozygous FH commonly die of CHD in childhood or early adult life |**2**|. They are usually poorly responsive to statin treatment, although atorvastatin is licensed in the UK for use in homozygotes. Extracorporeal removal of LDL has become a standard treatment but some patients have undergone drastic surgical interventions to ameliorate their serum cholesterol levels. Twenty to 30 years ago some homozygotes were treated with portocaval anastamosis with variable effects in reducing LDL cholesterol. This procedure was subsequently supplanted by liver transplantation as a means of transplanting LDL receptors |**29**|. The present study cautions against using portocaval anastamosis in infants with homozygous FH even as a method of delaying transplantation as it makes the latter technically much more difficult.

Prospective randomised cross-over comparison of three LDL-apheresis systems in statin pretreated patients with familial hypercholesterolaemia.

S Schmaldienst, S Banyai, T M Stulnig, *et al. Atherosclerosis* 2000; **151**: 493–9.

B**ACKGROUND. Various LDL-apheresis systems are used to treat severe FH, particularly patients with CAD. This prospective cross-over comparison of three commercially available LDL-apheresis systems (immunoadsorption, dextran sulphate adsorption, direct adsorption of lipoproteins) was done in three patients with homozygous and five with heterozygous FH.**

I**NTERPRETATION.** Removal of atherogenic lipoproteins was highly effective in all systems, for LDL cholesterol in particular. Lp(a) was reduced by about 63% with each device. The loss of HDL cholesterol was highest with immunoadsorption. Fibrinogen removal was significantly higher with dextran sulphate adsorption than with immunoadsorption and significantly higher with immunoadsorption than with direct adsorption of lipoproteins. Treatment duration was shortest with direct adsorption of lipoproteins. No side-effects were recorded during 96 treatments. Long-term observations are needed to assess whether the differences in efficacy are clinically relevant.

Comment

Extracorporeal removal of lipoproteins by plasma exchange or LDL apheresis has been a mainstay of the management of homozygous FH and an alternative to liver transplantation for more than 25 years |**28**|. It appears to be effective in pro-longing survival in these patients |**3,30**|. The present study was performed in a mixed group of homozygous and heterozygous FH patients. Direct absorption of lipoproteins (on to polyacrylate-coated polyacrylamide) was marginally less effective at removing LDL than the other two systems, but was the quickest to perform. All methods were comparable in

terms of reduction of Lp(a) but dextran sulphate adsorption removed more fibrinogen and less HDL cholesterol than the other methods. Only the immunoadsorption columns are reusable. A fourth method

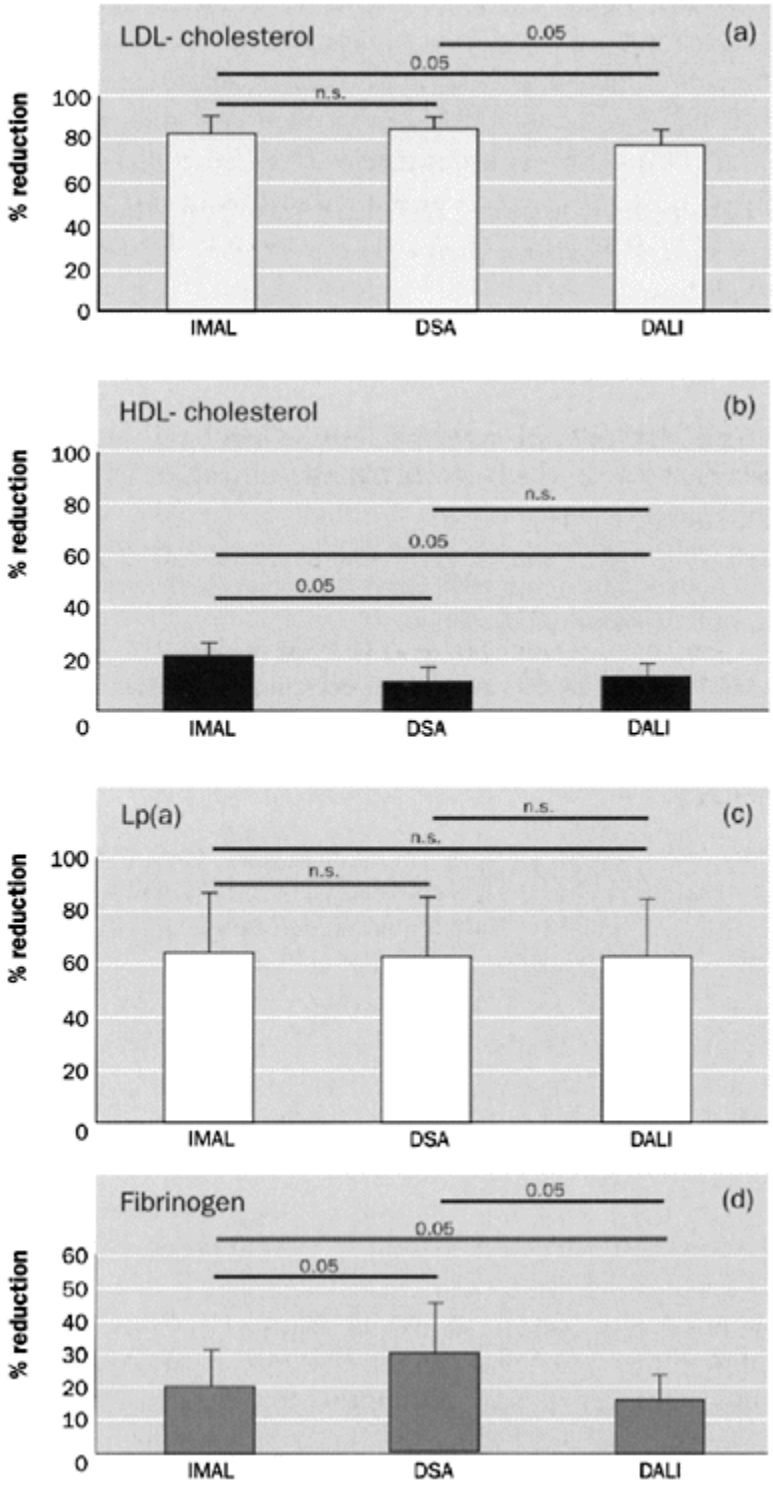

LDL- cholesterol
(a)
0.05
0.05
n.s.
% reduction
100
80
60
40
20
0
IMAL
DSA
DALI
HDL- cholesterol
(b)
n.s.
0.05
0.05
% reduction
100
80
60
40
20
0
IMAL
DSA
DALI
Lp(a)
(c)
n.s.
n.s.
n.s.
% reduction
100
80
60
40
20
0
IMAL
DSA
DALI
Fibrinogen
(d)
0.05
0.05
0.05
% reduction
60
50
40
30
20
10
0
IMAL
DSA
DALI

Fig. 1.4 Lipoprotein and fibrinogen reduction (%) by three different LDL-apheresis systems: IMAL, immunoadsorption; DSA, dextrane sulphate adsorption; DALI, direct adsorption of lipoproteins. Source: Schmaldienst *et al.* (2000).

(heparin-induced LDL precipitation) was not studied. It is not known if the differences observed between the methods used in this investigation are of prognostic significance.

Influence of LDL apheresis on LDL subtypes in patients with coronary heart disease and severe hyperlipoproteinemia.
B M Schamberger, H C Geiss, M M Ritter, P Schwandt, K G Parhofer. *J Lipid Res* 2000; **41**:727–33.

B**ACKGROUND. Small dense LDL subtypes are more atherogenic than large, buoyant LDL subtypes. LDL apheresis effectively lowers elevated LDL cholesterol but it is not known whether such therapy changes the LDL subtype distribution. The influence of LDL apheresis on LDL subtype distribution was evaluated in 22 patients with CHD and FH, six with combined hyperlipidaemia, and four with Lp(a)-hyperlipoproteinaemia who were regularly treated using various types of LDL apheresis.**

I**NTERPRETATION.** LDL apheresis not only decreases LDL mass, but also improves LDL density profile, particularly in patients with combined hyperlipidaemia. The proportion of larger LDL subfractions increased and that of small dense subfractions decreased concomitantly after apheresis. Small dense LDL reduction was more prominent in patients with combined hyperlipidaemia compared with patients with FH or Lp(a)-hyperlipoproteinaemia, but the type of apheresis technique had no effect.

Comment

Small dense LDL are thought to be particularly atherogenic |**31**|. This may relate partly to their association with the atherogenic lipoprotein phenotype characterized by high TGs and low HDL cholesterol. It is also likely to result from properties of the particles themselves, which are poorly cleared by the LDL receptor and consequently have a longer residence time and are more prone to oxidation. This work shows a favourable shift in LDL particle distribution after apheresis, which is independent of the type of apheresis method used. The mechanism is unclear and the phenomenon was more marked in patients with FCH than in FH, in which the effect was small.

Familial defective apolipoprotein B-100

In principle, defective LDL catabolism as a result of impaired binding of LDL to its receptor might be due to abnormalities in either the receptor or the ligand, namely the protein moiety of LDL, apoB-100. Receptor defects were identified by Brown

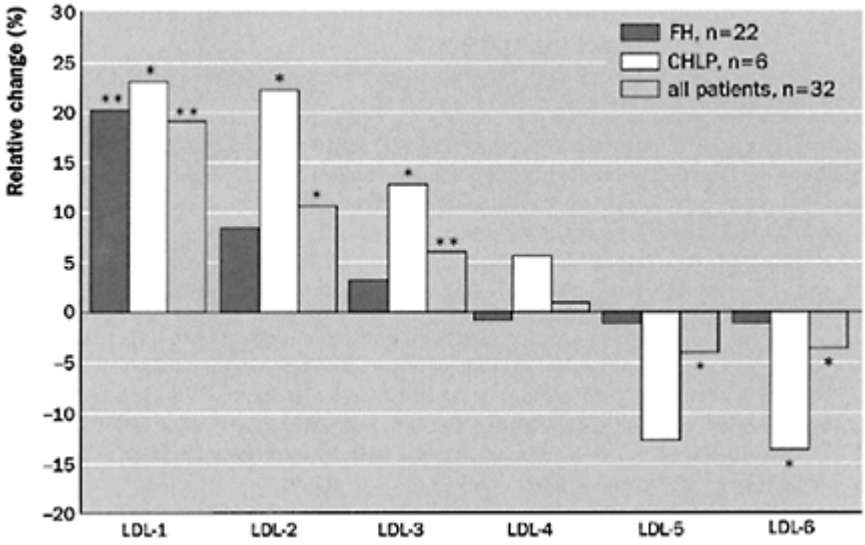


Fig. 1.5 Mean relative increase and decrease of LDL subfractions induced by apheresis according to the underlying hyperlipoproteinaemia. ***P*<0.01, **P*<0.05, Wilcoxon test. Source: Schamberger *et al.* (2000).

and Goldstein |**2**| as the basis of FH in the 1970s. It was not until the late 1980s that a specific defect in apoB-100 leading to hypercholesterolaemia was described by Grundy and colleagues |**32**|. A point mutation in the apoB-100 gene leads to substitution of glutamine for arginine at amino acid 3500 |**33**|.

The disorder seems to be common with a prevalence in the range 1 in 500–1000 in the UK population, not dissimilar from that of heterozygous FH. In a lipid clinic population of phenotypic FH, however, only some 2–5% have FDB |**34**|. FDB heterozygotes have more moderate elevations of LDL cholesterol than FH heterozygotes. This may be because of intact LDL receptor function allowing normal clearance of VLDL remnants and reducing conversion of VLDL to LDL. This milder phenotype may help to explain the under-representation of FDB among severely hypercholesterolaemic patients in specialist clinics and the apparently lower risk of CHD.

Although there are hundreds of different mutations in the LDL receptor leading to phenotypic FH, the number of functional defects in apoB-100 causing hypercholesterolaemia is small, with the arginine to glutamine substitution at position 3500

predominating. There are also two other much rarer mutations at positions 3500 and 3501 |**35**|.

Increased production of HDL ApoA-I in homozygous familial defective ApoB-100.

J R Schaefer, K Winkler, H Schweer, *et al. Arterioscler Thromb Vasc Biol* 2000; **20**:1796–9.

B**ACKGROUND. FDB is a frequent cause of hypercholesterolaemia, although hypercholesterolaemia in homozygous FDB is less severe than that in homozygotes for FH. Decreases in LDL apoB-100 fractional catabolism and in LDL production, due to enhanced removal of apoE-containing precursors, have been shown in a patient with homozygous FDB. The effects of defective apoB-100 on HDL metabolism are unknown. HDL apoA-I metabolism in the FDB patient and in six controls was studied using L-[2–^{3}H]-leucine as a tracer.**

I**NTERPRETATION.** The results show that defective apoB-100 may influence HDL kinetics. The fractional catabolic rate and the production rate of apoA-I were increased in the FDB patient. The increase in total HDL turnover might enhance reverse cholesterol transport and could contribute to the seemingly benign clinical course of FDB compared with that of FH.

Comment

This study of apoB-100 kinetics involves a single homozygous FDB patient. His total serum cholesterol was only 7.3 mmol/l, which is relatively modest even by FH heterozygote standards and underscores the point that the phenotype in FDB can be relatively mild. ApoB-100 kinetic studies in FH reveal an increase in production rate of LDL apoB and a decrease in its fractional catabolic rate. By contrast in FDB LDL apoB production rate and fractional catabolic rate are both decreased. There are also differences in HDL apoA-I kinetics. In FH, a decreased apoA-I production rate was associated with an increase in fractional catabolic rate. In homozygous FDB, however, this study shows for the first time that there is an increase in both the production and fractional catabolic rate of HDL apoA-I. The authors postulate that these findings suggest upregulation of reverse cholesterol transport, which may contribute to a reduced risk of premature CHD in FDB, when compared with FH. Their patient had a low HDL cholesterol concentration of 0.8 mmol/l, but there is independent evidence that hypoalphalipoproteinaemia associated with high apoA-I turnover may be less atherogenic |**36**|.

Autosomal dominant type IIa hypercholesterolemia: evaluation of the respective contributions of LDLR and APOB gene defects as well as a third major group of defects.
B Saint-Jore, M Varret, C Dachet, *et al. Eur J Hum Genet* 2000; **8**:621–30.

B**ACKGROUND. Numerous molecular defects in the LDL receptor and a few specific mutations in the apoB gene result in FH and FDB, respectively. To estimate the**

respective contribution of LDL receptor, apoB and other gene defects in autosomal dominant type IIa hypercholesterolaemia (ADH), we studied 33 French families with ADH, diagnosed over at least three generations, using the candidate gene approach.

I**NTERPRETATION.** The results demonstrate that the relative contributions of LDL receptor and apoB gene defects to ADH are very different, that genetic heterogeneity is generally underestimated in ADH, and that at least three major groups of defects are involved. The LDL receptor gene defect was estimated to be involved in roughly 50% of the families, whereas the estimated contribution of an apoB gene defect was only 15%. More surprisingly, 35% of the families in the sample were estimated to be linked to neither LDL receptor nor apoB genes.

Comment

The WHO classification provided a useful descriptive terminology for different hyperlipidaemic phenotypes but is falling progressively into disuse as the underlying pathogenesis is more frequently elucidated. It is possible for example to have a type II phenotype for many different reasons. It may be due to FH or FDB but more commonly it will be polygenic in origin. Type IIa or IIb patterns are common in FCH and may also be found in a number of secondary causes, including hypothyroidism and the nephrotic syndrome. Where there is no secondary cause and a severe type II pattern appears to be dominantly inherited, however, there is a tendency to suspect FH clinically, or perhaps FDB.

It is likely that any large group of such patients will, however, contain a significant number of patients with disorders of LDL metabolism that are poorly characterized. The literature is briefly reviewed in this paper. The authors have confined themselves in this study to families with a dominantly inherited IIa pattern to avoid confusion with FCH, which might have arisen if families with mixed IIa and IIb patterns had been included. The patients were severely hypercholesterolaemic (mean serum cholesterol 10.1 mmol/l). The study confirms other work suggesting that in selected populations with severe

hypercholesterolaemia FDB is much less common than FH, being found in about 15% of the families. FH accounted for hyperlipidaemia in half the families and in 35% the cause was unknown and due to neither FH nor FDB.

Conclusion

At the clinical level the most obvious deficit is the underdiagnosis of heterozygous FH in the general population and the insensitivity of a high-risk population screening strategy (of the type operating in the UK) for the detection of these individuals. The identification of a new case, or follow-up of a known case, represent, therefore, valuable opportunities to identify and investigate first-degree relatives, at least half of whom are likely to be affected. There remains much to be discovered about the mechanisms underlying the variability in risk in FH and also about the underlying defects in those subjects with dominantly inherited type II hyperlipidaemia who do not have FH or FDB.

References

1. Muller C. Angina pectoris in hereditary xanthomatasis. *Arch Intern Med* 1939; **64**: 675–700.
2. Goldstein JL, Hobbs HH, Brown MS. Familial hypercholesterolemia. In: Scriver CR, Beaudet AL, Sly WS, Valle D (eds). *The Metabolic and Molecular Bases of Inherited Disease*, 7th edn. New York: McGraw-Hill, 1995:1981–2030.
3. Thompson GR. Familial hypercholesterolaemia. In: Betteridge DJ, Illingworth DR, Shepherd J (eds). *Lipoproteins in Health and Disease*, 1st edn. London: Arnold, 1999: 675–92.
4. Marks D, Wonderling D, Thorogood M, Lambert H, Humphries SE, Neil HAW. Screening for hypercholesterolaemia versus case finding for familial hypercholesterolaemia: a systematic review and cost-effectiveness analysis. *Health Technol Assess* 2000; **4**: 1–123.
5. Kastelein JJP. Screening for familial hypercholesterolaemia. *Br Med J* 2000; **321**: 1483–4.
6. Kane JP, Malloy MJ, Ports TA, Phillips NR, Diehl JC, Havel RJ. Regression of coronary atherosclerosis during treatment of familial hypercholesterolemia with combined drug regimens. *JAMA* 1990; **264**:3007–12.
7. Bertolini S, Cassanelli R, Garuti M, Ghisellini M, Simone ML, Rolleri M, Masturzo P, Calandra S. Analysis of LDL receptor gene mutations in Italian patients with homozygous familial hypercholesterolemia. *Arterioscler Thromb Vasc Biol* 1999; **19**:408–18.
8. Heiberg A, Slack J. Family similarities in the age at coronary death in familial hypercholesterolaemia. *Br Med J* 1977; **2**:493–5.
9. O'Malley JP, Maslen CL, Illingworth DR. Angiotensin-converting enzyme and cardiovascular risk. *Curr Opin Lipidol* 1999; **10**:407–15.
10. O'Malley JP, Maslen CL, Illingworth DR. Angiotensin-converting enzyme DD genotype and cardiovascular risk in heterozygous familial hypercholesterolemia. *Circulation* 1998; **97**:1780–3.
11. Gaw A, Hobbs HH. Lipoprotein(a). In: Betteridge DJ, Illingworth DR, Shepherd J

(eds). *Lipoproteins in Health and Disease*, 1st edn. London: Arnold, 1999:87–109.
12. Lingenhel A, Kraft HG, Kotze MJ, Peeters AV, Kronenberg F, Kruse R, Utermann G. Concentrations of the atherogenic Lp(a) are elevated in familial hypercholesterolaemia: a sibpair and family analysis. *Eur J Hum Genet* 1998; **6**:50–60.
13. Boren J, Veniant MM, Young SG. Apo B100-containing lipoproteins are secreted by the heart. *J Clin Invest* 1998; **101**:1197–202.
14. Gordon DA. Recent advances in elucidating the role of the microsomal triglyceride transfer protein in apolipoprotein B lipoprotein assembly. *Curr Opin Lipidol* 1997; **8**: 131–7.
15. Celemajer DS, Sorensen KE, Gooch VM. Noninvasive detection of endothelial dysfunction in children and adults at risk of atherosclerosis. *Lancet* 1992; **340**:1111–15.
16. Chowienczyk PJ, Watts GF, Cockroft JR. Impaired endothelium-dependent vasodilation of forearm resistance vessels in hypercholesterolaemia. *Lancet* 1992; **340**: 1430–2.
17. Gerhard GT, Duell PB. Homocysteine and atherosclerosis. *Curr Opin Lipidol* 1999; **10**: 417–28.
18. Illingworth DR. Management of hypercholesterolemia. *Med Clin North Am* 2000; **84**: 23–42.
19. Wood D, Durrington P, Poulter N, McInnes G, Rees A, Wray R. Joint British recommendations on prevention of coronary heart disease in clinical practice. *Heart* 1998; **80** (Suppl 2): S1–S29.
20. Davignon J, Laaksonen R. Low-density lipoprotein-independent effects of statins. *Curr Opin Lipidol* 1999; **10**:543–59.
21. Jones P, Kafonek S, Laurora I. Comparative dose efficacy study of atorvastatin versus simvastatin, pravastatin, lovastatin and fluvastatin in patients with hypercholesterolemia (The CURVES study). *Am J Cardiol* 1998; **81**:582–7.
22. Mikhailidis DP, Wierzbicki AS. HDL-cholesterol and the treatment of coronary heart disease: contrasting effects of atorvastatin and simvastatin. *Curr Med Res Opin* 2000; **16**: 139–46.
23. Smilde TJ, van Wissen S, Wollersheim H, Trip MD, Kastelein JJP, Stahlenhoef AFH. Effect of aggressive versus conventional lipid lowering on atherosclerosis progression in familial hypercholesterolaemia (ASAP): a prospective, randomised, double-blind trial. *Lancet* 2001; **357**:577–81.
24. Rubins BH, Robins SJ, Collins D. Gemfibrozil for the secondary prevention of coronary heart disease in men with low levels of high-density lipoprotein cholesterol. *N Engl J Med* 1999; **341**:410–18.
25. Law M. Plant sterol and stanol margarines and health. *Br Med J* 2000; **320**:861–4.
26. Raal FJ, Pilcher GJ, Illingworth DR, Pappu AS, Stein EA, Laskarzewski P, Mitchel YB, Melino MR. Expanded-dose simvastatin is effective in homozygous familial hypercholesterolaemia. *Atherosclerosis* 1997; **135**:249–56.
27. Marais AD, Naoumova RP, Firth JC, Penny JC, Neuwirth CK, Thompson GR. Decreased production of low density lipoprotein by atorvastatin after apheresis in homozygous familial hypercholesterolemia. *J Lipid Res* 1997; **38**:2071–8.
28. Kajinami K, Mabuchi H. Therapeutic effects of LDL apheresis in the prevention of atherosclerosis. *Curr Opin Lipidol* 1999; **10**:401–6.
29. Bilheimer DW, Goldstein JL, Grundy SM, Starzl TE, Brown MS. Liver transplantation to provide low-density lipoprotein receptors and lower plasma cholesterol in a child with homozygous familial hypercholesterolaemia. *N Engl J Med*

1984; **311**:1658–64.
30. Thompson GR, Miller JP, Breslow JL. Improved survival of patients with homozygous familial hypercholesterolaemia treated with plasma exchange. *Br Med J* 1985; **291**: 1671–3.
31. Austin MA, Breslow JL, Hennekens CH, Buring JE, Willett WC, Krauss RM. Low-density lipoprotein subclass patterns and risk of myocardial infarction. *JAMA* 1988; **260**: 1917–21.
32. Soria LF, Ludwig EH, Clarke HRG, Vega GL, Grundy SM, McCarthy BJ. Association between a specific apolipoprotein B mutation and familial defective apolipoprotein B-100. *Proc Natl Acad Sci* 1989; **86**:587–91.
33. Tybjaerg-Hansen A. Familial defective apolipoprotein B-100. In: Betteridge DJ, Illing-worth DR, Shepherd J (eds). *Lipoproteins in Health and Disease*, 1st edn. London: Arnold, 1999:701–18.
34. Myant NB. Familial defective apolipoprotein B-100: a review, including some comparisons with familial hypercholesterolaemia. *Atherosclerosis* 1993; **104**:1–18.
35. Breslow JL. Genetics of lipoprotein abnormalities associated with coronary heart disease susceptibility. *Annu Rev Genet* 2000; **34**:233–54.
36. Rader DJ, Ikewaki K, Duverger N, Feuerstein I, Zech LA, Connor W, Brewer HB. Very low high-density lipoproteins without coronary atherosclerosis. *Lancet* 1993; **342**: 1455–8.

2
Hypertriglyceridaemia and mixed hyperlipidaemia

Introduction

The strong epidemiological relationship between total cholesterol and coronary heart disease (CHD) has dominated clinical thinking for many years. It is mediated through low-density lipoproteins (LDL), the major cholesterol carrying lipoprotein, which carries about 70% of the total serum cholesterol in normal subjects. This cholesterol-dominated view has been supported by the publication of the statin end-point trials beginning with the Scandinavian Simvastatin Survival Study (4S) in 1994 and subsequently in the Cholesterol and Recurrent Events (CARE), Long-term Intervention with Pravastatin in Ischaemic Disease (LIPID), West of Scotland Coronary Prevention Study (WOSCOPS) and Air Force/Texas Coronary Atherosclerosis Prevention Study (AFCAPS/TexCAPS) |**1**|. The principal effect of the statins is to lower LDL, although there are also usually modest reductions in serum tri-glycerides (TGs) and increases in high-density lipoprotein (HDL) cholesterol.

The clinical significance of hypertriglyceridaemia has been more contentious. Most studies have shown a significant univariate relationship between serum TGs and coronary events, but doubt was cast on any potential causal relationship when it was found that the significance disappeared if other variables, particularly HDL cholesterol (which varies inversely with serum TGs), were taken into account |**2**|. It has, however, been shown more recently that TGs remain a significant risk factor in some studies even after multivariate analysis |**3**|. TG-rich lipoproteins are very heterogeneous in composition and are likely to vary in their atherogenic potential.

Fibrates are more effective than statins at lowering TGs and the fibrate trials have generally shown a reduction in cardiovascular events, but so far evidence of ability to reduce total mortality, which is available for the statins, has not been demonstrated |**1,4,5**|.

The recent Veterans Administration HDL Intervention Trial (VAHIT) showed beneficial effects with gemfibrozil without any reduction in LDL cholesterol |**6**|. At first sight this seems to suggest that the benefit must relate to changes in serum TGs and/or HDL cholesterol, but the situation is more complex. Fibrates change LDL composition in favour of larger less dense particles, which are believed to be less atherogenic. This change in LDL composition may have contributed to the improved outcome even in the absence of any change in LDL cholesterol.

Postprandial lipaemia

One difficulty in assessing the role of TGs and TG-rich lipoproteins in vascular disease is

the marked diurnal variation in their concentration related to dietary fat intake. It is traditional to assess serum TGs on fasting samples because of this but unrepresentative because most individuals spend the majority of the time in the absorptive state. It is difficult to interpret non-fasting samples, however, because they represent different stages in the absorption of meals of differing composition.

Diurnal triglyceride profiles in healthy normolipidemic male subjects are associated to insulin sensitivity, body composition and diet. A J van Oostrom, M Castro Cabezas, J Ribalta, *et al. Eur J Clin Invest* 2000; **30**:964–71.

B**ACKGROUND. Raised fasting and postprandial TGs are established risk factors for CHD. Fasting plasma TGs are usually measured, although TGs are mainly produced in the postprandial state. This study investigated diurnal TG profiles, in 48 normolipidaemic healthy men, using serial capillary TG measurements.**

I**NTERPRETATION.** The results showed that diurnal TG profiles in healthy normolipidaemic males are not age dependent, but are associated with insulin sensitivity, fat mass and diet. Diurnal capillary TG profiles may be a valuable additional tool in estimating CHD risk as significant differences in diurnal TGs are not always reflected by elevated fasting plasma TGs.

Comment

This study examines diurnal capillary TG profiles in normotriglyceridaemic men eating a normal diet and represents a more physiological approach to the investigation of postprandial hyperlipaemia than the customary investigation of responses to a single meal often containing unusually large amounts of fat. It is well established that subjects with fasting hypertriglyceridaemia have increased postprandial lipaemia. In this study there is considerable variability of the integrated areas under the capillary TG concentration curves even in subjects with fasting TGs within the normal range. Area under the curve correlated with fasting insulin, insulin resistance, relative fat mass, dietary protein and saturated fat intake, but not with age.

In a thoughtful accompanying commentary Sniderman |**7**| makes a plea for expansion of the traditional concept of chylomicron metabolism characterized by the initial hydrolysis of TG in peripheral tissues followed by hepatic clearance of cholesterol-enriched remnants. The latter phase is usually seen as being of greater relevance to atherogenesis according to the classical Zilversmit hypothesis |**8**|. The free fatty acids (FFA) released during initial lipolysis are taken up into adjacent adipose tissue, or skeletal or cardiac muscle, but a variable amount spills into the circulation |**9**| and this may also be important in the genesis of vascular disease. Factors believed to be important in the regulation of fatty acid trapping include insulin and acylation stimulating protein.

Failure of trapping is likely to be associated with impaired lipolysis and the increased delivery of fatty acids to muscle for oxidation will lead to impaired glucose uptake. In addition greater availability of remnants and FFA to the liver will promote increased TG synthesis and secretion of apolipoprotein (apo)B-containing lipoproteins. This may be one mechanism underlying the overproduction of VLDL in patients with familial combined hyperlipidaemia (FCH) (see below).

Familial combined hyperlipidaemia

Almost 30 years ago it became apparent from studies of the families of myocardial infarction (MI) survivors that there were two common forms of familial hyperlipidaemia, which were apparently monogenic and dominantly inherited. FCH was found in about 10–20% of relatives of probands with premature MI and is characterized by multiple lipoprotein phenotypes (commonly IIa, IIb and IV) within the same family, or at different times in the same individual. Subjects with familial hypertriglyceridaemia (FHTG) usually have type IV or V phenotypes. The original cross-sectional studies suggested that FCH was associated with an excess risk of CHD, but the situation in FHTG was much less clear and depended upon whether the index case came to light because of CHD or because of hyperlipidaemia without clinical CHD.

A 20-year prospective study examining the risk of CHD death in families with FCH and FHTG has recently been published.

Cardiovascular disease mortality in familial forms of hypertriglyceridemia: A 20-year prospective study.
M A Austin, B McKnight, K L Edwards, *et al. Circulation* 2000; **101**: 2777–82.

B**ACKGROUND. FCH and FHTG are common familial forms of hyperlipidaemia but there is little prospective data on the risk of cardiovascular disease (CVD) in such families. This study aimed to estimate 20-year total and CVD mortality risk among relatives in these families and to evaluate plasma TG as a predictor of death. The study was based on lipid and medical history data from 101 families obtained in the 1970s, and mortality data obtained in the mid-1990s.**

I**NTERPRETATION.** The results confirm that relatives in FCH families are at increased risk for CVD mortality and that effective prevention strategies are needed in this group. Compared with spouse control subjects, 20-year CVD mortality risk was increased in siblings and offspring in FCH (relative risk 1.7) after adjusting for baseline covariates. In

Table 2.1 Risk of 20-year total mortality and cardiovascular mortality among first-degree relatives of probands compared with spouse control subjects

Comparison groups	Total mortality				Cardiovascular mortality			
	Sample size*	No. dead	Person-years	Relative risk† (95% CI)	Sample size‡	No. dead	Person-years	Relative risk† (95% CI)
Familial combined hyperlipidemia								
siblings and offspring	247	89	5100	1.4, *P*=0.06	240	37	4994	1.7, *P*=0.02
vs spouses	170	49	3586	(0.99, 1.9)	165	16	3502	(1.1, 2.7)
Familial hypertriglyceridemia								
siblings and offspring	124	32	2624	0.88, *P*=0.67	121	13	2574	1.7, *P*=0.39
vs spouses	81	25	1664	(0.51, 1.5)	79	8	1658	(0.50, 5.9)

* Twenty-nine subjects missing vital status and 21 additional study subjects missing baseline covariate data excluded.

† Cox regression analysis with adjustment for age, sex, baseline study, and diabetes, hypertension, smoking, and prior myocardial infarction at baseline.

‡ Nineteen additional subjects without cause of death classification excluded.
Source: Austin *et al.* (2000).

FHTG families, the relative risk was 1.7 but was not statistically significant. Baseline TG was correlated with increased CVD mortality risk independent of total cholesterol among relatives in FHTG families but not in FCH families after adjusting for baseline covariates.

Comment

First-degree relatives of subjects with both FCH and FTHG have an approximately 70% increase in cardiovascular mortality in this study but only in the FCH families does this reach statistical significance. Moreover, total mortality was increased about 40% in FCH and on the verge of significance, whereas there was no suggestion of this in the FHTG families. In addition, while the deaths occurred over a wide age range in the FCH

relatives none of the cardiovascular deaths were premature in the FHTG families. This prospective study confirms earlier cross-sectional data suggesting that FCH is a high-risk disorder. It very likely underestimates the risk as all available first-degree relatives were included whether or not they had dyslipidaemia. If this is an autosomal dominant disorder and the excess risk is mediated through dyslipidaemia then only half the subjects will have been responsible for the increased risk detected in the whole group. No HDL measurements were available at the time of the baseline assessments for this study so it is not possible to draw conclusions as to which aspect of the dyslipidaemic profile contributes most to the increase in risk. The risk status of subjects with FHTG remains unclear.

Although this is an important study the situation for the clinician confronted with an individual patient can be difficult. It is frequently not easy to determine whether a patient has either FCH or FHTG and if so which. There are no specific physical signs or laboratory tests, such as tendon xanthomas or apoE-2 homozygosity, which may provide the answer in FH and type III hyperlipoproteinaemia. A decision may be made by finding frequent dyslipidaemia with multiple phenotypes in close relatives, but often this is not feasible. Nevertheless identifying families likely to have FCH by screening the relatives of patients with premature CHD is a potentially important preventative measure and should be part of any high-risk screening strategy.

The initial studies performed in Seattle and Finland suggested that FCH was a dominantly inherited monogenic disorder with a frequency in the general population of about 1–2% and some 10-fold higher in survivors of premature MI. Full penetrance was not achieved until adult life. Unlike the situation in FH no single genetic locus, with a well-defined effect on lipoprotein metabolism, has emerged to explain the dyslipidaemia in FCH and it is now accepted that the disorder is likely to be heterogeneous |**10,11**|. A subset of patients are heterozygous for lipoprotein lipase deficiency but this is too infrequent to account for more than a small proportion of FCH overall.

Not only do subjects with FCH have an increase in serum cholesterol or TGs or both, they frequently have low serum HDL cholesterol levels and an excess of small, dense LDL. Thus they may exhibit the so-called atherogenic lipoprotein phenotype (ALP). Hypertension and insulin resistance may be features. Thus there is overlap with a variety of other atherogenic states, including hyperapobetalipoproteinaemia, familial dyslipidaemic hypertension and the insulin resistance syndrome. Despite the phenotypic overlap it is not suggested that these are all synonyms for a single condition. A large number of genes may ultimately prove to be involved and classification should become clearer as these are delineated |**12**|. An analysis of 40 FCH families has suggested for example that a common major gene is responsible for a substantial part of the variance in both apoB concentration and LDL particle size distribution |**13**|.

Contribution of the hepatic lipase gene to the atherogenic lipoprotein phenotype in familial combined hyperlipidemia.

H Allayee, K M Dominguez, B E Aouizerat, *et al. J Lipid Res* 2000; **41**: 245–52.

B**ACKGROUND. FCH is a common genetic lipid disorder characterized by hypercholesterolaemia and/or hypertriglyceridaemia, and often small, dense LDL particles and low HDL cholesterol. Families with FCH and families enriched for coronary artery disease (CAD) share genetic determinants for the ALP. The hepatic lipase (HL) gene is linked to HDL cholesterol levels, and polymorphism within the HL promoter is associated with increased HDL cholesterol levels and larger, more buoyant LDL particles. This study tested whether the HL gene locus also contributes to ALP, in Dutch FCH families.**

I**NTERPRETATION.** The results support the view that ALP is a multigenic trait and suggest that the relationship between small, dense LDL particles, HDL cholesterol and TG levels in FCH families is partly due to common genetic factors. Evidence for linkage of LDL particle size, HDL cholesterol and TG levels to the HL gene locus was observed. A genome scan in a subset of families also suggested a link between the HL gene locus and peak particle diameter (PPD) and HDL cholesterol levels. HL promoter polymorphism was significantly associated with higher HDL cholesterol levels in the unrelated males (but not females) of this population. No association was observed between the polymorphism and LDL particle size or TG levels.

Comment

Previous work from this group has shown that the ALP is 10 times more common in FCH subjects than in their unaffected spouses |**14**| and demonstrated linkage to several gene loci, including those for cholesterol ester transfer protein, lecithin: cholesterol acyltransferase and the apoAI-CIII-AIV complex. In the current study of 27 Dutch families with FCH evidence was found for linkage of the HL locus to HDL cholesterol and TG concentrations as well. HL has both TG hydrolase and phospholipase activities and is involved in lipoprotein metabolism at several stages. These include the conversion of remnants of TG-rich lipoproteins to LDL and their direct uptake by the liver. Its activity is inversely related to HDL cholesterol concentration and it plays a part in the recycling of HDL_2 back to HDL_3. Looked at teleologically, the situation is complex. Some actions of HL can be construed as pro-atherogenic and others as antiatherogenic. Familial HL deficiency is extremely rare but does appear to be associated with premature CHD |**15**|.

Evidence for a new pathophysiological mechanism for coronary artery disease regression: hepatic lipase-mediated changes in LDL density.

A Zambon, J E Hokanson, B G Brown, J D Brunzell. *Circulation* 1999; **99**: 1959–64.

B**ACKGROUND. Small dense LDL particles are associated with CAD and predict angiographic changes in response to lipid-lowering treatment. Intensive lipid-lowering therapy in the Familial Atherosclerosis Treatment Study (FATS) significantly improved CAD. This study examines the relationship between LDL density, HL and CAD progression. Participants in FATS (*n*=88) with documented CAD, apoB levels ≥125 mg/dl, and family histories of CAD were randomized to lovastatin (40 mg/day) and colestipol (30 g/day), niacin (4 g/day) and colestipol, or conventional therapy with placebo alone, or with colestipol in those with raised LDL cholesterol.**

I**NTERPRETATION.** LDL buoyancy increased and HL decreased with both lovastatin-colestipol and niacin-colestipol therapies. Changes in LDL buoyancy and HL activity were associated with changes in disease severity. These results support the hypothesis that therapy associated changes in HL alter LDL density, and retard CAD progression.

Comment

The work of Allayee *et al.* described above showing linkage of the HL gene locus to components of the ALP in FCH is of considerable interest in view of the current study of a subset of patients from the FATS. This suggests that HL activity may play a causal part in the progression of angiographic changes in a heterogeneous group of men (not only FCH) with dyslipidaemia and CHD.

Substantial reductions in serum and LDL cholesterol, induced by lovastatin or niacin in combination with colestipol, were associated with a reduction in HL activity. When their lipoproteins were perturbed with the trial medication regression of coronary stenoses was inversely and most strongly predicted by an increase in LDL buoyancy. Increased buoyancy was a much stronger predictor than reduction in LDL cholesterol or apoB levels, and was in turn inversely and highly significantly related to reduction in HL activity. This is plausible given that HL is thought to promote the formation of small dense LDL, although it is not easy to see how it is reconciled with the observation that familial HL deficiency is associated with premature CHD |**15**|. The results of the current study are very persuasive and make a significant contribution to the debate as to whether HL is pro- or antiatherogenic |**16**|.

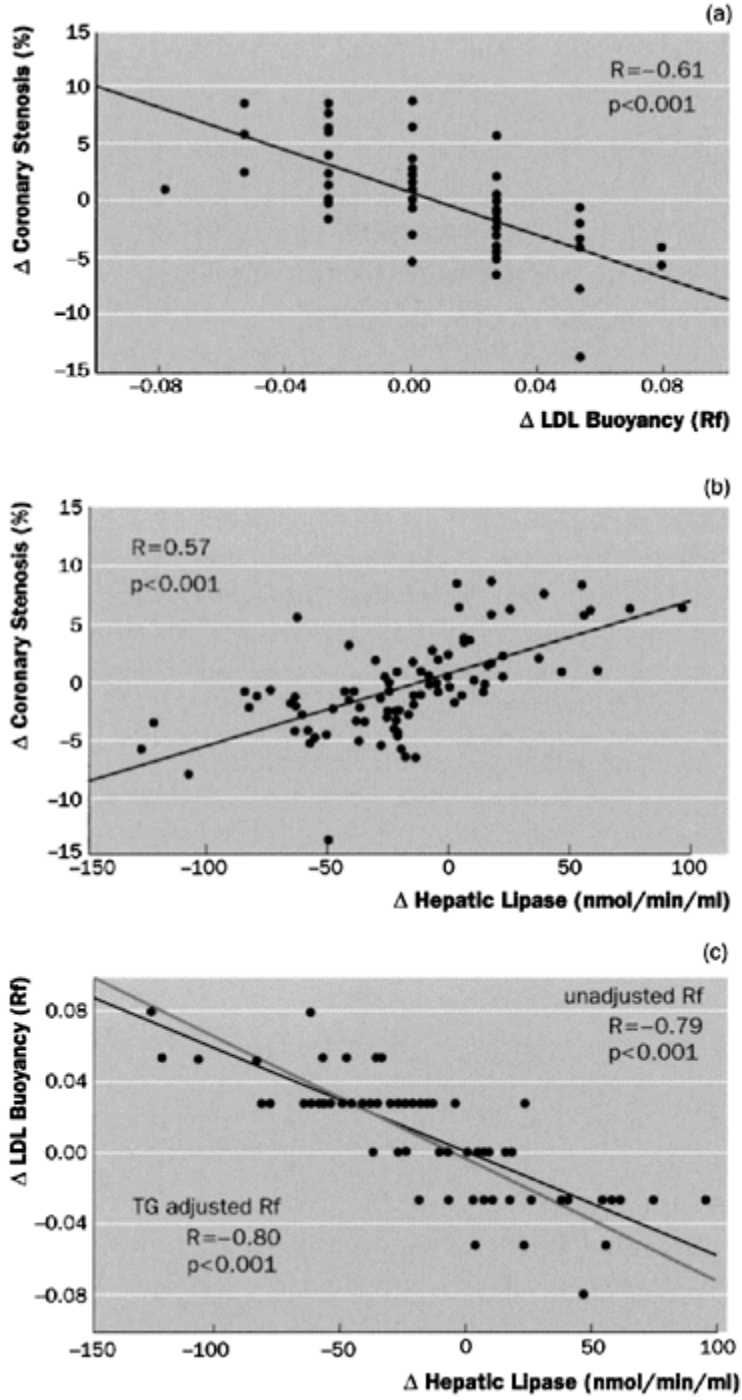

(a)
Δ Coronary Stenosis (%)
15
10
5
0
−5
−10
−15
R=−0.61
p<0.001
−0.08
−0.04
0.00
0.04
0.08
Δ LDL Buoyancy (Rf)
(b)
Δ Coronary Stenosis (%)
15
10
5
0
−5
−10
−15
R=0.57
p<0.001
−150
−100
−50
0
50
100
Δ Hepatic Lipase (nmol/min/ml)
(c)
Δ LDL Buoyancy (Rf)
0.08
0.04
0.00
−0.04
−0.08
unadjusted Rf
R=−0.79
p<0.001
TG adjusted Rf
R=−0.80
p<0.001
−150
−100
−50
0
50
100
Δ Hepatic Lipase (nmol/min/ml)

Fig. 2.1 (a) Linear regression analysis of changes in LDL buoyancy (LDL-Rf indicates relative flotation) and changes in percentage coronary stenosis. (b) Linear regression analysis of changes in HL activity and changes in percentage coronary stenosis. (c) Linear regression analysis of changes in HL activity and changes in LDL buoyancy (symbols and solid regression line). Light grey line represents regression line of linear regression analysis with TG-adjusted changes in LDL buoyancy. Symbols for this analysis are omitted. Source: Zambon *et al.* (1999).

In vivo evidence of defective postprandial and post-absorptive free fatty acid metabolism in familial combined hyperlipidemia.
S Meijssen, M C Cabezas, T B Twickler, H Jansen, D W Erkelens. *J Lipid Res* 2000; **41**:1096–102.

B**ACKGROUND. Patients with FCH overproduce very low density lipoprotein (VLDL). Enhanced FFA flux to the liver may be one of the determinants of VLDL overproduction. FFA changes and products of hepatic FFA metabolism, in response to a 24-h oral fat loading test (50 g/m^2) were studied in seven FCH patients and seven matched controls.**

I**NTERPRETATION.** The results provide *in vivo* evidence of impaired metabolism of postprandial FFA in FCH, which may explain, in part, the hepatic VLDL overproduction characteristic of FCH subjects.

Comment

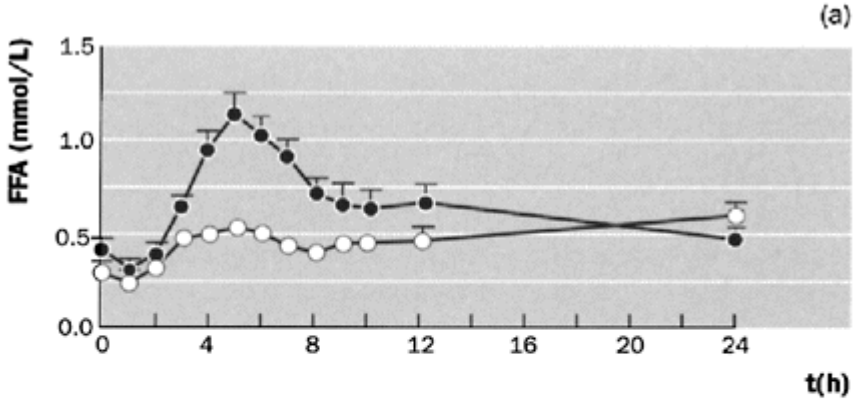
The increase in postprandial plasma FFA seen in FCH patients in this study might be due to increased release in peripheral tissues as a result of higher serum TG concentrations. The data would also fit with a failure of fatty acid trapping in adipose tissue in FCH as discussed above (see section on postprandial lipaemia). Either way this would lead to increased FFA delivery to the liver and would account for the increased generation of ketone bodies seen in FCH in the current study. Increased hepatic FFA uptake is likely to be an important contributor to VLDL overproduction, which is a feature of FCH.

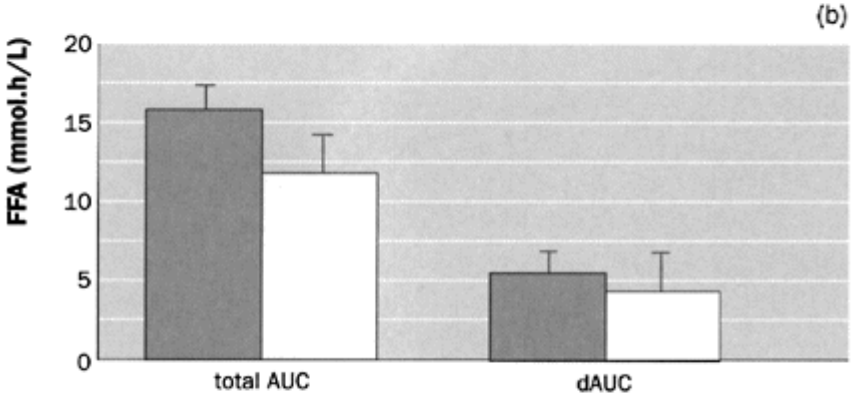
Impaired free fatty acid suppression during hyperinsulinemia is a characteristic finding in familial combined hyperlipidemia, but insulin resistance is observed only in hypertriglyceridemic patients.

J Pihlajamaki, L Karjalainen, P Karhapaa, I Vauhkonen, M Laakso. *Arterioscler Thromb Vasc Biol* 2000; **20**:164–70.

B**ACKGROUND. Insulin resistance has been linked with hypertriglyceridaemia, combined hyperlipidaemia and FCH. To investigate whether all types of FCH patients have low insulin sensitivity, insulin sensitivity was measured, by the hyperinsulinaemic euglycaemic clamp with indirect calorimetry, in healthy controls and non-diabetic FCH family members (50 without dyslipidaemia, 19 with hypercholesterolaemia, 22 with hypertriglyceridaemia and 14 with combined hyperlipidaemia).**

I**NTERPRETATION.** During the hyperinsulinaemic clamp, FCH family members had higher FFA levels than did controls. Relatives without dyslipidaemia and patients with hypertriglyceridaemia and with combined hyperlipidaemia had lower rates of


(a)
FFA (mmol/L)
1.5
1.0
0.5
0.0
0
4
8
12
16
20
24
t(h)


(b)
FFA (mmol.h/L)
20
15
10
5
0
total AUC
dAUC

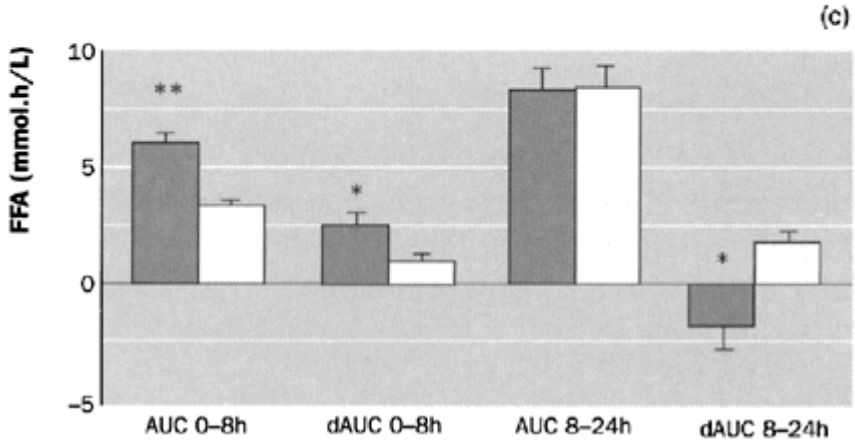

(c)
FFA (mmol.h/L)
10
5
0
−5
**
*
*
AUC 0–8h
dAUC 0–8h
AUC 8–24h
dAUC 8–24h

Fig. 2.2 (a) Mean changes in plasma FFAs in seven untreated FCH patients (solid symbols) compared with seven matched control subjects (open symbols). (b) Areas under the curve (AUC) and incremental AUC (dAUC) of total area. (c) AUC and dAUC of postprandial and post-absorptive periods. Data represent mean±SEM. **P*<0.05, ***P*<0.01 FCH compared with control subjects. Note: error bars for control subjects are so small that they are not visible at all times. Source: Meijssen *et al.* (2000).

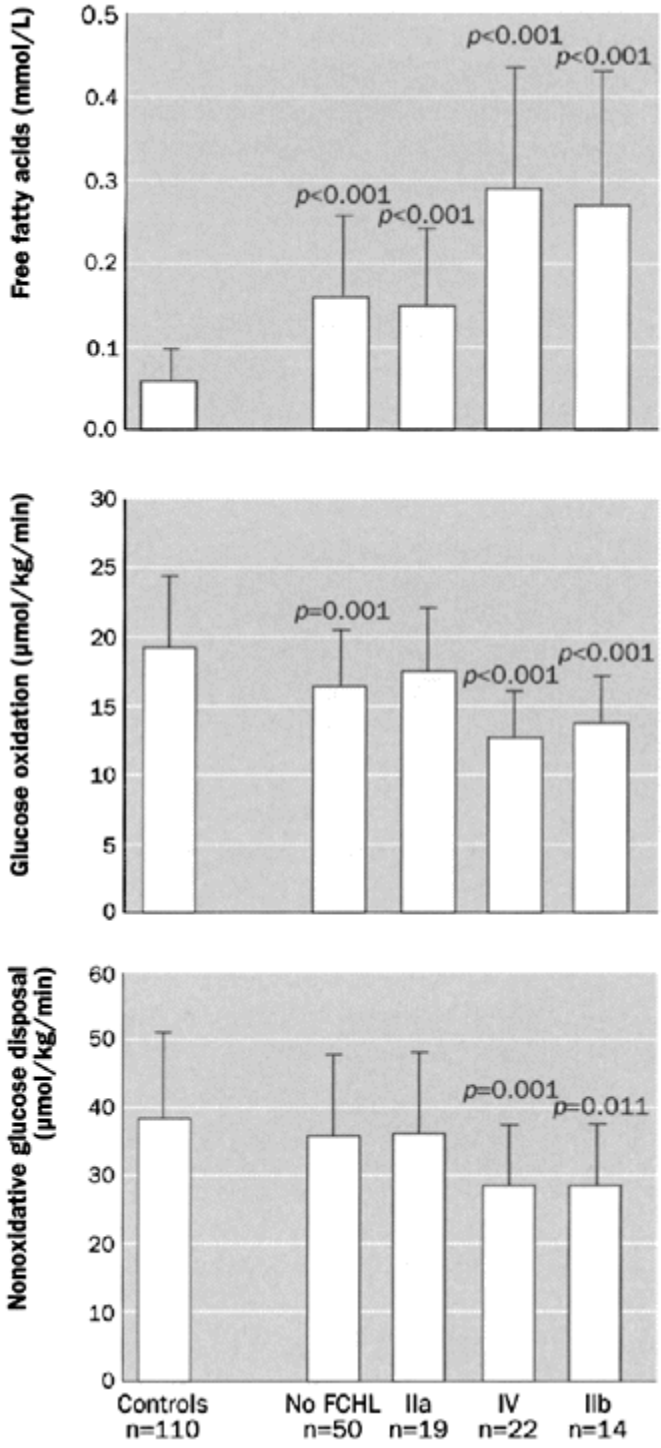

Free fatty acids (mmol/L)
0.5
0.4
0.3
0.2
0.1
0.0
p<0.001
p<0.001
p<0.001
p<0.001
Glucose oxidation (µmol/kg/min)
30
25
20
15
10
5
0
p=0.001
p<0.001
p<0.001
Nonoxidative glucose disposal (µmol/kg/min)
60
50
40
30
20
10
0
p=0.001
p=0.011
Controls n=110
No FCHL n=50
IIa n=19
IV n=22
IIb n=14

Fig. 2.3 FFA levels and the rates of oxidative and non-oxidative glucose disposal (mean±SD) in control subjects and familial combined hyperlipidaemia (FCH) family members without FCH, with hypercholesterolaemia (type IIa dyslipidaemia), with hypertriglyceridaemia (type IV), and with combined hyperlipidaemia (type IIb). Source: Pihlajamaki *et al.* (2000).

insulin-stimulated glucose oxidation than did controls. Rates of non-oxidative glucose disposal were lower in patients with hypertriglyceridaemia and combined hyperlipidaemia than in controls but those with hypercholesterolaemia and controls had similar rates of insulin-stimulated glucose uptake. The results indicate that all FCH patients have defective FFA suppression during hyperinsulinaemia, probably located in adipose tissue, whereas insulin resistance in skeletal muscle is seen only in FCH patients with raised TG levels.

Comment

FCH appears to be a heterogeneous disorder and a variety of metabolic defects have been described that may contribute to the phenotype to variable degrees in different families. As already discussed, insulin resistance is a common feature of FCH, as is impaired suppression of FFA levels by insulin, and in the current study the authors set out to discover if these were universal findings in such patients or confined to certain subgroups.

In addition to a group of healthy controls they studied 105 subjects from 33 FCH families, half of whom were normolipidaemic, the remainder exhibiting the multiple lipoprotein phenotypes characteristic of FCH. As judged by whole body glucose uptake using the hyperinsulinaemic euglycaemic clamp technique only those FCH subjects with hypertriglyceridaemia were insulin resistant. FFA concentrations during the clamp were elevated in both the normolipidaemic and the dyslipidaemic FCH family members, although the elevations were greatest in the hypertriglyceridaemic subjects. The finding of impaired FFA suppression in the normolipidaemic (i.e. ‘unaffected’) relatives is intriguing and presumably militates further against FCH being a single gene Mendelian disorder.

It appears that impaired FFA suppression by insulin may be a general characteristic of FCH, whereas impaired insulin-mediated glucose uptake is confined to hypertriglyceridaemic subjects. The impaired FFA suppression is likely to be the result of reduced uptake by adipose tissue. This may be an example of a defect in FFA trapping as discussed above |**7**|. Increased FFA release is unlikely because, if anything, lipoprotein and hormone-sensitive lipase activities tend to be reduced in FCH.

Familial dysbetalipoproteinaemia (type III)

Type III hyperlipoproteinaemia is characterized by the accumulation of the partially

metabolized remnants of TG-rich lipoproteins, which may under certain circumstances lead to severe mixed hyperlipidaemia. Patients may have characteristic changes in the palmar creases (orange discoloration, striate xanthomas) and tuberous xanthomas, and are at increased risk of premature cardiovascular, particularly peripheral vascular, disease. Affected individuals are usually homozygous for the E2 variant of apoE, which exhibits defective binding to lipoprotein receptors. A few have other rare mutations in apoE |**17**|. All E2 homozygotes exhibit dysbeta-lipoproteinaemia with the presence of remnants (β-VLDL), but the vast majority is not hyperlipidaemic. The presence of severe mixed hyperlipidaemia seems to need not only an abnormality in apoE but also an additional factor such as obesity, hypothyroidism or the presence of a second genetic disorder such as FCH. Rare instances of the coexistence of type III and FH have been reported (see below). In women oestrogen deficiency occurring at the time of the menopause may be the precipitant of hyperlipidaemia in apoE2 homozygotes.

LDL concentrations are usually reduced supporting the notion that some classes of TG-rich lipoprotein, particularly remnants, are atherogenic, although of course it may well not be the TG *per se* in these particles that does the damage.

Coexisting dysbetalipoproteinemia and familial hypercholesterolemia. Clinical and laboratory observations.
R Carmena, M Roy, G Roederer, A Minnich, J Davignon. *Atherosclerosis* 2000; **148**:113-24.

B**ACKGROUND. Type III dysbetalipoproteinaemia and familial hypercholesterolaemia (FH) cause severe disturbances of lipid homeostasis and premature atherosclerosis. Both metabolic abnormalities have a genetic basis and co-occurrence in the same patient has seldom been described. The unique structure of the French Canadian population, provided an opportunity to observe patients with both dysbetalipoproteinaemia (E2/2 homozygotes) and FH (*n*=14) and to compare their clinical data with that of patients with type III alone, or with FH.**

I**NTERPRETATION.** In the patients studied, the coexistence of dysbetalipoproteinaemia and heterozygous FH does not appear to increase the prevalence of cardiovascular complications above that observed among control type III or control E3/3 FH patients. The presence of two ε2 alleles in these patients affects the expression of the abnormal LDL-R allele and the resulting phenotype substantiates the non-additive effects of alleles at these two loci (epistasis).

Comment

This is probably the largest group of patients reported with coexisting FH and type III.

Even so the numbers are inevitably small and the results need to be interpreted with some caution. Surprisingly, the prevalence of CHD does not appear to be increased in the subjects with combined type III/FH compared with controls with type III alone. Moreover those with the combined disorders have a prevalence of CHD, which is less than half of that in patients with FH alone, despite matching for the LDL receptor mutation. Overall clinical atherosclerotic vascular disease was not greater in the subjects with the combined disorders than in type III or FH alone. Women with type III, which is often not fully expressed until the menopause, appeared to be at lower risk of vascular disease than those with FH.

Patients with the combined disorder also had LDL cholesterol and LDL apoB concentrations that were substantially lower than those with FH alone, confirming the well-documented effect of apoE2 homozygosity in reducing LDL cholesterol concentrations.

In broad terms the risk of CVD in individuals with both type III and FH is not additive and does not seem to exceed that of either condition alone. The likely explanation is that the defect in type III reduces LDL formation from TG-rich lipoproteins, despite concomitant defective LDL clearance, and thereby attenuates risk.

Chylomicronaemia syndrome

Severe hypertriglyceridaemia may be associated with abdominal pain or frank pancreatitis and a variety of spurious biochemical abnormalities, most notably hyponatraemia, due to dilution by the mass of circulating TGs. With some assay methods the serum amylase may be artificially depressed even in the presence of pancreatitis. Serial dilution of the sample leads to an apparent increase in amylase activity under these circumstances. This constellation of features is known as the chylomicronaemia syndrome |**18–20**| and is of considerable clinical importance, although relatively uncommon. If the true situation is not appreciated patients may be treated inappropriately with hypertonic saline or submitted to unnecessary laparotomy.

Chylomicronaemia syndrome may present in childhood as the result of deficiency of lipoprotein lipase or its activator apoC-II |**18**|. More common, however, is the patient who has mild or moderate primary hypertriglyceridaemia, exacerbated by a secondary cause, often alcohol or diabetes. A variety of drugs may also exacerbate primary hypertriglyceridaemia, notably oestrogens, particularly if given orally, and β-blockers. Cimetidine has also been implicated and there have been recent reports involving tamoxifen |**21**|, interferon-α |**22**| and protease inhibitors (PI) used to treat HIV infection (see below).

Initial treatment of patients with severe hypertriglyceridaemia is to deal with any superimposed secondary cause of hyperlipidaemia, particularly alcohol and uncontrolled diabetes, and where possible to withdraw drugs that are likely to be implicated. Treatment with fibrates, nicotinic acid and its derivatives or n-3 fatty acid concentrates may all be helpful. A very low total TG intake is the basis of treatment in severe genetic hypertriglyceridaemia. Some patients, however, are unable to comply or are refractory to all of these measures.

Prevention of recurrent pancreatitis in familial lipoprotein lipase deficiency with high-dose antioxidant therapy.
A P Heaney, N Sharer, B Rameh, J M Braganza, P N Durrington. *J Clin Endocrinol Metab* 1999; **84**:1203–5.

BACKGROUND. This report describes a dramatic response to antioxidant therapy in three patients with familial lipoprotein lipase deficiency, complicated by frequent severe episodes of pancreatitis that had not responded to other dietary and pharmacological measures.

I**NTERPRETATION.** Antioxidant therapy may be an important advance in the management of such patients.

Comment

Braganza *et al.* have pioneered the use of antioxidant therapy in recurrent pancreatitis generally. Although confined to three cases of severe genetic hypertriglyceridaemia this study provides impressive evidence of the ability of a mixture of antioxidants to prevent attacks of pancreatitis even when serum TGs cannot be reduced. It seems sensible to offer antioxidant therapy to all patients with severe genetic hypertriglyceridaemia and thought to be at risk of pancreatitis. It is unknown to what extent antioxidants may prove useful in refractory cases of chylomicronaemia involving a superimposed secondary cause of hypertriglyceridaemia but antioxidants might reasonably be tried.

In the acute attack of pancreatitis associated with chylomicronaemia, the TG concentration often falls rapidly if no fat is given either orally or parenterally for a few days. Withdrawal of precipitating factors, such as alcohol, and control of diabetes, also contribute. Indeed, insulin may produce a rapid reduction in TGs even in non-diabetic patients with chylomicronaemia |**23**|. Other approaches in refractory cases include plasma exchange and 'fat-free' parenteral nutrition.

Therapeutic plasma exchange in patients with chylomicronemia syndrome complicated by acute pancreatitis.
A Lennertz, K G Parhofer, W Samtleben, T Bosch. *Ther Apher* 1999; **3**: 227–33.

B**ACKGROUND. Chylomicronaemia syndrome is a rare disorder,**

characterized by the presence of chylomicrons in the fasting state, causing a milky appearance of plasma, eruptive xanthomas and hepatosplenomegaly; an acute and potentially life-threatening complication is severe acute pancreatitis. The underlying defects are inborn errors of metabolism such as deficiencies of lipoprotein lipase or apoC-II as well as FHTG. This study retrospectively analysed the results of therapeutic plasma exchange in five patients transferred to our hospital with severe acute pancreatitis due to chylomicronaemia syndrome.

I**NTERPRETATION.** One or two sessions of therapeutic plasma exchange substantially decreased the bulk of TGs in acutely exacerbated chylomicronaemia syndrome producing a rapid resolution of acute severe pancreatitis.

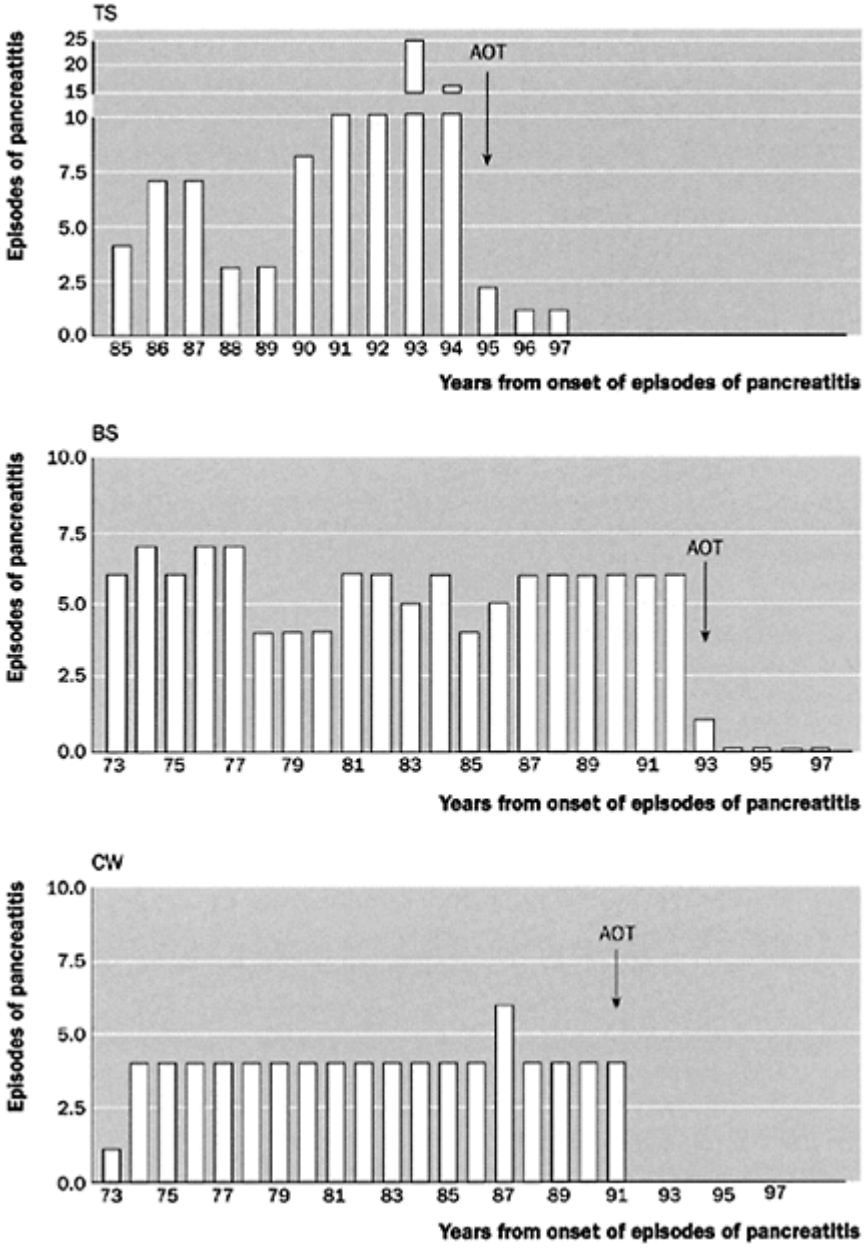


Fig. 2.4 The number of episodes of pancreatitis documented by hospital admissions each year in three patients before and after the introduction of antioxidant therapy (AOT).
Source: Heaney *et al.* (2000).

Comment

Plasma exchange and other methods of extracorporeal lipoprotein removal have been used extensively in homozygous FH and plasma exchange has also been described for the removal of TG-rich lipoproteins |**24**|. The current paper describes five patients, one of whom was 30 weeks pregnant, in which one or two plasma exchange sessions led to a rapid, major reduction in circulating TGs and was associated with resolution of the pancreatitis. This is not an area in which controlled clinical trials are likely to be practicable. In severely ill patients with chylomicronaemia syndrome, plasma exchange at an early stage should be seriously considered.

Total parenteral nutrition in the chylomicronemia syndrome and acute pancreatitis.
M A Crook, A Sankaralingam. *Nutrition* 1999; **15**:299–301.

B**ACKGROUND. Patients who are severely ill with pancreatitis associated with the chylomicronaemia syndrome may need a period of parenteral nutrition but the use of intravenous fat emulsions may exacerbate the hypertriglyceridaemia.**

I**NTERPRETATION.** 'Fat-free' parenteral nutrition is feasible in this situation, with the administration of only minimal amounts of intravenous lipid to prevent essential fatty acid deficiency.

Comment

This is a single case report of a 35-year old woman with serum TGs of 56 mmol/l and severe pancreatitis. She was acutely ill with septicaemia and needed ventilation and inotropic support. Because of probable paralytic ileus she needed parenteral nutrition, but it was unreasonable to give her intravenous fat emulsions in the presence of severe hypertriglyceridaemia and pancreatitis. A regimen of 'fat-free' total parenteral nutrition is described in which a small dose of intravenous fat was given twice weekly to meet essential fatty acid requirements.

Dyslipidaemia and HIV

Dyslipidaemia is now a well-established feature of HIV infection and its treatment |**25,26**| and increase in TGs is one of the most consistent abnormalities. Severe hypertriglyceridaemia and pancreatitis are not common, but can occur. As patients with HIV live longer the dyslipidaemia is likely to become increasingly relevant. There are several anecdotal reports of premature atherosclerotic vascular disease in patients with HIV treated with PI |**27,28**|, which may relate in part to the tendency of these drugs also

to increase LDL cholesterol and lipoprotein(a) (Lp(a)).

Atherogenic dyslipidemia in HIV-infected individuals treated with protease inhibitors. The Swiss HIV Cohort Study.
D Periard, A Telenti, P Sudre, *et al. Circulation* 1999; **100**:700–5.

B**ACKGROUND. Administration of PIs to HIV-infected individuals has been associated with hyperlipidaemia. In this study, we characterized the lipoprotein profile in HIV-infected adults receiving ritonavir, indinavir or nelfinavir, alone or in combination with saquinavir. Plasma lipoprotein levels in 93 HIV-infected adults receiving PIs were compared with pretreatment values and with those of non-PI-treated HIV-infected adults.**

I**NTERPRETATION.** Administration of PIs to HIV-infected individuals is associated with marked, compound-specific dyslipidaemia. Plasma cholesterol levels were increased in all PI-treated groups but particularly the ritonavir group. Ritonavir, but not indinavir or nelfinavir, increased plasma TG levels. Plasma HDL cholesterol levels were unchanged. Combinations of ritonavir or nelfinavir with saquinavir did not further increase plasma lipids. Plasma Lp(a) was increased by 48% in PI-treated individuals with pre-treatment Lp(a) values>20 mg/dl. Similar changes in plasma lipids were seen in six children receiving ritonavir. The risk of pancreatitis and premature atherosclerosis due to PI-associated dyslipidaemia is yet to be established.

Comment

Ritonavir, indinavir and nelfinavir all tended to increase LDL and hence total serum cholesterol but only ritonavir increased TGs. The effect on average was modest and insufficient to cause the chylomicronaemia syndrome, though in one child with baseline hypertriglyceridaemia the increase was of this order (TGs rose from 5.7 to 14 mmol/l). There are also case reports of massive hypertriglyceridaemia and pancreatitis attributed to ritonavir |**29,30**|.

Effect of ritonavir on lipids and post-heparin lipase activities in normal subjects.
J Q Purnell, A Zambon, R H Knopp, *et al. AIDS* 2000; **14**:51–7.

B**ACKGROUND. Intensive therapy of HIV infection with highly active**

antiretroviral therapy (HAART) dramatically reduces viral loads and improves immune status. Abnormalities of lipid levels, body fat distribution and insulin resistance have been commonly reported after starting HAART. Whether the lipid abnormalities result from changes in metabolism after an improvement in HIV status or are partly attributable to the effects of PI use is unknown. The effect of the PI ritonavir on total lipids, apolipoproteins and post-heparin plasma lipase activities was evaluated in a 2-week double-blind, placebo-controlled study in 21 healthy volunteers.

INTERPRETATION. Treatment with ritonavir, in the absence of HIV infection or changes in body composition, results in hypertriglyceridaemia that is apparently not mediated by impaired lipoprotein lipase activity or the defective removal of remnant lipoproteins, but could be caused by enhanced formation of VLDLs. Long-term studies of patients with HIV infection receiving HAART will be necessary to determine the impact of these drugs and associated dyslipidaemia on the risk of CAD.

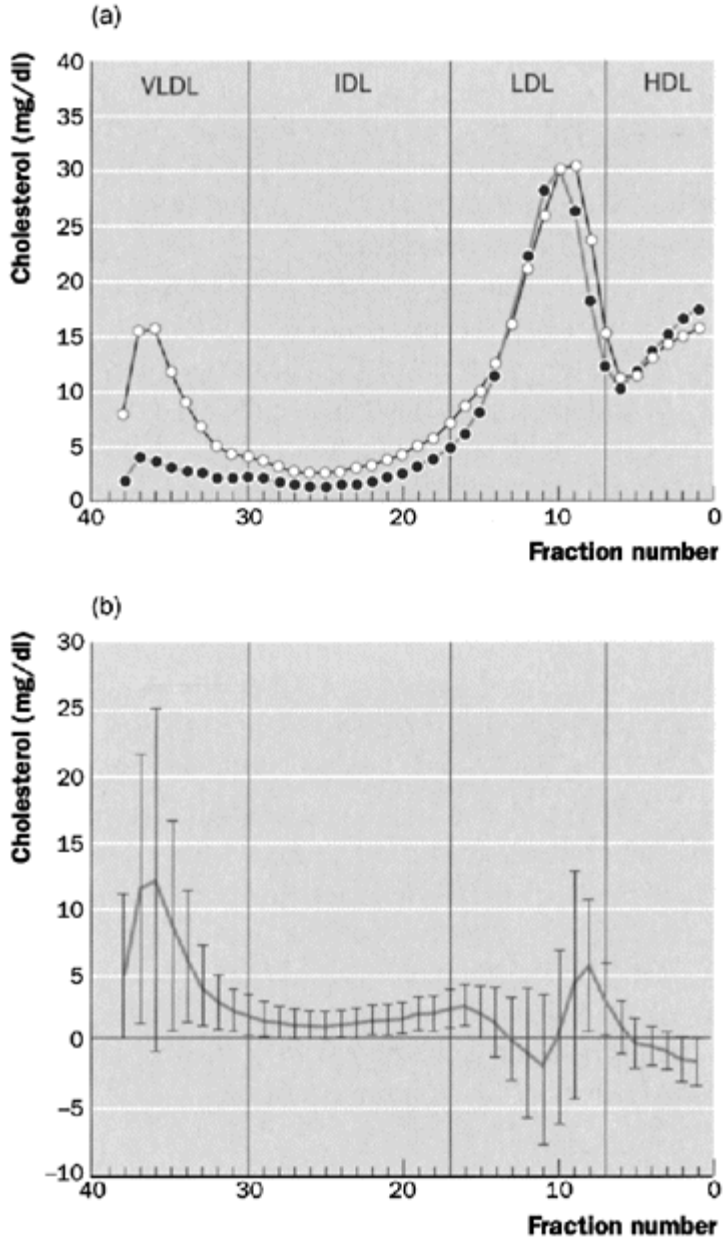


Fig. 2.5 Distribution of lipoprotein cholesterol across non-equilibrium density gradient ultracentrifugation comparing baseline (●) with follow-up (○) of the group receiving ritonavir (a) and difference plot of mean cholesterol levels of each fraction with 95% confidence interval (b). Source: Purnell *et al.* (2000).

Comment

It appears from this study of a short course of ritonavir in normal subjects that the increase seen in serum TGs is not the result of alterations in metabolic status due to the suppression of HIV infection but rather is directly due to the drug itself. This is supported by the observation that indinavir and nelfinavir did not cause hypertriglyceridaemia (see Periard *et al.* above). The mechanism does not seem to involve lipoprotein lipase and, although HL activity was reduced, the fact that most of the TG accumulated in VLDL and to a lesser extent in remnants may mean that the dominant effect is to increase VLDL secretion.

Use of fenofibrate in the management of protease inhibitor-associated lipid abnormalities.
J C Thomas, M F Lopes-Virella, V E Del Bene, *et al. Pharmacotherapy* 2000; **20**:727–34.

B**ACKGROUND. HIV PIs are associated with several metabolic abnormalities, including hypercholesterolaemia and hypertriglyceridaemia. Fenofibrate is a new lipid-lowering agent, for adults with very high TG levels, that was administered to two HIV-positive patients who developed hypertriglyceridaemia while taking PIs. Starting dosages of 134 and 201 mg/day, were increased to 268 mg/day in both patients.**

I**NTERPRETATION.** TG levels decreased from 1450 to 337 mg/dl (76.8%) and from 1985 to 322 mg/dl (83.8%), respectively, after 10 months of therapy. HDL levels increased in both patients.

Comment

Fenofibrate is effective over the medium term at reducing severe PI-induced hypertriglyceridaemia in these two case reports and would appear to be justified in those patients with a history, or at risk, of pancreatitis. It is less clear when drug treatment is indicated for other dyslipidaemic states in PI-treated patients with HIV infection. Henry *et al.* |**31**| investigated the use of atorvastatin and gemfibrozil, and suggested that dyslipidaemia in HIV may be managed with care according to NCEP guidelines. Others have advised particular caution because of the potential for drug interactions and because the risk of vascular disease is uncertain |**32**|.

Dyslipidaemia and non-alcoholic steatohepatitis (NASH)

Apparently healthy patients with mild to moderate abnormalities of liver function are commonplace in medical clinics. If investigated further many will prove to have fatty liver as judged by ultrasound or biopsy. Alcohol is usually suspected as the cause and can be difficult to exclude. Many of these patients, however, are not drinking excessively and have NASH **|20,33|,** a term coined by Ludwig *et al.* 20 years ago |**34**|. Even histologically, NASH and alcoholic steatosis can be indistinguishable. Overall, the serum aspartate aminotransferase exceeds the alanine aminotransferase in alcoholic fatty liver and vice versa in NASH, but this is not always a reliable distinction in the individual case. A serum aspartate aminotransferase/alanine aminotransferase ratio of 2 or greater is said to be strongly suggestive of alcoholic liver disease |**35**|.

There is a wide variety of causes of NASH that include drugs (such as amiodarone and perhexilene), exposure to volatile petrochemicals |**36**|, hepatitis C, morbid obesity (including its surgical treatment by jejuno-ileal bypass) and diabetes. Fatty liver is also a feature of the chylomicronaemia syndrome (see above) and rarely in genetic hypolipidaemias, such as abetalipoproteinaemia and hypobeta-lipoproteinaemia. It is increasingly apparent that NASH may be a feature of the metabolic or insulin resistance syndrome.

Fatty infiltration of liver in hyperlipidemic patients.
N Assy, K Kaita, D Mymin, *et al. Dig Dis Sci* 2000; **45:**1929–34.

B**ACKGROUND. Hyperlipidaemia is a known risk factor for fatty infiltration of the liver, a condition that can progress to cirrhosis and liver failure. This study documents the prevalence of fatty infiltration in the livers of hyperlipidaemic patients in an attempt to identify the predictor variables associated with this condition. Over an 18-month recruitment period, clinical, biochemical and radiological assessments were made of 95 adult patients referred to an urban hospital-based lipid clinic for evaluation and management of hyperlipidaemia.**

I**NTERPRETATION.** The results indicate that there is ultrasonographic evidence of fatty infiltration of the liver in roughly 50% of patients with hyperlipidaemia: hypertriglyceridaemia is the lipid profile most often seen. Serum aspartate aminotransferase values, hyperglycaemia and age, all independently predict the presence of fatty infiltration, while hypertriglyceridaemia and diabetes are the only risk factors that significantly increase the risk of fatty infiltration in hyperlipidaemic patients.

Comment

This study based on patients referred to a Lipid Clinic in Winnipeg confirms what many doctors working in Lipid Clinics will intuitively know, but have probably never formally evaluated—that many patients (64% in this instance) attending such clinics have

abnormal liver function tests (LFT). Half the patients referred had fatty change in the liver as assessed by ultrasound. Fatty change was found predominantly in patients with hypertriglyceridaemia or mixed hyperlipidaemia in whom it was about five times more common than in patients with pure hypercholesterolaemia. Alcohol is often blamed (at least by the doctor) for both abnormal liver function and liver fat deposition. In this study two-thirds of subjects were said to drink to excess on the basis of fairly stringent criteria (40 g/day for men and 20 g/day for women) but surprisingly alcohol was not a predictor of fatty change. Presumably in some cases alcohol may be superimposed on NASH and exacerbate the changes. Diabetes and hypertriglyceridaemia were the two principal predictors of fatty change. It is likely that many of the patients in this study had NASH and insulin resistance, even those who were not frankly diabetic.

Prospective evaluation of unexplained chronic liver transaminase abnormalities in asymptomatic and symptomatic patients. S Daniel, T Ben-Menachem, G Vasudevan, C K Ma, M Blumenkehl. *Am J Gastroenterol* 1999; **94**:3010–14.

B**ACKGROUND. Liver biopsy is recommended in patients with chronically elevated LFTs of unknown aetiology (marker negative) but the benefits of this are unclear. This prospective observational study was designed to determine the prevalence of marker-negative LFT in patients referred for evaluation of chronically raised LFTs; to determine the prevalence of diseases that may be associated with marker-negative abnormal LFT; and to assess whether a liver biopsy alters the management of such patients.**

I**NTERPRETATION.** In the setting of marker-negative elevated LFT, the most likely histological diagnosis is fatty metamorphosis of the liver with occasional associated fibrosis. Liver biopsies in the 81 marker-negative patients revealed: normal histology (eight), steatosis (41), steatohepatitis (26), fibrosis (four) and cirrhosis (two). All 73 abnormal biopsies showed some steatosis. Histological findings were not significantly associated with obesity, hyperlipidaemia, diabetes, gender or symptoms.

Comment

In this prospective study of marker-negative patients who had been referred because of chronically abnormal liver function 90% had some degree of fatty change on biopsy, while 10% had normal biopsies. Half had simple steatosis and a third steatohepatitis, defined as fatty change with superimposed inflammation, fibrosis or Mallory bodies. Only 7% had fibrosis or cirrhosis. Features of the metabolic (insulin resistance) syndrome, such as obesity, diabetes or dyslipidaemia, were not correlated with the type of histological change. The prevalence of insulin resistance is likely to have been

underestimated, perhaps greatly, as glucose metabolism was assessed only by fasting glucose levels. No biopsy revealed a new specific cause for the liver dysfunction apart from the fatty change.

There is some uncertainty about the prognosis in NASH. Some have reported an apparently benign course especially when there is fat deposition alone |**37,38**|. NASH is probably an important cause of cryptogenic cirrhosis, however |**39**|, and when there are superimposed abnormalities the risk of cirrhosis is increased as indicated in the following study.

Nonalcoholic fatty liver disease: a spectrum of clinical and pathological severity.

C A Matteoni, Z M Younossi, T Gramlich, *et al. Gastroenterology* 1999; **116**:1413–19.

B**ACKGROUND. The spectrum of non-alcoholic fatty liver disease ranges from fatty liver alone to non-alcoholic steatohepatitis. Most previous studies have short follow-up and have not carefully delineated the different histological types when determining clinical outcomes. This study aimed to compare clinical characteristics and outcomes of patients with different types of non-alcoholic fatty liver. Liver biopsy specimens with fat accumulation were assessed for inflammation, ballooning degeneration, Mallory hyaline, fibrosis, histological iron and hepatitis C RNA. Complete data were available in 132 patients.**

I**NTERPRETATION.** The outcome of cirrhosis and liver-related death is not uniform across the spectrum of non-alcoholic fatty liver. Poor outcomes are more common in patients in whom biopsies show ballooning degeneration and Mallory hyaline or fibrosis. Fatty liver (type 1) did not differ from the other three types combined, with respect to gender, race, age or obesity. Cirrhosis was more common in the other types combined (22%) than in fatty liver alone. Overall mortality, histological iron and hepatitis C did not differ between groups. Most of the liver-related deaths were in type 4.

Comment

In this large retrospective study mean follow-up was about 8–9 years with a maximum of 18 years. Patients with simple fatty liver did not have a better prognosis overall than those with additional lobular inflammation or ballooning degeneration. Those who had Mallory hyaline (which can be a feature of NASH as well as alcohol-induced disease) and fibrosis, however, did seem to have a worse prognosis and those with ballooning degeneration and/or Mallory hyaline and fibrosis were more likely to develop cirrhosis.

It is clear that NASH is a common feature of the insulin resistance syndrome and is present in large numbers of individuals who are only moderately obese and who have

insulin resistance, but not yet frank diabetes.

Fatty liver—an additional and treatable feature of the insulin resistance syndrome.

H Knobler, A Schattner, T Zhornicki, *et al. QJM* 1999; **92**:73–9.

B**ACKGROUND. To test the hypothesis that fatty liver coexists with other metabolic abnormalities of the insulin resistance syndrome and responds to their amelioration,**

48 patients with chronically elevated liver enzymes and clinical, ultrasound and histological findings consistent with fatty infiltration of the liver were studied prospectively. Most patients were overweight or obese (64%) with increased waist circumference, which closely relates to visceral fat.

I**NTERPRETATION.** Fatty liver was strongly associated with many features of the insulin resistance syndrome, and follow-up showed a high potential for reversibility and a benign course. Only 10% of patients had normal glucose tolerance: 44% had diabetes mellitus, 29% impaired glucose tolerance and 17% were hyperinsulinaemic. The most common dyslipidaemia was hypertriglyceridaemia and/or low HDL cholesterol (86%). Dietary intervention and follow-up (median 24 months), supplemented by oral hypoglycaemic or lipid-lowering drugs, resulted in weight loss (mean 3.7 kg), decreased fasting blood glucose, improved serum lipid profile and improved serum liver enzymes, which became normal in more than half of the patients.

Comment

The patients in this study were referred to a Gastroenterology Unit because of chronically raised liver enzymes. After alcohol and other specific causes had been excluded 48 patients with fatty liver were studied. About two-thirds were overweight and had an increased waist-hip ratio. Only 10% had no detectable abnormality of glucose and insulin metabolism and only 10% were normolipidaemic. Pure hypercholesterolaemia was unusual (4%) and most patients had abnormalities of TGs and/or HDL concentration.

Apart from emphasizing the relationship between NASH and the insulin resistance syndrome, the important feature of this study is the observation that weight reduction and the use of hypoglycaemic and hypolipidaemic] agents was associated both with an improvement in glucose and lipid metabolism and also in liver enzymes. This confirms earlier work that weight reduction can lead to improvement in LFTs and depletion of hepatic fat |**40,41**|. Improvement may also follow surgical intervention in patients with severe morbid obesity, although rapid, drastic weight reduction may led to an increase in

inflammatory changes |**42**|. There is relatively little work on the effect of lipid-lowering drugs on liver function and fat content in patients with NASH. One pilot study suggested a modest improvement in liver function using gemfibrozil |**43**|, whereas in another |**44**| ursodeoxycholic acid was said to reduce liver enzyme levels and the grade of hepatic steatosis, whereas clofibrate did not.

In summary NASH is an area of increasing interest for hepatologists. There remains a lot to be learnt about the factors determining its prognosis. On the one hand some patients may pursue a benign course, although a benign outcome over 10 years does not necessarily presage a similar result over 30 years. On the other hand a significant proportion of cases of cryptogenic cirrhosis may have evolved from patients with NASH. What proportion of cases, and which, will ultimately become cirrhotic? It is conceivable that a relatively small proportion of patients with NASH go on to cirrhosis but those who do constitute a substantial proportion of patients with cryptogenic cirrhosis. The relationship between NASH and the

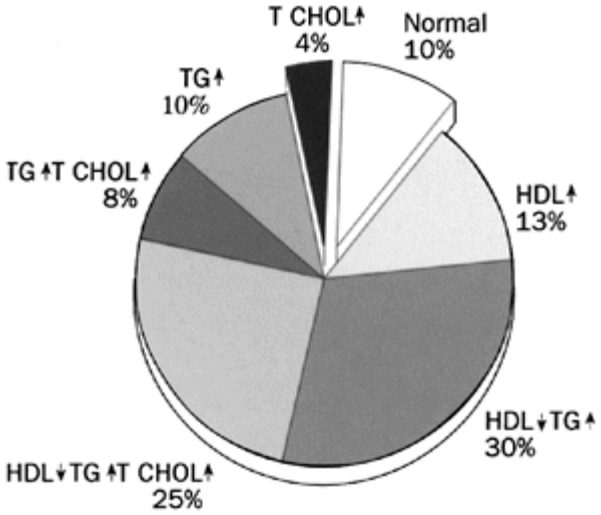


Fig. 2.6 Distribution of lipid profiles in patients with fatty liver. T CHOL, total cholesterol. Lipoprotein abnormalities were defined according to the Lipid Research Clinic data for age and tender (levels above the 90th percentile for T CHOL and TG levels or below the 10th percentile for HDL cholesterol level were considered abnormal). Source: Knobler *et al.* (1999).

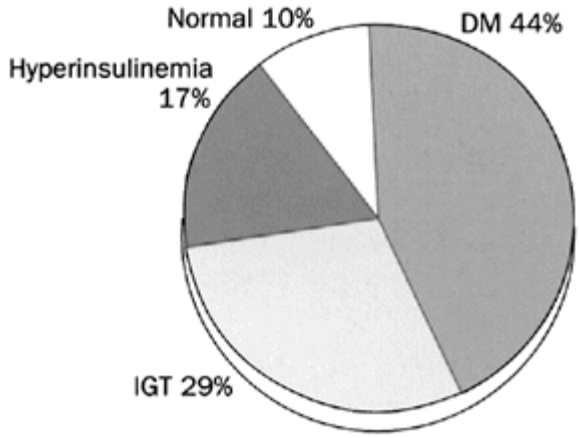


Fig. 2.7 Distribution of glucose tolerance in patients with fatty liver. Normal, normal response to oral glucose tolerance test; IGT, impaired glucose tolerance test; DM, diabetes mellitus. Hyperinsulinaemia was defined by fasting insulin level, exceeding the 90th percentile of its distribution among patients with normal glucose tolerance.
Source: Knobler *et al.* (1999).

insulin resistance syndrome is of great potential importance for physicians interested in metabolic medicine. To what extent the measures taken to reduce the risk of CHD will also prevent the development of cryptogenic cirrhosis remains to be determined.

Fish oil, dyslipidaemia and cardiovascular disease

Interest in fatty acids of marine origin stemmed from the perception that Greenland Eskimos and others eating a diet rich in oily fish were subject to a very low risk of CHD |**45**|. The Diet and Reinfarction Trial (DART) |**46**| showed that men who had suffered a previous myocardial infarct, and who were randomized to take at least two meals with a main course of fatty fish per week, had a reduction in all-cause mortality of 29% over 2 years. CHD mortality was reduced by a similar amount. These effects are dramatic for a comparatively low intake of n-3 fatty acids. The estimated consumption of eicosapentaenoic acid (EPA) was about 2.4 g/week. Surprisingly, non-fatal reinfarction was not reduced, in fact there was a small but non-significant increase. This was rationalized on the basis that the fish diet may have reduced the incidence of fatal arrhythmias but not influenced that of myocardial reinfarction.

The presumption is that diets rich in marine oils exert their beneficial effects through very long-chain polyunsaturated fatty acids, predominantly EPA and docosahexaenoic acid (DHA), which have a variety of effects on lipid and haemostatic variables, as well as anti-arrhythmic properties |**47**|.

Dietary supplementation with n-3 polyunsaturated fatty acids and vitamin E after myocardial infarction: results of the GISSI-Prevenzione trial.

Gruppo Italiano per lo Studio della Sopravvivenza nell'Infarto Miocardico. *Lancet* 1999; **354**:447–55.

B**ACKGROUND. There is conflicting evidence on the benefits of foods rich in vitamin E (α-tocopherol), n-3 polyunsaturated fatty acids (PUFA), and their pharmacological substitutes. The effects of these substances as supplements were investigated in patients who had survived recent (≤3 months) MI. Groups of almost 3000 patients were randomly assigned supplements of n-3 PUFA (1 g daily), vitamin E (300 mg daily), both, or neither for 3.5 years. The primary combined efficacy end-point was death, non-fatal MI, and stroke.**

I**NTERPRETATION.** Dietary supplementation with n-3 PUFA led to a clinically important and statistically significant benefit. Vitamin E was not beneficial. Treatment with n-3 PUFA, but not vitamin E, significantly lowered the risk of the primary end-point by decreasing the risks of death and cardiovascular death. The effect of the combined treatment was similar to that of n-3 PUFA for the primary end-point and for fatal events.

Comment

This study, like The Heart Outcomes Prevention Evaluation (HOPE) study |**48**|, is a setback for the antioxidant approach to atherosclerosis prevention, but it confirms the benefits of marine fatty acids. The subjects consumed 1 g capsules containing n-3 fatty acids of which about 870 mg were EPA and DHA (in a ratio of 1:2), equivalent to about 100 g fatty fish per day. This corresponds to an average weekly intake of about 2 g EPA, which is not greatly different from that taken in two fish meals in the DART study. The fact that benefit in terms of cardiovascular events was seen with capsules containing esters of marine fatty acids supports the belief that it is these fatty acids rather than some other constituent of the whole fish that confer the benefit seen in the DART study and in epidemiological observations.

As with the DART study no reduction in non-fatal cardiac events was seen, supporting the notion that the action of fish oil was to reduce fatal arrhythmias. There was a significant reduction in sudden death, which accounted for most of the benefit seen in cardiac deaths overall. Stroke was not reduced, and as the cause in most cases was not determined it is not possible to say if there was any increase in haemorrhagic stroke.

Omacor in familial combined hyperlipidaemia: effects on lipids and low density lipoprotein subclasses.

L Calabresi, D Donati, F Pazzucconi, C R Sirtori, G Franceschini. *Atherosclerosis* 2000; **148**:387–96.

B**ACKGROUND. Elevated plasma cholesterol and/or TGs, and the prevalence of small, dense LDL particles greatly increase coronary risk in patients with FCH. To assess the effects of Omacor, which contains the n-3 fatty acids eicosapentaenoic and DHA (EPA and DHA), on plasma lipid/lipoprotein levels and LDL particle distribution, 14 FCH patients received Omacor (3.4 g EPA+DHA daily) or placebo for 8 weeks in a randomized, double-blind, cross-over study.**

I**NTERPRETATION.** Omacor significantly lowered plasma TGs and VLDL cholesterol levels. Total cholesterol did not change but LDL cholesterol and apolipoprotein (apo)B concentrations increased. LDL particles were small and apoB-rich in the selected subjects. After Omacor treatment LDL became enriched in cholesterol (mainly cholesteryl esters) indicating accumulation in plasma of more buoyant and core-enriched LDL particles. Separation of LDL subclasses showed increased plasma concentrations of intermediate density lipoprotein and of the fast-floating LDL-1 and LDL-2 subclasses after Omacor, and a decrease in the denser, slow floating LDL-3 subclass but average LDL size did not change. The resistance of the small LDL pattern to drug-induced modifications implies that a maximal lipid-lowering effect must be achieved to reduce coronary risk in FCH patients.

Comment

Fish oil preparations have not been widely adopted in Lipid Clinics. There are probably a number of reasons for this. The predominant effect on serum lipids is to lower TGs, which have been a more contentious risk factor than serum or LDL cholesterol. Moreover, in hypertriglyceridaemic subjects fish oil may raise LDL cholesterol (as may fibrates). Depending on the preparation used compliance has also sometimes been a problem with the need to take a large number of capsules or drink a small volume of actual oil of questionable taste.

In this study of FCH patients, in common with some previous work, marine fatty acids not only reduced TGs but increased both LDL cholesterol and, to a lesser extent, apoB. The ratio of LDL cholesterol/apoB was therefore also increased implying on average less dense particles and this was reflected in an increase in concentration of LDL-2 and possibly LDL-1. Average LDL size measured on polyacrylamide gels, however, was said not to change.

The implications for cardiovascular risk are not easy to determine. A shift in LDL distribution in favour of more buoyant particles would normally be interpreted as favourable. Whether this would in fact be the case, when there was also an increase in apoB (i.e. an increase in particle number) and intermediate density lipoprotein and no significant reduction in the small dense LDL-3, is unclear. The results of studies like DART and the Gruppo Italiano per lo Studio della Supravvivenza nell'Infarto miocardico (GISSI) trial discussed above would imply likely benefit from the intake of n-3 fatty acids (at a lower dose than in the current study), although it is far from clear that the reduction in cardiovascular risk is mediated by effects on lipoproteins.

The effect of concentrated n-3 fatty acids versus gemfibrozil on plasma lipoproteins, low density lipoprotein heterogeneity and oxidizability in patients with hypertriglyceridemia.

A F Stalenhoef, J de Graaf, M E Wittekoek, *et al. Atherosclerosis* 2000; **153**:129–38.

B**ACKGROUND. This double-blind randomized double-dummy trial in 28 patients with primary hypertriglyceridaemia, compared the effects of gemfibrozil (1200 mg/day) and Omacor (4 g/day; n-3 fatty acids eicosapentaenoic and DHA) on lipid and lipoprotein levels, LDL subfraction profile and LDL oxidizability.**

I**NTERPRETATION.** Both Omacor and gemfibrozil had favourable effects on lipid and lipoprotein concentrations and on the LDL subfraction profile. Omacor increased the susceptibility of LDL to oxidation, but gemfibrozil did not affect the resistance of LDL to oxidation. The clinical relevance of these changes should be established in the light of other postulated favourable effects of n-3 fatty acids on the course of CVD.

Comment

The patients in this study were a heterogeneous group with relatively severe primary hypertriglyceridaemia (serum TG 4–28 mmol/l). Gemfibrozil and the marine fatty acid esters produced indistinguishable changes in serum lipids and lipoproteins with reductions in TG-rich lipoproteins and an increase in both LDL and HDL cholesterol. As Fig. 2.8 shows both agents produced a shift in favour of larger less dense LDL with a reduction in the absolute amount of cholesterol in the two most dense LDL fractions. According to current thinking this would constitute an improvement in LDL composition, which might outweigh the overall increase in LDL cholesterol. LDL-apoB concentration was not determined so there is no direct measure of the effect on the number of circulating LDL particles. It should be noted that LDL was subdivided into five subfractions in this study that are therefore not directly comparable with the three subfractions in the study of Calabresi *et al.* described above.

Studies of the effects of fish oil on LDL oxidizability are contradictory. Here the n-3 fatty acids were associated with an increase in the susceptibility of LDL lipid to oxidation, whereas gemfibrozil was not. Although this is potentially an unfavourable effect it has to be seen in the context of the failure of pharmacological doses of

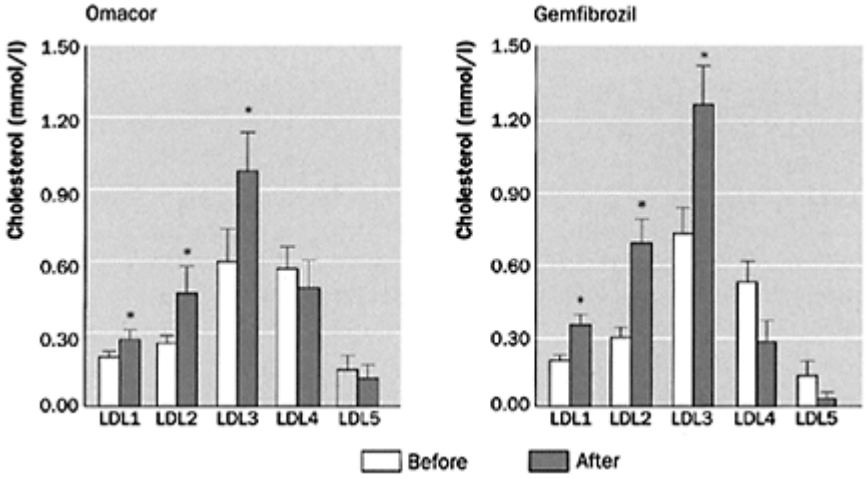


Fig. 2.8 Effect of treatment with either Omacor (n=15) or gemfibrozil (n=13) on the cholesterol content of five LDL subfractions (LDL1–5) of patients with hypertriglyceridaemia: 'before' denotes values at week 0; 'after' denotes values at week 12. *P<0.05 for the within-treatment group, Wilcoxon signed rank test (before versus after). No significant difference between the effect of Omacor and gemfibrozil treatment on the cholesterol concentration of the LDL subfractions was found.
Source: Stalenhoef *et al.* (2000).

vitamin E to reduce cardiovascular events in prospective trials (HOPE and GISSI), and of the beneficial effects of marine fatty acids identified in the DART and GISSI trials discussed above.

Effect of n-3 fatty acids on the composition and binding properties of lipoproteins in hypertriglyceridemic patients.
H C Hsu, Y T Lee, M F Chen. *Am J Clin Nutr* 2000; **71**:28–35.

B**ACKGROUND. Treatment of hyperlipidaemic patients with fish oil increases plasma LDL cholesterol despite a marked decrease in the LDL precursor, VLDL. To investigate the relationship between VLDL composition and LDL concentrations 14 hypertriglyceridaemic patients**

were treated with encapsulated fish oil (containing 1.45 g EPA and 1.55 g DHA) daily for 4 weeks. Eleven normolipidaemic individuals acted as controls.

I**NTERPRETATION.** Treatment of hypertriglyceridaemic patients with fish oil caused differential effects on VLDL subfractions and decreased LDL binding to fibroblast receptors, which may have contributed to the paradoxical increase in LDL cholesterol concentrations.

Comment

The well-known increase in LDL cholesterol often seen in hypertriglyceridaemic subjects treated with fibrates and fish oil discussed above is usually attributed to increased conversion of TG-rich remnants to LDL. This work suggests an additional mechanism in the case of fish oil, namely an alteration in LDL conformation leading to impaired clearance by the LDL receptor. This is consonant with the finding of an increase in apoB concentration in some studies showing that there is an increase in number of lipoprotein particles and not just an increase in LDL mass due to enrichment with lipid.

Dietary supplementation with marine omega-3 fatty acids improve systemic large artery endothelial function in subjects with hypercholesterolemia.

J Goodfellow, M F Bellamy, M W Ramsey, C J Jones, M J Lewis. *J Am Coll Cardiol* 2000; **35**:265–70.

B**ACKGROUND. Marine omega-3 fatty acids improve vascular function, but the underlying mechanism(s) are unclear. To determine whether dietary supplementation with marine omega-3 fatty acids improves systemic large artery endothelial function in patients with hypercholesterolaemia, hypercholesterolaemic individuals with no other known cause of endothelial dysfunction were treated with omega-3 fatty acids**

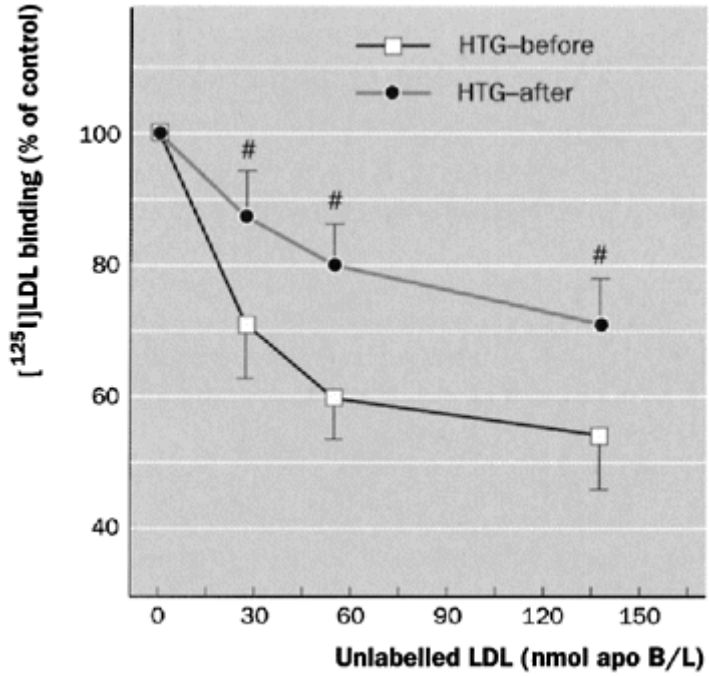


Fig. 2.9 Competition for fibroblast binding between [^{125}I]LDL and increasing concentrations of unlabelled LDL from hypertriglyceridaemic (HTG; n=14) patients before and after 4 weeks of supplementation with fish oil. $^{\#}$Significantly different from before treatment, P<0.05 (paired t-test). Source: Hsu *et al.* (2000).

(4 g/day) for 4 months in a placebo-controlled, randomized, double-blind, parallel-group study.

I**NTERPRETATION.** Marine omega-3 fatty acids improve large artery endothelium-dependent dilation in subjects with hypercholesterolaemia without affecting endothelium-independent dilation. The treatment significantly improved flow-mediated dilation and significantly reduced TGs, whereas with placebo there was no change in either. Responses to sublingual glyceryl trinitrate were unchanged.

Comment

The beneficial effects of cholesterol lowering on vascular reactivity, and in particular endothelial function, were discussed in Chapter 1. This study demonstrates similar improvement of endothelium-dependent vasodilatation produced by pharmacological doses of marine fatty acids. There are, however, conflicting data in the literature |**49**|.

ApoE polymorphism and fish oil supplementation in subjects with an atherogenic lipoprotein phenotype.

A M Minihane, S Khan, E C Leigh-Firbank, *et al. Arterioscler Thromb Vasc Biol* 2000; **20**:1990–7.

B**ACKGROUND. This study assessed the efficacy of fish oil supplementation in counteracting the classic dyslipidaemia of the ALP. The impact of the common apoE polymorphism on lipid profiles and on responsiveness to the fish oil was also investigated. Men with ALP (*n*=55) completed a randomized placebo-controlled cross-over trial of fish oil (3.0 g EPA/DHA daily) and placebo (olive oil) capsules with 6-week treatment arms separated by a 12-week washout period.**

I**NTERPRETATION.** Fish oil fatty acids partially counteracted the pro-atherogenic lipid profile of ALP, and responsiveness to this treatment was influenced by the apoE genotype. Fish oil supplementation reduced fasting TGs, the postprandial TG response and the percentage LDL-3. No change in HDL cholesterol was seen. Baseline HDL cholesterol levels were significantly lower in apoE4 carriers. In response to fish oil intervention those with an apoE2 allele showed a reduced postprandial incremental TG response and a trend towards increased lipoprotein lipase activity compared with non-E2 carriers. ApoE4 individuals, showed increased total cholesterol and a trend towards reduced HDL cholesterol, compared with the common homozygous E3/E3 profile.

Comment

As shown in Fig. 2.10 supplementation with fish oil (3 g EPA+DHA per day) not only produced the usual reduction in serum TGs and alimentary lipaemia in this group of subjects with an ALP, but also reduced the proportion of LDL contributed by small dense LDL-3. The reduction in area under the TG response curve partly relates to the lower baseline TG concentration but is also the result of a reduction in the postprandial increment in TG concentration.

It is common experience that patient responses to dietary or drug treatments vary considerably and compliance is, undoubtedly, often a factor in this. In addition, a multitude of genetic polymorphisms are likely to play a part and none has been more extensively investigated than the common variations in apoE structure |**12**|. This has a well-documented effect on LDL cholesterol concentration and accounts for some 10% of the population variance.

In this study apoE genotype had little effect on the reduction of fasting TG concentration (35% overall) but subjects with a copy of the apoE2 gene (there were no E2 homozygotes) showed the most marked suppression in the postprandial incremental

TG response. They constituted 16% of the whole group of subjects, a figure not very different from the 8% quoted for Caucasian populations in general. The effect of fish oil in reducing the incremental TG response in the common E3 homozygotes (44% of subjects in this study and about 60% of Caucasian populations) was tiny. The implication is that all genotypes benefit in terms of reduced

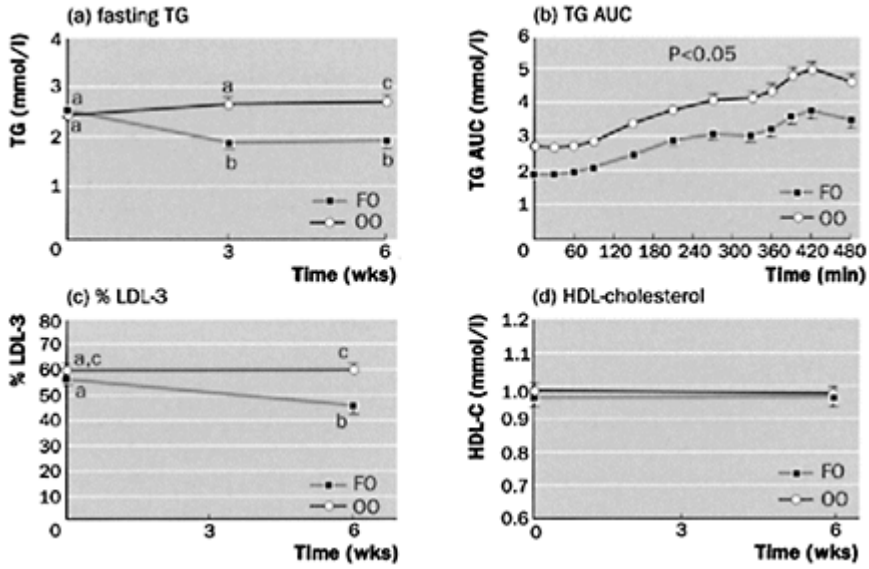


Fig. 2.10 Responsiveness of the characteristic features of ALP to fish oil (FO) supplementation. 00 indicates olive oil. Group means for fasting TG, per cent LDL-3, and HDL-C were compared by two-way repeated-measures ANOVA, with time and oil supplement (FO and 00) used as independent variables. *Post hoc* Tukey honestly significant difference tests were performed. Different lettering indicates that the mean values are significantly different (P<0.05). TG AUCs were compared by paired t-test.
Source: Minihane *et al.* (2000).

VLDL secretion leading to a reduction in fasting TG concentration, whereas in those with an E2 copy there is an additional increase in postprandial TG-rich lipoprotein clearance.

All genotypes appeared to benefit in terms of a reduction in the percentage of LDL in LDL-3. This was greater in those with an apoE4 gene than in E3 homozygotes. There was an increase of LDL cholesterol overall of 7.1% and this was most marked in those with an E4 allele. LDL-apoB was not determined so it is not possible to comment to what extent this reflects an increase in LDL particle number, as opposed to a favourable shift from dense to lighter LDL subfractions.

Conclusion

It is increasingly apparent that hypertriglyceridaemic subjects generally are at increased risk of vascular disease even when concomitant variables such as LDL cholesterol and HDL cholesterol are taken into account. The risk in individual subjects, however, is difficult to define given the heterogeneity of TG-rich lipoproteins. Most national guidelines do not set targets for serum TG concentrations and they are not one of the variables included in the Framingham risk equation. The incorporation of HDL cholesterol in the equation no doubt embraces elements of risk associated with serum TGs and LDL particle size given the relationships between these variables and this appears to function well for certain large population groups. Refinement of the process for the better definition of risk in the individual attending a routine Lipid Clinic is likely to be a considerable way away.

Familial combined hyperlipidaemia is common but is probably a rather woolly concept in the minds of many clinicians dealing with dyslipidaemic patients. This reflects the failure to identify a predominant metabolic or genetic defect such as those in FH and FDB. It continues, rightly, to be an area of intense research activity and it appears that several different underlying mechanisms will be confirmed. A defect in FFA metabolism seems to be fundamental. The association with the insulin resistance syndrome overlaps with the large body of dyslipidaemic subjects who have concomitant abnormalities of liver function and NASH. Most physicians see large numbers of these patients but they are under-recognized and the management is ill-defined, although most would agree that the usual life-style measures are appropriate if not always effective.

Patients with severe hypertriglyceridaemia and the chylomicronaemia syndrome represent perhaps the only medical emergency where management of the dyslipidaemia is crucial over a matter of hours or days. Unfortunately, when the situation arises the patients are often not under the care of clinicians with expertise in lipid disorders, although chemical pathologists can provide valuable advice. Plasma exchange and ‘fat-free’ parenteral nutrition may be useful adjuncts to mitigate attacks of pancreatitis in the very sick patient. Antioxidant supplementation appears to be a valuable prophylactic measure in those with genetic hypertriglyceridaemia and deserves investigation in other groups as well.

References

1. Illingworth DR. Management of hypercholesterolemia. *Med Clin North Am* 2000; **84**: 23–42.
2. Hulley SB, Rosenman RH, Bawol RD, Brand RJ. Epidemiology as a guide to clinical decisions. The association between triglyceride and coronary heart disease. *N Engl J Med* 1980; **302**:1383–9.
3. Hokanson JE, Austin ME. Plasma triglyceride level is a risk factor for cardiovascular disease independent of high-density lipoprotein cholesterol level: a meta-analysis of population-based prospective studies. *J Cardiovasc Risk* 1996; **3**:213–19.
4. Watts GF, Dimmit SB. Fibrates, dyslipoproteinaemia and cardiovascular disease. *Curr Opin Lipidol* 1999; **10**:561–74.
5. Gaw A, Shepherd J. Fibric acid derivatives. In: Betteridge DJ, Illingworth DR, Shepherd J (eds). *Lipoproteins in Health and Disease*, 1st edn. London: Arnold,

1999:1145–60.
6. Rubins BH, Robins SJ, Collins D. Gemfibrozil for the secondary prevention of coronary heart disease in men with low levels of high-density lipoprotein cholesterol. *N Engl J Med* 1999; **341**:410–18.
7. Sniderman AD. Postprandial hypertriglyceridemia(s): time to enlarge our pathophysiologic perspective. *Eur J Clin Invest* 2000; **30**:935–7.
8. Zilversmit DB. Atherogenesis: a postprandial phenomenon. *Circulation* 1979; **60**: 473–85.
9. Frayn KN. Non-esterified fatty acid metabolism and postprandial lipaemia. *Atherosclerosis* 1998; **141**(Suppl 1): S41–6.
10. Humphries SE, Peacock R, Gudnason V. Genetic determinants of hyperlipidaemia. In: Betteridge DJ, Illingworth DR, Shepherd J (eds). *Lipoproteins in Health and Disease*, 1st edn. London: Arnold, 1999:127–62.
11. Jarvik GP, Austin MA, Brunzell JD. Familial combined hyperlipidaemia. In: Betteridge DJ, Illingworth DR, Shepherd J (eds). *Lipoproteins in Health and Disease*, 1st edn. London: Arnold, 1999:692–9.
12. Breslow JL. Genetics of lipoprotein abnormalities associated with coronary heart disease susceptibility. *Annu Rev Genet* 2000; **34**:233–54.
13. Juo SH, Bredie SJH, Kiemeney LA, Demacker PNM, Stalenhoef AFH. A common genetic mechanism determines plasma apolipoprotein B levels and dense LDL subfraction distribution in familial combined hyperlipidaemia. *Am J Hum Genet* 1998; **63**: 586–94.
14. Allayee H, Aouizerat BE, Cantor RM, Dallinga-Thie GM, Krauss RM, Lanning CD, Rotter JI, Luis AJ, de Bruin TWA. Families with familial combined hyperlipidemia and families enriched for coronary heart disease share genetic determinants for the atherogenic lipoprotein phenotype. *Am J Hum Genet* 1998; **63**:577–85.
15. Connelly PW, Hegele RA. Hepatic lipase deficiency. In: Betteridge DJ, Illingworth DR, Shepherd J (eds). *Lipoproteins in Health and Disease*, 1st edn. London: Arnold, 1999: 829–40.
16. Santamarina-Fojo S, Haudenschild C, Amar M. The role of hepatic lipase in lipoprotein metabolism and atherosclerosis. *Curr Opin Lipidol* 1998; **9**:211–19.
17. Mahley RW, Rall SC. Type III hyperlipoproteinaemia (dysbetalipoproteinaemia; remnant particle disease). In: Betteridge DJ, Illingworth DR, Shepherd J (eds). *Lipoproteins in Health and Disease*, 1st edn. London: Arnold, 1999:719–36.
18. Brunzell JD. Familial lipoprotein lipase deficiency and other causes of the chylomicronemia syndrome. In: Scriver CR, Beaudet AL, Sly WS, Valle D (eds). *The Metabolic and Molecular Bases of Inherited Disease*, 7th edn, Vol. II. New York: McGraw-Hill, 1995: 1913–32.
19. Bhatnagar D. Hypertriglyceridaemia. In: Betteridge DJ, Illingworth DR, Shepherd J (eds). *Lipoproteins in Health and Disease*, 1st edn. London: Arnold, 1999:737–51.
20. Miller JP. Serum triglycerides, the liver and the pancreas. *Curr Opin Lipidol* 2000; **11**: 377–82.
21. Hozumi Y, Kawano M, Saito T, Miyata M. Effect of tamoxifen on serum lipid metabolism. *J Clin Endocrinol Metab* 1998; **83**:1633–5.
22. Fernandez-Miranda C, Castellano G, Guijarro C, Fernandez I, Schoebel N, Larumbe S, Gomez-Izquierdo T, del Palacio A. Lipoprotein changes in patients with chronic hepatitis C treated with Interferon-alpha. *Am J Gastroenterol* 1998; **93**:1901–4.
23. Jabbar MA, Zuhri-Yafi MI, Larrea J. Insulin therapy for a non-diabetic patient with severe hypertriglyceridemia. *J Am Coll Nutr* 1998; **17**:458–61.

24. Kollef MH, McCormack MT, Caras WE, Reddy VVB, Bacon D. The fat overload syndrome: successful treatment with plasma exchange. *Ann Intern Med* 1990; **112**:545–6.
25. Koppel K, Bratt G, Eriksson M, Sandstrom E. Serum lipid levels associated with increased risk for cardiovascular disease is associated with highly active antiretroviral therapy (HAART) in HIV-1 infection. *Int J STD AIDS* 2000; **11**:451–5.
26. Thiebaut R, Dabis F, Malvy D, Jacqmin-Gadda H, Mercie P, Valentin VD. Serum triglycerides, HIV infection, and highly active antiretroviral therapy, Aquitaine Cohort, France. 1996–98. *J Acquir Immune Defic Syndr* 2000; **23**:261–5.
27. Henry K, Melroe H, Huebesch J, Hermundson J, Levine C, Swensen L, Daley J. Severe premature coronary artery disease with protease inhibitors. *Lancet* 1998; **351**:1328.
28. Correspondence. *Lancet* 1998; **351**:1958–60.
29. Echevarria KL, Hardin TC, Smith JC. Hyperlipidemia associated with protease inhibitor therapy. *Ann Pharmacother* 1999; **33**:859–63.
30. Clark Perry R, Cushing HE, Deeg MA, Prince MJ. Ritonavir, triglycerides and pancreatitis. *Clin Infect Dis* 1999; **28**:161–2.
31. Henry K, Melroe H, Huebesch J, Hermundson J, Simpson J. Atorvastatin and gemfibrozil for protease-inhibitor-related lipid abnormalities. *Lancet* 1998; **352**:1031–2.
32. Wierzbicki AS, Reynolds TM, Crook MA, Tatler J, Peters BS. Lipid lowering therapy in patients with HIV infection. *Lancet* 1998; **352**:1782.
33. Diehl AM. Nonalcoholic steatohepatitis. *Semin Liver Dis* 1999; **19**:221–9.
34. Ludwig J, Viggiano TR, McGill DB, Ott BJ. Nonalcoholic steatohepatitis: Mayo Clinic experiences with a hitherto unnamed disease. *Mayo Clin Proc* 1980; **55**:434–8.
35. Sorbi D, Boynton J, Lindor KD. The ratio of aspartate aminotransferase to alanine aminotransferase: potential value in differentiating non-alcoholic steatohepatitis from alcoholic liver disease. *Am J Gastroenterol* 1999; **94**:1018–22.
36. Cotrim HP, Andrade ZA, Parana R, Portugal M, Lyra LG, Freitas LAR. Nonalcoholic steatohepatitis: a toxic liver disease in industrial workers. *Liver* 1999; **19**:299–304.
37. Teli MR, James OFW, Burt AD, Bennett MK, Day CP. The natural history of non-alcoholic fatty liver: a follow-up study. *Hepatology* 1995; **22**:1714–9.
38. Powell EE, Cooksley WGE, Hanson R, Searle J, Halliday JW, Powell LW. The natural history of nonalcoholic steatohepatitis: a follow-up study of forty-two patients for up to 21 years. *Hepatology* 1990; **11**:74–80.
39. Caldwell SH, Oelsner DH, Iezzoni JC, Hespenheide EE, Battle EH, Driscoll CJ. Cryptogenic cirrhosis: clinical characterization and risk factors for underlying disease. *Hepatology* 1999; **29**:664–9.
40. Eriksson S, Eriksson K-F, Bondesson L. Nonalcoholic steatohepatitis in obesity: a reversible condition. *Acta Med Scand* 1986; **220**:83–8.
41. Palmer M, Schaffner F. Effect of weight reduction on hepatic abnormalities in overweight patients. *Gastroenterology* 1990; **99**:1408–13.
42. Luyckx FH, Lefebvre PJ, Scheen AJ. Non-alcoholic steatohepatitis: association with obesity and insulin resistance, and influence of weight loss. *Diab Metab* 2000; **26**:98–106.
43. Basaranoglu M, Acbay O, Sonsuz A. A controlled trial of gemfibrozil in the treatment of patients with nonalcoholic steatohepatitis. *J Hepatol* 1999; **31**:384.
44. Laurin J, Lindor KD, Crippin JS, Gossard A, Gores GJ, Ludwig J, Rakela J, McGill DB. Ursodeoxycholic acid or clofibrate in the treatment of non-alcohol-induced

steatohepatitis: a pilot study. *Hepatology* 1996; **23**:1464–7.

45. Elwood PC, Burr ML, Sweetnam PM. Fish, fibre and heart disease. In: Marmot M, Elliott P (eds). *Coronary Heart Disease Epidemiology From Aetiology to Public Health*, 1st edn. Oxford: Oxford University Press, 1992:203–16.

46. Burr ML, Gilbert JF, Holliday RM, Elwood PC, Fehily AM, Rogers S, Sweetnam PM, Deadman NM. Effects of changes in fat, fish, and fibre intakes on death and myocardial reinfarction: diet and reinfarction trial (DART). *Lancet* 1989; **2**:757–61.

47. Angerer P, von Schacky C. n-3 fatty acids and the cardiovascular system. *Curr Opin Lipidol* 2000; **11**:57–63.

48. The Heart Outcomes Prevention Evaluation Study Investigators. Vitamin E supplementation and cardiovascular events in high-risk patients. *N Engl J Med* 2000; **342**: 154–60.

49. Kothny W, Angerer P, Stork S, von Schacky C. Short term effects of omega-3 fatty acids on the radial artery of patients with coronary heart disease. *Atherosclerosis* 1998; **140**: 181–6.

3
Other lipoprotein disorders

Introduction

In this chapter selected papers relating to several unrelated disorders of lipoprotein metabolism are discussed. These include renal disease and post-transplantation dyslipidaemia. Important new work on the initial stages of reverse cholesterol transport is briefly reviewed as well as two disorders that cause hypolipidaemia and that can have very similar phenotypic expression, even though genetically they are quite distinct: abetalipoproteinaemia (ABL) and familial hypobetalipoproteinaemia (FHBL).

Renal disease

The burden of cardiovascular disease in patients with chronic renal disease is generally acknowledged. As techniques of renal replacement therapy with haemodialysis and peritoneal dialysis as well as transplantation have improved, so has the prognosis of the patients allowing them increasingly to manifest the clinical complications of atherosclerotic disease. Dyslipidaemia is common in all forms of renal disease, although often not actively treated. There is some anxiety about prescribing additional medication in patients with impaired renal function, although the statins, which are excreted predominantly in the bile are increasingly widely used. Fibrates have significant renal excretion and can sometimes be associated with declining renal function. Nevertheless they too can be used, provided the dose is appropriately adjusted, and renal function is carefully monitored. They should probably be reserved for those patients in whom the principal abnormality is in triglycerides (TGs) and high-density lipoprotein (HDL). There is a lack of endpoint trial evidence, which will be difficult to obtain, and of specific guidelines for the primary and secondary prevention of vascular disease in chronic renal failure. Inevitably, treatment is based on extrapolation from evidence obtained in patients with normal renal function. There are several relevant current reviews |**1–6**|.

Chronic renal failure

Effects of atorvastatin on dyslipidaemia in uraemic patients on peritoneal dialysis.

G Hufnagel, C Michel, F Vrtovsnik, *et al. Nephrol Dial Transplant* 2000; **15**:684–8.

B**ACKGROUND. The efficacy and safety of atorvastatin, a potent cholesterol-and TG-lowering agent, was evaluated in 31 peritoneal dialysis patients with hypercholesterolaemia who were treated for 4 months with atorvastatin at a starting dose of 10 mg. The dose could be increased to 20 or 40 mg to achieve a plasma low-density lipoprotein (LDL) cholesterol of 130 mg/dl for primary prevention, or 100 mg/dl for secondary prevention of coronary heart disease (CHD), and plasma TGs of 200 mg/dl. Nineteen patients also had hypertriglyceridaemia and seven had diabetes; 20 had no coronary history (primary prevention), whereas nine had experienced a coronary event (secondary prevention).**

I**NTERPRETATION.** Atorvastatin appeared to be effective and safe for lipid lowering in peritoneal dialysis patients with mixed dyslipidaemia. In primary and secondary prevention patients, mean LDL cholesterol and TG levels decreased significantly: 19 primary and seven secondary prevention patients achieved the LDL cholesterol target while the TG target was achieved by 15 of the 19 hypertriglyceridaemic patients. Two patients stopped treatment because of gastrointestinal disturbances or allergic skin reaction.

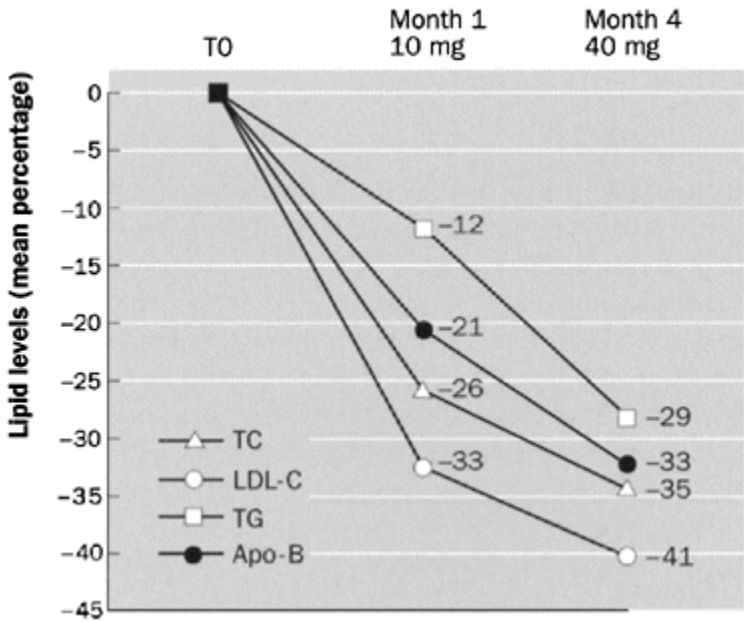


Fig. 3.1 Change in lipid levels in patients treated with atorvastatin 40 mg (n=14).

Source: Hufnagel *et al.* (2000).

Comment

Patients on peritoneal dialysis are believed to be particularly prone to hyperlipidaemia because of the absorption of glucose from the dialysis fluid and loss of protein into the peritoneal cavity. Cardiovascular disease is the principal cause of death |**7**|. There have been two previous small trials of statins in this population |**8,9**| and data are limited.

This relatively short-term study (4 months) had no comparison group, but shows clear-cut changes in lipid and lipoprotein variables compared with baseline with doses of atorvastatin up to 40 mg/day. One patient developed muscle weakness with a moderate increase in serum creatine kinase (CK), to four times the upper limit of normal, while taking 80 mg/day of atorvastatin and this dose was not used further in the trial. Two other patients were withdrawn because of treatment-related adverse effects (diarrhoea, rash), but the drug was generally well tolerated.

A randomized placebo-controlled double-blind trial of lipid-lowering strategies in patients with renal insufficiency: diet modification with or without fenofibrate.

A Levin, L Duncan, O Djurdjev, *et al. Clin Nephrol* 2000; **53**:140–6.

B**ACKGROUND. Renal insufficiency is characterized by lipoprotein abnormalities, including elevated TG levels. The safety and efficacy of micronized fenofibrate as a treatment for dyslipidaemia in patients with progressive renal insufficiency was evaluated in a randomized, placebo-controlled double-blind study comparing fenofibrate and dietary modification with dietary modification alone. After 3 months of dietary counselling patients with moderate renal insufficiency and TG levels 2.3 mmol/l or LDL/HDL ratio 5 were randomized to placebo or fenofibrate, with dietary counselling, for 6 months.**

I**NTERPRETATION.** Fenofibrate with dietary modification effectively reduced TG levels in renal insufficiency patients without serious adverse effects. Ten of 16 patients treated with fenofibrate achieved a 30% reduction in TG levels or LDL/HDL ratio reduction <5 compared with two of 12 in the placebo group. Triglyceride levels were significantly reduced in the fenofibrate group. Compliant patients also showed a significantly greater increase in HDL cholesterol levels in the fenofibrate group compared with placebo. Changes in creatinine clearance did not differ significantly between the groups.

Comment

The patients in this study had mild renal impairment with creatinine clearance in the range 20–74 ml/min as an entry criterion. Those in the active treatment group were initially given micronized fenofibrate 67 mg/day, which was titrated to a maximum of 201 mg/day according to response. The authors quote LDL cholesterol concentrations but do not specify how these were determined. As they do not

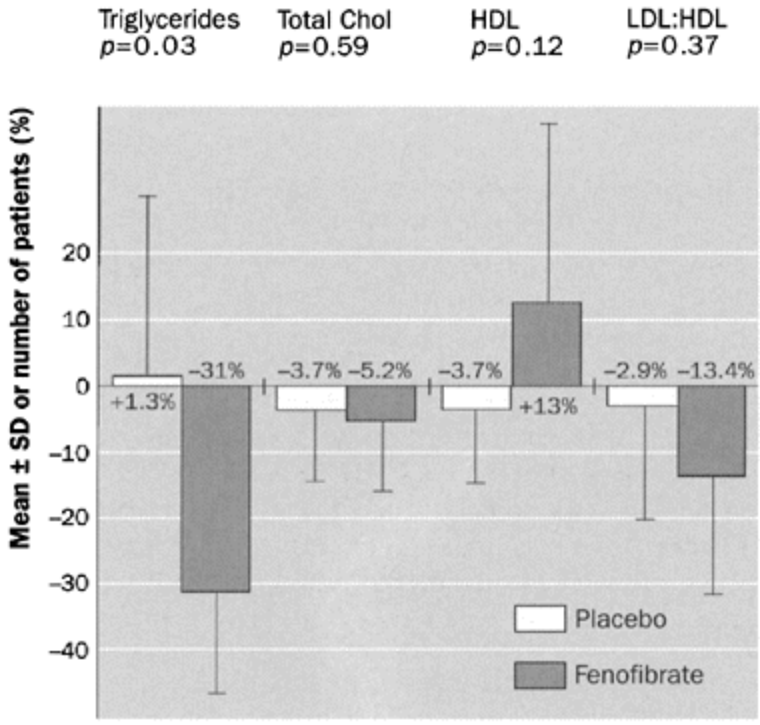


Fig. 3.2. Changes in lipid levels following treatment with placebo or fenofibrate. Mean changes and standard deviations shown. Source: Levin *et al.* (2000).

appear to have been directly measured they were presumably calculated using the Friedewald equation, which is generally regarded as inapplicable when serum TG concentrations exceed 4.5 mmol/l. Several subjects in this study will have had values in excess of this (the mean baseline concentration in the fenofibrate group was 5.3 mmol/l) and in them the calculated LDL cholesterol and LDL/HDL ratios are likely to be unreliable.

There has been concern about decline in renal function in patients with renal failure taking fibrate drugs. In this study there was an increase in plasma creatinine during treatment with both placebo and fenofibrate over the 6 months of the trial but this did not differ between the groups. Although fenofibrate did not produce a significant effect on HDL cholesterol compared with placebo on an intention-to-treat basis there was a highly significant increase in compliant patients.

The study provides useful data as to the safety and efficacy of fenofibrate in mild renal failure particularly when the dominant abnormality is in the TG/HDL axis.

Nephrotic syndrome

The most severe hyperlipidaemia in patients with renal disease is found in those with nephrotic syndrome. Total and LDL cholesterol may be grossly elevated even in those with normal renal function and serum TGs are also commonly increased. The underlying mechanisms include overproduction of TG-rich lipoproteins by the liver together with impairment of lipolysis and receptor-mediated LDL-catabolism |**10,11**|.

Patients with nephrotic-range proteinuria have apolipoprotein C and E deficient VLDL1.

C J Deighan, M J Caslake, M McConnell, J M Boulton-Jones, C J Packard. *Kidney Int* 2000; **58**:1238–46.

B**ACKGROUND. Impaired very low-density lipoprotein (VLDL) clearance contributes to dyslipidaemia in nephrotic-range proteinuria. VLDL can be subdivided into large light VLDL1 and smaller, denser VLDL2. In nephrotic-range proteinuria, VLDL1 clearance is delayed. VLDL1 lipolysis is influenced by apolipoprotein (apo) CII and apoCIII, whereas apoE regulates receptor-mediated clearance. To determine whether impaired VLDL1 clearance was related to a deficiency in apolipoproteins on VLDL1, VLDL subfraction concentrations and VLDL1 apolipoprotein and lipid compositions were measured in patients with glomerular disease and urinary albumin >2 g/24 h and matched controls.**

I**NTERPRETATION.** The results suggested that impaired VLDL1 clearance in nephrotic-range proteinuria is caused by the appearance of particles deficient in apoCII, apoCIII and apoE. VLDL1 apoC deficiency is associated with the formation of smaller particles with a high free cholesterol/phospholipid ratio, and is likely to cause inefficient lipolysis. VLDL1 apoE deficiency is associated with smaller VLDL1 particles but not altered VLDL1 surface lipid content, and may reduce receptor-mediated clearance of this lipoprotein.

Comment

Hypertriglyceridaemia in the nephrotic syndrome is associated with an increase in both large VLDL1, which is generally inefficiently converted to LDL, and smaller, more dense VLDL2, which is richer in cholesterol esters and more readily converted to LDL. Previous work from the same group suggests that the increase in VLDL1 is the result of

impaired clearance whereas that in VLDL2 is due to a combination of increased secretion and reduced catabolism. These changes seem to be a function of proteinuria and are independent of concomitant renal function and serum albumin concentration. An excess of lipid-rich VLDL1 is believed to be a prerequisite for the formation of atherogenic small, dense LDL, which the authors have previously shown to be a feature of nephrotic-range proteinuria. The present study was designed to examine the mechanisms underlying defective VLDL1 metabolism in nephrotic patients. As the same group had previously found such patients to have normal lipoprotein lipase activity *in vitro* they postulated that the defect in clearance was likely to lie in the composition of the VLDL particle.

VLDL1 were found to be relatively deficient in apoCII, apoCIII and apoE. The apoC deficiency seemed to relate to smaller particle size and enrichment of the surface with free cholesterol. ApoE deficiency related solely to change in size. ApoCII activates and apoCIII inhibits lipoprotein lipase activity, but no difference in the ratio of these cofactors was observed between patients and controls. It is the authors' belief that the apoCII content of VLDL1 is so low in these proteinuric subjects that impaired lipolysis is likely. Additionally, the reduction in apoE content was such that at least two-thirds of VLDL1 particles were totally lacking in apoE. As it is a ligand for receptor binding this will probably impair VLDL clearance further. It should be emphasized that this is a compositional study with no kinetic data, but the findings help to explain some of the previously described defects in VLDL metabolism in proteinuric subjects. It would appear that the liver secretes a smaller VLDL1 particle depleted in apoE, which contributes to, but does not totally explain all the observed abnormalities. Defective clearance of this particle is likely to favour formation of atherogenic small dense LDL.

Pravastatin treatment of very low density, intermediate density and low density lipoproteins in hypercholesterolemia and combined hyperlipidemia secondary to the nephrotic syndrome.
R D Toto, S M Grundy, G L Vega. *Am J Nephrol* 2000; **20**:12–17.

B**ACKGROUND. To determine whether pravastatin decreases LDL production and reduces levels of VLDL and intermediate density lipoprotein (IDL), it was used in nephrotic syndrome patients with hypercholesterolaemia and combined hyperlipidaemia (seven with high LDL alone and six with high VLDL, IDL and LDL). Patients, randomized in a placebo-controlled cross-over study were treated for 8 weeks with pravastatin (40 mg/day) or placebo.**

I**NTERPRETATION.** Pravastatin effectively reduced LDL levels in both types of dyslipidaemia by increasing LDL clearance. Treatment did not affect production of LDL or levels of VLDL+IDL-apo B. Thus, pravastatin increases LDL clearance. Statins do not seem to affect production rates of apoB-containing lipoproteins. Treatment of combined hyperlipidaemia may require pravastatin and another drug targeted to normalize levels of VLDL

and IDL

Comment

These authors have previously demonstrated differences in lipoprotein metabolism in nephrotic patients with pure hypercholesterolaemia (IIa phenotype) and those with combined hyperlipidaemia (IIb phenotype). In the former, kinetic studies imply that the defect is primarily one of LDL clearance, whereas in the latter there is a high rate of both LDL production and clearance.

The present study was devised to determine if the effects of pravastatin would differ in these two groups of nephrotic subjects. In the event LDL clearance was increased to a similar extent in both groups on average, without any effect on its production, although there was considerable individual variability. VLDL+IDL cholesterol was decreased only in the subjects with combined hyperlipidaemia and there was no significant effect on VLDL+IDL-apoB in either group. The authors accept that statins have a place in the treatment of combined hyperlipidaemia by reducing LDL concentrations, but they suggest that additional agents may be required to reduce secretion of TG-rich lipoproteins and improve the metabolism of their remnants. They do not specify what additional agents might be used but presumably a statin-fibrate combination is a possibility and the effects of such a combination could be investigated using the kinetic techniques here. This is still a stage removed from assessing the risks and benefits of combined treatments in end-point trials and we are unlikely to see such data for nephrotic patients in the foreseeable future.

LDL-apheresis in patients with nephrotic syndrome: effects on serum albumin and urinary albumin excretion.
P Stenvinkel, A Alvestrand, B Angelin, M Eriksson. *Eur J Clin Invest* 2000; **30**:866–70.

B**ACKGROUND. Hyperlipidaemia is a common feature of the nephrotic syndrome. Retrospective studies suggest that aggressive lipid-lowering with LDL apheresis (LDL-A) may improve dyslipidaemia, decrease urinary albumin excretion and increase serum levels of albumin in patients with focal segmental sclerosis. Seven patients with nephrotic syndrome of diverse aetiologies were investigated in a prospective study, using a fixed protocol of LDL-A designed for treatment twice-a-week for 3 weeks and then once a week for 7 weeks.**

I**NTERPRETATION.** The results of this uncontrolled prospective study show that LDL-A causes a rapid 30–40% decrease in serum cholesterol and plasma lipoprotein(a) (Lp(a)) levels in patients with nephrotic syndrome and

suggest that short-term LDL-A treatment may increase serum albumin levels in nephrotic patients.

Comment

Plasma exchange was first used for the management of hyperlipidaemia in subjects with painful xanthomatous neuropathy in primary biliary cirrhosis |**12**|, but methods of extracorporeal LDL removal have most often been used for the treatment of homozygous familial hypercholesterolaemia (FH) |**13,14**|. This is a short-term study (10 weeks) of intensive LDL-A (once or twice weekly), and contrasts with the longer-term use of the technique in FH where it is commonly used every 2 weeks in conjunction with statin treatment. As expected apheresis had profound effects on total and LDL cholesterol and Lp(a). A small but significant increase in serum albumin was also observed, which persisted in five of the seven patients who achieved a good initial response to apheresis and who were followed for a year. This is an uncontrolled study, however, and the increase in albumin was achieved without apparent alteration in urinary excretion.

This is essentially a pilot study of the effects of a short period of intensive LDL-A for the induction of remission, or at least amelioration, of the nephrotic syndrome, rather than for the long-term control of hyperlipidaemia. The results require confirmation in controlled studies. Statins are most often going to be the treatment of choice for nephrotic hyperlipidaemia. Only rarely will the resources required for prolonged extracorporeal LDL removal be justified in such patients.

Dyslipidaemia after renal and cardiac transplantation

Some manifestation of dyslipidaemia is present in the majority of patients after renal or cardiac transplantation and is thought to be a factor in the high incidence of cardiovascular disease and possibly in chronic graft rejection as well |**15**|. The lipid profile depends not only on renal function but is influenced by a variety of drugs commonly used in such patients, including diuretics and β-blockers, and in particular immunosuppressive agents. Cyclosporin, tacrolimus and corticosteroids all have adverse effects on serum lipids. Ischaemic heart disease is a major cause of morbidity and death after renal transplantation and is thought to play a causative role in chronic transplant rejection (graft coronary vascular disease), which accounts for about 25% of deaths in heart transplant recipients. Enthusiasm for aggressive reduction of serum lipids using statins in transplant patients has been tempered by reports of rhabdomyolysis and renal failure in subjects treated with cyclosporin and lovastatin in high doses |**16**|. It remains to be established whether this was primarily an effect of dose or whether there are differences between the statins in their propensity to cause this complication in transplant recipients. Cyclosporin, which is metabolized by cytochrome P4503A, can cause an increase in the plasma levels of certain statins and this may be the basis of the clinical interaction |**17**|. This has led some authors to suggest that pravastatin and fluvastatin, which are not metabolized by the same cytochrome enzymes as the other statins, may be

less likely to cause this particular adverse interaction |**18,19**|.

Pravastatin has the most extensive evidence base in terms of end-point studies for the primary and secondary prevention of CHD. In patients who had undergone cardiac transplantation it was shown to improve 1-year survival and reduce graft coronary vascular disease and the prevalence of acute rejection |**20**|. Similar benefits have been demonstrated over 4 years with simvastatin |**21**|. At a given dose simvastatin reduces LDL more than pravastatin but atorvastatin is generally regarded as the most potent of the currently marketed statins |**22**|.

As the following trials emphasize, albeit mostly in rather short-term studies, there is an increasing body of evidence for several different statins that they can be used safely in renal and cardiac allograft recipients. The reductions in total and LDL cholesterol are comparable with those in the general population. The tendency still is to use low dosages because of previous reports of rhabdomyolysis in a small number of patients on statin-cyclosporin combinations. The largest reduction in cholesterol is usually obtained with the initial dose of statins, with subsequent doubling of the dose producing only relatively small additional benefits—usually estimated at about 6% on average. It seems prudent in routine practice to continue to use low-dose statins in transplant recipients for the time being until more evidence is available about the longer-term use of higher dosages in carefully supervised studies. A 20% reduction in LDL cholesterol represents a very significant reduction in risk assuming that one can extrapolate to this patient group from the general population.

A double-blind placebo-controlled trial of simvastatin for the treatment of dyslipidemia in renal allograft recipients.

F Lepre, R Rigby, C Hawley, *et al. Clin Transplant* 1999; **13**:520–5.

B**ACKGROUND. Renal failure patients can now regain near-normal health after renal transplantation, but premature cardiovascular disease is a major problem. Dyslipidaemia may be an important contributor to this but the use of lipid-lowering agents in renal allograft recipients has been limited by potential interactions with the widely used immunosuppressive agent, cyclosporin. This double-blind, placebo-controlled study investigated the efficacy and safety of simvastatin (5 mg daily for 6 weeks then 10 mg for 6 weeks) in patients (*n*=32) taking either cyclosporin or azothioprine after renal transplantation. The 12-week double-blind phase, was followed by a 36-week open phase when all subjects were treated with 10 mg simvastatin/day for 36 weeks.**

I**NTERPRETATION.** Low-dose simvastatin is effective and well-tolerated in the treatment of dyslipidaemia in renal allograft recipients. Compared with placebo, 5 mg simvastatin significantly decreased total cholesterol, LDL cholesterol and apoB. Increasing simvastatin to 10 mg daily did not lead to further significant changes but HDL cholesterol and apoA1

increased only on 10 mg simvastatin/day. The benefits of simvastatin were maintained to the end of the study. Three patients withdrew, all from the simvastatin/cyclosporin group, two with musculoskeletal pain and one with abdominal pain.

Comment

In this study the authors found 5 mg/day as effective as 10 mg/day in lowering total (20%) and LDL cholesterol (29%) in patients with renal grafts. Only the higher dose had a significant effect in raising HDL cholesterol (9%), however. Including the randomized and open-label phases subjects in the simvastatin group were followed for almost a year. Two subjects on active treatment withdrew because of musculoskeletal pain but in neither was the serum CK raised.

Role of statins in the management of dyslipidemia after cardiac transplant: randomized controlled trial comparing the efficacy and the safety of atorvastatin with pravastatin.
G Magnani, V Carinci, C Magelli, *et al. J Heart Lung Transplant* 2000; **19**: 710–15.

B**ACKGROUND. Cardiac transplant patients are at increased risk of dyslipidaemia, a known pathogenetic factor in chronic rejection. This study compared the efficacy and the safety of treatment with atorvastatin and pravastatin in dyslipidaemic transplant patients. Transplant patients (*n*=39) were randomized to receive a 4-month cycle of therapy with atorvastatin or pravastatin, in a cross-over sequence.**

I**NTERPRETATION.** Atorvastatin was significantly more effective than pravastatin in reducing total cholesterol, LDL cholesterol and TGs, at lower doses and with comparable tolerability and safety.

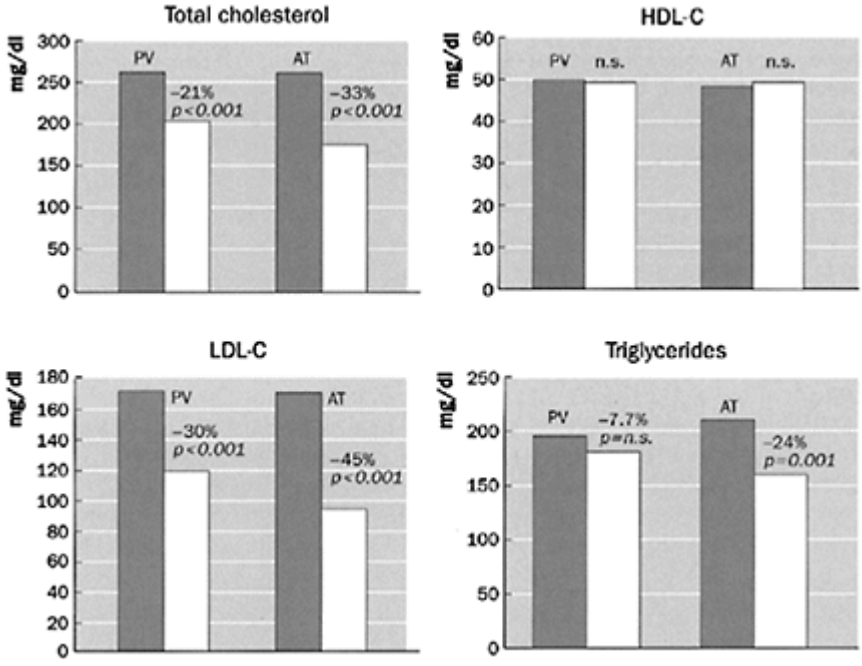


Fig. 3.3 Changes in lipid and lipoprotein variables in patients after cardiac transplantation taking pravastatin (PV), 20 mg/day, and atorvastatin (AT), 10 mg/day.
Source: Magnani *et al.* (2000).

Comment

This cross-over study demonstrates greater efficacy of atorvastatin (10–20 mg/day) than pravastatin (20–40 mg/day) in lowering total and LDL cholesterol and serum TGs in 39 heart transplant recipients who were also receiving cyclosporin, azathioprine and prednisolone. An increase of serum CK to more than twice the upper limit of normal, even in the absence of muscle symptoms was a criterion for withdrawal and was seen in only two patients on each drug. No cases of myositis or serious toxicity were seen. The results are encouraging but this was a relatively short-term study with the subjects taking each statin for only 4 months.

Beneficial effects of atorvastatin in the treatment of hyperlipidemia after renal transplantation.
D Demetriou, A Shabpar, G Bohmig, *et al. Wien Klin Wochenschr* 2000;

212:358–61.

B**ACKGROUND. Despite the availability of various lipid-lowering drugs, treatment of hyperlipidaemia, an important risk factor for morbidity and mortality after organ transplantation, remains a therapeutic challenge. The safety and efficacy of atorvastatin (10 mg/day) were studied for 3 months in 24 renal transplant patients whose serum lipids were insufficiently controlled by diet and treatment with other lipid-lowering drugs.**

I**NTERPRETATION.** Low-dose atorvastatin is effective and appears safe in the treatment of post-transplant hyperlipidaemia. Its long-term effects should be investigated in larger and more prolonged prospective trials. In the 18 patients who completed the study, low-dose atorvastatin therapy led to a significant reduction in total cholesterol and LDL cholesterol and a modest reduction in serum TG levels.

Table 3.1 Serum lipid profiles at baseline and 1 and 3 months after conversion to atorvastatin

	Baseline	**Month 1**	**Month 3**
Total cholesterol	304.6±13.2	262.7±22.9	247.6±12.0*
LDL cholesterol	191.9±9.0	135.2±11.2**	141.8±14.7**
HDL cholesterol	43.2±1.5	43.7±2.1	45.8±2.7
Serum triglycerides	389.4±36.0	360.9±43.5	358.1±50.3
Serum creatinine	1.9±0.2	1.9±0.2	2.0±0.2
CPK	57.3±7.7	58.1±12.9	50.2±7.4

* P=0.007, ** P<0.0001
Source: Demetriou *et al.* (2000).

Comment

The reductions in total (19%) and LDL cholesterol (26%) seen with low-dose atorvastatin in this study may be underestimates as the patients were switched, without a washout period, from a variety of other lipid-lowering medications, to which they had made an inadequate response. Although serum TGs were generally raised at baseline (mean 4.4 mmol/l) they were not significantly reduced by atorvastatin. The drug was well tolerated and there were said to be no episodes of myositis or significant increases in serum CK. One patient complained of myalgia.

This again is a short-term study (3 months) and longer studies of the safety and efficacy of atorvastatin in transplant recipients are required. If confirmed to be safe then the use of higher doses might be carefully explored.

Disorders of high-density lipoprotein metabolism

Low concentrations of HDL cholesterol have been reported to be associated with a high risk of CHD for about 50 years and the relationship has been extensively investigated over the last quarter of a century |**23**|. It has not been established beyond doubt that this is a causal relationship, however, to the extent that a wealth of pathophysiological studies and the cholesterol-lowering end-point trials are regarded as having established the causal role of LDL in atherogenesis. Both statins and fibrates have complex effects, which involve TG-rich lipoprotein and HDL metabolism as well as that of LDL. The recent Veterans Affairs HDL Intervention Trial (VA-HIT) using gemfibrozil reduced coronary events without any change in LDL cholesterol |**24**|. It is not clear, however, that the benefits were mediated through changes in HDL cholesterol as there were marked reductions in serum TG concentration. There are also likely to have been changes in LDL composition with fibrate treatment, away from small dense LDL to lighter less atherogenic particles, despite lack of any change in LDL cholesterol.

If there is, as many believe, a causal relationship between HDL metabolism and coronary disease, additional to that mediated through LDL, then there is also uncertainty about the mechanism(s). The traditional view has been that HDL is a key agent in reverse cholesterol transport |**25**,**26**|, acting as an acceptor for tissue, including arterial wall, cholesterol, which is returned to the liver for excretion. HDL may, however, have several antiatherogenic properties. One alternative, or additional, view is that HDL exerts its beneficial effects by protecting LDL from oxidation |**27**|.

Isolated low HDL cholesterol or primary hypoalphalipoproteinaemia (i.e. in the absence of hypertriglyceridaemia or hypercholesterolaemia) can be a high-risk situation, although is not the subject of formal recommendations in most national guidelines. Intervention to raise HDL cholesterol seems reasonable at least in those subjects with established vascular disease. Weight loss, exercise and avoidance of cigarette smoking, together with HRT in postmenopausal women all have the potential to raise HDL cholesterol. While some or all of these measures may be desirable in any event the effect may be small in many subjects with primary hypoalphalipoproteinaemia. Nicotinic acid (NA) can have substantial effects on HDL cholesterol, but is often poorly tolerated. Fibrates may also be helpful |**19**|. The following study investigates them in combination.

Gemfibrozil, nicotinic acid and combination therapy in patients with isolated hypoalphalipoproteinemia: a randomized, open-label, crossover study.

M J Zema. *J Am Coll Cardiol* 2000; **35**:640–6.

B**ACKGROUND. Isolated hypoalphalipoproteinaemia (low HDL cholesterol alone) accounts for a significant percentage of patients with**

premature atherosclerosis. To assess the effects of NA, gemfibrozil and combination therapy on their lipid profiles, 23 patients with clinically well-defined atherosclerosis and isolated hypoalphalipoproteinaemia were randomized to receive gemfibrozil, NA or combination therapy in an open cross-over study.

I**NTERPRETATION.** In the majority of patients with clinical atherosclerotic disease and isolated hypoalphalipoproteinaemia, pharmacological therapy to raise HDL cholesterol is not only feasible but is also effective with currently available agents, particularly when used in combination. In the 14 patients able to tolerate all forms of pharmacotherapy, HDL cholesterol increased while taking gemfibrozil, NA and both drugs combined. Statistically significant favourable alterations in LDL cholesterol, LDL/HDL cholesterol, non-HDL/HDL cholesterol, apoB and apoB/apoA1 were also observed.

Comment

It is estimated that some 4–19% of patients undergoing coronary angiography have hypoalphalipoproteinaemia as an isolated lipid abnormality. This can be familial and not necessarily associated with other factors commonly linked to low HDL cholesterol, such as hypertriglyceridaemia, obesity or cigarette smoking.

The patients in this study all had atherosclerotic disease and were current non-smokers. They had isolated hypoalphalipoproteinaemia (HDL cholesterol 0.89 mmol/l) without significant hypercholesterolaemia (total and LDL cholesterol 4.87 and 3.40 mmol/l) or hypertriglyceridaemia (1.28 mmol/l). Gemfibrozil and NA produced roughly additive effects on HDL cholesterol, which was increased by 45% on the combination. About three-quarters of this increase could be attributed to the NA. There were additional benefits to LDL cholesterol (2.33 mmol/l on the combination) and serum TGs (0.65 mmol/l).

Combined gemfibrozil and NA is clearly a useful combination in patients with isolated hypoalphalipoproteinaemia, but its use was limited by adverse effects, particularly of the NA, as is often the case. Of 23 patients originally selected only 17 tolerated NA as monotherapy in low dose (1.0–1.5 g/day) and a further three were withdrawn on the higher dose (2.0–3.0 g/day).

Table 3.2 Comparison of responses to gemfibrozil, nicotinic acid and combination pharmacotherapy in patients (*n*=14) with isolated hypoalphalipoproteinemia

	Baseline	**Gemfibrozil**	**Nicotinic acid**	**Combined therapy**
Level in mmol/litre (mg/dl)				

Total cholesterol	4.87±0.69 (189±27)	4.49±0.70 (174±27)*	4.33±0.74 (168±29)*	3.91±0.76 (152±29)*†‡
LDL-C	3.40±0.60 (132±23)	3.06±0.64 (119±25)*	2.67±0.64 (104±25)*†	2.33±0.72 (91±28) *†‡
HDL-C	0.89±0.17 (35±7)	1.02±0.18 (40±7)*	1.20±0.21 (47±8) *†	1.29±0.19 (50±8) *†
Triglycerides	1.28±0.43 (113±38)	0.87±0.45 (78±40) *	1.02±0.58 (91±52)	0.65±0.22 (58±20) *
Apo B	2.31±0.51 (90±20)	1.94±0.42 (75±16) †	1.69±0.51 (66±20)*†	1.51±0.54 (59±21) *†
Apo A1	2.97±0.44 (115±17)	3.14±0.36 (122±14)	3.38±0.36 (131±14)	3.58±0.56 (139±22)*†
Ratio				
LDL-C/HDL-C	3.9±0.9	3.1±0.9*	2.3±0.9*†	1.9±0.7*†‡
Non-HDL-C/HDL-C	4.6±1.2	3.5±1.1*	2.8±1.1*	2.1±0.8*†‡
Apo B/Apo A1	0.82±0.21	0.63±0.11	0.53±0.16*	0.43±0.13*†

* Significantly different from baseline. †Significantly different from gemfibrozil. ‡Significantly different from nicotinic acid. Data are presented as the mean value±SD. Apo=apolipoprotein; HDL-C=high density lipoprotein cholesterol; LDL-C=low density lipoprotein cholesterol; TC=total cholesterol.
Source: Zema (2000).

Tangier disease, adenosine triphosphate binding cassette transporter and reverse cholesterol transport

Work of the greatest importance published over the last 2–3 years has identified the genetic defect in Tangier disease (TD) and familial HDL deficiency (FHD) as well as shedding light on the initial stages of the reverse cholesterol transport process. These developments are well reviewed in the following paper.

In simplistic terms cholesterol efflux from cells is an important determinant of HDL concentration (see Clee *et al.* below) and this suggests that the association between high HDL levels and a low incidence of CHD is not because high HDL levels promote reverse cholesterol transport. Indeed the principal acceptors of tissue cholesterol are small lipid-poor complexes and free apolipoproteins, which contribute little to total HDL cholesterol. High concentrations of HDL cholesterol are more likely the result of active reverse cholesterol transport and a marker for it. If, as it seems, HDL also exerts anti-atherogenic effects by protecting LDL from oxidation, then the extent to which HDL cholesterol concentration reflects its antioxidant capability, determined largely by its paraoxonase-1 activity, will also be important in understanding the epidemiological relationship between HDL cholesterol and CHD |**27**|.

Tangier disease and ABCA1.

Comment

Tangier disease, named after an island in Chesapeake Bay, was first reported by Fredrickson *et al.* |**28**| 40 years ago. A young boy and his sister had large orange tonsils, stuffed with cholesteryl esters, and almost undetectable HDL. Less than 100 cases have since been described and in addition to the tonsils reticuloendothelial cells elsewhere are also engorged with cholesteryl esters. Tangier patients seem to be prone to premature vascular disease, although not to the same extent as FH homozygotes. They may be partially protected by their concomitant low levels of LDL. This is probably the result of a lack of cholesteryl esters, normally transferred to VLDL-LDL from HDL under the influence of cholesteryl ester transfer protein, and upregulation of LDL receptors as a result of impaired HDL-derived cholesterol to the liver.

The initial acceptors of tissue cholesterol are believed to be lipid-poor apolipoproteins, which with progressive lipidation and modification by LCAT give rise to spherical HDL particles. Tangier patients have normal apoAI, which is present in very low concentrations, and is rapidly catabolized when it cannot acquire cellular lipid. It and other apolipoproteins are unable to take up cholesterol and phospholipids from fibroblasts of patients with TD, the defect lying in the cells rather than the apolipoproteins. A similar but less severe defect is found in FHD.

The molecular defect in TD and FHD has been shown by several groups to lie in the gene for a member of the large group of ATP binding cassette transporters known as ABCA1 (Fig. 3.4). Mutations in other ABC genes are responsible for certain types of macular degeneration and retinitis pigmentosa, familial intra-hepatic cholestasis, Dubin-Johnson syndrome and cystic fibrosis. The gene products use energy derived from ATP to transfer their respective substrates across cell

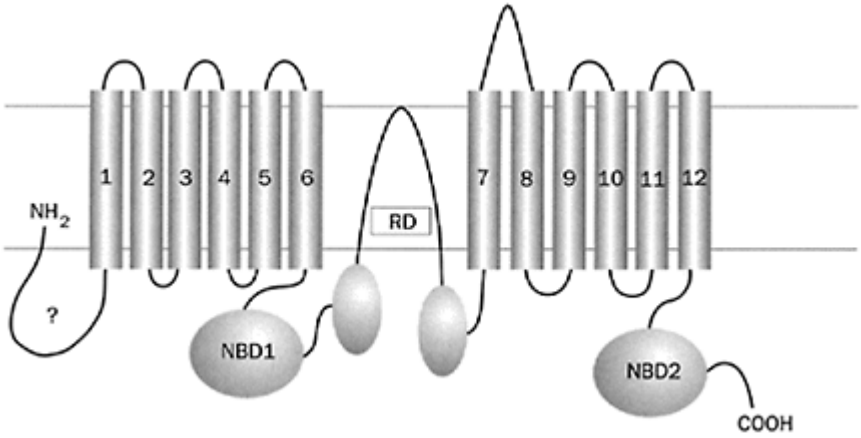


Fig. 3.4 The predicted topology of ABCA1. The protein consists of 2261 amino acids. There are two transmembrane sections, each encoding six transmembrane helices (1–6 and 7–12). After each transmembrane

section there are two nucleotide binding domains (NBD1 and NBD2), where ATP is bound and utilized as energy for substrate transport across the membrane. The central part of the protein is the regulatory domain (RD) which contains a highly hydrophobic segment that is thought to directly interact with the plasma membrane (shown as a looped structure in the membrane). The membrane orientation of the N-terminus is unknown. Source: Oram (2000).

membranes and between cellular compartments. Multiple mutations have been found in the ABCA1 to lead to TD and FHD. The latter are Tangier heterozygotes and are more mildly affected, with about half-normal levels of HDL cholesterol. Tangier patients can be either true homozygotes or compound heterozygotes. Other mutations and polymorphisms at the ABCA1 locus, and at other sites involved in the cholesterol efflux pathway, may lead to low HDL levels in the general population.

The findings in TD and FHD imply the ABCA1 pathway for lipid efflux from cells is rate-limiting in HDL formation. The heterozygotes with FHD do not have orange tonsils and lipid-laden macrophages, so it appears that one normal ABCA1 allele is sufficient to prevent these. Nevertheless, FHD patients are still prone to develop premature atherosclerosis.

The proposed cellular lipid excretion pathway is activated by the binding of lipid-free apolipoproteins to sites on the cell surface membrane, probably in the region of ABCA1. It involves transport of cholesterol and phospholipids (mainly lecithin) in vesicles from the Golgi apparatus to the cell surface, where transmembrane transport is facilitated by the ABCA1 protein (Fig. 3.5). Inactivation of the

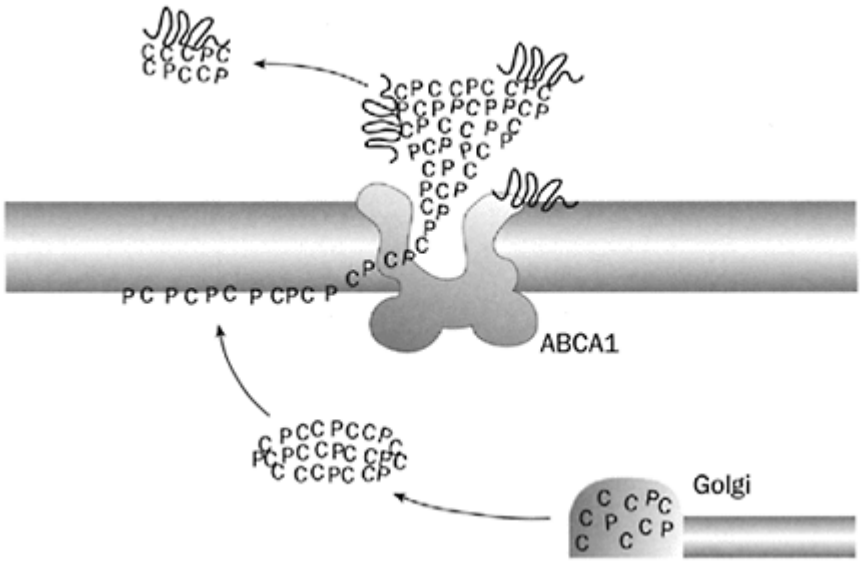


Fig. 3.5 Model for the ABCA1 lipid secretory pathway. Cholesterol (C) and phospholipids (P) are transported into the Golgi, where they are

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packaged into vesicles and transported to the plasma membrane. The vesicles and their cargo are incorporated into the inner leaflet of the plasma membrane. ABCA1 translocates this cargo into the aqueous chamber of the protein, forming structures that protrude from the cell and become solubilized by apolipoproteins (squiggles) for removal from the cell. Source: Oram (2000).

ABCA1 gene also increases absorption of dietary cholesterol, implying that it also serves to limit this by resecreting absorbed cholesterol back into the lumen of the intestine.

Finding pharmacological methods to upregulate the activity of ABCA1 produced by the normal allele in FHD patients looks a promising way to reduce the risk of CHD in a significant number of patients with familial hypoalphalipo-proteinaemia.

Decreased cellular cholesterol efflux is a common cause of familial hypoalphalipoproteinemia: role of the ABCA1 gene mutations.
S Mott, L Yu, M Marcil, *et al*. *Atherosclerosis* 2000; **152**:457–68.

B**ACKGROUND. HDLs are complex lipoprotein particles involved in reverse cholesterol transport and are negatively associated with the risk for coronary artery disease. In FHD cellular cholesterol efflux is abnormal. This study investigated cellular cholesterol efflux on skin fibroblasts from 15 probands with moderate to severe hypoalphalipoproteinaemia, including one with TD and family studies were done on eight probands (269 individuals) with familial hypoalphalipoproteinaemia (HDL cholesterol<5th centile, with no known cause of HDL deficiency). Four of the FHD patients and patients with TD have mutations at the ABC1 gene, demonstrating that FHD is a heterozygous form of TD.**

I**NTERPRETATION.** Familial hypoalphalipoproteinaemia syndromes are phenotypically heterogeneous; one form is associated with abnormal cellular cholesterol efflux caused by heterozygous mutations at the ABCA1 gene, which defines FHD while homozygous mutations or compound heterozygosity causes TD. Other forms are primary hypoalphalipoproteinaemia of unknown cause, while the remaining cases are associated with hypertriglyceridaemia with or without elevated apoB levels. Thus a cellular cholesterol defect is a relatively frequent cause of FHD and a mutation at the ABCA1 gene can be identified in half of these patients.

Comment

Low levels of HDL cholesterol may arise from a variety of mechanisms and are often, but not invariably associated with a predisposition to atherosclerotic disease. They are

commonly a feature of the metabolic syndrome in association with central obesity, hypertension, hypertriglyceridaemia and insulin resistance. HDL cholesterol may be increased by exercise, weight loss and alcohol and is reduced by cigarette smoking and high carbohydrate/low fat diets.

In this study the authors have confined their attention to familial cases of low HDL cholesterol. The probands had serum TGs below the 95th centile and no evidence of diabetes. Of 17 subjects studied from 14 families with unexplained hypoalphalipoproteinaemia, six (from four families) had decreased cholesterol efflux from their skin fibroblasts in culture and were deemed to have FHD. In these four families a mutation in the ABCA1 gene was identified, suggesting that this is a relatively common cause of familial hypoalphalipoproteinaemia. Three additional probands had cholesterol efflux reduced to about half control levels but were not assigned to FHD because of lack of family data and yet others had reduced HDL but no defect in cholesterol efflux.

If FHD patients (Tangier heterozygotes) are relatively common then why is classical TD so vanishingly rare? Presumably there is a spectrum of phenotypic severity for Tangier patients, just as there is in familial hypercholesterolaemia, which depends upon the nature of the underlying mutation so that a significant number of Tangier patients go unrecognized.

Age and residual cholesterol efflux affect HDL cholesterol levels and coronary artery disease in ABCA1 heterozygotes.
S M Clee, J J Kastelein, M van Dam, *et al. J Clin Invest* 2000; **106**: 1263–70.

B**ACKGROUND. Mutations in the ABCA1 gene are the underlying cause of TD and of a dominantly inherited form of familial hypoaiphalipoproteinaemia associated with reduced cholesterol efflux. 13 ABCA1 mutations have been identified in 11 families (five TD, six familial hypoalphalipoproteinaemia) and the phenotypes of 77 individuals heterozygous for mutations in the ABCA1 gene have been examined.**

I**NTERPRETATION.** ABCA1 heterozygotes have decreased HDL cholesterol and increased TGs. Age is an important modifier of the phenotype in heterozygotes, with a higher proportion of older heterozygotes having HDL cholesterol>5th percentile for age and sex compared with younger carriers. Levels of cholesterol efflux are highly correlated with HDL cholesterol levels. ABCA1 heterozygotes show a more than threefold increase in the frequency of coronary artery disease, with earlier onset than unaffected family members. Coronary artery disease is more frequent in heterozygotes with lower cholesterol efflux values. These data provide direct evidence that impaired cholesterol efflux, and consequently reverse cholesterol transport, is associated with reduced plasma HDL cholesterol levels and increased risk of

coronary artery disease.

Comment

This study makes a major contribution to our appreciation of phenotypic variability in patients with FHD (heterozygosity for mutations in the ABCA1 gene). This was possible because of the ability to identify such heterozygotes with certainty by defining the mutations they possessed. On average the heterozygotes had HDL cholesterol levels (0.74 mmol/l), which were 56% of those in unaffected family members. The difference between males and females was reduced (0.70 versus 0.76 mmol/l) compared with their normal relatives. There is, however, considerable phenotypic variability and 5% of heterozygotes had HDL cholesterol greater than the 20th centile for age and sex.

Heterozygotes had a 3.5-fold relative risk, albeit with a wide confidence interval (1.1–11.1), for prevalence of vascular disease compared with their relatives and its manifestations seemed more severe. By determining HDL cholesterol and *in vitro* cholesterol efflux from cultured fibroblasts for each of the heterozygotes the authors were able to show a phenomenally close correlation with more than 80% of the variability in HDL cholesterol determined by efflux. The nature of the ABCA1 mutation was not a major determinant of phenotypic severity in the heterozygotes, which is probably more dependent upon residual cholesterol efflux, determined by the normal ABCA1 allele. Other factors, which correlate with HDL cholesterol in the general population, such as body mass index, were also important in affected subjects.

As pointed out in an accompanying commentary |**29**| we are at the beginning of a new era in HDL research. Better understanding of the reverse cholesterol transport pathway should lead to new therapeutic opportunities.

Abetalipoproteinaemia and hypobetalipoproteinaemia

ABL, or the Bassen-Kornzweig syndrome, was first described 50 years ago and is characterized by deficiency of apoB-containing lipoproteins and consequently very low concentrations of serum and LDL cholesterol. The patients are protected against the development of atherosclerotic disease but may have ataxia from spinocerebellar degeneration, retinitis pigmentosa, steatorrhoea and abnormal red blood cells (acanthocytes) |**30**|. The neurological and ocular abnormalities can be greatly delayed or prevented if patients are adequately supplemented with vitamin E. Patients with ABL may also develop vitamin K deficiency but deficiency of vitamin D does not seem to have been reported.

ABL is inherited as an autosomal recessive disorder. Almost a decade ago it became apparent that ABL is the result of a variety of mutations, not at the apoB100 locus, but in the gene for microsomal TG transfer protein (MTP) |**31,32**|. MTP plays a crucial part in the lipidation of newly synthesized apoB to form lipoproteins. In the absence of functional MTP the apoB is rapidly degraded. Interference with MTP function would seem to be a promising approach for the reduction of atherosclerotic disease by reducing

secretion of TG-rich lipoproteins and their subsequent conversion to remnants and LDL. The difficulty has been to achieve this without inducing fatty liver and the potential for chronic liver disease.

FHBL is an autosomal codominant disorder. The homozygotes may be clinically indistinguishable from subjects with ABL. FHBL proves in many instances to be the result of mutations in the gene for apoB which lead in different families to truncations of apoB100 of varying length from the C-terminal end. These are designated according the length of the residual apoB expressed as a percentage of that of apoB100, in the same way that apoB48, normally produced in the intestine in humans, and a constituent of chylomicrons, is the N-terminal 48% of apoB100.

Deletions of varying lengths of the C-terminal end of apoB100 lead to the loss of functional domains. The minimum size to allow secretion of chylomicrons and VLDL seems to be apoB29 or apoB30. Molecules shorter than apoB70 are incapable of complexing with apo(a) to form Lp(a).

Not all patients with FHBL have truncated apoB and in several instances no link-age to the apoB gene can be demonstrated |**33**|. The precise defect in most of these cases remains to be determined. When this is achieved then a new nomenclature will be needed that clearly describes the underlying abnormality. At present FHBL appears to be a more heterogeneous condition than ABL.

Novel mutations in the microsomal triglyceride transfer protein gene causing abetalipoproteinemia.
K Ohashi, S Ishibashi, J Osuga, *et al. J Lipid Res* 2000; **41**:1199–204.

B**ACKGROUND. ABL is an inherited disease characterized by the virtual absence of apoB-containing lipoproteins from plasma. Few families have been screened for mutations in the MTP gene. To clarify the genetic basis of clinical diversity of ABL, mutations of the MTP gene were screened in four unrelated patients with ABL.**

I**NTERPRETATION.** The results indicated that defects of the MTP gene are the proximal cause of ABL. Three novel mutations were identified: a frameshift mutation caused by a single adenine deletion at position 1389 of the cDNA, and a mis-sense mutation, Asn780Tyr, each in homozygous forms; and a splice site mutation, 2218–2A→G, in a compound heterozygous form.

Comment

At the time of writing this paper 14 different mutations in the MTP gene, giving rise to ABL, had been described. It does not follow that the ABL phenotype is always due to mutations at this locus, however, as other proteins in the assembly or secretion of apoB-containing particles might theoretically be implicated. This study investigated four

further cases, two from Japan and two from the USA. Five mutations were identified, three of which were new. Three patients were true homozygotes and one a compound heterozygote. All the mutations were at the MTP locus. Two first-degree relatives of two different probands, who were presumed obligate heterozygotes, were found to have low levels of serum cholesterol (3.6 mmol/l) and apoB (46 and 61 mg/dl). This suggests that, at least in some families, ABL may be inherited as a codominant disorder, rather than as a true recessive.

A study of fatty liver disease and plasma lipoproteins in a kindred with familial hypobetalipoproteinemia due to a novel truncated form of apolipoprotein B (APO B-54.5).
P Tarugi, A Lonardo, G Ballarini, *et al. J Hepatol* 2000; **33**:361–70.

B**ACKGROUND. FHBL is a co-dominant disorder characterized by reduced plasma levels of LDLs. It can be caused by mutations in the gene encoding apoB-100, leading to the formation of truncated apoBs with a reduced capacity to export lipids from the hepatocytes as lipoprotein constituents. Case reports suggest the occurrence of liver disease in FHBL. The presence of fatty liver disease was investigated in 16 members of a FHBL kindred. The proband, a male non-obese heavy drinker with FHBL, had steatohepatitis with fibrosis. He was heterozygous for a novel non-sense mutation of apoB gene producing a truncated apoB of 2745 amino acids.**

I**NTERPRETATION.** In this kindred apoB-54.5 predisposes to fatty liver, but this may need additional factors to become clinically relevant. Seven other members of this kindred carried apoB-54.5. They were all hypolipidaemic, but their lipid levels showed much inter-individual variability, not accounted for by polymorphisms of genes involved in apoB metabolism. Four carriers (two heavy drinkers and two teetotallers), irrespective of their plasma lipid levels, had ultrasonographic evidence of fatty liver but the other four carriers did not.

Comment

Fatty liver is known to be a feature of both ABL and FHBL but has not been extensively investigated. In this study eight heterozygotes from a single kindred with a new mutation giving rise to FHBL were studied. These subjects make normal apoB100 from the normal allele and a truncated form, which is just over half the size of the normal protein (apoB54.5), from the mutated gene. Only four of the eight subjects had evidence of fatty liver and two of these consumed substantial amounts of alcohol. Fatty liver, and with it the possibility of significant liver disease culminating in cirrhosis |**34**|, is not inevitable in FHBL patients and may require additional factors to precipitate its development.

Hepatocytes secrete truncated apoB-containing lipoproteins at rates proportional to the length of the apoB |**35**|. The likelihood of developing fatty liver probably depends in part on the size of the mutated apoB.

References

1. Burke SW, Solomon AJ. Cardiac complications of end-stage renal disease. *Adv Renal Replacement Ther* 2000; **7**: 210–19.
2. Keane WF. The role of lipids in renal disease: future challenges. *Kidney Int* 2000; 57 (Suppl.75):S27–31.
3. Lechleitner M. Dyslipidaemia and renal disease—pathophysiology and lipid lowering therapy in patients with impaired renal function. *J Clin Basic Cardiol* 2000; **3**: 3–6.
4. Levin A, Foley RN. Cardiovascular disease in chronic renal insufftciency. *Am J Kidney Dis* 2000; **36**(Suppl. 3): S24–30.
5. Locatelli F, Marcelli D, Conte F, D'Amico M, Del Vecchio L, Limido A, Malberti F, Spotti D. Cardiovascular disease in chronic renal failure: the challenge continues. *Nephrol Dial Transplant* 2000; **15**(Suppl. 5): 69–80.
6. Wanner C. Importance of hyperlipidaemia and therapy in renal patients. *Nephrol Dial Transplant* 2000*;* **15**(Suppl. 5): 92–6.
7. Prichard S. Major and minor risk factors for cardiovascular disease in peritoneal dialysis. *Peritoneal Dialysis Int* 2000; **20**(Suppl. 2): S154–9.
8. Mattys E, Schurgers M, Lamberigts G. Effects of simvastatin treatment on the dyslipoproteinaemia in CAPD patients. *Atherosclerosis* 1991; **86**:183–92.
9. Li PK, Mak TW, Chiu K. Effect of lovastatin on serum lipid profile in the treatment of dyslipoproteinaemia in uraemic patients on continuous ambulatory peritoneal dialysis. *Aust N Z J Med* 1993; **23**: 252–7.
10. Warwick GL, Packard CJ. Lipoprotein metabolism in the nephrotic syndrome. *Nephrol Dial Transplant* 1993; **8**: 385–96.
11. Short CD, Durrington PN. Renal disorders. In: Betteridge DJ, Illingworth DR, Shepherd J (eds). *Lipoproteins in Health and Disease,* 1st edn. London: Arnold, 1999: 943–66.
12. Turnberg LA, Mahoney MP, Gleeson MH, Freeman CB, Gowenlock AH. Plasmapheresis and plasma exchange in the treatment of hyperlipaemia and xanthomatous neuropathy in patients with primary biliary cirrhosis. *Gut* 1972; **13**: 976–81.
13. Thompson GR. Plasma exchange for hypercholesterolaemia. *Lancet* 1981; **1**: 1246–8.
14. Seidel D. Non-pharmacological procedures: treatment of severe hypercholesterolaemia in patients with coronary heart disease by means of heparin-induced extracorporeal low-density lipoprotein plasmapheresis. In: Betteridge DJ, Illingworth DR, Shepherd J (eds). *Lipoproteins in Health and Disease,* 1st edn. London: Arnold, 1999:1255–66.
15. Wanner C, Quaschning T, Weingarnter K. Impact of dyslipidaemia in renal transplant recipients. *Curr Opin Urol* 2000; **10**: 77–80.
16. Markell MS. Transplantation. In: Betteridge DJ, Illingworth DR, Shepherd J (eds). *Lipoproteins in Health and Disease,* 1st edn. London: Arnold, 1999:1049–67.
17. Arnadottir M, Eriksson LO, Thysell H, Karkas JD. Plasma concentration profiles of simvastatin 3-hydroxy-3-methyl-glutaryl-coenzyme A reductase inhibitory activity in kidney transplant recipients with and without ciclosporin. *Nephron* 1993; **65**: 410–13.
18. Christians U, Jacobsen W, Floren LC. Metabolism and drug interactions of 3-hydroxy-3-methylglutaryl coenzyme A reductase inhibitors in transplant patients: are the statins mechanistically similar? *Pharmacol Ther* 1998; **80**: 1–34.

19. Rader DJ, Rosas S. Management of selected lipid abnormalities. *Med Clin North Am* 2000; **84**: 43–61.
20. Kobashigawa JA, Katznelson S, Laks H, Johnson JA, Yeatman L, Xiu MW, Chia D, Terasaki PI, Sabad A, Cogert GA, Trosian K, Hamilton MA. Effect of pravastatin on outcomes after cardiac transplantation. *N Engl J Med* 1995; **333**: 621–7.
21. Wenke K, Meiser B, Thiery J, Nagel D, Von Scheidt W, Steinbeck G, Seidel D, Reichart B. Simvastatin reduces graft vessel disease and mortality after heart transplantation. A four-year randomised trial. *Circulation* 1997; **96**:1398–402.
22. Jones P, Kafonek S, Laurora I. Comparative dose efficacy study of atorvastatin versus simvastatin, pravastatin, lovastatin and fluvastatin in patients with hypercholesterolemia (The CURVES study). *Am J Cardiol* 1998; **81**: 582–7.
23. Miller NE, Miller GJ. Plasma-high-density-lipoprotein concentration and development of ischaemic heart-disease. *Lancet* 1975; **i**: 16–19.
24. Rubins BH, Robins SJ, Collins D. Gemfibrozil for the secondary prevention of coronary heart disease in men with low levels of high-density lipoprotein cholesterol. *N Engl J Med* 1999; **341**: 410–18.
25. Glomset JA. The plasma lecithin: cholesterol acyltransferase reaction. *J Lipid Res* 1968; **9**: 155–67.
26. Glomset JA. High-density lipoproteins in human health and disease. *Adv Int Med* 1980; **25**: 91–116.
27. Durrington PN, Mackness B, Mackness MI. Paraoxonase and atherosclerosis. *Arterioscler Thromb Vasc Biol* 2001; **21**: 473–80.
28. Fredrickson DS, Altrocchi PH, Avioli LV, Goodman DS, Goodman HC. Tangier Disease—Combined Clinical Staff Conference at the National Institutes of Health. *Ann Intern Med* 1961;**55**:1016–31.
29. Tall AR, Wang N. Tangier disease as a test of the reverse cholesterol transport hypothesis. *J Clin Invest* 2000; **106**: 1205–7.
30. Malloy MJ, Kane JP. Disorders involving deficiencies of lipoproteins that contain B apolipoproteins. In: Betteridge DJ, Illingworth DR, Shepherd J (eds). *Lipoproteins in Health and Disease*, 1st edn. London: Arnold, 1999: 863–77.
31. Wetterau JR, Aggerbeck LP, Bouma M-E, Eisenberg C, Munck A, Hermier M, Schmitz J, Gay G, Rader DJ, Gregg RE. Absence of microsomal triglyceride transfer protein in individuals with abetalipoproteinemia. *Science* 1992; **258**: 999–1001.
32. Gordon DA. Recent advances in elucidating the role of the microsomal triglyceride transfer protein in apolipoprotein B lipoprotein assembly. *Curr Opin Lipidol* 1997; **8**: 131–7.
33. Elias N, Patterson BW, Schonfeld G. In vivo metabolism of apoB, apoA-I, and VLDL triglycerides in a form of hypobetalipoproteinemia not linked to the apoB gene. *Arterioscler Thromb Vasc Biol* 2000; **20**: 1309–15.
34. Caldwell SH, Oelsner DH, Iezzoni JC, Hespenheide EE, Battle EH, Driscoll CJ. Cryptogenic cirrhosis: clinical characterization and risk factors for underlying disease. *Hepatology* 1999; **29**: 664–9.
35. Parhofer KG, Barrett PHR, Aguilar-Salinas CA, Schonfeld G. Positive linear correlation between the length of truncated apolipoprotein B and its secretion rate: in vivo studies in human apo B-89, apo B-75, apo B-54.8 and apo B-31. *J Lipid Res* 1996; **37**: 844–52.

4
Alcohol, lipids and lipoproteins

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Introduction

There are excellent reviews of this topic by Taskinen *et al.* |**1**|, Savolainen and Kesaniemi |**2**| and Lieber's group, the latter having contributed so much to this field over many years |**3,4**|. Changes in the lipoprotein profile of subjects with established alcohol-induced cirrhosis of the liver resemble those in cirrhosis from other causes and are beyond the scope of this review.

Much of the interest in the influence of alcohol on lipoprotein metabolism relates to the apparent protective effect of moderate alcohol consumption against atherosclerotic events |**5,6**|. A recent meta-analysis suggests that consumption of 30 g alcohol (2–3 units) per day is associated with a reduction in coronary heart disease (CHD) risk of about 25% |**7**|. Although low to moderate alcohol consumption is associated with a favourable effect on cardiovascular disease, heavy intake is linked to an increase in haemorrhagic stroke and non-cardiovascular morbidity and mortality. The threshold at which overall benefit changes to hazard is a matter of debate, but a recent study suggests that this may be at about 14 units/week in women |**8**|.

Increases in high-density lipoprotein (HDL) cholesterol and serum triglycerides (TGs), with a reduction in low-density lipoprotein (LDL) cholesterol, are the most frequent changes in routinely measured lipid variables in response to alcohol consumption |**9**|. They are usually rapidly reversible during abstinence. In a Japanese observational dose-response study |**10**| serum HDL cholesterol increased progressively over the whole range of alcohol consumption. TGs were significantly increased only in the heavier consumers (75 g alcohol per day or more). LDL cholesterol tended to be reduced in those taking more than 25 g alcohol per day. The precise changes in lipids and lipoproteins induced by alcohol probably depend not only upon the quantity but also on the pattern of alcohol consumption, the concomitant diet and the gender and genetic constitution of the individual |**11**|. It has been suggested that binge or episodic drinkers have less favourable lipoprotein profiles than regular consumers, but this has not been confirmed in a recent study |**12**|. Binge drinkers have also been reported to be more likely to experience progression of carotid intima-media thickness |**13**|.

The mechanisms of the putative benefit of alcohol on cardiovascular disease |**14**, **15**| is only briefly discussed here. It is suggested that about half of it may relate to lipoprotein changes, particularly in HDL |**7,16**|. A small additional benefit probably relates to a reduction in LDL but this is largely offset by an increase in systolic blood pressure |**16**|.

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Effects on coagulation factors may also play a part |**7,17–19**|, although these are not apparent at low levels of consumption in some individual studies. Plasminogen activator inhibitor-1 activity has been found to be increased |**20,21**|. Other possible relevant mechanisms include the ability of alcohol to inhibit smooth muscle cell proliferation 221 and to improve insulin sensitivity |**23**|.

There has been much interest in the hypothesis that alcohol reduces coronary artery disease events by protecting lipoproteins against oxidation |**5**|. The fact that red wine is rich in antioxidants |**24,25**| has been suggested as an explanation for the 'French paradox', in which the French have lower rates of coronary disease than would be predicted from their saturated fat intake and serum lipids |**26,27**|. It remains to be established, however, that there is protective effect of alcoholic drinks, which is independent of alcohol itself |**28–30**|. Goldberg *et al.* |**31**| were unable to demonstrate that there was a major differential effect between red and white wine on lipoprotein concentrations, although TGs increased with red wine. Grape juice, either alone or enriched with trans-resveratrol (a candidate antioxidant), was without apparent effect. A failure to show a quantitative effect does not, however, exclude a qualitative effect on lipoproteins or elsewhere, which might influence the progression of atherosclerosis. The situation is complex as alcohol has also been shown to have pro-oxidant effects |**32,33**|.

The clinician concerned with the management of dyslipidaemia for the prevention of atherosclerotic vascular disease has a variety of opposing factors to consider in relation to the effects of alcohol. On the one hand moderate alcohol intake is found in epidemiological studies to confer some protection against vascular events. Hypertriglyceridaemia, which may be exacerbated by alcohol, is, however, associated with an increased propensity to vascular disease in almost all studies on univariate analysis. It ceases to be an independent risk factor after allowance for other risk factors, particularly HDL, in many studies, although not in all |**34**|. Moreover profound hypertriglyceridaemia, to which alcohol may be a major contributor, can be a cause of pancreatitis (the chylomicronaemia syndrome) |**35**|.

Triglyceride-rich lipoproteins

It is widely assumed that alcohol increases plasma TG concentrations. In practice these are often normal in light or moderate drinkers and even in alcoholics |**36,37**|. TGs may even rise during abstention in alcoholics |**36**|. At intakes of greater than 60–80 g alcohol per day there is a tendency to increased secretion of very low-density lipoprotein (VLDL) from the liver, which may be compensated in terms of plasma TGs by an increase in lipoprotein lipase (LPL) activity |**1**|. This is not observed in all studies. Alcohol added to cultured HepG2 cells promotes secretion of TG, cholesteryl esters and apolipoprotein (apo) B into the medium in the short term, but not on more chronic exposure |**38**|. The hypertriglyceridaemic effect of alcohol seems to be idiosyncratic and more readily seen in obese subjects |**39**|. It may be transient |**40**|, although in other subjects it can persist |**41**|. In terms of hypertriglyceridaemia some subjects are carbohydrate and others alcohol inducible. The two do not necessarily coexist.

The normal increase of TGs seen in late pregnancy is exaggerated in alcohol abusers

|**42**|. In the short term, some increase in fasting TGs is seen in subjects taking alcohol (0.75 g/kg) before their evening meal |**43**|. Postprandial lipaemia is greater in normal subjects who take alcohol with food |**44–47**|, although it was found to be unaffected by alcohol taken at other times |**48**|.

In normotriglyceridaemic subjects an acute alcohol load, in the absence of food, induces a modest rise in plasma TGs over a period of a few hours |**49–51**|, but has little effect in hypertriglyceridaemic individuals |**51**|. The ingestion of alcohol was accompanied by a fall in plasma non-esterified fatty acid concentrations over 4-6 h, which mirrored the rise in plasma acetate, the oxidation product of alcohol. The effects of alcohol in reducing plasma non-esterified fatty acid concentrations and fatty acid oxidation in the liver are similar in alcoholics and normal subjects |**52**|.

Chylomicronaemia syndrome

In some subjects alcohol can be the precipitating cause of massive hypertriglyceridaemia. They may exhibit the features of the chylomicronaemia syndrome |**35,53, 54**| with abdominal pain or frank pancreatitis and pseudohyponatraemia. The plasma sodium, and other plasma constituents, are diluted by the large volume of circulating fat, but the concentration of sodium in the plasma water is normal and the temptation to give hypertonic saline should be resisted. The serum amylase may seem to be normal, depending upon the method of assay, its true activity becoming apparent on progressive dilution of the samples before measurement |**55**–**57**|.

Development of chylomicronaemia in response to heavy alcohol consumption is idiosyncratic and probably most likely to occur in those who have a primary cause for more moderate hypertriglyceridaemia. Indeed in general those who develop the most severe secondary hyperlipidaemias usually also have a primary hyperlipidaemia. In those who are prepared to abstain from alcohol the TGs fall rapidly towards normal |**58**| (Fig. 4.1). A 2-week period of abstinence is a useful way of determining if alcohol is a major contributor to raised serum TGs.

In most subjects with chylomicronaemia secondary to alcohol the treatment is to abstain from or at least moderate alcohol consumption. In those who are unable to comply, consideration might be given to prescribing high-dose antioxidant treatment, which appears to be a promising treatment for prophylaxis against pancreatitis in patients with primary chylomicronaemia |**59**|. In the severely ill patient with pancreatitis in whom the TGs are not falling rapidly, consideration may be given to plasma exchange |**60,61**| and to ‘fat-free’ total parenteral nutrition |**62**|.

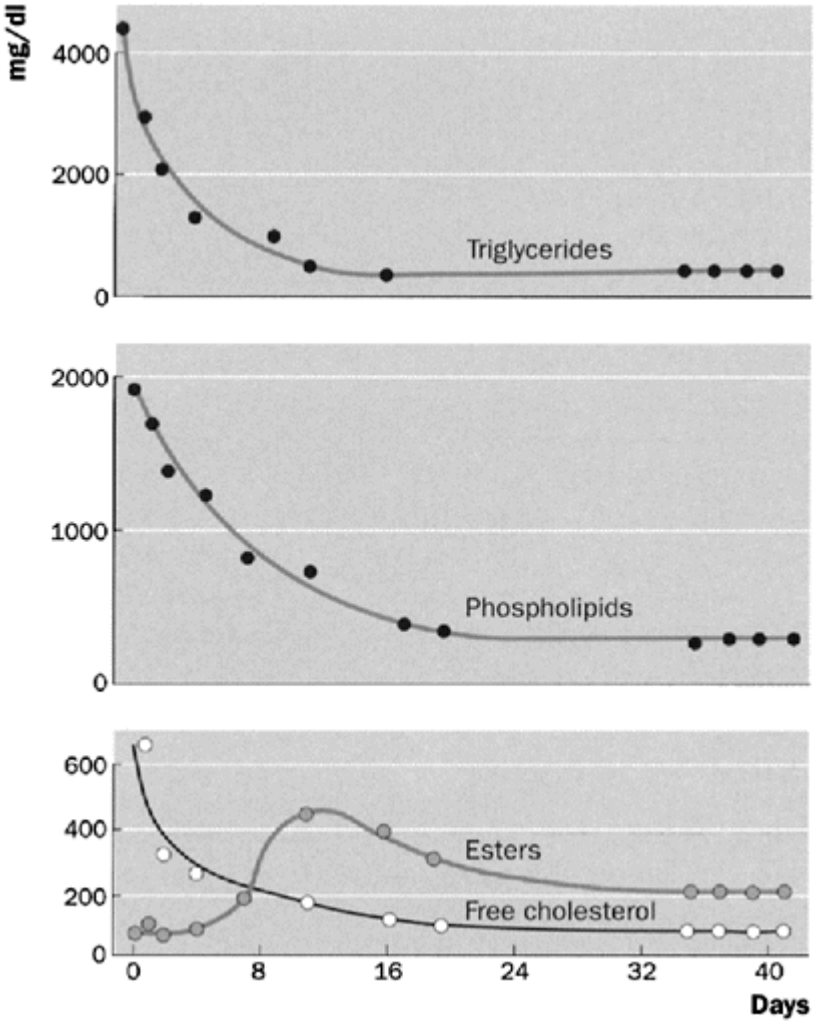

Fig. 4.1 Rapid resolution of alcohol-induced hyperlipidaemia upon abstinence. Source: Losowsky *et al.* (1963).

Low-density lipoproteins and lipoprotein(a)

LDL cholesterol tends to be reduced in moderate drinkers and in chronic alcoholics |**1,9,63,64**| and this may contribute to the reduction in vascular events associated with alcohol ingestion |**16**|. There is, however, evidence in heavy drinkers with raised HDL concentrations that LDL particles are smaller with reduced affinity for the LDL receptor |**64**|. These are regarded as unfavourable features with regard to atherogenesis |**65**|. Both synthesis and fractional catabolic rate of apoB have been shown to be increased |**66**|.

Suggested mechanisms include decreased conversion of VLDL to LDL and increased LDL clearance because of the development of acetaldehyde adducts of apoB-containing lipoproteins, which are immunogenic |**67**|. Acetaldehyde modification of LDL has been shown to increase its fractional catabolic rate in humans |**68**|.

Lipoprotein(a) (Lp(a)), which is found predominantly in the LDL density range, tends to be reduced by alcohol consumption with the lowest levels in heavy drinkers and cirrhotics |**69**|. Red wine appears more potent than white |**70**|. The changes can be rapid with a significant increase in Lp(a) within 1–4 weeks of abstention in alcoholics |**71,72**|. It has been suggested that the effect of alcohol in suppressing Lp(a) concentrations is transient |**73**|, but this is not a general finding. Some studies have failed to demonstrate an effect of moderate alcohol intake on Lp(a) in women |**74,75**|. Suggested mechanisms for the suppression of Lp(a) include effects of alcohol on sialylation of proteins |**76**| and on insulin-like growth factor binding protein-1 |**77**|.

High-density lipoproteins

An increase in HDL cholesterol is the most common and consistent lipoprotein change in moderate drinkers |**9,78**|. However, when alcohol is substituted isocalorically for carbohydrate in subjects on a low saturated fat diet no effect on HDL may be seen |**79,80**|. Isocaloric substitution of larger quantities of alcohol (75 g/day) for all three major macronutrients did, however, still lead to increases of HDL cholesterol |**81**|. Smoking even a small number of cigarettes may also offset the putative beneficial effects of alcohol on HDL |**37**|. The change in HDL cholesterol seems to be more marked in women than in men at low levels of consumption |**82**|. The increased susceptibility of women to the effects of alcohol may relate to a lower activity of alcohol dehydrogenase in gastric mucosa, increasing its bioavailablity, rather than to the usually cited smaller volume of distribution |**83**|.

It has been proposed that HDL cholesterol concentrations, particularly in combination with γ-glutamyl transpeptidase estimations, may be useful in screening for alcohol abuse and recidivism |**84**|. Normally, there is an inverse relationship between plasma TG and HDL cholesterol concentrations. When both are elevated this should alert one to the possibility of excess alcohol consumption.

A retrospective study suggested that alcohol exerted effects on HDL that were additional to those conferred by exercise |**85**| and short-term changes in alcohol consumption were reflected in both HDL2 and HDL3 subfractions in runners |**86**|. This contrasts with previous work from the same group in which the changes in HDL were limited to sedentary individuals |**87**|. In a recent meta-analysis the effects of alcohol on HDL were weaker in the physically fit than in the sedentary |**7**|.

Although clinically HDL is usually quantitated by measuring its cholesterol component after precipitation of the other (apoB-containing) lipoproteins, the other major components of HDL, namely phospholipids, apoA-I and particularly apoA-II, are also increased. There have been conflicting reports as to whether the predominant change is in HDL2 or HDL3. In addition to the separation of HDL into subclasses based upon density (HDL2 and HDL3) they can also be separated according to the apolipoproteins they

contain. Some particles contain only apoA-I (known as LpAI) and others both apoA-I and apoA-II (LpAI:AII). The effect of 50 g of alcohol daily as red wine is predominantly to increase LpAI:AII, by virtue of a decrease in the catabolic rate of apoA-II, with little effect on the concentration of LpAI or the metabolism of apoA-I |**88**|. Previous work has shown a variable effect on LpAI concentration |**73,89,90**|. There is general agreement that the increase in apoA-II concentration exceeds that in apoA-I |**3,4**|. The dominant effect of alcohol on apoA-II tends to accord with the work of those who have observed a more pronounced effect on HDL3 than HDL2, as LpAI:AII makes up the greater part of HDL3, whereas the majority of HDL2 comprises LpAI.

Although there are favourable changes seen in HDL metabolism with heavy (100 g/day) alcohol ingestion, it has been suggested that the changes seen with moderate consumption are too small to exert a beneficial effect on vascular risk |**36**|. This is not the general view. Meta-analysis suggests that on average consumption of 30 g alcohol per day raises HDL cholesterol by about 8% |**7**|. No difference could be discerned between the effects of wine, beer or spirits.

Changes in HDL metabolism can be observed within hours of a meal, depending on whether or not it is accompanied by alcohol. Forty grams of alcohol with an evening meal attenuate the postprandial fall in HDL TGs. Levels of HDL phospholipids and HDL cholesterol are higher than control measurements some 9–13 h after the meal, although all the differences are small |**47**|. ApoA-II was also higher the following morning, although this was not seen during a previous study by the same group |**46**|. It seems likely that the attenuated fall in HDL TGs after a meal with alcohol is the result of greater postprandial lipaemia, which promotes exchange of TGs for cholesteryl esters between TG-rich lipoproteins and HDL, under the influence of cholesteryl ester transfer protein (CETP).

Mechanisms for the effects of alcohol on high-density lipoproteins

An increase in secretion of hepatic HDL probably contributes to the increase in HDL cholesterol that usually accompanies regular alcohol intake |**3**|. ApoA-I concentration is increased in alcoholic patients with normal livers or steatosis but falls in those with more advanced liver damage |**91–93**| and apoA-I synthesis has been found to be increased by alcohol in cultured hepatocytes |**94**|. Malmendier and Delcroix |**66**| found apoA-I turnover is stimulated with increases in both its synthesis and fractional catabolic rate. Patients with steatosis who have raised apoA-I levels also have increased hepatic apoA-I mRNA levels but the correlation is lost in patients with fibrosis who have not yet reached end-stage disease, suggesting that in these patients levels are regulated at a post-transcriptional stage |**95**|. Exposure of cultured HepG2 cells to alcohol stimulated the accumulation of apoA-I in the medium on chronic exposure, and also acutely but then only at high, possibly non-physiological, concentrations |**38**|.

Modulation of the activity of a variety of enzymes and lipid transfer factors might be involved in the alterations in HDL metabolism induced by alcohol. Increases in LPL and lecithin: cholesterol acyltransferase (LCAT) activity and reduction in CETP activity could all in principle contribute to the elevation of HDL cholesterol concentration.

Changes in the activity of hepatic lipase (HL) have the potential to influence HDL lipid clearance by the liver and also the recycling of HDL2 back to HDL3.

Increased LPL activity is a common, although not universal finding, in alcohol drinkers. Not only does it tend to compensate for increased hepatic VLDL secretion and prevent an increase in serum TGs, but it contributes to the mass of circulating HDL by promoting the transfer of surface components from TG-rich lipoproteins to HDL during lipolysis. With moderate short-term alcohol consumption LPL activity has been reported to be increased |**96**| or unchanged |**86**|. Acute heavy consumption of alcohol has also been reported to increase |**1**| or leave LPL activity unchanged |**39**|. With chronic heavy consumption LPL activity is usually increased and falls rapidly upon abstention |**1,63,72,97**|. Paradoxically, Chait *et al.* |**98**| could find no change in TG clearance as judged by the intravenous fat tolerance test when heavy consumers abstained.

The data for HL are confusing. In the short term it has been reported not to change |**86,96**|, although a single 40 g dose produced a transient marked decrease |**99**|. Heavier short-term consumption has also been found to reduce HL activity |**1**| or lead to no change |**39**|. Abstention after heavy chronic consumption was followed by either a fall in HL activity |**63,72**| or no change |**97**|.

LCAT and CETP are key proteins in the process of reverse cholesterol transport. LCAT esterifies tissue cholesterol on HDL. This may then be transferred to TG-rich lipoproteins under the influence of CETP and thence to LDL, a significant proportion of which is removed through the apoB/E receptor in the liver. Moderate alcohol consumption seems to produce little change in LCAT activity |**46,96,100**|. Neither was any change reported during abstinence after heavy chronic consumption |**97**|.

With moderate consumption CETP activity has been found to be unchanged |**96,100**| in normal subjects, although lipid transfer may be increased acutely in the postprandial phase probably as a result of changes in the lipoprotein substrates |**46**|. In one series 30 g alcohol daily for 14 days depressed CETP activity in normal subjects but not in the obese |**101**|. Several groups have found alcohol consumption, especially if heavy to depress cholesteryl ester transfer. Not only may CETP activity and concentration be reduced by alcohol |**64,72,102,103**|, but the net mass transfer of cholesteryl esters may actually be reversed (i.e. from the apoB-containing lipoproteins, VLDL and LDL, to HDL), more commonly than is seen in normal subjects |**104**|. In fact the net mass transfer of cholesteryl esters was not correlated with CETP activity and seems to depend more on the concentration and composition of the substrate lipoproteins |**104,105**|. Specifically increased VLDL concentrations promote net transfer of cholesteryl esters from HDL to VLDL-LDL, whereas increasing HDL concentrations inhibit this process and may even lead to its reversal. The question arises, therefore, as to what extent the changes observed in HDL cholesterol concentration in heavy drinkers are the result or the cause of changes in cholesteryl ester transfer.

Genetic factors are also likely to be important. A polymorphism of the CETP gene (B2) is associated with lower activity of CETP in plasma and higher HDL cholesterol levels, although these two associations do not seem to be linked. The effect of the polymorphism on HDL cholesterol was seen only in heavy drinkers in whom there was a marked reduction in risk of myocardial infarction |**106**|. This study confirmed previous observations that CETP activity and HDL cholesterol are not correlated.

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Phospholipid transfer protein promotes the transfer of phospholipids from the surface of TG-rich lipoproteins to HDL. Chronic abusers have been shown to have both increased phospholipid transfer protein activity and net mass transfer of phospholipids |**104**|.

References

1. Taskinen M-R, Nikkila EA, Valimaki M, Sane T, Kuusi T, Kesaniemi YA, Ylikahri R. Alcohol-induced changes in serum lipoproteins and in their metabolism. *Am Heart J* 1987; **113**: 458–64.

2. Savolainen MJ, Kesaniemi YA. Effects of alcohol on lipoproteins in relation to coronary heart disease. *Curr Opin Lipidol* 1995; **6**: 243–50.

3. Baraona E, Lieber CS. Alcohol and lipids. *Recent Dev Alcohol* 1998; **14**: 97–134.

4. Baraona E, Lieber CS. Alcohol. In: Betteridge DJ, Illingworth DR, Shepherd J (eds). *Lipoproteins in Health and Disease*, 1st edn. London: Arnold, 1999:1011–1036.

5. Goldberg DM, Hahn SE, Parkes JG. Beyond alcohol: beverage consumption and cardiovascular mortality. *Clin Chim Acta* 1995; **237**:155–87.

6. Criqui MH. Alcohol and coronary heart disease: consistent relationship and public health implications. *Clin Chim Acta* 1996; **246**: 51–7.

7. Rimm EB, Williams P, Fosher K, Criqui M, Stampfer MJ. Moderate alcohol intake and lower risk of coronary heart disease: meta-analysis of effects on lipids and haemostatic factors. *Br Med J* 1999; **319**: 1523–8.

8. Nanchahal K, Ashton WD, Wood DA. Alcohol consumption, metabolic cardiovascular risk factors and hypertension in women. *Int J Epidemiol* 2000; **29**: 57–64.

9. Castelli WP, Doyle JT, Gordon T, Hames CG, Hjortland MC, Hulley SB, Kagan A, Zukel WJ. Alcohol and blood lipids. The Cooperative Lipoprotein Phenotyping Study. *Lancet* 1977; **2**: 153–5.

10. Nagaya T, Yoshida H, Takahashi H, Matsuda Y, Kawai M. Dose-response relationships between drinking and serum tests in Japanese men aged 40–59 years. *Alcohol* 1999; **17**: 133–8.

11. Frohlich JJ. Effects of alcohol on plasma lipoprotein metabolism. *Clin Chim Acta* 1996; **246**: 39–49.

12. Rakic V, Puddey IB, Dimmitt SB, Burke V, Beilin LJ. A controlled trial of the effects of pattern of alcohol intake on serum lipid levels in regular drinkers. *Atherosclerosis* 1998; **137**: 243–52.

13. Kauhanen J, Kaplan GA, Goldberg DE, Salonen R, Salonen JT. Pattern of alcohol drinking and progression of atherosclerosis. *Arterioscler Thromb Vasc Biol* 1999; **19**: 3001–6.

14. Gaziano JM, Buring JE. Alcohol intake, lipids and risks of myocardial infarction. In: *Alcohol and Cardiovascular Diseases Novartis Foundation Symposium V 216.* Chichester: Wiley, 1998: 86–95.

15. Chick J. Alcohol, health and the heart: implications for clinicians. *Alcohol Alcohol* 1998; **33**: 576–91.

16. Langer RD, Criqui MH, Reed DM. Lipoproteins and blood pressure as biological pathways for effect of moderate alcohol consumption on coronary heart disease. *Circulation* 1992; **85**: 910–5.

17. Meade TW, Chakrabarti R, Haines AP, North WRS, Stirling Y. Characteristics affecting fibrinolytic activity and plasma fibrinogen concentrations. *Br Med J* 1979; **1**:153–6.

18. Mikhailidis DP, Jeremy JY, Barradas MA, Green N, Dandona P. Effect of ethanol on vascular prostacyclin (prostacyclin 12) synthesis, platelet aggregation, and platelet thromboxane release. *Br Med J* 1983; **287**:1495–8.
19. Seigneur M, Bonnet J, Dorian B, Benchimol D, Drouillet F, Gouverneur G, Larrue G, Crockett R, Boisseau M-R, Ribereau-Gayon P, Bricaud H. Effect of the consumption of alcohol, white wine, and red wine on platelet function and serum lipids. *J Appl Cardiol* 1990; **5**: 215–22.
20. Hendricks HF, Veenstra J, Velthuis-te Wierick EJ, Schaafsma G, Kluft C. Effect of moderate dose of alcohol with evening meal on fibrinolytic factors. *Br Med J* 1994; **308**: 1003–6.
21. Marques-Vidal P, Cambou JP, Nicaud V, Luc G, Evans A, Arveiler D, Bingham A, Cambien F. Cardiovascular risk factors and alcohol consumption in France and Northern Ireland. *Atherosclerosis* 1995; **115**: 225–32.
22. Locher R, Suter PM, Vetter W. Ethanol suppresses smooth muscle cell proliferation in the postprandial state: a new antiatherosclerotic mechanism of ethanol? *Am J Clin Nutr* 1998; **67**: 338–41.
23. Kiechl S, Willeit J, Poewe W, Egger G, Oberhollenzer F, Muggeo M, Bonora E. Insulin sensitivity and regular alcohol consumption: large prospective, cross sectional population study (Bruneck study). *Br Med J* 1996; **313**:1040–4.
24. Frankel EN, Kanner J, German JB, Parks E, Kinsella JE. Inhibition of oxidation of human low-density lipoprotein by phenolic substances in red wine. *Lancet* 1993; **341**: 454–7.
25. Frankel EN, Waterhouse AL, Kinsella JE. Inhibition of human LDL oxidation by resveratrol. *Lancet* 1993; **341**:1103–4.
26. Renaud S, Lorgeril M. Wine, alcohol, platelets, and the French paradox for coronary heart disease. *Lancet* 1992; **339**:1523–6.
27. Criqui MH, Ringel BL. Does diet or alcohol explain the French paradox? *Lancet* 1995; **344**: 1719–23.
28. Hennekens CH, Willett W, Rosner B, Cole DS, Mayrent SL. Effects of beer, wine, and liquor in coronary deaths. *JAMA* 1979; **242**: 1973–4.
29. Hegsted DM, Ausman LM. Diet, alcohol and coronary heart disease in men. *J Nutr* 1988; **118**:1184–9.
30. Rimm EB, Giovannucci EL, Willett WC, Colditz GA, Ascherio A, Rosner B, Stampfer MJ. Prospective study of alcohol consumption and risk of coronary heart disease in men. *Lancet* 1991; **338**: 464–8.
31. Goldberg DM, Garovic-Kocic V, Diamandis EP, Pace-Asciak CR. Wine: does the colour count? *Clin Chim Acta* 1996; **246**: 183–93.
32. Bjorneboe A, Bjorneboe GE. Antioxidant status and alcohol-related diseases. *Alcohol Alcohol* 1993; **28**: 111–6.
33. Croft KD, Puddey IB, Rakic V, Abu-Amsha R, Dimmitt SB, Beilin LJ. Oxidative susceptibility of low-density lipoproteins—influence of regular alcohol use. *Alcohol Clin Exp Res* 1995; **20**: 980–4.
34. Hokanson JE, Austin ME. Plasma triglyceride level is a risk factor for cardiovascular disease independent of high-density lipoprotein cholesterol level: a meta-analysis of population-based prospective studies. *J Cardiovasc Risk* 1996; **3**: 213–19.
35. Brunzell JD. Familial lipoprotein lipase deficiency and other causes of the chylomicronemia syndrome. In: Scriver CR, Beaudet AL, Sly WS, Valle D (eds). *The Metabolic and Molecular Bases of Inherited Disease*, 7th edn. Vol. II. New York: McGraw-Hill, 1995: 1913–32.

36. Lecomte E, Herbeth B, Paille F, Steinmetz J, Artur Y, Siest G. Changes in serum apolipoprotein and lipoprotein profile induced by chronic alcohol consumption and withdrawal: determinant effect on heart disease? *Clin Chem* 1996; **42**:1666–75.
37. Whitehead TP, Robinson D, Allaway SL. The effects of cigarette smoking and alcohol consumption on blood lipids: a dose-related study on men. *Ann Clin Biochem* 1996; **33**: 99–106.
38. Dashti N, Franklin FA, Abrahamson DR. Effect of ethanol on the synthesis and secretion of apoA-I- and apoB-containing lipoproteins in HepG2 cells. *J Lipid Res* 1996; **37**: 810–24.
39. Crouse JR, Grundy SM. Effects of alcohol on plasma lipoproteins and cholesterol and triglyceride metabolism in man. *J Lipid Res* 1984; **25**: 486–96.
40. Belfrage P, Berg B, Hagerstrand I, Nilsson-Ehle P, Tornquist H, Wiebe T. Alterations of lipid metabolism in healthy volunteers during long-term ethanol intake. *Eur J Clin Invest* 1977; **7**: 127–31.
41. Kudzma DJ, Schonfeld G. Alcoholic hyperlipidemia: induction by alcohol but not by carbohydrate. *J Lab Clin Med* 1971; **77**: 384–95.
42. Valimaki M, Halmesmaki E, Keso L, Ylikorkala O, Ylikahri R. Serum lipids and lipoproteins in alcoholic women during pregnancy. *Metabolism* 1990; **39**:486–93.
43. Barboriak JJ, Hogan WJ. Preprandial drinking and plasma lipids in man. *Atherosclerosis* 1976; **24**: 323–5.
44. Barboriak JJ, Meade RC. Enhancement of alimentary lipemia by preprandial alcohol. *Am J Med Sci* 1968; 255: 245–51.
45. Wilson DE, Schreibman PH, Brewster AC, Arky RA. The enhancement of alimentary lipemia by ethanol in man. *J Lab Clin Med* 1970; 75: 264–74.
46. Van Tol A, Groener JEM, Scheek LM, Van Gent T, Veenstra J, Van De Pol H, Hendriks HFJ, Schaafsma G. Induction of net mass lipid transfer reactions in plasma by wine consumption with dinner. *Eur J Clin Invest* 1995; **25**: 390–5.
47. Hendriks HFJ, Veenstra J, Van Tol A, Groener JEM, Schaafsma G. Moderate doses of alcoholic beverages with dinner and postprandial lipoprotein composition. *Alcohol Alcohol* 1998; **33**: 403–10.
48. Superko HR. Effects of acute and chronic alcohol consumption on postprandial lipemia in healthy normotriglyceridemic men. *Am J Cardiol* 1992; **69**: 701–4.
49. Avogaro P, Cazzolato G. Changes in the composition and physico-chemical characteristics of serum lipoproteins during ethanol-induced lipaemia in alcoholic subjects. *Metabolism* 1975; **24**: 1231–42.
50. Siler SQ, Neese RA, Parks EJ, Hellerstein MK. VLDL-triglyceride production after alcohol ingestion, studied using [2–13C1] *glycerol J Lipid Res* 1998; **39**: 2319–28.
51. Pownall HJ, Ballantyne CM, Kimball KT, Simpson SL, Yeshurun D, Gotto AM. Effect of moderate alcohol consumption on hypertriglyceridemia. *Arch Int Med* 1999; **159**: 981–7.
52. Hirsch S, Pia de la Maza M, Petermann M, Bunout D. Lipid turnover in alcoholics before and after an ethanol load. *Nutrition* 1998; **14**: 437–42.
53. Miller JP. The chylomicronaemia syndrome. In: Neil HAW, Rees A, Taylor C, eds. *Hyperlipidaemia in Childhood*, 1st edn. London: Royal College of Physicians, 1996: 17–26.
54. Bhatnagar D. Hypertriglyceridaemia. In: Betteridge DJ, Illingworth DR, Shepherd J (eds). *Lipoproteins in Health and Disease*, 1st edn. London: Arnold, 1999: 737–751.
55. Fallat RW, Vester JW, Glueck CJ. Suppression of amylase activity by hypertriglyceridemia. *JAMA* 1973; **225**: 1331–4.

56. Warshaw AL, Bellini CA, Lesser PB. Inhibition of serum and urine amylase activity in pancreatitis with hyperlipemia. *Ann Surg* 1975; **182**: 72–5.
57. Lesser PB, Warshaw AL. Diagnosis of pancreatitis masked by hyperlipemia. *Ann Intern Med* 1975; **82**: 795–8.
58. Losowsky MS, Jones DP, Davidson CS, Lieber CS. Studies of alcoholic hyperlipemia and its mechanism. *Am J Med* 1963; **35**: 794–803.
59. Heaney AP, Sharer N, Rameh B, Braganza JM, Durrington PN. Prevention of recurrent pancreatitis in familial lipoprotein lipase deficiency with high-dose antioxidant therapy. *J Clin Endocrinol Metab* 1999; **84**: 1203–5.
60. Kollef MH, McCormack MT, Caras WE, Reddy VVB, Bacon D. The fat overload syndrome: successful treatment with plasma exchange. *Ann Intern Med* 1990; **112**: 545–6.
61. Lennertz A, Parhofer KG, Samtleben W, Bosch T. Therapeutic plasma exchange in patients with chylomicronemia syndrome complicated by acute pancreatitis. *Ther Apheresis* 1999; **3**: 227–33.
62. Crook MA, Sankaralingam A. Total parenteral nutrition in the chylomicronemia syndrome and acute pancreatitis. *Nutrition* 1999; **15**: 299–301.
63. Taskinen M-R, Valimaki M, Nikkila EA, Kuusi T, Enholm C, Ylikahri R. High density lipoprotein subfractions and postheparin plasma lipases in alcoholic men before and after ethanol withdrawal. *Metabolism* 1982; **31**:1168–74.
64. Hirano K, Yamashita S, Sakai N, Hiraoka H, Ueyama Y, Funahashi T, Matsuzawa Y. Low-density lipoproteins in hyperalphalipoproteinemic heavy alcohol drinkers have reduced affinity for the low-density lipoprotein receptor. *Clin Biochem* 1992; **25**: 357–62.
65. Austin MA, Breslow JL, Hennekens CH, Buring JE, Willett WC, Krauss RM. Low-density lipoprotein subclass patterns and risk of myocardial infarction. *JAMA* 1988; **260**: 1917–21.
66. Malmendier CL, Delcroix C. Effect of alcohol intake on high and low density lipoprotein metabolism in healthy volunteers. *Clin Chim Acta* 1985; **152**: 281–8.
67. Wehr H, Rodo M, Lieber CS, Baraona E. Acetaldehyde adducts and autoantibodies against VLDL and LDL in alcoholics. *J Lipid Res* 1993; **34**: 1237–44.
68. Kesaniemi YA, Kervinen K, Miettinen TA. Acetaldehyde modification of low density lipoprotein accelerates its catabolism in man. *Eur J Clin Invest* 1987; **17**: 29–36.
69. Marth E, Cazzolato G, Bittolo Bon G, Avogaro P, Kostner GM. Serum concentrations of Lp(a) and other lipoprotein parameters in heavy alcohol consumers. *Ann Nutr Metab* 1982; **26**: 56–62.
70. Sharpe PC, McGrath LT, McLean E, Young IS, Archbold GP. Effect of red wine consumption on lipoprotein(a) and other risk factors for atherosclerosis. *Q J Med* 1995; **88**: 101–8.
71. Huang CM, Elin RJ, Ruddel M, Schmitz J, Linnoila M. The effect of alcohol withdrawal on serum concentrations of Lp(a), apolipoproteins A-I and B, and lipids. *Alcohol Clin Exp Res* 1992; **16**:895–8.
72. Valimaki M, Kahri J, Laitinen K, Lahdenpera S, Kuusi T, Enholm C, Jauhiajnen M, Bard JM, Fruchart J-C, Taskinen M-R. High density lipoprotein subfractions, apolipoprotein A-I containing lipoproteins, lipoprotein(a), and cholesterol ester transfer protein activity in alcoholic women before and after ethanol withdrawal. *Eur J Clin Invest* 1993; **23**: 406–17.
73. Valimaki M, Laitinen K, Ylikahri R. The effect of moderate alcohol intake on serum apolipoprotein A-I-containing lipoproteins and lipoprotein(a). *Metabolism* 1991; **40**:

1168–72.
74. Clevidence BA, Reichman ME, Judd JT, Muesing RA, Schatzkin A, Schaefer EJ, Li Z, Jenner J, Brown CC, Sunkin M. Effects of alcohol consumption on lipoproteins of pre-menopausal women. A controlled diet study. *Arterioscler Thromb Vasc Biol* 1995; **15**: 179–84.
75. Nago N, Kayaba K, Hiraoka J, Matsuo H, Goto T, Kario K, Tsutsumi A, Nakamura Y, Igarashi M. Lipoprotein(a) levels in the Japanese population: influence of age and sex, and correlation to atherosclerotic risk factors. The Jichi Medical School Cohort Study. *Am J Epidemiol* 1995; **141**: 815–21.
76. Delarue J, Husson M, Schellenberg F, Tichet JS, Couet C, Lamisse F. Serum lipoprotein(a) [Lp(a)] in alcoholic men: effect of withdrawal. *Alcohol* 1996; **13**: 309–14.
77. Paassilta M, Kervinen K, Linnaluoto M, Kesaniemi YA. Alcohol withdrawal-induced change in lipoprotein(a): association with the growth hormone/insulin-like growth factor-I (IGF-I) /IGF-binding protein-1 (IGFBP-1) axis. *Arterioscler Thromb Vasc Biol* 1998; **18**: 650–4.
78. Hulley SB, Gordon S. Alcohol and high-density lipoprotein cholesterol. Causal inference from diverse study designs. *Circulation* 1981; **64**(III): 57–63.
79. Glueck CJ, Hogg E, Allen C, Gartside PS. Effects of alcohol ingestion on lipids and lipoproteins in normal men: isocaloric metabolic studies. *Am J Clin Nutr* 1980; **33**:2287–93.
80. Rumpler WV, Clevidence BA, Muesing RA, Rhodes DG. Changes in women's plasma lipid and lipoprotein concentrations due to moderate consumption of alcohol are affected by dietary fat level. *J Nutr* 1999; **129**:1713–17.
81. Contaldo F, D'Arrigo E, Carandente V, Cortese C, Coltori A, Mancini M, Taskinen M-R, Nikkila EA. Short-term effects of moderate alcohol consumption on lipid metabolism and energy balance in normal men. *Metabolism* 1989; **38**:166–71.
82. Weidner G, Connor SL, Chesney MA, Burns JW, Connor WE, Matarazzo JD, Mendell NR. Sex differences in high density lipoprotein cholesterol among low-level alcohol consumers. *Circulation* 1991; **83**:176–80.
83. Frezza M, Di Padova C, Pozzato G, Terpin M, Baraona E, Lieber CS. High blood alcohol levels in women. The role of decreased gastric alcohol dehydrogenase activity and first-pass metabolism. *N Engl J Med* 1990; **322**:95–9.
84. Barboriak JJ, Jacobson GR, Cushman P, Herrington RE, Lipo RF, Daley ME, Anderson AJ. Chronic alcohol abuse and high density lipoprotein cholesterol. *Alcohol Clin Exp Res* 1980; **4**:346–9.
85. Willett WC, Hennekens CH, Siegel AJ, Adner MM, Castelli WP. Alcohol consumption and high density lipoprotein cholesterol in marathon runners. *N Engl J Med* 1980; **303**: 1159–61.
86. Hartung GH, Foreyt J, Reeves R, Crock LP, Patsch W, Patsch JR, Gotto AM. Effect of alcohol dose on plasma lipoprotein subfractions and lipolytic enzyme activity in active and inactive men. *Metabolism* 1990; **39**:81–6.
87. Hartung GH, Foreyt JP, Mitchell RE, Mitchell JG, Reeves RS, Gotto AM. Effect of alcohol intake on high-density lipoprotein cholesterol levels in runners and inactive men. *JAMA* 1983; **249**:747–50.
88. Gottrand F, Beghin L, Duhal N, Lacroix B, Bonte J-P, Fruchart J-C, Luc G. Moderate red wine consumption in healthy volunteers reduced plasma clearance of apolipoprotein AII. *Eur J Clin Invest* 1999; **29**:387–94.
89. Puchois P, Ghalim N, Zylberberg G, Fievet P, Demarquilly C, Fruchart J-C. Effect of

alcohol intake on human apolipoprotein A-I-containing lipoprotein subfractions. *Arch Intern Med* 1990; **150**:1638–41.

90. Branchi A, Rovellini A, Tomella C, Sciariada L, Torri A, Molgora M, Sommariva D. Association of alcohol consumption with HDL subpopulations defined by apolipoprotein A-I and apolipoprotein A-II content. *Eur J Clin Nutr* 1997; **51**:362–5.

91. Duhamel G, Nalpas B, Goldstein S, Laplaud PM, Berthelot P, Chapman MJ. Plasma lipoprotein and apolipoprotein profile in alcoholic patients with and without liver disease: on the relative roles of alcohol and liver injury. *Hepatology* 1984; **4**:577–85.

92. Poynard T, Abella A, Pignon J-P, Naveau S, Leluc R, Chaput J-C. Apolipoprotein AI and alcoholic liver disease. *Hepatology* 1986; **6**:1391–5.

93. Bedossa P, Poynard T, Abella A, Aubert A, Pignon J-P, Naveau S, Leluc R, Lemaigre G, Martin ED, Chaput J-C. Apolipoprotein AI is a serum and tissue marker of liver fibrosis in alcoholic patients. *Alcohol Clin Exp Res* 1989; **13**:829–33.

94. Okamoto Y, Fujimori Y, Nakano H, Tsujii T. Role of the liver in alcohol-induced alteration of high-density lipoprotein metabolism. *J Lab Clin Med* 1988; **111**:482–5.

95. Mathurin P, Vidaud D, Vidaud M, Bedossa P, Paradis V, Ratziu V, Chaput J-C, Poynard T. Quantification of apolipoprotein A-I and B messenger RNA in heavy drinkers according to liver disease. *Hepatology* 1996; **23**: 44–51.

96. Nishiwaki M, Ishikawa T, Ito T, Shige H, Tomiyasu K, Nakajima K, Kondo K, Hashimoto H, Saitoh K, Manabe M, Miyajima E, Nakamura H. Effects of alcohol on lipoprotein lipase, hepatic lipase, cholesteryl ester transfer protein, and lecithin: cholesterol acyltransferase in high-density lipoprotein cholesterol elevation. *Atherosclerosis* 1994; **111**:99–109.

97. Ekman R, Fex G, Johansson BG, Nilsson-Ehle P, Wadstein J. Changes in plasma high density lipoproteins and lipolytic enzymes after long-term, heavy ethanol consumption. *Scand J Clin Lab Invest* 1981; **41**: 709–15.

98. Chait A, Mancini M, February A, Lewis B. Clinical and metabolic study of alcoholic hyperlipidaemia. *Lancet* 1972; **2**: 62–4.

99. Goldberg CS, Tall AR, Krumholz S. Acute inhibition of hepatic lipase and increase in plasma lipoproteins after alcohol intake. *J Lipid Res* 1984; **25**: 714–20.

100. Ito T, Nishiwaki M, Ishikawa T, Nakamura H. CETP and LCAT activities are unrelated to smoking and moderate alcohol consumption in healthy normolipidemic men. *Jpn Circ J* 1995; **59**: 541–6.

101. Hagiage M, Marti C, Rigaud D, Senault C, Fumeron F, Apfelbaum M, Girard-Globa A. Effect of a moderate alcohol intake on the lipoproteins of normotriglyceridemic obese subjects compared with normoponderal controls. *Metabolism* 1992; **41**: 856–61.

102. Savolainen MJ, Hannuksela M, Seppanen S, Kervinen K, Kesaniemi YA. Increased high-density lipoprotein cholesterol concentration in alcoholics is related to low cholesteryl ester transfer protein activity. *Eur J Clin Invest* 1990; **20**: 593–9.

103. Hannuksela M, Marcel YL, Kesaniemi YA, Savolainen MJ. Reduction in the concentration and activity of plasma cholesteryl ester transfer protein by alcohol. *J Lipid Res* 1992; **33**: 737–44.

104. Liinamaa MJ, Hannuksela ML, Kesaniemi YA, Savolainen MJ. Altered transfer of cholesteryl esters and phospholipids in plasma from alcohol abusers. *Arterioscler Thromb Vasc Biol* 1997; **17**: 2940–7.

105. Liinamaa MJ, Kesaniemi YA, Savolainen MJ. Lipoprotein composition influences cholesteryl ester transfer in alcohol abusers. *Ann Med* 1998; **30**: 316–22.

106. Fumeron F, Betoulle D, Behague I, Ricard S, Poirier O, Jemaa R, Evans A, Arveiler D, Marques-Vidal P. Alcohol intake modulates the effect of a polymorphism of the

:
cholesteryl ester transfer protein gene on plasma high density lipoprotein and the risk of myocardial infarction. *J Clin Invest* 1995; **96**: 1664–71.

Part II
Lipid-lowering trials

Lipid-lowering trials

Introduction

The first two chapters in this section focus on the findings of large-scale intervention trials with lipid-lowering agents with clinical end-points and lipoprotein end-points. Of outstanding importance in the last year has been the increasing evidence of benefit from statins to lower low-density lipoprotein cholesterol and the emerging evidence that triglyceride-lowering will reduce coronary heart disease (CHD) risk as the result of new fibrate trials. Evidence that omega-3 fatty acids can decrease new CHD events in patients who have survived myocardial infarction has also been published. There are, however, yet more negative trials of antioxidant vitamins. There then follows a chapter, which includes recent publications on how clinical practice should be modified to take account of evidence from clinical trials and epidemiology. Reports on methods of prioritizing patients for primary prevention, screening and triglycerides as a CHD risk factor are among these. The final chapter contains publications on the effect of lipid-lowering drugs on vascular endothelial dysfunction, C reactive protein, fibrinogen and paraoxonase, which has recently been proposed as a possible protective factor against CHD. Some reports suggest that diet and alcohol may modify paraoxonase activity and others that statins may restore its activity to normal in hypercholesterolaemia. Finally, two papers documenting the unexpected effect to statins in decreasing the risk of bone fracture are reviewed.

5
Trials with clinical end-points

Introduction

During the last year there was reinforcement of the evidence that statins can prevent coronary heart disease (CHD) in secondary prevention |**1**| by the publication of the results of pravastatin treatment in patients presenting with unstable angina as opposed to acute myocardial infarction (MI) in the Long Term Intervention with Pravastatin in Ischaemic Disease Study. Evidence that statins may influence the outcome of cardiac transplantation was also strengthened by the publication of the results of Keogh *et al.*

In primary prevention, evidence that statin treatment can prevent CHD |**2**| took on an even more dramatic turn with the publication of the Air Force/Texas Coronary Atherosclerosis Prevention Study in which CHD incidence was decreased by using lovastatin in a population whose annual CHD risk was about 1%. In Britain, the National Service Framework is still only advocating the use of statin drugs in primary prevention at levels of risk of ≥3% |**3**| and in Europe recommendations are for a 2% CHD risk threshold |**4**|. A consortium of the British societies with an interest in preventing cardiovascular disease has, however, recommended a move towards 1.5% on the basis of the scientific evidence, including the Air Force/Texas Coronary Atherosclerosis Prevention Study findings |**5**|.

The evidence for the importance of measuring and treating serum triglycerides (TG) certainly in secondary prevention has also been considerably boosted this year by the publication of two fibrate drug trials. The first of these was the Veterans Affairs High-Density Lipoprotein Cholesterol Intervention Trial (VAHIT) in which patients with CHD whose high-density lipoprotein (HDL) was low and whose serum cholesterol was on average 4.5 mmol/l were randomized to receive gemfibrozil or placebo. There was a 22% reduction in new CHD events in those on active treatment, the most obvious other effect of which was to produce a 31% decrease in TGs and a 6% increase in HDL cholesterol. The study may provide some answers to the old chestnut about what to do with patients with CHD whose cholesterol is <5 mmol/l. In a study of bezafibrate versus placebo in Israel in MI survivors, the Bezafibrate Infarction Prevention (BIP) study, active treatment decreased new CHD event rates overall by only 7.3% (non-significant). However, in patients whose serum TGs exceeded 2.3 mmol/l the decrease was 39.5% (highly significant). The study may have been different from VAHIT because the average cholesterol in BIP was higher. BIP may therefore herald treatment of raised levels of TGs as well as cholesterol in treating secondary prevention.

Omega 3 fatty acids have also become more prominent in the last year with evidence from the Lyon Heart Study that plant omega 3 fatty acids can decrease new CHD events after MI and from the Gruppo Italiano per lo Studio della Supravvivenza nell'Infarto miocardico (GISSI) study that those from fish are similarly effective. Interestingly, in

neither study was the effect associated with any major decrease in TGs and in GISSI the reduction in event rate was largely due to a reduction in sudden death, suggesting the omega 3 fatty acids were decreasing fatal cardiac dysrhythmias.

While statins, fibrate and long-chain omega 3 fatty acids have risen in the firmament this year, the same is not true of antioxidant vitamins. Two major trials, GISSI and the Heart Outcomes Prevention Evaluation (HOPE) study produced negative results. While some have attempted to put a brave face on this it must be recognized that the evidence is telling us that antioxidant vitamins are neither the best way to prevent CHD nor perhaps are clinical trials of their effects on cardiovascular disease the best way to test the oxidant hypothesis of atherosclerosis. In another study Arrol *et al.*, for example, found that while antioxidant vitamins may protect low-density lipoprotein (LDL) against oxidation for a short while, they also have the potential adverse effect of increasing cholesteryl ester transfer |**6**|. Cholesteryl ester transfer protein (CETP) activity [which catalyses the movement of cholesteryl ester out of HDL into very low density lipoprotein (VLDL) and may also be associated with the generation of small dense LDL] has moved a step closer to influencing clinical practice. Inhibitors of CETP have been under development for some time and, although there are as yet no publications on their influence in humans, one of them has been shown by Okamoto *et al.* to retard diet-induced atheroma in rabbits. This study complements two earlier ones. In one of these antibodies to CETP were shown to have a similar effect |**6**| and the other anti-sense oligonucleotides to the CETP gene discouraged atherogenesis in the diet-induced rabbit model |**7**|.

While antioxidant vitamins have not been successful in preventing new CHD events, there was a report in 1999 by Heaney *et al.* suggesting that in patients with extreme hypertriglyceridaemia, they may in high doses prevent acute pancreatitis. This was a clinical report based on three patients with familial lipoprotein lipase deficiency who were experiencing frequent episodes of acute pancreatitis and confirmation is necessary. This can be a very difficult problem in lipid clinic practice and it deserves further attention.

Clinical impression has been varied as to whether the outcome of coronary artery bypass grafting is as successful in patients with familial hypercholesterolaemia as in other patients. The study by Kawasuji *et al.* appears to dispel this.

Effects of pravastatin in 3260 patients with unstable angina: results from the LIPID study.

A M Tonkin, D Colquhoun, J Emberson, *et al. Lancet* 2000; **356**:1871–5.

B**ACKGROUND. The Long-Term Intervention with Pravastatin in Ischaemic Disease (LIPID) study is a major trial of secondary prevention of CHD events that includes**

hospital admission with unstable angina (as well as MI) as a qualifying

event. In this substudy of LIPID, both the subsequent cardiovascular risks and the response to pravastatin were compared in patients with previous unstable angina (*n*=3260) or with previous MI (*n*=5754). Patients were randomized to 40 mg pravastatin daily or placebo over a mean of 6 years.

I**NTERPRETATION.** Patients who have survived acute MI or unstable angina have similar long-term prognoses with a high frequency of subsequent unstable angina, and both groups benefit similarly from pravastatin therapy. Pravastatin reduced the relative risk of mortality by 21% in the MI group and 26% in the unstable angina group.

Comment

This is an important study, first, because in many hospitals the intensity of management of patients presenting with acute cardiac ischaemic chest pain is greater if they are proved to have a definite MI compared with those labelled as unstable angina. The prognosis in this study was similar in the two groups and thus the urgency with which further investigations and measures to prevent further cardiac events are instituted should be the same. Cholesterol lowering is an important component of the prevention of future ischaemic events.

Efficacy and safety of pravastatin *vs* simvastatin after cardiac transplantation.

A Keogh, P Macdonald, A Kaan, *et al. J Heart Lung Transplant* 2000; **19**(6): 529–37.

B**ACKGROUND. Previous studies of cardiac transplant recipients have shown that pravastatin reduces 12-month rejection and mortality after cardiac transplantation and that simvastatin reduces 4-year mortality, LDL cholesterol levels and intimal thickening. In this 12-month open observational study, cardiac transplant recipients received pravastatin 40 mg or simvastatin 20 mg daily on an alternating basis from the time of transplantation.**

I**NTERPRETATION.** For safety and pharmacokinetic reasons, pravastatin should be considered the statin of choice after heart transplantation. Pravastatin was associated with a trend toward superior survival, attributable to fewer immunosuppression-related deaths. Survival at 12 months on an actuarial treatment basis was 97.6% for patients on pravastatin and 83.7% for those on simvastatin. One immunosuppression-related death occurred on pravastatin compared with seven on simvastatin. Rhabdomyolysis or myositis

occurred only in patients on simvastatin (13.3%). Both drugs resulted in similar lipid profiles.

Comment

This is a relatively small study, but it does indicate that there is a need to gather together a larger data set on the various statins and immunosuppressant regimens, particularly as larger doses of statins and newer statins are introduced into the transplant field. It is important that the favourable effects of some statin drugs are balanced against their potential to cause serious side-effects in transplant patients.

Primary prevention of acute coronary events with lovastatin in men and women with average cholesterol levels: results of AFCAPS/TexCAPS. Air Force/Texas Coronary Atherosclerosis Prevention Study.

J R Downs, M Clearfield, S Weis, *et al. JAMA* 1998; **279**:1615–22.

B**ACKGROUND. Although cholesterol-reducing treatment reduces fatal and non-fatal coronary disease in patients with CHD, it is not known whether the benefits of reducing LDL cholesterol in patients without CHD extend to those with average serum cholesterol levels, women and older persons. This randomized double-blind placebo-controlled trial compared lovastatin with placebo for prevention of the first acute major coronary event in men and women without clinically evident atherosclerotic cardiovascular disease with average total and LDL cholesterol levels and below-average HDL cholesterol levels. Both groups were on a diet low in saturated fat and cholesterol.**

I**NTERPRETATION.** Lovastatin reduces the risk of the first acute major coronary event in men and women with average total and LDL cholesterol levels and below-average HDL cholesterol levels. These findings support the inclusion of HDL cholesterol in risk factor assessment, confirm the benefit of LDL cholesterol reduction to a target goal and suggest that the National Cholesterol Education Program guidelines on pharmacological intervention should be reassessed.

Comment

In this successful trial the annual CHD event rate in the placebo group was only 1.07%. Compare this with the recommended threshold for intervention with statins to prevent CHD of 3% in the NHS Framework and 2% in the joint European Guidelines. In the USA

the National Cholesterol Education Program recommendations would, however, target many people at similar levels of risk to those in this trial. Of course, 3304 people had to receive statin therapy for 5.2 years to prevent 67 events, giving an NNT of 49. The trial has been criticised for this, but it is a much better NNT than for many other interventions including the treatment of high blood pressure.

Use of lipid lowering drugs for primary prevention of coronary heart disease: meta-analysis of randomised trials.
M Pignone, C Phillips, C Mulrow. *Br Med J* 2000; **321**:983–6.

B**ACKGROUND. The aim of this meta-analysis was to summarize the effect of primary prevention with lipid-lowering drugs on CHD events, CHD mortality and all-cause**

mortality. All randomized trials, lasting at least 1 year, of drug treatments for patients with no known CHD, cerebrovascular disease or peripheral vascular disease and that measured clinical end-points, including all-cause mortality, CHD mortality and non-fatal MIs were included.

I**NTERPRETATION.** Treatment with lipid-lowering drugs lasting 5–7 years reduced the odds of CHD events by 30% but not the odds of all-cause mortality in people with no

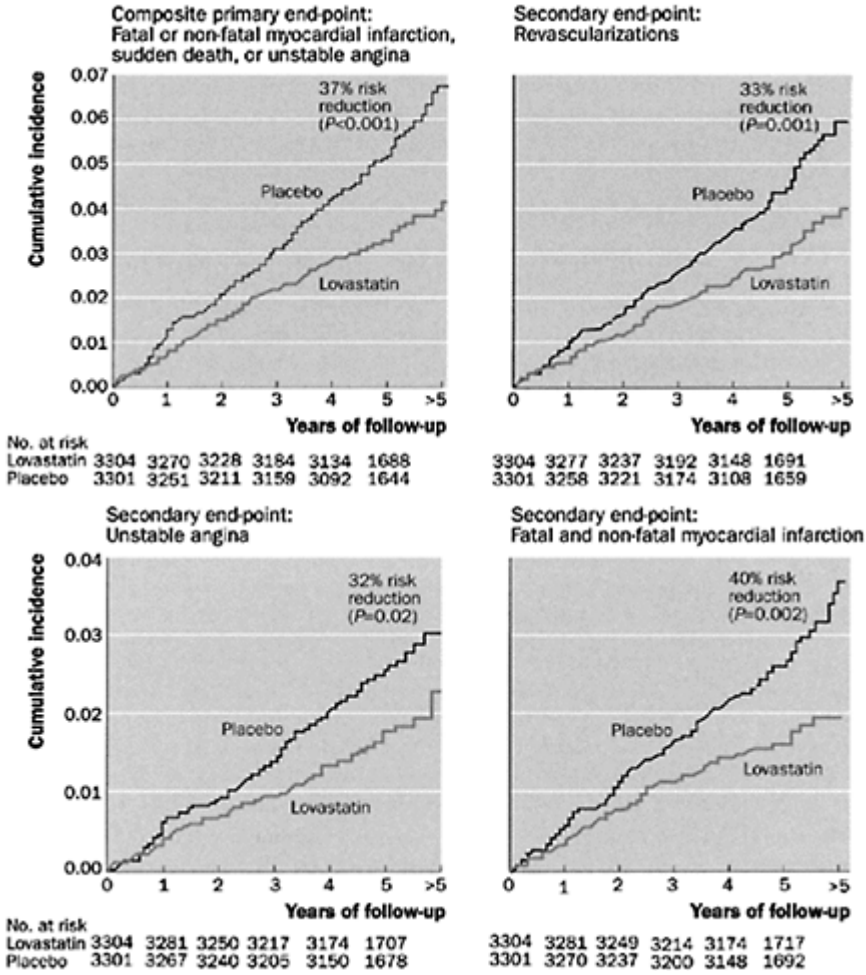

Fig. 5.1 Cumulative incidence of primary end-points (composite of fatal and non-fatal MI, sudden death and unstable angina) and secondary end-points (composite of fatal and non-fatal MI, unstable angina and coronary revascularizations) by treatment group.
Source: Downs *et al.* (1998).

known cardiovascular disease. When statin drugs were considered alone, no substantial differences in results were found.

Comment

This is a valuable study confirming that lowering cholesterol with statin drugs can decrease CHD events in primary prevention. The finding of a lack of effect on all-cause

mortality should be interpreted with care. For an intervention that decreases CHD mortality to have a significant impact on all-cause mortality, CHD mortality must comprise a large proportion of the overall death rate as it does in secondary prevention trials. Otherwise a much larger number of people must be studied than were included in this meta-analysis or followed for longer than in this meta-analysis.

Gemfibrozil for the secondary prevention of coronary heart disease in men with low levels of high-density lipoprotein cholesterol. Veterans Affairs High-Density Lipoprotein Cholesterol Intervention Trial Study Group.

H B Rubins, S J Robins, D Collins, *et al. N Engl J Med* 1999; 5 **341**: 410–18.

B**ACKGROUND. It is generally accepted that lowering elevated serum LDL cholesterol in patients with CHD is beneficial, but there are few data to guide decisions about therapy for patients whose primary lipid abnormality is a low HDL cholesterol. This double-blind trial aimed to compare gemfibrozil (1200 mg daily) with placebo in 2531 men with CHD. The primary outcome was non-fatal MI or death from coronary causes and the median follow up was 5.1 years.**

I**NTERPRETATION.** Gemfibrozil significantly reduced the risk of major cardiovascular events in patients with coronary disease whose primary lipid abnormality was low HDL cholesterol. The findings suggest that the rate of coronary events is reduced by raising HDL cholesterol levels and lowering levels of TGs without lowering LDL cholesterol levels.

Comment

This was a secondary CHD prevention trial in men whose serum HDL cholesterol was≤1 mmol/l and whose LDL cholesterol was≤3.6 mmol/l, the average serum cholesterol at entry was thus only 4.5 mmol/l. The mean serum TG concentration was 1.8 mmol/l so only a third or so of the men had hypertriglyceridaemia (≥2.3 mmol/l). Nonetheless, the effect of treatment with gemfibrozil was to decrease future CHD events (Fig. 5.2) to a similar extent to that achieved in statin trials conducted in patients with higher cholesterol levels. Because the men were selected for low HDL cholesterol this trial is often interpreted as suggesting that the rise in HDL (from 0.83 to 0.88 mmol/l) with gemfibrozil explains the benefit.

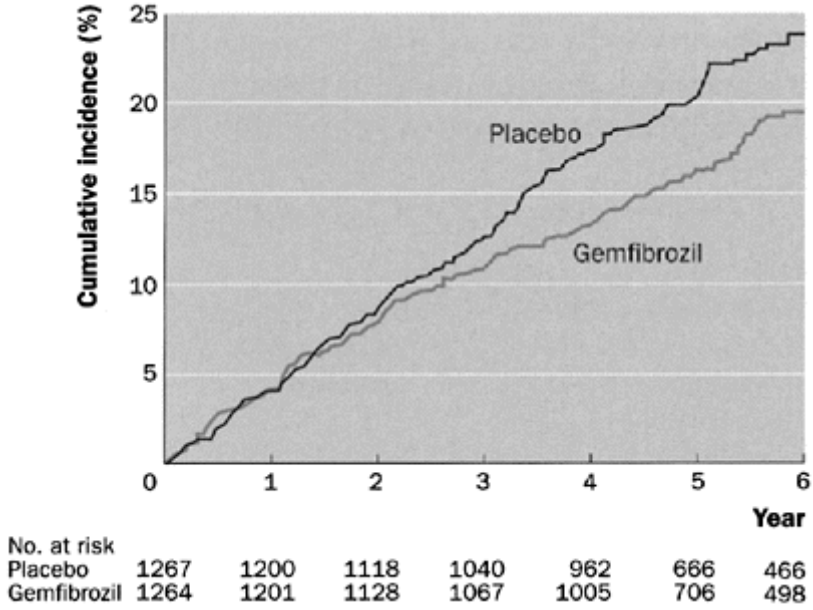

Fig. 5.2 Kaplan-Meier estimates of the incidence of death from coronary heart disease and non-fatal myocardial infarction in the gemfibrozil and placebo groups. The relative risk was 22% (P=0.006), as derived from a Cox model. Source: Rubins *et al.* (1999).

However, now that TGs are much more clearly established as a CHD risk factor it is hard to ignore the substantial decrease in serum TGs with gemfibrozil. Although the majority of the patients had serum TG levels in the 'normal range' for most laboratories, the patients were selected by virtue of low serum cholesterol and established CHD, which makes it likely that they had increased levels of small, dense LDL, which is detectable in the circulation of such patients even when TGs are as low as 1.5 mmol/l |**8**|. The effect may thus have been mediated through a reduction in small, dense LDL secondary to the decrease in serum TGs. However, this interpretation must be cautious. Bezafibrate, another fibrate drug, was used in the BIP trial and the major benefit in terms of preventing further CHD incidents was seen only in patients with serum TGs exceeding 2.3 mmol/l, whereas in VAHIT there is the same relative reduction in new CHD events in men from the upper half of the TG concentration range as in the lower half. Apart from the different drugs used in these two trials, the lipoprotein entry criteria differed with many of the men in BIP having higher cholesterol and slightly higher HDL cholesterol values. Thus it may be that when cholesterol is elevated fibrates are most successful when TGs are clearly raised. In such patients, of course, a statin would also be required in practice to lower the LDL cholesterol. On the other hand in the unusual situation of a patient with CHD despite low cholesterol levels even relatively 'normal' levels of TGs can be worth reducing because they may indicate the presence of small dense LDL in this highly selected group.

Secondary prevention by raising HDL cholesterol and reducing triglycerides in patients with coronary artery disease: the Bezafibrate Infarction Prevention (BIP) study.

Israeli Society for Prevention of Heart Attacks. *Circulation* 2000; **102**: 21–7.

B**ACKGROUND. CHD patients with low HDL cholesterol levels, high TG levels or both are at an increased risk of cardiovascular events. Whether raising HDL cholesterol or decreasing TGs reduces this excess risk remains to be confirmed.**

I**NTERPRETATION.** Bezafibrate, 400 mg daily for a mean 6.2 years, was safe and effective in elevating HDL cholesterol levels and lowering TGs in this double-blind trial of 3090 patients with a previous MI or stable angina. An overall trend towards reduction of the primary end-point (incidence of fatal or non-fatal MI or sudden death) was observed. The reduction in the primary end-point in patients with high baseline TGs (≤200 mg/dl) needs further confirmation.

Comment

The patients in this trial had established CHD and were randomized to receive bezafibrate or placebo for an average of 6.2 years. The average serum cholesterol at entry was 5.5 mmol/l and HDL cholesterol 0.9 mmol/l and serum TG concentration 1.7 mmol/l. So the major difference between these patients and those in VAHIT was their cholesterol level, which was on average 1 mmol/l higher in BIP. The outcome of the trial was different. In BIP the overall decrease in new CHD events (Fig. 5.3) was only 7.3% (not significant), but in those with serum TG level ≥2.3 mmol/l there was a 39.5% reduction in these events. In VAHIT the entry TG level did not affect the relative decrease in new CHD risk. Although the mechanism for the favourable effect of gemfibrozil in VAHIT is uncertain, it should be recognized that the patients in this trial were somewhat unusual for people with established CHD in that their serum cholesterol was relatively low. They will not be as commonly encountered in clinical practice as patients similar to those in BIP. Thus in the more typical CHD patient fibrate drugs would seem to have a place when TGs are clearly elevated, although, of course, statins will be necessary to reduce cholesterol levels to≤5 mmol/l (LDL cholesterol≤3.5 mol/l). In those unusual patients who develop CHD with cholesterol levels of<5 mmol/l VAHIT provides powerful evidence for the use of gemfibrozil. An alternative is, of course, to consider a statin. A subgroup of patients in the Cholesterol and Recurrent Events (CARE) (Fig. 5.3) trial did have low cholesterol levels. However, those with LDL cholesterol levels of≤125 mg/dl (<3.2 mmol/l) did not appear to show any benefit.

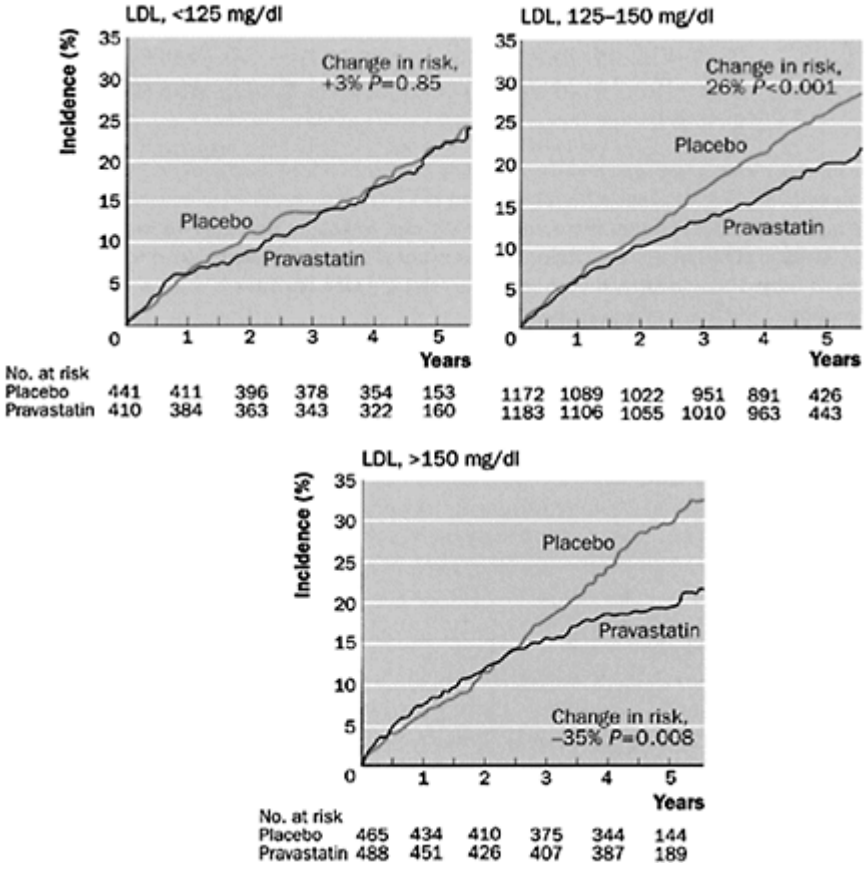

Fig. 5.3 Kaplan-Meier estimates of the incidence of fatal coronary heart disease, non-fatal myocardial infarction, coronary bypass surgery or angioplasty in the study groups. According to baseline LDL cholesterol level. Changes in risk are those attributable to pravastatin. P=0.03 for the interaction between baseline LDL cholesterol level and treatment, by Cox proportional hazards analysis.

Source: Sacks *et al.* (1996).

Mediterranean diet, traditional risk factors, and the rate of cardiovascular complications after myocardial infarction: final report of the Lyon Diet Heart Study.

M de Lorgeril, P Salen, J L Martin, *et al. Circulation* 1999; **6**:779–85.

B**ACKGROUND. The Lyon Diet Heart Study is a randomized secondary prevention trial aimed at testing whether a Mediterranean-type diet may reduce the rate of recurrence after a first MI. An intermediate analysis showed a striking protective effect after 27 months of follow-up. This report presents results of extended follow-up (mean of 46 months per patient) and considers the relationships between dietary patterns, traditional risk factors and recurrence.**

I**NTERPRETATION.** The protective effect of the Mediterranean dietary pattern was maintained up to 4 years after the first infarction, confirming previous intermediate analyses. Major traditional risk factors, such as high blood cholesterol and blood pressure, were shown to be independent and joint predictors of recurrence, indicating that the Mediterranean dietary pattern did not alter, at least qualitatively, the usual relationships between major risk factors and recurrence. A comprehensive strategy to decrease cardiovascular morbidity and mortality should include a cardioprotective diet combined with other (pharmacological?) means of reducing modifiable risk factors.

Comment

Clearly on the one hand those who have berated the importance of diet in preventing CHD need radically to revise their views, whereas on the other what constitutes a diet that should be advocated to prevent CHD also requires revision. The Mediterranean diet in this study was one that was richer in monounsaturated fatty acids, particularly, oleic acid and in the plant omega 3 long-chain fatty acid, linolenic acid, present in olive oil and rapeseed oil.

It was compared with a 'prudent Western diet', which is similar to that commonly advised for the prevention of CHD, which would contain the omega 6 long-chain fatty acid, linoleic acid, present in sunflower and corn oil. This does not mean that linoleic acid is ineffective as a means of CHD prevention when substituted for saturated fatty acids, because, of course, the study did not have a control group who maintained a typical Western diet rich in saturated fats. Indeed one earlier review found that linoleic acid was likely to decrease CHD risk |**9**|. However, given the successful outcome of the Lyon study and that of the GISSI study using fish omega 3 fatty acids, omega 3 fatty acids from either plant or marine sources should probably receive more attention.

The continuing influence of risk factors, such as cholesterol, on the cardiac outcome of the study emphasizes the importance of a combined dietary and pharmacological approach to the modification of CHD risk in secondary prevention.

Dietary supplementation with n-3 polyunsaturated fatty acids

and vitamin E after myocardial infarction: results of the GISSI-Prevenzione trial.
Gruppo Italiano per lo Studio della Sopravvivenza nell'Infarto miocardico. *Lancet* 1999; **354**:447–55.

B**ACKGROUND. There is conflicting evidence on the benefits of diets rich in vitamin E (α-tocopherol), n-3 polyunsaturated fatty acids (PUFA), and their pharmacological substitutes. The effects of these substances as supplements were investigated in 11 324 patients surviving recent MI who were randomly assigned to supplements of n-3 PUFA (1 g daily), vitamin E (300 mg daily), both, or none for 3.5 years.**

I**NTERPRETATION.** Dietary supplementation with n-3 PUFA led to a clinically and statistically significant reduced risk of the primary combined end-point (death, non-fatal MI and stroke). Vitamin E had no benefit. Its effects on fatal cardiovascular events require further exploration.

Comment

This trial examined the effects in a large population of MI survivors of a concentrate of the omega-3 long-chain polyunsaturated fatty acids, eicosapentaenoic acid and docosahexaenoic acid, found in fish oil and/or vitamin E. The 10% reduction in cardiovascular end-points over the 3.5 years of the trial associated with the administration of omega-3 long-chain fatty acids was principally due to a decrease in sudden deaths suggesting that it resulted from a reduction in fatal dysrhythmias. This finding together with that of the Lyon diet-heart study clearly emphasizes the need to examine the role of fatty acids in the prevention of CHD both in the diet and as pharmacological agents. Vitamin E on the other hand was without effect on the prognosis of the heart attack survivors.

Vitamin E supplementation and cardiovascular events in high-risk patients.
The Heart Outcomes Prevention Evaluation Study Investigators. *N Engl J Med* 2000; **342**:154–60.

B**ACKGROUND. Observation and experimental studies suggest that the amount of vitamin E ingested in food and in supplements is associated with a lower risk of CHD and atherosclerosis.**

I**NTERPRETATION.** Enrolled were a total of 2545 women and 6996 men 55 years of age or older who were at high risk for cardiovascular events because they had cardiovascular disease or diabetes in addition to one other risk factor. These patients were randomly assigned according to a two-by-two

factorial design to receive either 400 IU of vitamin E daily from natural sources or matching placebo and either an

angiotensin-converting-enzyme inhibitor (ramipril) or matching placebo for a mean of 4.5 years (the results of the comparison of ramipril and placebo are reported in a companion article published in the same number of the New England Journal of Medicine). The primary outcome was a composite of MI, stroke and death from cardiovascular causes. The secondary outcomes included unstable angina, congestive heart failure, revascularization or amputation, death from any cause, complications of diabetes and cancer. A total of 772 of the 4761 patients assigned to vitamin E (16.2%) and 739 of the 4780 assigned to placebo (15.5%) had a primary outcome event (relative risk, 1.05; 95% confidence interval, 0.95–1.16; *P*=0.33). There were no significant differences in the numbers of deaths from cardiovascular causes (342 of those assigned to vitamin E versus 328 of those assigned to placebo; relative risk 1.05; 95% confidence interval, 0.90–1.22), MI (532 versus 524; relative risk, 1.02; 95% confidence interval, 0.90–1.15), or stroke (209 versus 180; relative risk, 1.17; 95% confidence interval, 0.95–1.42). There were also no significant differences in the incidence of secondary cardiovascular outcomes or in death from any cause. There were no significant adverse effects of vitamin E. In patients at high risk for cardiovascular events, treatment with vitamin E for a mean of 4.5 years has no apparent effect on cardiovascular outcome.

Comment

A similar negative result for vitamin E as that seen in the previous study from GISSI. The authors of this paper also review earlier trials that give a similar overall negative picture.

Oxidants and antioxidants in atherogenesis. An appraisal.
S Parthasarathy, N Santanam, S Ramachandran, O Meilhac. *J Lipid Res* 1999; **40**(12):2143–57.

B**ACKGROUND. Oxidized LDL (Ox-LDL) has many components that are not in native LDL. Their presence and quantity depends on the nature and extent of oxidation. Lipids esterified to oxidized fatty acids are formed during early oxidation and show various proatherogenic properties in *in vitro* cell cultures. Recent evidence suggests that some of these oxidized lipids could also elicit 'antioxidant, antiatherogenic'**

responses from cells. Some cellular effects of Ox-LDL, previously interpreted as atherogenic, could also be reinterpreted as antiatherogenic. Further, the antioxidants that are carried in lipoproteins could have anomalous behaviour attributable to their metabolism, ability to be internalized by arterial cells and the presence of oxidative systems that could render them pro-oxidants.

I**NTERPRETATION.** Improved understanding of the many contributing factors is needed before antioxidant therapy becomes a treatment option for cardiovascular diseases.

Comment

The experimental evidence that LDL must undergo oxidative modification before it becomes atherogenic is vast and contrasts sharply with the disappointing results of clinical trials of antioxidants in the prevention of CHD. Thus, while oxidation of LDL is likely to be an important atherogenic mechanism, antioxidant vitamins are unlikely to be an effective means of protecting LDL from oxidation. Other systems that prevent LDL from oxidation such as HDL, which contains the enzyme paraoxonase (PON1), may be more important in protecting against lipid peroxidation |**10**|.

Vitamin E supplementation increases the resistance of both LDL and HDL to oxidation and increases cholesteryl ester transfer activity.

S Arrol, M I Mackness, P N Durrington. *Atherosclerosis* 2000; **150**: 129–34.

B**ACKGROUND. There is increasing evidence that lipid peroxidation and oxidative modification of LDL are important in atherogenesis. Evidence that antioxidant therapy decreases mortality is, however, inconclusive. The effects of vitamin E for 50 days (in 37 healthy volunteers) on the susceptibility of LDL and HDL to oxidation, and on cholesteryl ester heteroexchange were examined *in vitro* using autologous serum lipoproteins.**

I**NTERPRETATION.** Vitamin E (200 and 400 mg daily) significantly decreased the susceptibility of LDL and HDL to oxidation *in vitro*. There was also an increase in cholesteryl ester transfer activity (CETA), however. There is some evidence that increased CETA is potentially deleterious and might therefore counteract the beneficial effects of vitamin E and other antioxidants on the susceptibility of lipoproteins to oxidation.

Comment

Vitamin E administered orally does protect both LDL and HDL against the early phase of lipid peroxidation by delaying the formation of conjugated dienes, which are the precursors of lipid peroxides. However, this is unlikely to have much effect on the overall formation of lipid peroxides over longer periods. Indeed, because vitamin E protects lipids against oxidation by itself undergoing oxidation, the oxidized vitamin could contribute to later free radical attack on the lipoprotein phospholipids. Furthermore, this study shows that vitamin E like another antioxidant drug, probucol, increases the activity of CETP, which may be pro-atherogenic and thus counteract any benefit from the antioxidant properties of vitamin E.

A cholesteryl ester transfer protein inhibitor attenuates atherosclerosis in rabbits.

H Okamoto, F Yonemori, K Wakitani, *et al. Nature* 2000; **406**(6792): 203–7.

B**ACKGROUND. The plasma protein CETP mediates the exchange of cholesteryl ester in HDL for TG in VLDL. This process decreases antiatherogenic HDL cholesterol and**

increases pro-atherogenic VLDL and LDL cholesterol. Thus CETP is potentially atherogenic, but it could also be antiatherogenic because it participates in reverse cholesterol transport (from peripheral cells through the plasma to the liver). To clarify the role of CETP in atherosclerosis the development of a potent and specific CETP inhibitor was attempted.

I**NTERPRETATION.** CETP inhibitors that form a disulphide bond with CETP are described, and one such inhibitor (JTT-705) that increases HDL cholesterol, decreases non-HDL cholesterol and inhibits the progression of atherosclerosis in rabbits is presented. The findings indicate that CETP may be atherogenic *in vivo* and that JTT-705 may be a potential antiatherogenic drug.

Comment

An exciting paper that conjures the prospect of a new class of antiatherogenic drugs, inhibiting the heteroexchange of cholesteryl ester between lipoprotein classes and markedly increasing HDL cholesterol. Further research is required to establish the safety and efficacy of such drugs in humans, and also to explore their antiatherogenic mechanism further, in particular their effects on the formation of small, dense LDL, which is also linked with low HDL levels, hypertriglyceridaemia and increased CETP activity.

Prevention of recurrent pancreatitis in familial lipoprotein lipase deficiency with high-dose antioxidant therapy.

A P Heaney, N Sharer, B Rameh, J M Braganza, P N Durrington. *J Clin Endocrinol Metab* 1999; **84**:1203–5.

B**ACKGROUND/INTERPRETATION. This report describes a dramatic response to antioxidant therapy in three patients with familial lipoprotein lipase deficiency complicated by frequent severe episodes of pancreatitis who had failed to respond to other dietary and pharmacological measures. Antioxidant therapy may be an important advance in the management of such patients.**

Comment

In three patients with extreme hypertriglyceridaemia due to familial lipoprotein lipase deficiency and particularly frequent episodes of acute pancreatitis and its complications in the previous years in whom all other attempts to prevent it had failed, the antioxidant vitamin supplement Bioantox in higher doses than in CHD prevention trials had a dramatic effect in abolishing further attacks of pancreatitis. This was an uncontrolled open study, but the observation merits further study and may already be worth trying in this particular group of patients who present an extremely difficult problem in many lipid clinics.

Coronary artery bypass surgery with arterial grafts in familial hypercholesterolemia.

M Kawasuji, N Sakakibara, S Fujii, T Yasuda, Y Watanabe. *J Thorac Cardiovasc Surg 2000;* **119**:1008–14.

B**ACKGROUND. Familial hypercholesterolaemia is a dominantly inherited disorder caused by mutations at the locus for the LDL receptor and is frequently associated with premature coronary artery disease. The aim of this study was to determine whether arterial grafting was associated with long-term benefits for patients with familial hypercholesterolaemia (*n*=101), all of whom received diet therapy and intensive cholesterol-lowering drugs after operation.**

I**NTERPRETATION.** Arterial grafting improved the long-term freedom from reoperation in patients with familial hypercholesterolaemia (mean follow

up 95 months). Further benefit from multiple arterial grafting was not identified.

Comment

Many years ago there was a feeling among some cardiac surgeons that patients with familial hypercholesterolaemia did badly after coronary artery bypass grafting. The overall impression from this study is that this is not the case now that we have effective cholesterol-lowering therapy and certainly when the internal thoracic (mammary) artery is used.

References

1. Hebert PR, Gaziano JM, Chan KS, Hennekens CH. Cholesterol lowering with statin drugs, risk of stroke, and total mortality. An overview of randomised trials. *JAMA* 1997; **278**:313–21.

2. Shepherd J, Cobbe SM, Ford I, Isles CG, Lorimer AR, MacFarlane PW, McKillop JH, Packard C, for the West of Scotland Coronary Prevention Study Group. Prevention of coronary heart disease with pravastatin in men with hypercholesterolaemia. *N Engl J Med* 1995; **333**:1301–7.

3. Standing Medical Advisory Committee on use of statins. NHS. Executive. London: Department of Health, May 1997.

4. Wood D, De Backer G, Faergeman O, Graham I, Mancia G, Pyörälä K. with members of the Task Force. Prevention of coronary heart disease in clinical practice: recommendations of the Second Joint Task Force of European and other Societies on Coronary Prevention. *Atherosclerosis* 1998; **140**:199–270.

5. Wood D, Durrington PN, Poulter N, McInnes G, Rees A, Wray R. Joint British Recommendations on prevention of coronary heart disease in clinical practice. *Heart* 1998; **80**(Suppl 2): S1–29.

6. Rittershaus CW, Miller DP, Thomas LJ, Picard MD, Honan CM, Emmett CD, Pettey CL, Adari H, Hammond RA, Beattie DT, Callow AD, Marsh HC, Ryan US. Vaccine-induced antibodies inhibit CETP activity *in vivo* and reduce aortic lesion in a rabbit model of atherosclerosis. *Arterioscler Thromb Vasc Biol*, 2000; **20**(9): 2106–12.

7. Sugano M, Makino N, Sawader S, Otsuka S, Watanabe M, Okamoto H, Kamada M, Mizushima A. Effect of antisense oligonucleotides against cholesteryl ester transfer protein on the development of atherosclerosis in cholesterol-fed rabbits. *J Biol Chem* 1998; **273**:5033–6.

8. Griffin BA, Freeman DJ, Tait G, Thomson J, Caslake MJ, Packard CJ, Shepherd J. Role of plasma triglyceride in the regulation of plasma low density lipoprotein (LDL) subfractions. Relative contribution of small dense LDL to coronary heart disease risk. *Atherosclerosis* 1994; **106**:241–9.

9. Oliver MF. It is more important to increase the intake of unsaturated fats than to decrease the intake of saturated fats: evidence from clinical trials relating to ischaemic heart disease. *Am J Clin Nutr* 1997; **66**:980S–6S.

10. Durrington PN, Mackness B, Mackness MI. Paraoxonase and atherosclerosis.

Arterioscler Thromb Vasc Biol 2001, **21**:473–80.

6
Trials of therapy to modify serum lipids and lipoproteins

Introduction

In most patients with hypercholesterolaemia statins can generally achieve reductions in cholesterol and low-density lipoprotein (LDL) cholesterol to the target values recommended in most guidelines. The principal reason for the failure to do so in clinical practice stems largely from a reluctance by clinicians to prescribe adequate doses. In familial hypercholesterolaemia (FH), however, where the initial levels are particularly high even a 40 or 50% reduction may leave patients with cholesterol levels that are still unacceptably high. Combination of a statin with a bile acid sequestrating agent, although capable of producing even greater reductions in LDL cholesterol than with statin alone, is in practice only rarely acceptable to patients. Extracorporeal LDL removal can also achieve greater LDL lowering, but is expensive and also poses considerable difficulties for patients. However, certainly in homozygotes and for exceptionally high-risk heterozygotes it has an important place in treatment. Articles in this chapter compare some of the methods available.

Toto *et al.* provided an important study adding to our knowledge of how statins work by confirming in patients with nephrotic syndrome that their action is almost exclusively on the catabolism of apolipoprotein B (apoB)-containing lipoproteins. This mechanism probably largely involves the induction of hepatic LDL receptors and would account for why in receptor-negative FH patients atorvastatin was generally found by Yamamoto *et al.* to be without effect in lowering LDL in contrast to those who were receptor-defective. Increased removal of remnant-like particles is also likely to be the mechanism by which a statin lowered the circulating levels of these lipoproteins in the study by Twickler *et al.* in heterozygotes for FH. This chapter also contains a report on the efficacy of a statin in patients with chronic renal failure treated by peritoneal dialysis.

An important clinical question is whether plant stanol esters can cause any further decrease in LDL cholesterol once patients are established on statin therapy. This is addressed by the study of Blair *et al.* Papers on statin-fibrate, statin-oestrogen and statin-marine omega-3 long chain polyunsaturated fatty acid combinations are also included. Marine omega-3 fatty acids have received a lot of attention this year now that they are available in a concentrated preparation (Omacor) that avoids some of the difficulties posed by raw fish oil preparations, the most important of which clinically was their lack of tolerability.

This chapter ends with an interesting subgroup analysis from the Heart and Estrogen/Progestin Replacement Study investigation showing a decrease in serum lipoprotein(a) [LP(a)] most marked in women with higher levels, which was associated

with a decrease in coronary heart disease (CHD) events. This was unlike the overall finding of the study that showed neither benefit nor detriment of hormone replacement in the postmenopausal women all of whom had established CHD.

Direct adsorption of lipoproteins (DALI) from whole blood: first long-term clinical experience with a new LDL-apheresis system for the treatment of familial hypercholesterolaemia.

M Jansen, S Banyai, S Schmaldienst, *et al. Wien Klin Wochenschr* 2000; **112**:61–9.

B**ACKGROUND. The DALI (direct adsorption of lipoproteins) LDL-apheresis system is a novel device for the removal of lipoproteins from whole blood. We report the first long-term treatments (at weekly or 2-weekly intervals for a mean 16.7 months) using different DALI adsorber sizes in seven patients with homozygous or severe heterozygous FH.**

I**NTERPRETATION.** Sufficient reductions in LDL cholesterol and Lp(a) were achieved using the DALI-750 system (but not the DALI-500 system) and the treatment was well tolerated. The easy use and short time (mean 153 min) needed for each treatment are the major advantages of the BALI system over other available LDL-apheresis devices. Potential particle release from the adsorber into the circulation must be ruled out before the system can be introduced in clinical routine.

Comment

The development of extracorporeal systems for the removal of LDL in patients with severe hypercholesterolaemia, which are easier to use than the present ones could make this type of treatment more widely available.

Prospective randomised cross-over comparison of three LDL-apheresis systems in statin pretreated patients with familial hypercholesterolaemia.

S Schmaldienst, S Banyai, T M Stulnig, *et al. Atherosclerosis* 2000; **151**: 493–9.

B**ACKGROUND. Various LDL-apheresis systems have gained increasing clinical acceptance for treating patients with severe FH, especially those with coronary artery disease. For each device data on**

efficacy have been provided, but as yet there has

been no comparative analysis that has included the novel direct adsorption of lipoproteins from whole blood. This prospectively designed cross-over comparison of three LDL-apheresis systems (immunoadsorption; dextran sulphate adsorption; and DALI) was done in three patients with homozygous and five with heterozygous (*n*=5) FH.

I**NTERPRETATION.** All three systems effectively removed atherogenic lipoproteins, LDL cholesterol in particular. The reduction in Lp(a) was about 63% for each device. The loss of high density lipoprotein (HDL) cholesterol was higher with immunoadsorption compared with dextran sulphate adsorption and the DALI system. Significant differences between the systems were found for the removal of fibrinogen: dextran sulphate adsorption was most effective followed by immunoadsorption and then DALI. The shortest treatment duration was with the DALI system. No side-effects were recorded. Long-term observations have yet to prove whether these differences in efficacy are clinically relevant.

Comment

Extracorporeal removal of LDL is an essential part of the treatment of patients with homozygous FH. This type of comparison is of considerable interest. Many patients, however, continue to receive conventional plasmapheresis and it would be important to know how this compares.

The effect of atorvastatin on serum lipids and lipoproteins in patients with homozygous familial hypercholesterolemia undergoing LDL-apheresis therapy.

A Yamamoto, M Harada-Shiba, A Kawaguchi, *et al. Atherosclerosis* 2000; **153**:89–98.

B**ACKGROUND. The efficacy of atorvastatin in reducing serum lipid levels, modifying lipoprotein composition, and suppressing cholesterol synthesis was assessed in nine patients with homozygous FH undergoing LDL-apheresis therapy. Atorvastatin was given in escalating doses (10, 20 and 40 mg/day).**

I**NTERPRETATION.** Five of nine patients responded well to atorvastatin; four of whom were receptor defective, the other being receptor negative. In the receptor-defective patients LDL cholesterol was reduced by about 20% at

the highest atorvastatin dose. Of five receptor-negative patients, only one responded well to atorvastatin (LDL cholesterol reduced by 14.9%) but the other four showed considerable increases in HDL cholesterol. All patients showed reduced urinary excretion of mevalonic acid, suggesting that atorvastatin decreases LDL cholesterol by inhibiting cholesterol biosynthesis, even in the absence of LDL receptor activity. Atorvastatin also decreased serum triglycerides (TG) in both receptor-negative and -defective patients, especially in the latter. As cholesterol level rebounds quickly after each apheresis procedure, combined therapy with atorvastatin and apheresis may increase the efficacy of apheresis, improving cost-benefit effectiveness by reducing the frequency of apheresis treatment.

Comment

This confirms the efficacy of atorvastatin in patients with homozygous FH as an adjunct to the extracorporeal removal of LDL. The larger dose of 80 mg/day is more typically used in Western patients when they are old enough to receive it.

Efficacy of atorvastatin compared with simvastatin in patients with hypercholesterolemia.
M Farnier, J J Portal, P Maigret. *J Cardiovasc Pharmacol Ther* 2000; **5**: 27–32.

B**ACKGROUND. Atorvastatin markedly reduces LDL cholesterol at doses ranging from 10 to 80 mg/day. This trial was designed to compare the efficacy of atorvastatin 10 mg with simvastatin 10 mg and 20 mg.**

I**NTERPRETATION.** In this 6-week parallel-group randomized study, in 272 patients with primary hypercholesterolaemia, atorvastatin 10 mg was more effective than was simvastatin 20 mg and significantly more effective than was simvastatin 10 mg for reducing LDL cholesterol levels. Both drugs were well tolerated.

Comment

The debate about which statin to use as a first-line agent, which involves considerations of the results of trials with cardiovascular efficacy, effectiveness in lowering LDL, possible benefits achieved with statins by non-LDL lowering mechanisms and cost, will continue.

High dose of simvastatin normalizes postprandial remnant-like particle response in patients with heterozygous familial hypercholesterolemia.

T B Twickler, G M Dallinga-Thie, H W de Valk, *et al. Arterioscler Thromb Vasc Biol* 2000; **20**:2422–7.

B**ACKGROUND. FH and disturbances in postprandial lipoprotein metabolism are both associated with premature atherosclerosis. Heterozygous FH patients have increased fasting and postprandial remnant lipoprotein concentrations. The effect of statins on plasma cholesterol levels in patients with FH is well established but it is not known whether postprandial lipoproteins are also influenced. This case-controlled study investigated the effects of high-dose simvastatin 80 mg daily on postprandial lipoproteins in seven FH patients.**

I**NTERPRETATION.** Treatment with simvastatin for 3 months significantly reduced the fasting and postprandial remnant lipoprotein cholesterol concentrations but did not improve the postprandial retinyl ester response.

Comment

This is a very interesting study that revives interest in the increased levels of chylomicron remnants and intermediate density lipoprotein (IDL) as well as LDL in FH. It is difficult to know how much these other lipoproteins contribute to atheroma in FH, but it could be considerable. A similar LDL catabolic defect in familial defective apoB_{3500}, which does not result in an increase in IDL, is generally accompanied by less aggressive CHD.

Pravastatin treatment of very low density, intermediate density and low density lipoproteins in hypercholesterolemia and combined hyperlipidemia secondary to the nephrotic syndrome.

R D Toto, S M Grundy, G L Vega. *Am J Nephrol* 2000; **20**:12–17.

B**ACKGROUND. Pravastatin was used in nephrotic syndrome patients with hypercholesterolaemia and combined hyperlipidaemia to assess whether the drug decreases production of LDL and reduces levels of very low-density lipoprotein (VLDL) and IDL. Seven patients with high LDL alone and six with high VLDL, IDL and LDL were treated**

with pravastatin 40 mg daily in a placebo-controlled cross-over study.

I**NTERPRETATION.** Pravastatin effectively reduced LDL levels in both types of dyslipidaemia by increasing LDL clearance. Treatment did not affect production of LDL or levels of VLDL+IDL-apoB. Thus, pravastatin increases LDL clearance. Statins do not seem to affect production rates of apoB-containing lipoproteins. Treating combined hyperlipidaemia may require another drug to normalize levels of VLDL and IDL as well as pravastatin.

Comment

This study provides further evidence that the mechanism by which statins act is to increase the removal of apoB-containing lipoproteins from the circulation. This occurs most probably by increasing the hepatic expression of LDL receptors in response to the inhibition of hepatic cholesterol biosynthesis decreasing intrahepatic cholesterol levels and triggering LDL receptor synthesis. This receptor has both apoB and apoE as its ligands and can thus increase the fraction of both circulating LDL, IDL and smaller VLDL removed from the circulation both through binding to the apoB, which is incorporated into these lipoproteins during their synthesis, and the apoE, which VLDL acquires after its secretion into the circulation. This, therefore, provides an explanation for both the cholesterol- and TG-lowering properties of statins.

Effects of atorvastatin on dyslipidaemia in uraemic patients on peritoneal dialysis.

G Hufnagel, C Michel, F Vrtovsnik, *et al. Nephrol Dial Transplant* 2000; **15:** 684–8.

B**ACKGROUND. To evaluate the efficacy and safety of atorvastatin, in peritoneal dialysis patients with dyslipidaemia, 31 dialysis patients with hypercholesterolaemia were treated for 4 months with atorvastatin (starting dose 10 mg). The dose could be increased to 20 or 40 mg to achieve the targets: plasma LDL cholesterol of 130 mg/dl for primary prevention of CHD; plasma LDL cholesterol of 100 mg/dl for secondary prevention; and plasma TGs of 200 mg/dl.**

I**NTERPRETATION.** Atorvastatin is an effective and safe lipid-lowering agent for peritoneal dialysis patients with mixed dyslipidaemia. Mean LDL cholesterol and TG levels were significantly decreased. Nineteen of 20 primary prevention and seven of nine secondary prevention patients achieved their LDL cholesterol targets while 15 of 19 hypertriglyceridaemic patients achieved their TG targets. Two patients stopped treatment (one because of gastrointestinal disturbances, the other because of an allergic skin reaction).

Comment

Patients with chronic renal failure treated by peritoneal dialysis often have the most severe hyperlipoproteinaemias encountered in renal disease. The case for statin therapy in this group of patients who are at high CHD risk is largely from extrapolation from clinical trials in patients, who do not have renal failure, but whose level of CHD risk is also high. That statins can produce substantial improvements in the lipoprotein profile with relative safety is encouraging for those renal physicians who are persuaded that their use is justified.

Effects of combination therapy with estrogen plus simvastatin on lipoprotein metabolism in postmenopausal women with type IIa hypercholesterolemia.

A Wakatsuki, Y Okatani, N Ikenoue. *Atherosclerosis* 2000; **150**:103–11.

B**ACKGROUND. We investigated the effects of oestrogen and simvastatin, administered both alone and in combination, on plasma lipid levels and lipoprotein-related enzymes in 45 postmenopausal women with type IIa hypercholesterolaemia. The patients received conjugated equine oestrogen 0.625 mg, simvastatin 5 mg, or the combination daily for 3 months.**

I**NTERPRETATION.** Findings indicate that combination therapy with oestrogen plus simvastatin favourably affected lipid metabolism by reducing the concentrations of very low density lipoprotein and IDL particles as well as large and small LDL particles, by

increasing the concentration of HDL particles, and by preventing oestrogen-induced increases in plasma TG levels.

Comment

In the climate of controversy surrounding the disappointing outcome of hormone replacement therapy using combined oestrogen and progestin in postmenopausal women with CHD, the results of the oestrogen alone arm of this trial are interesting. It remains, however, difficult to advise on the value of oestrogen/progestin combinations in dyslipidaemic high-risk postmenopausal women. Are they better off on statin therapy alone, when one takes into account possible adverse effects of oestrogens on thrombosis and of progestins on the lipoprotein profile?

Bezafibrate and simvastatin combination therapy for diabetic dyslipidaemia: efficacy and safety.
D Gavish, E Leibovitz, I Shapira, A Rubinstein. *J Intern Med* 2000; **247**: 563–9.

B**ACKGROUND. This open study in 148 patients was designed to determine the efficacy and safety of a statin-fibrate combination in patients with type 2 diabetes mellitus. Each patient received either bezafibrate slow release 400 mg daily or simvastatin 20 mg daily for 6 months and then a combination of the two for 1 year.**

I**NTERPRETATION.** The statin and fibrate combination was more efficacious than either of the single medications for the treatment of diabetic dyslipidaemia, as shown by improvements in the lipoprotein profile and reductions in Lp(a), fibrinogen and the cardiovascular event rate. The cardiovascular event rate was significantly reduced, from 9.5% during the first 6 months of the study to less than 2% during the last year while patients were on the combination treatment.

Comment

Increasing use of the combination of statin and fibrate therapy is being made in high-risk diabetic patients with combined hyperlipidaemia. The results of this trial are encouraging in this regard, although the need to inform patients of the risk of myositis and to monitor them carefully remains prudent.

Incremental reduction of serum total cholesterol and low-density lipoprotein cholesterol with the addition of plant stanol ester-containing spread to statin therapy.
S N Blair, D M Capuzzi, S O Gottlieb, *et al. Am J Cardiol* 2000; **86**:46–52.

B**ACKGROUND. This study compared the effect of plant stanol ester spread with a placebo spread on cholesterol in patients taking statin therapy, but who still had elevated LDL cholesterol.**

INTERPRETATION. Consumption of spread that provided 5.1 g/day of plant stanol esters effectively reduced elevated total and LDL cholesterol

levels in participants on a stable regimen of a statin. This was a randomized, double-blind, placebo-controlled clinical trial, with 67 women and 100 men with LDL cholesterol (130 mg/dl) and TGs (350 mg/dl), who had been taking a stable dose of a statin for at least 90 days. For 8 weeks, participants consumed three servings of plant stanol ester spread daily. The plant stanol ester spread significantly reduced serum total cholesterol and LDL cholesterol compared with placebo spread.

Comment

This is a particularly helpful study because hitherto it has been difficult to advise patients about whether they are likely to derive any additional benefits from plant stanol ester food products once they are receiving statin therapy. What we really need to know is how many of those patients who have not achieved their cholesterol target with maximum statin therapy would do so if they consumed plant stanol ester products.

Effect of a fish-oil concentrate on serum lipids in postmenopausal women receiving and not receiving hormone replacement therapy in a placebo-controlled, double-blind trial.
K D Stark, E J Park, V A Maines, B J Holub. *Am J Clin Nutr* 2000; **72**: 389–94.

B**ACKGROUND. n-3 Fatty acid supplementation lowered serum triacylglycerol concentrations in studies in which most of the subjects were male. The effects of such supplements in postmenopausal women have received little attention. The effects of a fish-oil-derived n-3 fatty acid concentrate on serum triacylglycerol concentrations and on the ratio of triacylglycerol to HDL cholesterol were determined in 36 postmenopausal women some of whom were receiving hormone replacement therapy (HRT). The supplement used provided 2.4 g eicosapentaenoic acid (EPA) plus 1.6 g docosahexaenoic acid (DHA) daily.**

I**NTERPRETATION.** The results show that supplementation with a fish-oil-derived concentrate can favourably influence selected cardiovascular disease risk factors: there were significant reductions in serum triacylglycerol concentrations and in the triacylglycerol/HDL cholesterol ratio in postmenopausal women receiving and not receiving HRT. Fish oil supplements may have the potential to reduce the risk of CHD by as much as 27% in postmenopausal women.

Comment

The dose of omega-3 fatty acids used in this trial was greater than in the Gruppo Italiano per lo Studio della Sopravvivenza nell'Infarto miocardico (GISSI) study (see Chapter 5, page 125). In the latter study very little effect on serum TG levels was observed. However, there was benefit probably due to a decrease in fatal cardiac dysrhythmias. The essential question now given the results of the study by Stark *et al.* is whether larger doses of omega-3 marine fatty acids would have an even greater benefit by combining the favourable effect on atherogenic end-points, perhaps related to TG lowering, observed in the VAHIT (see Chapter 5, page 120) and BIP (see Chapter 5, page 122) studies with the anti-dysrhythmic action of these fatty acids.

Purified eicosapentaenoic and docosahexaenoic acids have differential effects on serum lipids and lipoproteins, LDL particle size, glucose, and insulin in mildly hyperlipidemic men.
T A Mori, V Burke, I B Puddey, *et al. Am J Clin Nutr* 2000; **71**:1085–94.

B**ACKGROUND. Regular consumption of n-3 fatty acids of marine origin can improve serum lipids and reduce cardiovascular risk. This study aimed to determine whether EPA and DHA have differential effects on serum lipids and lipoproteins, glucose and insulin in humans. In a double-blind, placebo-controlled parallel trial, 59 overweight, non-smoking, mildly hyperlipidaemic men were randomized to receive supplements of 4 g purified EPA, DHA, or olive oil (placebo) daily for 6 days.**

I**NTERPRETATION.** EPA and DHA had differential effects on lipids, fatty acids and glucose metabolism in overweight men with mild hyperlipidaemia. Compared with the olive oil group, triacylglycerols fell by slightly more in the DHA group than in the EPA group. Neither EPA nor DHA affected total cholesterol. LDL, HDL, and HDL(2) cholesterol were not affected significantly by EPA, but HDL(3) cholesterol decreased significantly. HDL cholesterol was not significantly increased by DHA but HDL(2) cholesterol showed a significant increase and LDL increased. Adjusted LDL particle size increased with DHA but not with EPA. Both EPA and DHA increased fasting insulin significantly. EPA, but not DHA, tended to increase fasting glucose, but not significantly so.

Comment

A study showing the effects of the omega-3 fatty acids, EPA and DHA individually is welcome. Whether the apparent differences would be confirmed in a larger study is open

to question, because the statistical power of this investigation is relatively low, but it is to be hoped that this important first study will lead to other investigations of the comparative effects of different omega-3 fatty acids.

Effect of omega-3 fatty acids and simvastatin on hemostatic risk factors and postprandial hyperlipemia in patients with combined hyperlipemia.

A Nordoy, K H Bonaa, P M Sandset, J B Hansen, H Nilsen. *Arterioscler Thromb Vasc Biol* 2000; **20**:259–65.

B**ACKGROUND. Patients with combined hyperlipaemia have lipid abnormalities together with an increased tendency to develop atherosclerosis and thrombosis. This tendency may be accelerated during postprandial hyperlipaemia. In the present double-blind parallel study, 41 patients with combined hyperlipaemia were treated with simvastatin at 20 mg daily for at least 10 weeks; and then randomized to receive simvastatin plus omega-3 fatty acids 3.36 g daily or placebo (corn oil) for a further 5 weeks.**

I**NTERPRETATION.** Omega-3 fatty acids given in addition to simvastatin to patients with combined hyperlipaemia had potential beneficial effect on the haemostatic risk profile in this patient group. The fatty acids reduced the free tissue factor pathway inhibitor fraction in the fasting state, reduced the degree of postprandial hyperlipaemia and inhibited the activation of factor VII during postprandial lipaemia.

Comment

Again a study showing the potential benefits of marine omega-3 fatty acids, this time in patients on a statin. This is an important study design, because most of the patients who will be considered for omega-3 fatty acid supplementation following the GISSI trial (see Chapter 5, page 125) are likely to have established CHD and will thus also be receiving statin therapy. They will also, of course, mostly also receive aspirin treatment so perhaps combined effects of aspirin and marine omega-3 fatty acids on haemostasis are also relevant. Nonetheless, the additional benefit of omega-3 fatty acids on postprandial TG metabolism is of great interest because this is one aspect of lipoprotein metabolism relatively unaffected by statin treatment.

Omacor in familial combined hyperlipidemia: effects on lipids and low density lipoprotein subclasses.
L Calabresi, D Donati, F Pazzucconi, C R Sirtori, G Franceschini. *Atherosclerosis* 2000; **148**:387–96.

B**ACKGROUND. Elevations of plasma cholesterol and/or TGs, and the prevalence of small, dense LDL particles remarkably increase coronary risk in patients with familial combined hyperlipidaemia (FCHL). The potential of Omacor, which contains the n-3 fatty acids EPA and DHA, to correct plasma lipid/lipoprotein levels and LDL particle distribution was studied in 14 FCHL patients. Patients received Omacor, providing**

3.4 g EPA+DHA daily, or placebo for 8 weeks in a randomized, double-blind, cross-over study.

I**NTERPRETATION.** Omacor significantly lowered plasma TGs and VLDL cholesterol levels, by 27 and 18%, respectively. Total cholesterol did not change but LDL cholesterol and apoB concentrations increased by 21 and 6%. LDL particles were small and apoB-rich in selected subjects. After Omacor treatment LDL was enriched in cholesterol, mainly cholesteryl esters, indicating accumulation in plasma of more buoyant core-enriched LDL particles. Plasma concentrations of IDL and LDL-1 and LDL-2 subclasses increased after Omacor, and that of LDL-3 decreased, but average LDL size did not change. The resistance of the small LDL pattern to drug-induced modifications implies that a maximal lipid-lowering effect must be achieved to reduce coronary risk in FCHL patients.

Comment

The small dense LDL particles are potentially the most atherogenic particles. Thus the decrease with omega-3 fatty acids is encouraging. However, in this particular disorder the increase in IDL, which could also be potentially atherogenic requires further study. It is unlikely, however, that the omega-3 fatty acid concentrate would in practice be used alone and its combination with statins or fibrates both of which lower IDL may overcome any increase in IDL.

Estrogen and progestin, lipoprotein(a) and the risk of recurrent coronary heart disease events after menopause.

M G Shlipak, J A Simon, E Vittinghoff, *et al. JAMA* 2000; **283**:1845–52.

B**ACKGROUND. Lp(a) has been identified as an independent risk factor for CHD events. However, few data exist on the clinical importance of Lp(a) lowering for CHD prevention. Hormone therapy with oestrogen has been found to lower Lp(a) levels in women. Objectives were to determine the relationships among treatment with oestrogen and progestin, serum Lp(a) levels and subsequent CHD events in postmenopausal women.**

I**NTERPRETATION.** The Heart and Estrogen/progestin Replacement Study, a randomized, blinded placebo-controlled secondary prevention trial conducted from January 1993 through July 1998 with a mean follow-up of 4.1 years at 20 centres. A total of 2763 postmenopausal women younger than 80 years with coronary artery disease and an intact uterus participated; their mean age was 66.7 years. Participants were randomly assigned to receive either conjugated equine oestrogens, 0.625 mg, plus medroxyprogesterone acetate, 2.5 mg, in 1 tablet daily (*n*=1380), or identical placebo (*n*=1383). Main outcome measures: Lp(a) levels and CHD events (non-fatal myocardial infarction and CHD death). Increased baseline Lp(a) levels were associated with subsequent CHD events among women in the placebo arm. After multivariate adjustment, women in the second, third and fourth quartiles of baseline Lp(a) level had

relative hazards (RHs) (compared with the first quartile) of 1.01 [95% confidence interval (CI), 0.64–1.59], 1.31 (95% CI, 0.85–2.04) and 1.54 (95% CI, 0.99–2.39), respectively, compared with women in the lowest quartile (P for trend=0.03). Treatment with oestrogen and progestin reduced mean (SD) Lp(a) levels significantly [−5.8 (15) mg/dL] [−0.20 (0.53) µmol/I] compared with placebo [0.3 (17) mg/dl] [0.01 (0.60) µmol/L] (*P*<0.001). In a randomized subgroup comparison, women with low baseline Lp(a) levels had less benefit from oestrogen and progestin than women with high Lp(a) levels; the RH for women assigned to oestrogen and progestin compared with placebo were 1.49 (95% CI, 0.97–2.26) in the lowest quartile and 1.05 (95% CI, 0.67–1.65), 0.78 (0.52–1.18) and 0.85 (0.58–1.25) in the second, third and fourth quartiles, respectively (P for interaction trends=0.03). Our data suggest that Lp(a) is an independent risk factor for recurrent CHD in postmenopausal women and that treatment with oestrogen and progestin lowers Lp(a) levels. Oestrogen and progestin therapy appears to have a more favourable effect

(relative to placebo) in women with high initial Lp(a) levels than in women with low levels. This apparent interaction needs confirmation in other trials.

Comment

Overall, the Heart and Estrogen/progestin Replacement Study showed neither a beneficial or detrimental effect of conjugated equine oestrogen 0.625 mg daily and

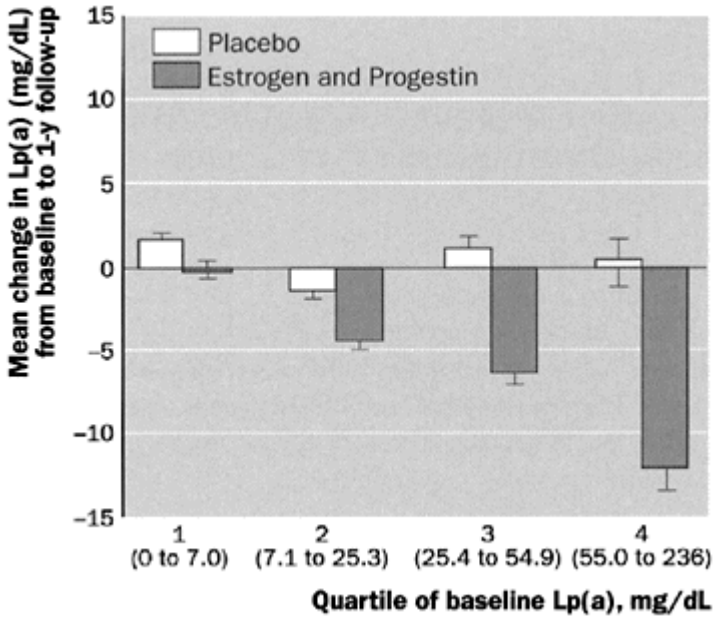

Fig. 6.1 Data are presented in mg/dl (to convert mg/dl to pmol/l, multiply by 0.0357), stratified by quartile of baseline level of Lp(a). Within each quartile, women assigned to estrogen and progestin had a significantly greater reduction in Lp(a) level than women assigned to placebo ($P<0.001$). A trend of greater reduction from oestrogen progestin with increasing quartile of baseline Lp(a) was observed ($P<0.001$). Error bars indicate SD. Source: Shlipak *et al.* (2000).

medroxyprogesterone acetate 2.5 mg compared with placebo in 2763 post-menopausal women with established CHD followed for 4.1 years. This subgroup analysis suggests that there may have been a decrease in CHD events in women treated with hormone replacement in the upper quartile of the serum Lp(a) distribution (55–236 mg/dl) who showed the greatest decrease in Lp(a) (Fig. 6.1). Lp(a) is an LDL-like lipoprotein that contains apo(a), a member of the plasminogen supergene family, disulphide linked to its apoB moiety.

7
Clinical trials in the context of epidemiology and clinical practice

Introduction

The foundation for intervening to prevent coronary heart disease (CHD) with statin therapy in people who have not yet developed clinical CHD has been strengthened by a detailed comparison of the two secondary prevention trials of pravastatin with a primary prevention trial employing the same agent in the Prospective Pravastatin Pooling Project. As the most cost-effective means of extending the use of statins into primary CHD prevention the estimation of absolute risk of CHD assessed by a method employing the Framingham risk equation has been widely recommended in Europe. However, our group found worrying inconsistencies in the applicability and the faithfulness with which the results of some of these methods compared with those obtained with the original equation. The measurement of HDL cholesterol was found to be obligatory if such methods are to have validity. In the USA where issues of cost-effectiveness are less often considered, no doubt because the cost of treatment is more directly borne by the patient rather than by a socialized health service, concerns about the use of absolute risk rather than the relative risk have been expressed most elegantly in a review by Grundy *et al.* Certainly methods based on the Framingham equation will all grossly underestimate risk in heterozygous familial hypercholesterolaemia (FH). The cost-effectiveness of other possible approaches to identifying these patients before they have developed CHD have therefore been evaluated by Marks *et al.*

Triglycerides (TG) once relegated to the dustbin of risk factors by epidemiologists now emerge as particularly important in the Caerphilly Heart Study, which was analysed in such a way as to take account of the greater biological variation in serum TG levels than, for example, high-density lipoprotein (HDL) cholesterol with which they are strongly inversely correlated. It now appears that earlier negative findings were due to a lack of understanding of the effects of regression dilution bias on multivariate analysis. So where does this leave the Framingham equation, which does not include TGs? The answer is that it can still be used to give an approximation of risk in many situations, because much of the predictive information embodied in TGs is included in the HDL cholesterol term. This does not, however, imply that serum TGs are not causally related to CHD: an important intervention in many patients at high CHD risk may be to lower TG levels (see Chapter 5, page 115.

One mechanism by which raised TGs lead to CHD may be because of their close relationship to the generation of highly atherogenic small, dense low-density lipoprotein (LDL). In the study by Kulkarni *et al.* increased levels of small dense LDL were found in Asian Indians in whom hypertriglyceridaemia secondary to insulin resistance is prevalent

when they move to cities and particularly to countries with a high fat diet. Larmarche *et al.* provide a detailed review on the subject of small, dense LDL and its relationship to hypertriglyceridaemia and CHD.

Effect of pravastatin on coronary disease events in subgroups defined by coronary risk factors. The Prospective Pravastatin Pooling Project.

F M Sacks, A M Tonkin, J Shepherd, *et al.* for the prospective Pravastatin Pooling Project Investigators Group. *Circulation* 2000; **102**:1893–1900.

B**ACKGROUND. Previous trials have had insufficient numbers of coronary events to address definitively the effect of lipid-modifying therapy on CHD in subgroups of patients with varying baseline characteristics. The data from three large randomized trials with pravastatin 40 mg were pooled and analysed with the use of a prospectively defined protocol.**

I**NTERPRETATION.** Included were 19 768 patients, 102 559 person-years of follow-up, 2194 primary end-points (coronary death or non-fatal myocardial infarction) and 3717 expanded end-points (primary end-points, coronary artery bypass grafting or percutaneous transluminal coronary angioplasty). Pravastatin significantly reduced relative risk in younger (<65 years) and older (≥65 years) patients, men and women, smokers and non-smokers, and patients with or without diabetes or hypertension. The relative effect was smaller, but absolute risk reduction was similar in patients with hypertension compared with those without hypertension. Relative risk reduction was significant in predefined categories of baseline lipid concentrations. Tests for interaction were not significant between relative risk reduction and baseline total cholesterol (5–95% range 177–297 mg/dl, 4.6–7.7 mmol/l). HDL cholesterol (27–58 mg/dl, 0.7–1.5 mmol/l) and TG (74–302 mg/dl, 0.8–3.4 mmol/l), concentrations, analysed as continuous variables. However, for LDL cholesterol, the probability values for interaction were 0.068 for the prespecified primary end-point and 0.019 for the expanded end-point. Relative risk reduction was similar throughout most of the baseline LDL cholesterol range (125–212 mg/dl, 3.2–5.5 mmol/l) with the possible exception of the lowest quintile of Cholesterol and Recurrent Events/Long Term Intervention with Pravastatin in Ischaemic Disease trials (<125 mg/dl) (relative risk reduction 5%, 95% CI 19–12%).

Comment

An important study pooling the results of clinical trials of pravastatin in the prevention of CHD events, because it has implications for primary prevention. First, this is because the

similar reduction in relative CHD risk with pravastatin in primary and secondary trials supports the concept of using high absolute CHD risk as an indication for statin treatment regardless of whether it arises because the patient has already manifested CHD, from a particularly high cholesterol level or from a combination of several cardiovascular risk factors. Secondly, the value of lowering LDL cholesterol with a statin regardless of its level once it rises above 125 mg/dl (3.2 mmol/l) in patients at high CHD is confirmed.

Conclusion

Pravastatin treatment is effective in reducing CHD events in patients with high- or low-risk factor status and across a wide range of pretreatment lipid concentrations.

Indications for cholesterol-lowering medication: comparison of risk-assessment methods.

P N Durrington, H Prais, D Bhatnagar, *et al. Lancet* 1999; **353**:278–81.

Erratum in: *Lancet* 1999; **354**:166.

B**ACKGROUND. Recommendations for prescribing lipid-lowering drug semphasize the importance of assessing absolute CHD risk based on all factors, rather than serum cholesterol concentration alone. This study compares guidelines for such assessment in 570 patients without evidence of atherosclerosis referred to a lipid clinic. The guidelines compared were those of: the US National Cholesterol Education Program; the joint guidelines of the European Societies of Cardiology, Atherosclerosis, and Hypertension; the report of the UK Standing Medical Advisory Committee; and the Framingham risk equation programmed into a computer.**

I**NTERPRETATION.** Guidelines for the use of statin treatment in patients with CHD differ in their assessment of CHD risk. The method of risk assessment recommended in future guidelines for CHD prevention should be critically tested. Of 386 patients 62% of men and 72% of women met National Cholesterol Education Program criteria for lipid-lowering medication, whereas only 9% of men and less than 1% of women met the UK criteria. The Framingham equation estimated a CHD risk of above 3% per year in 22% of men and 7% of women, which shows that the UK tables underestimated CHD risk. European guidelines were fairly accurate in assessing a CHD risk of 2% per year.

Comment

A disturbing finding from this study was that the Standing Medical Advisory Committee to the Chief Medical Officer of Health |**1**| (whose advice was supposedly evidence-based)

had recommended a means of CHD risk assessment called the Sheffield tables that had not been subject to any form of evaluation in the patients it was supposed to detect as at≥ 3% annual CHD risk. In this study it proved to underestimate risk dramatically. This was principally due to the exclusion of HDL cholesterol from the Sheffield tables. Low HDL cholesterol clusters with other CHD risk factors (Fig. 7.1) and, if this is not taken into account, many high-risk patients

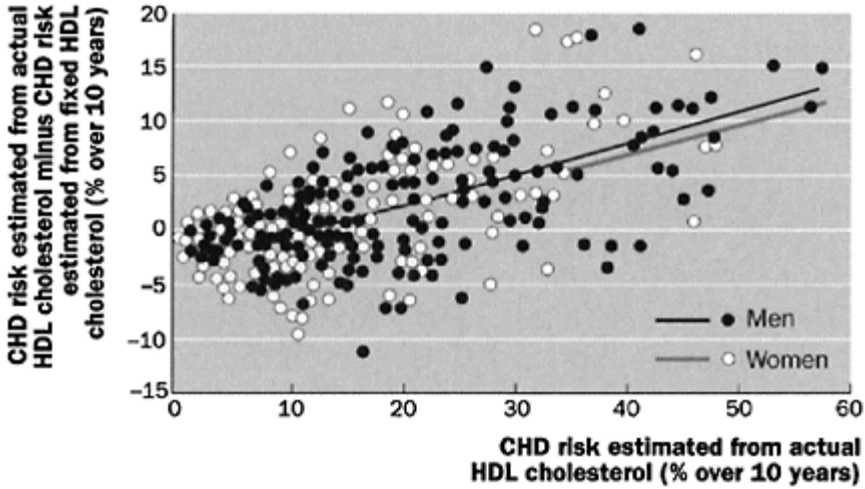

Fig. 7.1 Regression analysis of difference in CHD risk (% over 10 years) between that calculated from actual HDL cholesterol concentration and that calculated from standard value for HDL cholesterol (1.15 mmol/l in men, 1.4 mmol/l in women) plotted against CHD risk calculated from measured HDL cholesterol Men: SE=0.03, r=0.60, P<0.001. Women: SE=0.03, r=0.57, P<0.001. Source: Durrington *et al.* (erratum 1999).

are deprived of an important risk factor and their overall CHD risk is thus underestimated. This applies particularly to diabetic patients and to women. Later studies reveal that the most accurate |**2**| and convenient |**3**| method of estimating CHD risk as an aid to clinical judgement in making the clinical decision as to which primary British Guidelines |**4**|. Another disturbing finding was the disparity between prevention patients should receive statin therapy is that provided in the Joint societies with a socialized healthcare system and those where the patient more directly bears the cost of their own treatment. It is not at all clear that the reluctance to provide evidence-based medication, such as statins, in countries with socialized medical care, such as Britain, is for the greater benefit of society or simply because their healthcare systems are so unresponsive to new evidence and so determined to maintain the *status quo* that funds cannot be directed from less cost-effective, less evidence-based treatments to accommodate new effective ones.

Assessment of cardiovascular risk by use of multiple-risk-factor assessment equations: a statement for healthcare professionals from the American Heart Association and the American College of Cardiology.

S M Grundy, R Pasternak, P Greenland, S Smith, V Fuster. *Circulation* 1999; **100**:1481–92.

B**ACKGROUND/INTERPRETATION. A valuable American view of the advantages and disadvantages of the European enthusiasm for using absolute CHD risk assessment**

based on the Framingham equation as a guide to decide who should receive statin therapy in primary prevention.

Comment

The principal benefit of using absolute risk is that it bridges the evidence gap between primary and secondary prevention trials. The major disadvantage is that it leads to an excessive preoccupation with short-term gain and neglects individuals whose life-time risk, perhaps from a single treatable risk factor such as high cholesterol, is considerable. Grundy *et al.* provide an interesting method of jointly assessing relative and absolute CHD risk from a score derived from a points system based on the Framingham risk equation. However, because the estimate of relative risk gives no indication as to the prevalence of various levels of relative risk in the US population, it is unclear how the physician or health economist can judge from this the advisability of treating a particular level of relative risk at any age. If relative risk is to be of value, it should be related to the frequency distribution of risk at different ages.

Screening for hypercholesterolaemia versus case finding for familial hypercholesterolaemia: a systematic review and cost-effectiveness analysis.

D Marks, D Wonderling, M Thorogood, *et al. Health Technol Assess* 2000; **4**:1–123.

B**ACKGROUND. FH is usually caused by a mutation that impairs the functioning of the LDL receptor gene, resulting in very high levels of**

plasma cholesterol. Such levels result in early and severe atherosclerosis, and hence substantial excess mortality from CHD. Most people with FH are undiagnosed or only diagnosed after their first coronary event, but early detection and treatment with statins can reduce morbidity and mortality. The prevalence of FH in the UK population is estimated at 1 in 500, so roughly 110 000 people are affected. This review/model was intended to investigate the appropriateness of screening, and the relative costs and effectiveness of universal or opportunistic screening and case-finding screening (screening relatives of known FH cases).

I**NTERPRETATION.** The results show that case finding in the relatives of known FH patients is probably cost-effective, as is a universal screening strategy in young people, and screening patients admitted to hospital with premature myocardial infarction. Primary data on the effectiveness, psychosocial effects and cost implications of screening, and on the effectiveness of education and counselling at the time of screening are needed.

Comment

Heterozygous FH is generally due to a mutation of the LDL receptor gene. It produces the clinical phenotype of high cholesterol, tendon xanthomas and high CHD risk. Most patients are unrecognized by clinicians before the onset of CHD and frequently even afterwards. This lack of diagnosis and thus of appropriate management is a serious cause for concern, because, untreated, most male heterozygotes will manifest CHD and half of the females will do so before the age of 60, frequently many years earlier. This extensive study demonstrates the cost-effectiveness of screening the population before the age of 16 when there are few other causes of high cholesterol. Later in life polygenic hypercholesterolaemia becomes so prevalent in societies, such as those of the UK and USA, that the vast majority of people with high cholesterol do not have FH. Then the most cost-effective means of identifying people with FH is to undertake cascade family screening of known index cases with tendon xanthomas or identified LDL receptor mutations in the hope of finding other members of the family before they have developed clinical CHD. All physicians should include examination for tendon xanthomas as part of their evaluation of patients with CHD and not simply measure serum cholesterol. The importance of effective family screening for raised serum cholesterol in the families of patients with FH cannot be overestimated. Tendon xanthomas are frequently evident in the third decade of life, but may still be absent in 10–20% of patients in their fifth decade. Thus the finding of an elevated cholesterol in a relative of a known patient with the FH is generally sufficient to diagnose FH. If the LDL receptor mutation in a particular family can be identified then genetic screening becomes possible. Unfortunately, however, in countries such as the UK and USA, literally hundreds of different mutations of the LDL receptor can produce the syndrome. Genetic screening may, however, be possible in areas of the world where a smaller number of mutations produce the syndrome usually due to a founder gene effect, such as in South Africa.

Triglyceride as a risk factor for ischaemic heart disease in British men: effect of adjusting for measurement error.
M Egger, G D Smith, D Pfluger, E Altpeter, P C Elwood. *Atherosclerosis* 1999; **143**:275–84.

B**ACKGROUND. This study aimed to assess the influence of differential precision in the measurements of the correlated variables total cholesterol and HDL cholesterol on estimates of the risk of ischaemic heart disease associated with plasma TG levels. The results from two substudies of the Caerphilly Heart Disease Study, a prospective study of 2512 middle-aged men, were used to estimate the degree of imprecision (laboratory error and within-person variation) in measurements of TGs, total cholesterol and HDL cholesterol.**

I**NTERPRETATION.** In contrast to other cohort studies, TG concentration in the Caerphilly Heart Disease Study is associated with the risk of ischaemic heart disease independently of total and HDL cholesterol. This effect was pronounced after adjustment for measurement imprecision but was reduced when adjusted for other factors. Insulin resistance is probably the underlying metabolic disturbance.

Comment

The controversy about whether or not TGs are a risk factor for CHD is being resolved and it seems inescapable that they are. Intervention studies with drugs such as fibrates that lower serum TG levels with very little effect on LDL cholesterol clearly show a decrease in CHD risk. Meta-analysis of epidemiological studies has also shown that TGs are a risk factor |**5**|. However, they are a much more important risk factor than this suggests. This is because in epidemiological studies the importance of serum TG concentration was artificially degraded as a CHD risk factor in multivariate risk models after inclusion of HDL cholesterol with which it is strongly inversely correlated. Because the biological variation in serum TG levels is considerably greater than that of HDL cholesterol, the latter frequently emerged as explaining much of the CHD risk that should have been attributed to TGs. In the study by Egger *et al.* the effects of biological variation were controlled by estimating the true mean values of TG and HDL (and this really mirrors more closely clinical practice in which a decision is based on a series of measurements rather than the epidemiologist's approach in which a measurement is made on a single occasion only). The TGs emerged as a stronger risk factor than the HDL or serum cholesterol. The authors also concluded that insulin resistance may be the underlying metabolic disturbance associated with hypertriglyceridaemia when it is associated with CHD.

The important message here is that in the prediction of CHD risk, once the cholesterol

and HDL cholesterol have been included, TGs may not contribute a great deal more if the prediction is based on a multivariate equation from an epidemiological study such as Framingham. The risk is, however, likely to be an underestimate if it is established that the patient's TG levels are persistently elevated. Most importantly it should not be assumed that, because TG levels are not used in the risk prediction, it is not important to treat them, particularly in high-risk patients.

Increased prevalence of smaller and denser LDL particles in Asian Indians.

K R Kulkarni, J H Markovitz, N C Nanda, J P Segrest. *Arterioscler Thromb Vasc Biol* 1999; **19**(11): 2749–55.

B**ACKGROUND. There is evidence that Asian Indians are at an increased risk of CHD that cannot be attributed to the common risk factors. Individuals with small, dense LDL phenotype are also known to be at increased risk of CHD. This study examined whether the prevalence of smaller and denser LDL particles is increased in Asian Indians compared with white people.**

I**NTERPRETATION.** Small dense LDL type was significantly more prevalent in Asian Indians compared with white subjects. This increased prevalence appears to be due to increased TGs, fasting insulin being an important determinant of TG levels. In addition, fasting insulin was significantly increased in Asian Indians with small, dense LDL type compared with other Asian Indians, suggesting a significant role of insulin resistance. The increased prevalence of small, dense LDL observed in Asian Indians might contribute to their increased CHD risk.

Comment

Many patients with hypertriglyceridaemia and insulin resistance have an increased circulating level of LDL that is not apparent from measurements of serum cholesterol or LDL cholesterol. This is due to the presence of a small, dense LDL particle, which is depleted of cholesterol. It appears to be closely associated with CHD risk. In Asian Indians the increased risk of CHD associated with urbanization and migration is greater than can be explained in terms of the increase in cholesterol and blood pressure that they undergo. They are, however, exposed to a marked increase in insulin resistance and frank diabetes. This study suggests a likely mechanism by which these are linked with their increased CHD risk, because both the increased insulin resistance and its attendant hypertriglyceridaemia were closely correlated with the presence of small, dense atherogenic LDL. This study is part of a growing body of evidence for small, dense LDL

as a valuable predictor of CHD for which therapeutic intervention may be justified. At present methods for its detection are, however, too complex for its introduction into routine clinical practice.

The small, dense LDL phenotype and the risk of coronary heart disease: epidemiology, patho-physiology and therapeutic aspects.
B Lamarche, I Lemieux, J P Despres. *Diabetes Metab* 1999; **25**:199–211.

B**ACKGROUND. The presence of small, dense LDL particles has been associated with more than a threefold increase in the risk of CHD. The small, dense LDL phenotype is usually accompanied by hypertriglyceridaemia, reduced HDL cholesterol levels, abdominal obesity, insulin resistance and other metabolic alterations. Whether the small, dense LDL phenotype is an independent CHD risk factor is not yet clear. Individuals with three features of insulin resistance (elevated plasma insulin and apolipoprotein B concentrations, and small, dense LDL particles) seem to show a marked increase in CHD risk. The increased risk of CHD associated with having small, dense LDL particles may be significantly modulated by the presence/absence of insulin resistance, abdominal obesity and increased LDL particle concentration.**

I**NTERPRETATION.** Treatment of the small, dense LDL phenotype should aim at improving all features of the insulin resistance syndrome, particularly body weight loss and mobilization of abdominal fat, as well as reducing plasma TG levels. Interventions that reduce fasting TG levels will increase LDL particle size and contribute to reduced CHD risk, particularly if plasma apolipoprotein B is also reduced.

Comment

These authors summarize the evidence linking small, dense LDL with the metabolic abnormalities associated with insulin resistance and increased CHD risk. Both statins and fibrates can decrease small, dense LDL levels. Attention has frequently been drawn to the apparently greater decrease in CHD risk with statins than can be explained in terms of LDL cholesterol reduction and in the case of fibrates CHD risk is often decreased particularly in patients with hypertriglyceridaemia without any detectable decrease in LDL cholesterol. This may be due to the effects of both these classes of drugs on small, dense LDL, reduction in the concentration of which cannot be detected by measurement of serum or LDL cholesterol. We should thus be wary of those who seek effects of these drugs on non-lipoprotein parameters to explain their beneficial effects on CHD risk—their effect may well be to improve lipoprotein metabolism in ways that are not detected by the type of measurement of LDL made in most clinical trials.

References

1. Standing Medical Advisory Committee. The use of statins. London: Department of Health 1997 (11061 HCD Aug 97 (04)).
2. Isles CG, Ritchie LD, Murchie P, Norrie J. Risk assessment in primary prevention of coronary heart disease: randomised comparison of three scoring methods. *Br Med J* 2000; **320**:690–1.
3. Jones AF, Walker J, Jewkes C, Game FL, Bartlett WA, Marshall T, Bayly GR. Comparative accuracy of cardiovascular risk prediction in primary care patients. *Heart* 2001; **85**: 37–43.
4. Wood D, Durrington PN, Poulter N, McInnes G, Rees A, Wray R. Joint British Recommendations on prevention of coronary heart disease in clinical practice. *Heart* 1998; **80**(Suppl 2): S1–29.
5. Hokanson JE, Austin MA. Plasma triglyceride level is a risk factor for cardiovascular disease independent of high-density lipoprotein cholesterol level: a meta-analysis of population-based prospective studies. *J Cardiovasc Risk* 1996; **3**:213–19.

8
Effects of lipid-lowering drugs on non-lipid risk factors for coronary heart disease and other diseases

Introduction

The mind of the medical scientist can indeed be strange. There seems to have been almost as much interest in trying to find effects of statins that do not appear to be mediated through their effects on lipoprotein metabolism as in actually finding out what precisely they do to lipoprotein metabolism that is so beneficial. Clinical trials measure low-density lipoprotein (LDL) cholesterol using relatively crude methods suitable for large-scale analyses and what is reported as LDL cholesterol contains a wealth of LDL subclasses. Furthermore, there has been no validation of the Friedewald formula nor of the various methods of precipitating apolipoprotein (apo)B-containing lipoprotein in patients whose LDL composition has been altered by statin therapy. In clinical trials, effects on more subtle measures of lipoprotein biochemistry, if they are undertaken, are done on subgroups only. It is thus not altogether surprising that there are reports (often inconsistent) of effects of statins on coagulation factors, etc. and clinical outcomes that appear unrelated to LDL cholesterol changes as such. Some of these so-called pleiotropic effects should be viewed with caution. Nonetheless, statins could have the capacity to alter a variety of cellular metabolic processes. If those are postulated to occur outside the liver then there is, of course, also the difficulty of knowing whether the exposure of extrahepatic tissues to statins or their metabolites is high enough for such effects to occur *in vivo*.

The rapid improvement in endothelial function with pravastatin introduced following the clinical presentation of acute coronary syndromes reported by Dupuis *et al.* may explain some of the beneficial effects of statins on vascular outcome that seem to occur too early for them to have any appreciable effect on atheroma progression. The effect is highly likely to be in response to decreases in LDL or some lipoprotein parameter closely related to it rather than to a direct effect of statin. C-reactive protein (CRP) is attracting considerable attention as a risk factor for coronary heart disease (CHD) events. In a secondary prevention study Ridker *et al.* report that pravastatin diminishes its concentration whereas in another study by Certellero *et al.* fluvastatin failed to have the same effect. Besafibrate too did not lower CRP.

High-density lipoprotein (HDL) can protect LDL against oxidation *in vitro* and the enzyme paraoxonase (PON1) present only on HDL seems to mediate the effect probably by hydrolysis of phospholipid and chlolesteryl ester lipid peroxides |**1**|. Dietary constituents, such as used cooking fat and alcohol, have been shown recently respectively to decrease and increase serum PON1 activity. PON1 activity is decreased in

heterozygous familial hypercholesterolaemia (FH). Recently, studies have also shown that simvastatin can increase serum PON1 activity in heterozygous FH.

Cholesterol reduction rapidly improves endothelial function after acute coronary syndromes. The RECIFE (Reduction of Cholesterol in Ischaemia and Function of the Endothelium) Trial.

J Dupuis, J-C Tardif, P Cernacek, P Theroux. *Circulation* 1999; **99**: 3227–33.

B**ACKGROUND. Cholesterol lowering reduces coronary events. One mechanism could be improved endothelial function. In line with this hypothesis, this study investigates whether cholesterol reduction can result in rapid improvement of endothelial function after acute coronary syndromes.**

I**NTERPRETATION.** Patients with acute myocardial infarction or unstable angina and total cholesterol levels at admission≥5.2 mmol/l or LDL≥ 3.4 mmol/l were randomized to placebo (*n*=30) or pravastatin 40 mg daily (*n*=30) for 6 weeks. Brachial ultrasound was used to measure endothelium-dependent flow-mediated dilatation and response to endothelium-independent nitroglycerine. Changes in the levels of markers if platelet activation, coagulation factors and plasma endothelin levels were also assessed. Total and LDL cholesterol levels were similar at admission and before randomization in both groups. With pravastatin, but not with placebo, they decreased by 23% ($P<0.05$) and 33% ($P<0.01$), respectively. Flow-mediated dilatation was unchanged with placebo 5.43±0.74% (mean±SEM) to 5.84±0.81%, but nitroglycerine were similar during the time course of the study in the two groups. Markers of platelet activity, coagulation factors, and endothelin levels were not affected by pravastatin. Cholesterol reduction with pravastatin initiated early after acute coronary syndromes rapidly improves endothelial function after 6 weeks of therapy.

Comment

An interesting study that reveals how rapidly endothelial function can improve when statin treatment is initiated following an acute coronary syndrome. The most likely explanation is that this is due to LDL cholesterol lowering.

Long-term effects of pravastatin on plasma concentration of

C-reactive protein.
P M Ridker, N Rifai, M A Pfeffer, F Sacks, E Braunwald for the Cholesterol and Recurrent Events (CARE) Investigators. *Circulation* 1999; **100**: 230–5.

B**ACKGROUND. Elevated plasma concentrations of C-reactive protein (CRP) area associated with increased cardiovascular risk. We evaluated whether long-term therapy with pravastatin, an agent that reduces cardiovascular risk, might alter levels of this inflammatory parameter.**

I**NTERPRETATION.** CRP levels were measured at baseline and at 5 years in 472 randomly selected participants in the Cholesterol and Recurrent Events (CARE) trial who remained free of coronary events during follow-up. Overall, CRP levels at baseline and at 5 years were highly correlated (r=0.60, P<0.001). However, among those allocated to placebo, median CRP levels and the mean change in CRP tended to increase over time (median change, +4.2%; P=0.2 and mean change, ±0.07 mg/dl; P=0.04). By contrast, median CRP levels and the mean change in CRP decreased overtime among those allocated to pravastatin (median change, −17.4%; P=0.004 and mean change, −0.07 mg/dl; P=0.002). Thus, statistically significant differences were observed at 5 years between the pravastatin and placebo groups in terms of median CRP levels (difference, −21.6%; P=0.007), mean CRP levels (difference, −37.8%; P=0.002), and absolute mean change in CRP (difference, −0.137 mg/dl; P=0.003). These effects persisted in analyses stratified by age, body mass index, smoking status, blood pressure and baseline lipid levels. Attempts to relate the magnitude of change in CRP to the magnitude of change in lipids in both the pravastatin and placebo groups did not reveal any obvious relationships. Among survivors of myocardial infarction on standard therapy plus placebo, CRP levels tended to increase over 5 years of follow-up. In contrast, randomization to pravastatin resulted in a significant reduction in this inflammatory marker that was not related to the magnitude of lipid alterations observed. Thus, these data further support the potential for non-lipid effects of this agent.

Comment

CRP levels decreased in the pravastatin treated patients in this study of patients with established CHD. Many questions remain unanswered about CRP; however, it is emerging as a potentially important risk factor for CHD perhaps because it is simply a marker of inflammation or perhaps because it is more intimately involved in the production of cytokines promoting atherogenesis. It is probably too soon, however, to conclude that the effect of pravastatin on CRP is mediated through some non-lipid effect, because in a clinical trial such as this one the measurement of LDL cholesterol is a relatively crude affair and takes no account of other changes in lipoprotein metabolism, such as, for example, in the circulating levels of small, dense LDL that does not contribute to LDL cholesterol.

Effects of fluvastatin and bezafibrate combination on plasma fibrinogen, t-plasminogen activator inhibitor and C reactive protein levels in coronary artery disease patients with mixed hyperlipidaemia (FACT study). Fluvastatin alone and in combination treatment.
M Cortellaro, E Cofrancesco, C Boschetti, *et al. Thromb Haemost* 2000; **83**: 549–53.

B**ACKGROUND. The effects of fluvastatin and bezafibrate in monotherapy and in combination on plasma fibrinogen, t-plasminogen activator inhibitor (PAI-1) and CRP were studied in patients with coronary artery disease (CAD) and mixed hyperlipidaemia.**

I**NTERPRETATION.** The combined effects on fibrinogen and plasma lipids achieved by fluvastatin and bezafibrate combination treatment might be more useful than the simple reduction of cholesterol in preventing ischaemic cardiovascular disease. In this double blind trial 333 patients with stable angina pectoris or previous myocardial infarction or coronary revascularization and mixed hyperlipidaemia were randomized to fluvastatin 40 mg, bezafibrate 400 mg or a combination of the two drugs for 24 weeks. Plasma fibrinogen decreased significantly after combination treatments and after bezafibrate monotherapy but not after fluvastatin monotherapy. No significant changes were observed in PAI-1 and CRP plasma levels. Combination therapy significantly decreased both LDL cholesterol and triglycerides, and significantly increased HDL cholesterol.

Comment

Neither fluvastatin or bezafibrate altered CRP in this study. The decrease in fibrinogen seen here with bezafibrate has also been reported with other fibrates such as fenofibrate, ciprofibrate and clofibrate, although interestingly it does not occur with gemfibrozil |**2**|. Because this latter agent is equally effective at lowering tri-glycerides (the primary effect of fibrates) it seems likely that gemfibrozil has some hepatic effect in increasing fibrinogen production or that the other fibrates have an hepatic effect in decreasing its production.

Reduced postprandial serum paraoxonase activity after a meal rich in used cooking fat.

W H Sutherland, R J Walker, S A de Jong, *et al*. *Arterioscler Thromb Vasc Biol* 1999; **19**:1340–7.

B**ACKGROUND. PON1 is an enzyme associated with HDL in human serum that hydrolyses oxidized phospholipids and inhibits LDL oxidation, an important step in atherogenesis. In animals, adding oxidized lipids to the circulation reduces PON1 activity, and diets rich in oxidized fat accelerate the development of atherosclerosis.**

This randomized, cross-over study was designed to compare the effects, in 12 healthy men, of a meal rich in oxidized lipids with a control meal, rich in similar but non-oxidized fat, on postprandial serum PON1 (arylesterase) activity, on the peroxide content of LDL, and on LDL susceptibility to copper-ion catalysed oxidation.

I**NTERPRETATION.** The results suggest that in the postprandial period after a meal rich in used cooking fat, the enzymatic protection of LDL against accumulation of peroxides and atherogenic oxidative modification may be reduced, possibly due to factors associated with apoA1, without acutely affecting the intrinsic resistance of LDL to *in vitro* oxidation.

Comment

Serum PON1 activity varies greatly between different populations and different individuals within these populations. These differences are only partly explicable on the basis of the different genetically determined isoenzymes of PON1. Nutritional effects are highly likely to be important and this is one of the first studies to show the potential for such effects. It was unfortunate that the authors studied the susceptibility of LDL to oxidation in the absence of HDL, because PON1 is exclusively located on HDL. None is present in LDL.

Daily moderate alcohol consumption increases serum paraoxonase activity; a diet-controlled, randomised intervention study in middle-aged men.

M S van der Gaag, A van Tol, L M Scheek, *et al*. *Atherosclerosis* 1999; **147**:405–10.

B**ACKGROUND. Moderate alcohol consumption is associated with a reduced risk of CHD. Part of this inverse association may be explained by its effects on HDL. It has been suggested that PON1, an HDL-associated enzyme protects against LDL oxidation. The effects on serum**

PON1 activity, of moderate consumption of red wine, beer or spirits compared with mineral water, were assessed in a diet-controlled, cross-over study in 11 healthy middle-aged men who consumed each beverage for 3 weeks.

I**NTERPRETATION.** The results suggest that increased serum PON1 may be one of the mechanisms underlying the reduced CHD risk in moderate alcohol consumers. Fasting PON1 activity was higher after intake of wine, beer and spirits than after water consumption but did not differ significantly between the alcoholic beverages. The increases in PON1 activity were strongly correlated with increases in serum HDL cholesterol and apoAI.

Comment

The observation that alcohol can increase serum PON1 activity may explain some of the population and interindividual differences in the activity of this enzyme. It would be of interest to know whether these alcohol-induced changes in PON1 activity increased the capacity of HDL to protect LDL against oxidative modification.

Effect of simvastatin therapy on paraoxonase activity and related lipoproteins in familial hypercholesterolemic patients.

M Tomas, M Senti, F Garcia-Faria, *et al. Arterioscler Thromb Vasc Biol* 2000; **20**:2113–19.

B**ACKGROUND. Human PON1 is a calcium-dependent esterase closely associated with HDL-containing apoAI, which confers antioxidant properties to HDL, and has been implicated in the pathogenesis of atherosclerosis. Low PON1 activities have been found in FH and diabetes mellitus. We studied the effect of the lipid-lowering drug simvastatin on serum PON1 activity, on apoAI-containing and apoB-containing lipoproteins, and on lipid peroxide concentrations in 64 unrelated FH patients, and analysed the influence of the PON1-192 and PON1-55 genetic polymorphisms on the response of PON1 activity to simvastatin therapy.**

I**NTERPRETATION.** The major effect of simvastatin on lipid traits was to decrease serum cholesterol, LDL cholesterol, and lipid peroxide concentrations. We conclude that simvastatin may have important antioxidant properties through increasing serum PON1 activity, perhaps as a consequence of reducing oxidative stress, by a mechanism independent of apoAI-containing lipoprotein concentration and without the influence of PON1–192 and PON1–55 genetic polymorphisms. Further studies are needed to clarify

the mechanisms involved.

Comment

Serum PON1 activity was increased regardless of PON1 genotype by simvastatin therapy in patients with FH. Serum PON1 activity tends to be low in FH patients. Given the greatly increased circulating LDL levels in FH patients, the low PON1 activity, by diminishing the capacity of their HDL to protect LDL against oxidative modification, could contribute to their propensity to atherosclerosis.

Inhibitors of hydroxymethylglutaryl-coenzyme A reductase and risk of fracture among older women.

K A Chan, S E Andrade, M Boles, *et al. Lancet* 2000; **355**:2185–8.

B**ACKGROUND. Statins increase new bone formation in rodents and in human cells *in vitro;* their use is associated with increased bone mineral density of the femoral neck. This population-based case-control study at six health-maintenance organizations in the USA further investigates the relationship between statin use and fracture risk among women aged 60 years or older (928 cases and 2747 controls).**

INTERPRETATION. Statins seem to be protective against non-pathological fractures among older women (odds ratio 0.48). No association was found between fracture risk and fewer than 13 doses of statins or between fracture risk and use of non-statin lipid-lowering drugs. These findings are compatible with the hypothesis that statins increase bone mineral density in human beings and thereby decrease the risk of osteoporotic fractures.

Comment

At last a potentially clinically important and indisputably pleiotropic effect of statins that is most unlikely to involve lipoprotein effects.

HMG-CoA reductase inhibitors and the risk of hip fractures in elderly patients.

P S Wang, D H Solomon, H Mogun, J Avorn. *JAMA* 2000; **283**(24): 3211–16.

B**ACKGROUND. Recent animal studies have shown that statins substantially increase bone formation. Whether statin use in humans results in clinically meaningful bone formation or reduces the risk of osteoporotic fractures is not known. The aim of this case-control study of 6110 patients (1222 cases of hip fracture) aged 65 years or older was to determine whether statin use is associated with a reduced risk of hip fracture.**

I**NTERPRETATION.** The findings support an association between statin use by elderly patients and reduction in the risk of hip fracture. Use of statins in either the preceding 180 days or the preceding 3 years was associated with a significant reduction in the risk of hip fracture, even after controlling for confounding factors. No significant relationship was observed between use of non-statin lipid-lowering agents and hip fracture risk. There were clear relationships between the degree of reduction in hip fracture risk and the extent of statin use. Controlled trials are needed to exclude the possibility of unmeasured confounders.

Comment

Again a fascinating observation. The effect appears quite marked, but could be confounded. Clinical trial evidence in a population at high risk of fractures would be of great interest. Many other substances besides cholesterol are synthesized from mevalonic acid, the production of which is inhibited by statins, including isoprenoids and other cellular regulators.

References

1. Durrington PN, Mackness B, Mackness MI. Paraoxonase and atherosclerosis. *Arterioscl Thromb Vasc Biol* 2001; **21**:473–80.

2. Durrington PN, Mackness MI, Bhatnagar D, Julier K, Prais H, Arrol S, Morgan J, Wood GNI. Effects of two different fibric acid derivatives on lipoproteins, cholesteryl ester transfer, fibrinogen, plasminogen activator inhibitor and paraoxonase activity in type IIb hyperlipoproteinaemia. *Atherosclerosis* 1998; **138**:217–25.

9
Coronary heart disease risk factors and primary prevention

Introduction

Cholesterol, blood pressure and cigarette smoking are mutable coronary heart disease (CHD) risk factors, which when modified can change risk. Other CHD risk factors, although they may not themselves be modifiable also determine overall CHD risk and thus the degree of benefit that can result from blood pressure of cholesterol reduction. There is agreement that pre-existing CHD or other atherosclerotic disease identifies patients whose risk is so high that they should receive active treatment to decrease serum cholesterol to below 5 mmol/l or blood pressure to below 140/85. In primary prevention of CHD risk factors other than blood pressure or cholesterol must often be considered to estimate overall CHD risk before deciding to introduce antihypertensive or statin therapy. Many recent guidelines for the primary prevention of CHD have included methods for the identification of patients with the highest priority for drug treatment of raised blood pressure or cholesterol. These are based on an estimate of absolute coronary or cardiovascular risk from the equation derived from the Framingham study. The method of choice is a computer program based on this equation. Charts and tables have also been devised to obviate the need for a computer and of these the charts produced by the Joint British Societies has the greatest clinical utility.

Coronary risk factors

Pre-existing clinical coronary heart disease

Without doubt the most potent factor predicting the likelihood of a future coronary event is that clinically overt CHD is already present. In the British Regional Heart Study half of the coronary events observed occurred in men with pre-existing CHD |**1**|. This is not to say that the effect of pre-existing CHD in determining the risk of a future event excludes the influence of other risk factors. It does not. Indeed the effects of serum cholesterol or high-density lipoprotein (HDL) cholesterol, for example, are even greater in patients with pre-existing CHD |**2,3**|. It is interesting to speculate about whether the increased susceptibility of patients with CHD to these risk factors existed before the development of clinically overt disease. It is likely that it did and, had such susceptible individuals been identifiable at that stage, they would perhaps have benefited from intensive intervention to modify their risk factors. Guidelines for CHD prevention do not usually advocate the calculation of risk in patients with clinical evidence that CHD is already

present. This is because of the strong clinical trial evidence that statins, for example, will decrease mortality in this group of patients |**4–6**| and do so cost-effectively |**7**|. In two of these trials |**4, 6**| the relative decrease in risk of recurrent CHD events was the same regardless of the initial cholesterol level and in the other |**5**| there was evidence of some attenuation of the statin-induced risk reduction when the initial cholesterol was≤4.8 mmol/l. Thus the Joint British Guidelines advocate the use of statins in any patients with pre-existing CHD when the serum cholesterol exceeds 5 mmol/l |**8**|. There is, however, still considerable reluctance on the part of clinicians to prescribe statins to patients with cholesterol levels of<6.5 mmol/l |**9**|. There has never been a trial of blood-pressure lowering in established CHD. Nonetheless, such treatment is advocated on the basis of the high CHD risk in these patients and extrapolation from primary prevention trials.

Other pre-existing non-coronary manifestations of atherosclerosis

Patients with previous cerebral infarction and symptomatic femoral atherosclerosis are more at risk of dying of CHD than any other cause and their risk is similar to that of myocardial infarction (MI) survivors |**10**|. Thus, although the evidence is incomplete that lipid-lowering therapy decreases the likelihood of progression of cerebrovascular or peripheral arterial disease, the evidence that it decreases coronary events is generally considered to justify the use of such therapy in these patients |**8**|. An exception might be patients whose stroke was the result of cerebral haemorrhage, the risk of which may be increased by spontaneously occurring low cholesterol levels |**11**|.

Age

After pre-existing CHD, the most potent risk factor for a coronary incident is age |**12**|. CHD risk rises exponentially with age in both men and women. The incidence of CHD in women never achieves that in men of the same age, probably because their HDL cholesterol is higher than men throughout life (see later). There is no abrupt increase in CHD in women at the menopause and, although serum cholesterol usually rises around the time of the menopause, HDL cholesterol typically does not decline |**13**|.

The substantial effect of age means that prevention strategies based on absolute risk assessment will inevitably be weighted towards the elderly unless some upper age limit is imposed. (Absolute risk is the number of events per 100 people whereas relative risk is the number of times risk is increased above the average for people of that age and gender.) Most recommendations for statin therapy impose the age limitation that applied to entry of patients to the successful clinical trials. Thus, for example, in secondary prevention, patients as old as 75 years have been randomized |**5,6**| and in primary prevention trials of statins the oldest patients randomized were 73 years old |**14**|. This approach, while economically convenient is, however, scientifically unsound, because within the trials the relative decrease in CHD incidence was similar in younger and older patients |**4–6,14,15**| and because of the greater absolute risk in older patients the numbers who required treatment to prevent an event (i.e. the number who benefit from treatment), therefore, became progressively less with advancing age.

In the case of antihypertensive therapy most recent guidelines seek to impose no upper

age limit for treatment |**16**|.

Cholesterol

Serum cholesterol takes centre stage among the cast of mutable risk factors for CHD, and it is hard to comprehend the controversy in which until recently it was shrouded. Anitschow as early as 1913 wrote that 'there can be no atherosclerosis without cholesterol' |**17**|. He based his view on animal experiments, but there can be no doubt that subsequent epidemiology has confirmed his prescience. There is an enormous international variation in coronary rates, the highest being in North America, northern Europe, Australia and New Zealand, with rural Africa and most of Asia having much lower rates. Southern European countries are intermediate. The risk factor that is most closely associated with these differences in CHD incidence is serum cholesterol |**18**|.

Other risk factors such as cigarette smoking, diabetes mellitus and hypertension have little impact on CHD risk in countries such as Japan where the population cholesterol levels are low. Cholesterol thus appears to be the permissive factor without which these other factors operate to a much lesser extent. This does not, of course, mean that they are not important to eliminate if CHD rates are to be decreased in high cholesterol societies such as Britain, but neither should reliance be placed on their correction to the exclusion of cholesterol lowering. Furthermore, they will come to assume greater importance in low cholesterol countries as diets rich in fat are adopted, particularly saturated fat, and energy intake in the form of carbohydrate diminishes. That is exactly the dietary change, which has, for example, taken place in Britain during the twentieth century, a time when coronary death rates have risen to epidemic proportions |**19**|. Acute MI was evidently uncommon before that, not being diagnosed by a physician until 1912 |**20**|.

Clinical trial evidence that lowering cholesterol reduces CHD incidence whether by diet or by fibrate or bile acid sequestrating drug therapy or partial ileal bypass existed before the advent of statins |**21,22**|. There was also evidence from metaanalysis that in secondary prevention all-cause mortality was decreased |**21,22**|, but this was disputed particularly in primary prevention |**23**|. The clinical development of statin drugs, however, coincided with a greater understanding of clinical trial design, particularly statistical power, and this led to trials that establish beyond doubt that CHD can be decreased by cholesterol reduction without adverse effects on non-cardiac mortality |**4–6,14,15**|. These studies establish that statin therapy decreases CHD risk by one-third and stroke risk by 20–30% in the first 5 years of therapy |**11,14**|. Unfortunately, in countries such as Britain, hypercholesterolaemia is so prevalent that for economic reasons it can seldom be the only factor leading to the prescription of a statin. Two-thirds of men and women between the age of 25 and 64 years in Britain have serum cholesterol levels exceeding 200 mg/dl (5.2 mmol/l) |**24**|, a level that seems to separate countries where CHD is relatively uncommon from those where it is a major problem. Britain has consistently and, incredibly for a country where CHD is its major health problem, failed to institute any nutritional policy to overcome its high prevalence of hypercholesterolaemia. Hypercholesterolaemia is too common in Britain to tackle clinically, because of the high cost of screening and providing dietary advice and statin therapy. Most nations, including Britain, have adopted the strategy that statin treatment

should be limited to those members of the population who can benefit most |**8,25–27**|, defined according to the number of people who must receive treatment to prevent an event |**28**|. In order to follow this strategy the clinician would need to be able to predict the absolute risk of an event in a particular patient. If this were possible, then a decision could be made as to whether to institute treatment based on whether a decrease, of say one-third, in CHD risk was worth the inconvenience and cost to the patient of taking medication or, in the case of a state health service, whether the likelihood that an event would be prevented justified the cost when a similar spending in some other area of clinical practice might benefit more people. The latter argument is thus about cost-effectiveness. Thus, for example, it could be argued that treating patients at a 2% and greater risk of a CHD event each year would be cost-effective compared with many other medical interventions, including the treatment of mild-moderate hypertension with a generic thiazide |**7,29–32**|. This argument was, however, shifted in the case of statin recommendations in Britain away from issues of cost-effectiveness towards issues of total cost to the NHS |**33**|. The reason for this is that not only is high cholesterol prevalent in Britain, but so also is high CHD risk. It was calculated that 26.9% of men and 8.6% of women aged 35–69 years were at≥1.5% annual CHD risk |**34**| (Table 9.1). It was, therefore, proposed that a decision should be made about how much of the NHS budget should be devoted to statin therapy and then to work backwards to the degree of risk that this level of expenditure would permit to be treated. This cannot be seen as entirely logical unless the purpose of the NHS is to ration CHD prevention in order to protect its current level of expenditure on diseases that affect fewer people and are less serious than CHD and for which the treatments are less cost-effective. Nonetheless, the Standing Medical Advisory Committee to the Minister of State for Health adopted apparently this position when it accepted the premise that NHS expenditure on statin therapy in primary CHD prevention should be the same as that in secondary prevention (assuming that every patient with CHD aged less than 70 years received statin treatment) |**33**|. This led to an annual CHD risk of 3% being adopted as the indication for statin therapy in primary prevention in men (5.7% of men aged between 35 and 69 years in England are thought to have this level of risk;

Table 9.1 Estimate of the prevalence of coronary risk (in % per annum, i.e. the number of events per 100 people each year) of the population of England aged 35–69 years not already known to have CHD |**34**|

Men (CHD risk/per year)		**Women (CHD risk/year)**	
>3%	>1.5%	>3%	>1.5%
5.7%	26.9%	0.4%	8.6%

Table 9.1). Virtually no women, however, achieve this level of risk, unless they have symptomatic CHD. Thus the adoption of the 3% annual risk for statin therapy in primary prevention in women means that the use of statin therapy in women would be confined to secondary prevention. This would lead to the combined expenditure on men and women

for primary prevention being less than that spent on secondary prevention. The degree of annual CHD risk targeted for statin therapy in primary CHD prevention in Europe as a whole in the Joint European guidelines was 2% |**25**|. The Joint British Societies' recommendations were that an annual CHD risk of 3% for statin therapy should be regarded as the minimum level of acceptable care and that progress should be made to reducing this down to 1.5% as soon as resources permitted |**8**|. Both the Joint British and European guidelines are thus more in keeping with the scientific evidence of benefit.

Familial hypercholesterolaemia

All the recommendations make familial hypercholesterolaemia (FH) a special category that will generally require statin therapy from at least the age of 20 years in men and 30 years in women |**13**|. Familial hypercholesterolaemia is dominantly inherited and is associated with tendon xanthomas, although these may not have developed by the age at which statin therapy should ideally be commenced. Family screening should therefore be undertaken when an older patient with the clinical syndrome is encountered. Usually, the cholesterol level exceeds 9 mmol/l in adulthood and 6.0 mmol/l in childhood |**13,35**|. CHD risk is greatly increased, probably because the high cholesterol is present from birth and untreated most affected men and over half of the affected women will have died or have symptomatic CHD before the age of 60 years. Referral to a Lipid Clinic is generally to be recommended.

High blood pressure

The latest recommendations for the treatment of high blood pressure from the British Hypertension Society |**16**| are that the absolute cardiovascular risk (CHD and stroke) should be assessed before making the decision to introduce antihypertensive therapy for primary prevention in mild-moderate hypertension (systolic 140–159 and diastolic 90–99 mmHg) for the reasons discussed earlier in the context of cholesterol. In the case of antihypertensive medication an annual CHD risk of 1.5% (or an annual cardiovascular risk of 2%) is considered high enough. The cost-effectiveness of treating mild-moderate hypertension even with a generic thiazide is not particularly high compared with statin therapy |**31,32**| so it is hard to comprehend why on any grounds, other than total cost, a distinction should exist between the absolute risk to be targeted for blood pressure treatment as opposed to statin therapy. This is why the Joint British Societies' guidelines suggested that we should eventually aim to target the same absolute CHD or cardiovascular disease (CVD) risk for both antihypertensive and statin therapy |**8**|.

Clinical trial evidence that lowering blood pressure will prevent CHD is restricted to primary prevention trials and results of these with respect to CHD prevention are less consistent than the statin trials. Nonetheless, a meta-analysis of trials of antihypertensive drugs shows an average reduction in CHD risk of 16% |**36**|. It is, of course, the case that antihypertensive treatment is more effective in decreasing the relative risk of stroke, which in the same meta-analysis, for example, was reduced by 38%. It is frequently, however, not realized that in all but the most extreme cases of hypertension CHD risk outweighs stroke risk several-fold. Thus the major benefit from antihypertensive therapy

in absolute terms (i.e. number of events prevented per patient treated) will actually result from the 16% decrease in CHD events. Furthermore, because statin therapy decreases CHD risk by at least double this amount |**11,14**| statin treatment is likely to be more clinically effective than antihypertensive therapy even in mild to moderate hypertension. Take the example of a man aged 55 years whose blood pressure is 180/110 mmHg who is a non-smoker and does not have diabetes. Let us give him a cholesterol of 7.8 mmol/l and an HDL cholesterol of 1.4 mmol/l. His CHD risk over the next 10 years is 21% and his stroke risk 6%, giving him a CVD risk of 27%. He qualifies for antihypertensive therapy because of his raised blood pressure |**16**|, but not for lipid-lowering therapy according to the Standing Medical Advisory Committee, and British Hypertension Society guidelines (CHD risk ≥30% over 10 years) |**16,33|,** although he would be considered for it according to the Joint European and Joint British recommendations (CHD risk ≥20% or ≥15% over 10 years, respectively) |**8,25**|. Antihypertensive therapy alone would only decrease his CHD risk to 18% and his stroke risk to 4% (overall CVD risk 22%) |**36**|. On the other hand a statin would decrease his CHD risk to 14% and his stroke risk to 5% (overall CVD risk 19%) |**11**|. Prescribing both antihypertensive and lipid-lowering therapy would decrease his CHD risk to 12% and his stroke risk to 3% (CVD risk 15%).

These calculations were made using the computer program based on the Framingham risk equation |**12**| issued with the Joint British Societies' guidelines |**8**| (see later).

Cigarette smoking

Smoking is a major CHD risk factor. It is universally agreed that stopping smoking decreases this risk. It would be wrong to dissent from this view, but it does urge us to re-examine, whether the type of 'evidence-based medicine' in which randomized clinical trials are the most highly prized form of evidence, which it is fashionable to accept, is necessarily going to yield the greatest benefit. The evidence, for example, that smoking cessation is beneficial is exclusively extrapolated from observational trials of people who choose to continue to smoke and who choose to stop. It is only logical, therefore, to give at least equal prominence to the treatment of high blood pressure and cholesterol, which is strictly 'evidence-based', as to the advice to stop smoking. The major clinical importance of smoking in CHD prevention is that, together with cholesterol and blood pressure, it is not only a predictive risk factor, but also a mutable one.

Serum high-density lipoprotein cholesterol

Serum HDL cholesterol is inversely associated with CHD risk and its omission from risk prediction causes considerable error |**37**|. In women and the elderly it is a stronger predictor of risk than serum cholesterol |**38**|: indeed it is generally pointless to measure serum cholesterol without measuring HDL cholesterol, if the test is being done to predict CHD risk. Serum HDL cholesterol is typically lower in men than women from puberty onwards |**13**|. It does not generally decline at the menopause in women, which may explain why their CHD risk continues to be lower than that of men, although their serum cholesterol postmenopausally is on average greater than that in men of a similar age. The lower levels of HDL in men are more a consequence of androgens, rather than the higher

HDL in women occurring as the result of oestrogen.

There is an abundance of evidence that the best way to use the information contained in serum cholesterol and HDL cholesterol in the prediction of CHD risk is to express them as a ratio |**12,39**|. It is perhaps, therefore, somewhat illogical that indications and targets for cholesterol-lowering therapy even in the most recent guidelines continue to be couched in terms of LDL cholesterol (calculated according to the Friedewald formula |**40**| i.e. LDL cholesterol=total cholesterol−[tri-glycerides (TG)/2.19+HDL cholesterol] mmol/l).

Diabetes

Diabetes is associated with dyslipidaemia and with hypertension, but even after adjustment for these factors continues to exert an independent effect in predicting CHD risk |**12,41,42**|. This does not necessarily mean that there are specifically diabetic factors that need to be corrected in order to decrease CHD risk in diabetes. Indeed the opposite may be the case with diabetic patients often experiencing benefit from statin treatment (perhaps indicating they are more susceptible to cholesterol) |**43,44**| whereas they show no significant decrease in CHD risk from improvements in specifically diabetic factors such as glycaemic control |**45,46**|. Treatment of hypertriglyceridaemia may prove of particular importance in diabetes, but results of trials to demonstrate this are still awaited |**47**|.

The dyslipidaemia (raised TGs and low HDL) of diabetes and the increased CHD risk in type 2 diabetes may be present for many years before glycaemia of diabetic proportions develops |**48**|. Indeed, CHD may occur before glycaemia develops

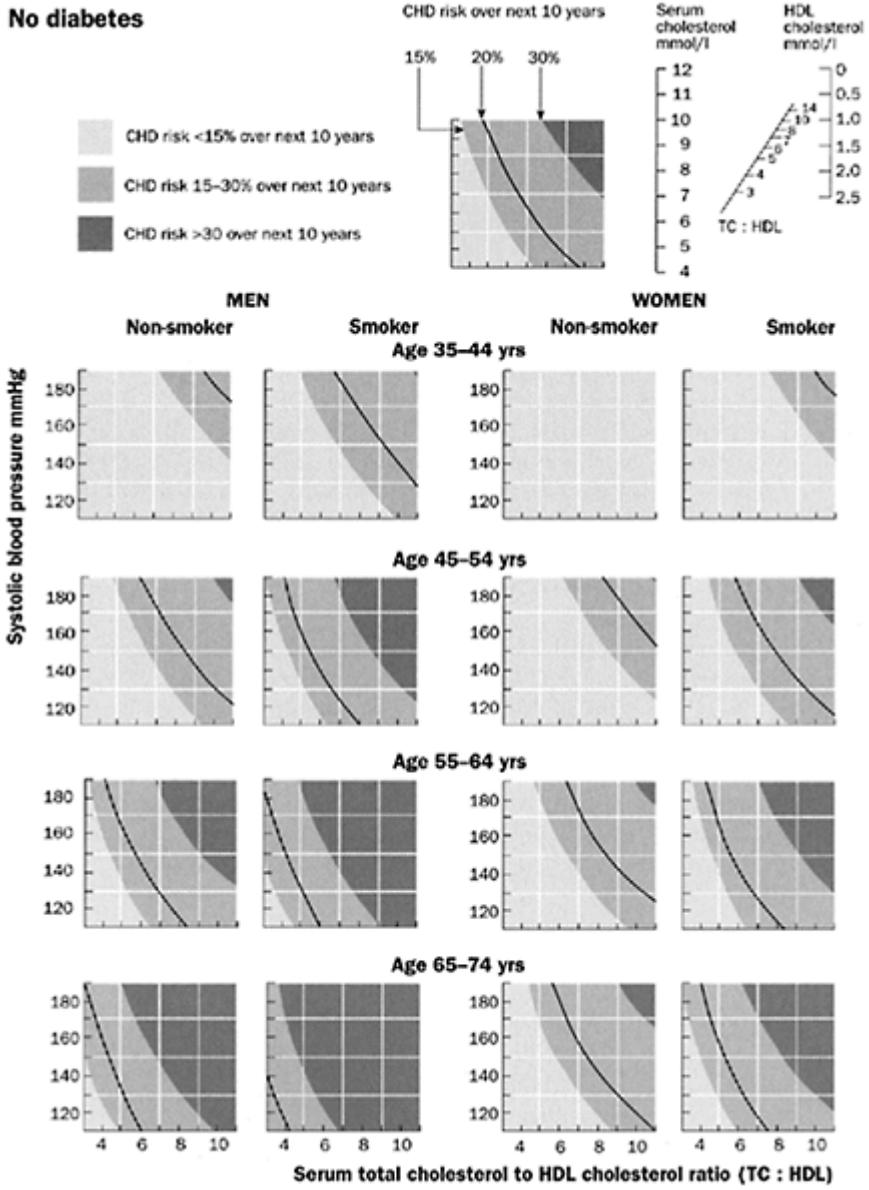

Fig. 9.1 The British Heart Foundation Joint British Societies' chart for the prediction of coronary risk in non-diabetic men and women (© the University of Manchester).

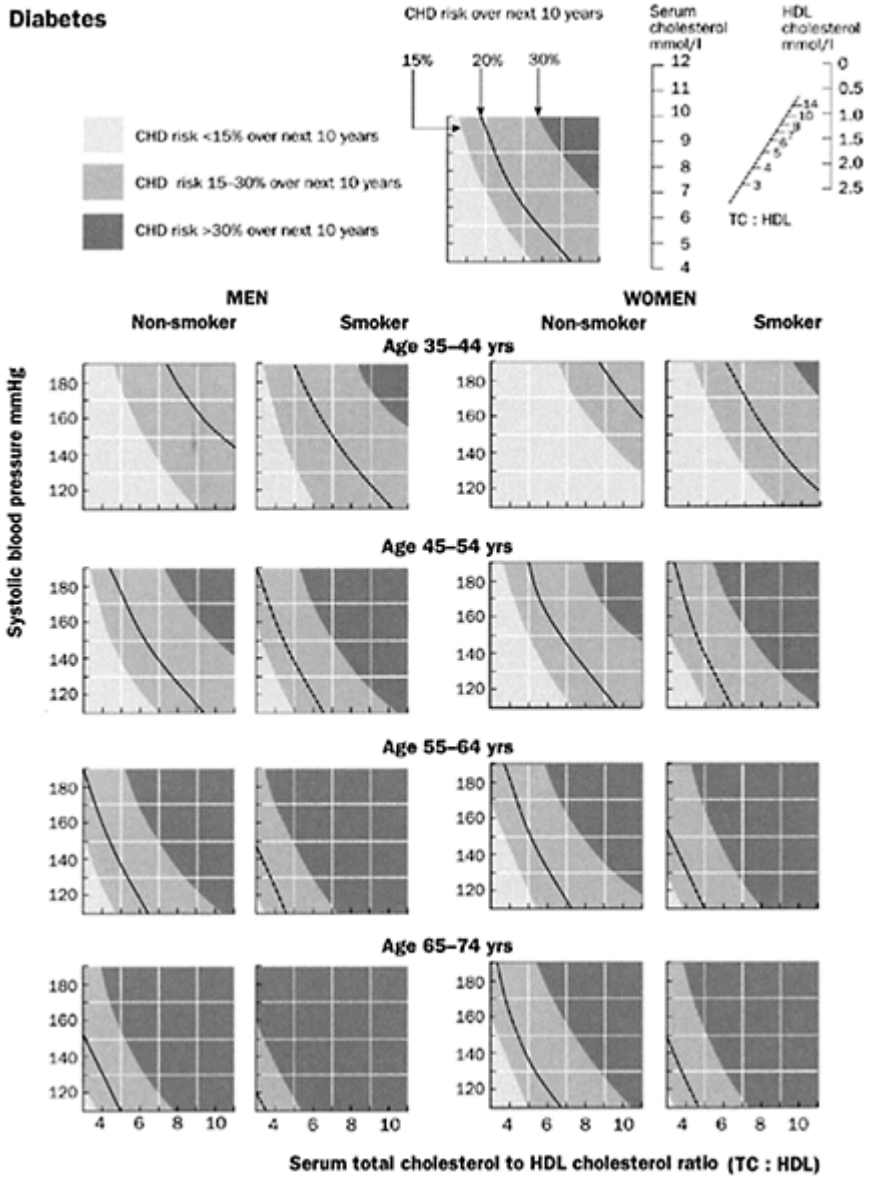

Fig. 9.2 The British Heart Foundation Joint British Societies' chart for the prediction of coronary risk in men and women with type 2 diabetes mellitus (© the University of Manchester).

explaining the high incidence of diabetes in the years following MI |**49**|. Insulin resistance and hyperinsulinaemia are closely associated with hypertriglyceridaemia and low HDL |**50**| and both are present in people destined to become diabetic many years

before they do so |**48**|, perhaps explaining their predisposition to CHD even at that stage.

As diabetes develops the HDL cholesterol declines in women to low levels |**51**| more typical of men and this may be the main reason that they lose the relative protection from CHD enjoyed by most other women.

Family history

It is the everyday experience of clinicians that patients presenting with CHD at an early age often have a family history of premature CHD. This is borne out by a number of studies |**13,38**|, but because hyperlipidaemia, high blood pressure, diabetes and cigarette smoking run in families, as the result of shared habits and genes, it is difficult to know whether family history makes a contribution to risk independently of these and is thus in itself of value in predicting CHD risk. Perhaps the most comprehensive study suggesting it does is a prospective study underway in Munster [the Prospective Cardiovascular Münster (PROCAM) study] |**38**|. The definition of what constitutes a family history of CHD is notoriously difficult, because the age at which it occurs in first-degree relatives is critical. The definition employed in PROCAM was a known MI in a first-degree relative before the age of 60 years.

Triglycerides, atherosclerosis and regression dilution bias

The other problem about risk factors, which is not commonly appreciated, is that a factor that may be important in the aetiology of atherosclerosis may not itself be a particularly good predictor of risk in multivariate analysis. Generally, this occurs when risk factors are correlated with each other, but vary in their biological and/or analytical variation |**52,53**|. A good example of this is the serum TG concentration, which is highly inversely correlated with HDL cholesterol, but shows substantially greater variation. In univariate analysis serum TG levels are often correlated more closely with CHD risk than cholesterol |**54**|, but with multivariate analysis they explain only a small, often insignificant proportion of CHD risk |**55**| when HDL cholesterol is included. In epidemiological studies variables are generally measured on a single occasion. Thus when variation is considerable a high or low value encountered on a single occasion is more likely to misclassify an individual whose true mean value is lower or higher, respectively, than is the case for a variable with less variation. This has the effect of flattening the slope of the relationship between a risk factor with high variation, such as the TG concentration, and CHD risk. This effect is known as regression dilution bias |**36,56**| and, when allowance is made for it, TGs can assume an even greater predictive power than HDL cholesterol |**52,57**|. Thus hypertriglyceridaemia, is highly likely to be involved in the aetiology of CHD and its treatment may decrease risk |**58**|, but this would not be evident from multivariate analysis.

This effect of regression dilution bias is important more often than is appreciated in the wider context of multiple risk factor equations based on epidemiological studies, because guidelines quite rightly emphasize the importance of basing treatment decisions (which may be life-long and so should be based on secure data) on more than one measurement of lipids or blood pressure. This, however, does mean that the risk attached to a raised

blood pressure or cholesterol value, which is the average of several measurements (and is thus more accurate and less variable) is going to be greater than that predicted by a risk equation, which was derived from measurements made on a single occasion |**12**|.

Newer risk factors for atherosclerosis

There are literally hundreds of factors that have been reported to be associated with CHD either in prospective studies or case-control studies. Many of these, it has been claimed, will permit the more accurate prediction of CHD risk. However, such claims frequently rest on case-control evidence, which is much weaker than results obtained in prospective studies or if prospective studies have been undertaken, they have not included one or more of the major CHD risk factors already discussed so that the part of the variation in CHD risk apparently explained by the potential new risk factor may be better described by the risk factor omitted. Most often the established risk factor omitted has been HDL cholesterol. Genetic hyperlipidaemias, such as FH, family history, insulin-dependent diabetes mellitus (IDDM), proteinuria and renal failure, and ethnicity, undoubtedly do increase CHD risk independently of the risk factors included in the Framingham risk equation |**12**|. It is, however, uncertain to what extent, if at all other potential risk factors for CHD, such as fibrinogen |**59**|, lipoprotein(a) |**60**|, homocysteine |**61**| or coronary artery calcification detected by ultrafast computed tomography |**62**| would contribute to risk prediction once the Framingham model has been applied. This, of course, does not mean they are unimportant for the aetiology of CHD as has been explained with regard to TGs.

Methods for coronary heart disease risk prediction

Guidelines for CHD prevention have generally relied on risk predictions from the risk equation based on the results of the Framingham Study |**12**|. It might at first seem odd that methods of risk prediction in Britain |**8**| and other European countries |**25**| should be based on the outcome of a prospective study begun 50 years ago and conducted largely in middle-class Americans from a single town. However, few other prospective studies have included a range of variables that match those routinely available to the clinician. Thus the equation from Framingham has as its variables the serum cholesterol to HDL cholesterol ratio, systolic (or diastolic blood hypertrophy on ECG criteria. The more recent PROCAM equation |**42**|, which also pressure), age, gender, smoking (yes/no), diabetes (yes/no) and left ventricular includes the Framingham variables (except LVH) as well as family history, TGs and angina, is based on German men, but there have been too few events in women as yet for a risk prediction equation to be devised for them. Thus for good or ill there are at present no alternatives to the use of the Framingham equation. In men the coronary event rates observed in PROCAM |**42**| and WOSCOPS |**63**| do agree reasonably well with the equation from Framingham.

The Framingham equation is too complex for the routine calculation of risk unless a programmable calculator or a computer is available. A computer program to run on Windows Excel was issued with the Joint British Guidelines |**8**| and is to be found on the British Hypertension Society and the British National Formulary websites

(www.hyp.ac.uk/bhs/management.html and http://bnf.org respectively). Generally, however, guidelines have been issued with simplified versions of the equation in the form of tables or charts. The Sheffield tables |**34**| recommended by the Standing Medical Advisory Committee to the Chief Medical Officer of Health in Britain |**33**| and the European Atherosclerosis Society (EAS) chart |**25**| included serum cholesterol rather than the serum cholesterol to HDL cholesterol ratio. This was a serious omission in the case of the original Sheffield tables because they underestimated risk in a high proportion of high-risk patients |**37**|. Low serum HDL clusters with other CHD risk factors. Women and diabetic patients fare exceptionally badly in the Sheffield tables. This is accentuated because of the high level of CHD risk (3% per annum) they were designed to detect |**37**|. The EAS chart which also omits HDL underestimates risk less frequently, partly because of the lower annual CHD risk of 2% recommended for statin treatment in the rest of Europe as opposed to Britain. However, 32.1% of high-risk patients cannot be accommodated on the axes of the chart |**37**| and the recommendations accompanying the chart are unclear about how these should be managed.

Two charts employ the serum cholesterol to HDL cholesterol ratio as one axis and blood pressure as the other. These are the New Zealand charts |**27**| and the Joint British Guidelines charts |**8**|. The Sheffield Risk table has also been revised with the serum to HDL cholesterol ratio replacing total cholesterol |**64**|. The various methods currently recommended have been compared with respect to how frequently they agree with the Framingham equation |**12**| (upon which they are based) programmed into a computer |**65, 66**|. Both the full Joint British chart published in *Heart* |**8**| and the version |**67**| produced for the British Heart Foundation leaflet entitled Joint British Recommendations on Prevention of Coronary Heart Disease in Clinical Practice (Part 2) Calculating absolute risk for primary prevention, Fact file 9/99, now published in the British National Formulary, were included in our comparison. The Joint British chart and the new Sheffield table could be applied to the greatest proportion of patients (Table 9.2). The large proportion of patients whose blood pressure or lipid values could not be accommodated on the Joint European and New Zealand Charts (Table 9.2) is likely to limit their usefulness in clinical practice. Agreement between the risk prediction of the charts and tables and the risk equation upon which they were based |**12**| was least good for the Joint European and New Zealand Charts (Table 9.3). In both cases this was largely because of overestimation of risk. In the case of the New Zealand charts this is not

Table 9.2 Proportion of patients (*n*=386) referred to a Lipid Clinic whose risk factors were outside the range covered by charts or tables for coronary or cardiovascular risk prediction

Joint British charts *(Heart)* |**8**|	11.9%
Joint British charts (British Heart Foundation) |**67**|	20.5%
New Zealand charts |**27**|	40.4%
Joint European charts |**25**|	32.1%
New Sheffield tables |**64**|	16.6%

Table 9.3 Proportion of 386 patients referred to a Lipid Clinic to whom the various charts or tables could be applied whose risk assessment agreed with the Framingham risk equation |**12**| on which they are all based. CHD risk was calculated for comparison in all cases except the New Zealand charts when cardiovascular risk, which they are designed to predict, was calculated. The total number of patients studied was 386. The percentages are only for the patients *(n)* to whom the charts or tables were applicable

	n	**Agreed**	**Underestimated**	**Overestimated**
Joint British charts *(Heart)* |**8**|	340	87.9%	5.0%	7.1%
Joint British charts (British Heart Foundation) |**67**|	307	88.3%	5.5%	6.2%
New Zealand charts |**27**|	230	63.0%	2.2%	34.8%
Joint European charts |**25**|	262	65.6%	6.5%	27.9%
New Sheffield tables |**64**|	322	81.1%	5.9%	13.0%

because they predict cardiovascular rather than coronary disease risk, because they were compared with cardiovascular risk predicted by the Framingham equation. That the New Zealand charts predict cardiovascular risk (CHD and stroke risk combined) does, however, in itself also impose difficulties in the use of the New Zealand tables in nations such as Britain where recommendations for statin therapy are currently based on CHD risk while their exact role in stroke prevention is being more fully evaluated.

The New Zealand charts also include both diastolic and systolic blood pressure (the other charts and tables use only systolic or include hypertension as an all-or-none phenomenon). This might be perceived as a benefit by some clinicians with a preference for diastolic blood pressure, unaware that diastolic blood pressure, generally is a less good predictor of CHD than systolic. The Framingham equation allows CHD or cardiovascular risk (CHD and stroke) to be calculated both for diastolic and systolic blood pressure. However, the curves for similar levels of CHD or cardiovascular risk for

systolic and diastolic blood pressure as a function of the serum to HDL cholesterol ratio are not parallel. There is thus no value of diastolic blood pressure which corresponds to systolic blood pressure in terms of CHD risk prediction across the whole range of serum to HDL cholesterol ratios. The use of 180/105, 160/95, 140/85 and 120/75mmHg as the blood pressure intervals in the New Zealand tables must therefore contribute to some of their loss of agreement with the Framingham risk equation. The inclusion of hypertension as an all-or-none phenomenon accounts for any lack of agreement between the new Sheffield tables and the Framingham equation. The Sheffield table has the advantage that it includes age by yearly intervals. Both the Joint British and New Zealand charts give risk for 10-year age bands so that, in younger patients in each of these bands, risk will be overestimated, whereas in the older ones it will be underestimated.

Overall, the Joint British charts agree more closely with the results of the Framingham equation than any of the other charts or tables |**65, 66**|. In another assessment of the New Sheffield tables doctors and nurses found the Joint British Societies' chart and the New Zealand Chart easier to use |**68**|.

The recommendations for the treatment of raised cholesterol levels in the USA provide the clinician with an algorithm based loosely on the Framingham equation. This leads to treatment at much lower levels of CHD risk than in the European or British recommendations, probably at a level of CHD risk of about 1% per annum or less and for women at lower levels of risk than men |**37**|. A case has recently been made for the introduction of risk assessment charts into the USA to assist in deciding who will benefit most from statin therapy |**69**|. However, the authors of this report voiced reservations about excessive reliance on absolute CHD risk as an arbiter of who should receive treatment because of the disadvantage this posed to younger patients whose present absolute risk may be low, but whose lifetime exposure to risk may be considerable.

Computer program for predicting CHD and stroke risk

The computer program (Cardiac Risk Assessor Programme) recommended in the Joint British guidelines |**8**| is true to the Framingham equation. It makes no assumption about age, which is treated as a continuous variable rather than in intervals as in the charts. Both systolic and diastolic blood pressures are also treated as continuous variables and risk of both stroke and CHD is given for each so that the clinician may choose the highest risk. The knowledge of a patient's stroke risk is also valuable in making therapeutic decisions and instructive in allowing the clinician to appreciate the relative benefit of antihypertensive and statin therapy (see earlier example under sub-heading High Blood Pressure). In addition the risk attached to the presence of left ventricular hypertrophy can also be computed, which is only otherwise possible with the Sheffield tables. All in all the computer program offers the best approach to the calculation of absolute risk.

Limitations of risk prediction methods

Methods of risk prediction based on the equation derived from epidemiological data in the Framingham study are likely to be inaccurate in patients with additional risk factors not included in the equation. Family history (see earlier) can operate independently of the

risk factors included in the equation and it has been suggested that when this is present (see earlier section for definition) the Framingham risk should be multiplied by 1.5 times |**8**|. Hyperlipidaemias due to pronounced single gene effects were also not included in the Framingham risk equation. Thus it should not be used for predicting risk in FH or type III hyperlipoproteinaemia. The risk of CHD in both these conditions means that lipid-lowering drug treatment in adults should generally begin as soon as these syndromes are detected |**13**|. The Framingham equation includes diabetes, but this is type 2 diabetes. There is no method of risk prediction in type 1 diabetes, although it substantially increases the risk of early onset CHD |**47**|. Furthermore HDL cholesterol in type 1 diabetes is frequently high or normal, unlike type 2 diabetes in which it is generally diminished |**47**|. It is uncertain whether the negative association between HDL cholesterol and atherosclerosis is preserved in type 1 diabetes, but, if it is, it is set at a different level from non-diabetic patients and the Framingham equation is likely to give falsely reassuring results. In both types of diabetes nephropathy will greatly compound risk |**70**| and it is recommended that in patients with proteinuria, hyperlipidaemia and blood pressure should be treated as if CHD is already present. Indeed, the American Diabetes Association does not recommend stratification of risk in either type of diabetes and advocates the treatment of diabetic patients generally as if they already had CHD |**71**|. Patients with renal disease are also at much higher CHD risk than predicted by the Framingham equation |**72**| and so also are people who have migrated from the Indian subcontinent to countries such as Britain |**73**|. Left ventricular hypertrophy too, although it was included in the Framingham equation, is often omitted from risk prediction charts and increases CHD risk substantially. Generally, the risk in patients with left ventricular hypertrophy justifies treatment of both blood pressure and cholesterol according to secondary prevention criteria.

Relative risk

The reason why most recent recommendations have been based on absolute risk is because this can readily be translated into the number of patients who must be treated to prevent one event, which allows medical resources to be directed towards people who can derive most benefit from intervention. This, however, has the effect, as has previously been discussed, of favouring older people as recipients of the most intense intervention. This may not always be appropriate. Greater benefit for particularly high-risk younger people may accrue if intervention is commenced earlier rather than later.

The whole frequency distribution of cardiovascular risk will be shifted downwards in younger people so that a risk factor combination, which may confer only an annual risk of 1% at the age of 40 years, may produce a risk of 3% per year at the age of 60. The difficulty is how to prioritize younger patients for treatment.

Relative risk is the number of times risk is increased compared with the average for people of that age and gender. Relative risk is an alternative to absolute risk, but has generally found little favour as a means of prioritization, because a given change in it can produce widely different health benefits in people of different age or gender. For example, in a woman of 30 years of age, who has FH, the relative risk of CHD may be 100 times greater than average, whereas a 60-year-old man with diabetes and

hypertension may have a relative risk only 10 times greater than average. The average annual absolute CHD risk in 30-year-old women is, however, 0.01% so her risk is 1%, whereas the average absolute risk in 60-year-old men is 0.5% so his risk is 5%.

It is possible to overcome this difficulty, if, instead of expressing relative risk in terms of the average risk, it is expressed as the percentile of risk for a given age and gender. This allows an assessment of the size of the population who will be treated, if intervention is practised at a particular percentile and above. The Dundee risk score was devised for this purpose |**74**|. In simple terms the percentile score tells the clinician how close the patient is to the front of an imaginary queue of men or women of similar age waiting for a coronary event. It would allow treatment to be targeted to younger patients most at risk, who might not receive treatment, if a single high level of absolute risk was employed as a treatment threshold regardless of age. The limitation of the Dundee risk score is that it includes only age, gender, smoking, blood pressure and cholesterol, but not HDL or diabetes as risk factors. Nonetheless, this approach is worthy of future investigation.

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References

1. Shaper AG, Pocock SJ, Phillips AN, Walker M. Identifying men at high risk of heart attacks: strategy for use in general practice. *Br Med J* 1986; **293**:474–9.
2. Pekkanen J, Linn S, Heiss G, Suchindran CM, Leon A, Rifkind BM, Tyroler HA. Ten-year mortality from cardiovascular disease in relation to cholesterol level among men with and without pre-existing cardiovascular disease. *N Engl J Med* 1990; **322**:1700–7.
3. Linden T, Bondjers G, Karlsson T, Wikhund O. Serum triglycerides and HDL cholesterol-major predictors for long-term survival after coronary surgery. *Eur Heart J* 1994; **15**:747–52.
4. Scandinavian Simvastatin Survival Study Group. Randomised trial of cholesterol lowering in 4444 patients with coronary heart disease; the Scandinavian Survival Study. *Lancet* 1994; **344**:1383–9.
5. Sacks FM, Pfeffer MA, Moye LA, Rouleau JL, Rutherford JD, Cole TG, Brown L, Warnica JW, Arnold JM, Wun CC, Davis BR, Braunwald E. The effect of pravastatin on coronary events after myocardial infarction in patients with average cholesterol levels. *N Engl J Med* 1996; **335**:1001–9.
6. The Long-Term Intervention with Pravastatin in Ischaemic Disease (LIPID) Study Group. Prevention of cardiovascular events and death with pravastatin in patients with coronary heart disease and a broad range of initial cholesterol levels. *N Engl J Med* 1998; **339**:1349–57.
7. Reckless JPD. Cost-benefit analysis of lipid lowering management. In: Betteridge DJ, Illingworth DR, Shepherd J (eds). *Lipoproteins in Health and Disease.* London: Arnold, 1999:1230–54.

8. Wood D, Durrington PN, Poulter N, McInnes G, Rees A, Wray R. Joint British Recommendations on prevention of coronary heart disease in clinical practice. *Heart* 1998; **80**(Suppl 2): S1–29.
9. Illingworth DR, Durrington PN. Dyslipidaemia and atherosclerosis: how much more evidence do we need? *Curr Opin Lipidol* 1999; **10**:383–6.
10. Kannel WB. Epidemiologic relationship of disease among the different vascular territories. In: Fuster V, Ross R, Topol EJ (eds). *Atherosclerosis and Coronary Artery Disease.* Philadelphia: Lippincott-Raven, 1996:1591–9.
11. Hebert PR, Gaziano JM, Chan KS, Hennekens CH. Cholesterol lowering with statin drugs, risk of stroke, and total mortality. An overview of randomised trials. *JAMA* 1997; **278**:313–21.
12. Anderson KM, Odell PM, Wilson PWF, Kannel WB. Cardiovascular disease risk profiles. *Am Heart J* 1990; **121**:293–8.
13. Durrington PN. Hyperlipidaemia. *Diagnosis and Management,* 2nd edn. London: Butterworth Heinemann, 1995.
14. Downs GR, Clearfield M, Weiss S, Whitney E, Shapiro DR, Beere PA, Lagendorfer A, Stein EA, Kruyer W, Gotto Jr AM. Primary prevention of acute coronary events with lovastatin in men and women with average cholesterol levels: results of the AFCAPS/ TEXCAPS (Air Force/Texas Coronary Atherosclerosis Prevention Study). *J Am Med Assoc* 1998; **279**:1615–22.
15. Shepherd J, Cobbe SM, Ford I, Isles CG, Lorimer AR, MacFarlane PW, McKillop JH, Packard C, for the West of Scotland Coronary Prevention Study Group. Prevention of coronary heart disease with pravastatin in men with hypercholesterolaemia. *N Engl J Med* 1995; **333**:1301–7.
16. Ramsay LE, Williams B, Johnston GD, MacGregor GA, Paston L, Potter JF, Poulter NR, Russell G. Guidelines for management of hypertension: report of the third working party of the British Hypertension Society. *J Hum Hypertension* 1999; **13**:569–92.
17. Klimov AN, Nagornev VANN. Anitschkow, his contribution to the doctrine of atherosclerosis (in commemoration of the centennial of his birth) in: Fidge NH, Nestel PJ (eds). *Atherosclerosis,* Vol. VII. Amsterdam: Elsevier Science, 1986:371–4.
18. Keys A. Coronary heart disease—the global picture. *Atherosclerosis* 1975; **22**:149–92.
19. Charlton J, Quaife K. Trends in diet 1841–1994. In: Charlton J, Murphy M (eds). *The Health of Adult Britain: 1841–1994.* London: The Stationery Office, 1997:93–113.
20. Herrick JB. Certain clinical features of sudden obstruction of the coronary arteries. *JAMA* 1912; 59:2015–20.
21. Law MR, Wald NJ, Thompson SG. By how much and how quickly does reduction in serum cholesterol concentration lower risk of ischaemic heart disease? *Br Med J* 1994; **308**:367–73.
22. Gould AL, Rossouw JE, Santanello NC, Heyse JF, Furberg CD. Cholesterol reduction yields clinical benefits. A new look at old data. *Circulation* 1995; **91**:2274–82.
23. Smith GD, Pekkanen J. Should there be a moratorium on the use of cholesterol lowering drugs? *Br Med J* 1992; **304**:431–4.
24. Dong W, Colhoun H, Lampe F. Blood analytes. In: Colhoun H, Prescott-Clarke P (eds). *Health Survey for England 1994.* London: Her Majesty's Stationery Office, 1996:369–419.
25. Wood D, De Backer G, Faergeman O, Graham I, Mancia G, Pyörälä K with members

of the Task Force. Prevention of coronary heart disease in clinical practice: recommendations of the Second Joint Task Force of European and other Societies on Coronary Prevention. *Atherosclerosis* 1998; **140**:199–270.
26. Expert Panel on Detection Evaluation, Treatment of High Blood Cholesterol in Adults. Summary of the second report of the National Cholesterol Education Program (NCEP) Expert Panel on Detection. Evaluation and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel II). *JAMA* 1993; **269**:3015–23.
27. Dyslipidaemia Advisory Group on behalf of the Scientific Committee of the National Heart Foundation of New Zealand 1996 National Heart Foundation Guidelines for the Assessment and Management of Dyslipidaemia. *N Z Med J* 1996; **109**:224–32.
28. Laupucis A, Sackett DL, Roberts RA. An assessment of clinically useful measures of the consequences of treatment. *N Engl J Med* 1988; **318**:1728–33.
29. West of Scotland Coronary Prevention Group. West of Scotland Coronary Prevention Study: identification of high-risk groups and comparison with other cardiovascular intervention trials. *Lancet* 1996; **348**:1339–42.
30. Johannesson M, Jonsson B, Kjekshus J, Olsson AG, Pedersen TR, Wedel H. Cost effectiveness of simvastatin treatment to lower cholesterol levels in patients with coronary heart disease. *N Engl J Med* 1997; **336**:332–6.
31. Tengs TO, Adams ME, Pliskin JS, Safran DG, Siegel JE, Weinstein MC, Graham JD. Five-hundred life-saving interventions and their cost effectiveness. *Risk Anal* 1995; **15**:369–90.
32. Edelson JT, Weinstein MC, Tosteson A, Williams L, Lee TH, Goldman L. Long-term cost-effectiveness of various monotherapies for mild to moderate hypertension. *JAMA* 1990; **263**:40–7–13.
33. NHS Executive. Standing Medical Advisory Committee Statement on the use of statins. Executive Letter EL (97), 41. Wetherby: Department of Health, 1997.
34. Haq IU, Ramsay LE, Pickin JN, Yeo WW, Jackson PR, Payne JN. Lipid lowering for prevention of coronary heart disease: what policy now? *Clin Sci* 1996; **91**:399–413.
35. Durrington PN. Normal lipid and lipoprotein levels in childhood and adolescence. In: Neil A, Rees A, Taylor C (eds). *Hyperlipidaemia in Childhood.* London: Royal College of Physicians, 1996:9–16.
36. Collins R, MacMahon S. Blood pressure, antihypertensive drug treatment and the risks of stroke and of coronary heart disease. *Br Med Bull* 1994; **50**:272–98.
37. Durrington PN, Prais H, Bhatnagar D, France M, Crowley V, Khan J, Morgan J. Indications for cholesterol-lowering medication: comparison of risk-assessment methods. *Lancet* 1999; **353**:278–81.
38. Assmann G, Schulte H, Funke H, van Eckardstein A, Seedorf U. The Prospective Cardiovascular Münster (PROCAM) study: identification of high-risk individuals and the role of high-density lipoprotein. In: Miller NE (ed.). *High-density Lipoproteins Reverse Cholesterol Transport and Coronary Heart Disease.* Amsterdam: Excerpta Medica, 1989:46–59.
39. Simons LA. Interrelations of lipids and lipoproteins with coronary artery disease mortality in 19 countries. *Am J Cardiol* 1986; **57**:5–10G.
40. Friedewald WT, Levy RI, Frederickson DS. Estimation of the concentration of LDL-cholesterol in plasma, without the use of preparative ultracentrifuge. *Clin Chem* 1972; **18**:499–502.
41. Stamler J, Vaccaro O, Neaton JD, Wentworth D. Diabetes other risk factors, and 12-year cardiovascular mortality for men screened in the Multiple Risk Factor Intervention Trial. *Diabetes Care* 1993; **16**:434–44.

42. Schulte H, Assmann G. CHD risk equations, obtained from the Framingham Heart Study, applied to PROCAM Study. *Cardiovasc Risk Factors* 1991; **1**:126–33.
43. Goldberg RB, Mellies MJ, Sacks FM, Moye LA, Howard BV, Howard WJ, Davies BR, Cole TG, Pfeffer MA, Braunwald E, for the CARE Investigators. Cardiovascular events and their reduction with pravastatin in diabetic and glucose-intolerant myocardial infarction survivors with average cholesterol levels. Subgroup analyses in the Cholesterol and Recurrent Events (CARE) trial. *Circulation* 1998; **98**:2513–19.
44. Pyörälä K, Pedersen TR, Kjekshus J, Faergeman O, Olsson AG, Thorgeirsson G. The Scandinavian Simvastatin Survival Study (4S) Group. Cholesterol lowering with simvastatin improves prognosis of diabetic patients with coronary heart disease. *Diabetes Care* 1997; **20**:614–25.
45. UK, Prospective Diabetes Study Group. Intensive blood-glucose control with sulphonylureas or insulin compared with conventional treatment and risk of complications in patients with type 2 Diabetes (UKPDS 33). *Lancet* 1998; **352**:837–53.
46. Diabetes Control, Complications Trial (DCCT) Research Group. Effect of intensive diabetes management on macrovascular events and risk factors in the diabetes control and complications trial. *Am J Cardiol* 1995; 75:894–903.
47. Durrington PN. Diabetic dyslipidaemia. *Ballière's Clin Endocrinol Metab* 2000; **13**: 265–78.
48. Haffner SM, Stern MP, Hazuda HP, Mitchell BD, Patterson JK. Cardiovascular risk factors in confirmed prediabetic individuals: does the clock for coronary disease start ticking before the onset of clinical diabetes. *JAMA* 1990; **263**:2893–8.
49. Farrer M, Fulcher G, Albers CJ, Neil HAW, Adams PC, Alberti KGMM. Patients undergoing coronary artery bypass surgery are at a high risk of impaired glucose tolerance and diabetes mellitus during the first post-operative year. *Metabolism* 1995; **44**:1016–27.
50. Wannamethee SG, Shaper AG, Durrington PN, Perry IJ. Hypertension, serum insulin, obesity and the metabolic syndrome. *J Hum Hypertension* 1998; **12**:735–41.
51. Barrett-Connor EL, Cohn BA, Wingaard DL, Edelstein SL. Why is diabetes mellitus a stronger risk factor for fatal ischaemic heart disease in women than in men? The Rancho Benardo Study. *JAMA* 1991; **265**:627–31.
52. Phillips AN, Davey Smith G. How independent are 'independent' effects? Relative risk estimation when correlated exposures are measured imprecisely. *J Clin Epidemiol* 1991; **44**:1223–31.
53. Abbot RD, Carroll RJ. Interpreting multiple logistic regression coefficients in prospective observations studies. *Am J Epidemiol* 1984; **119**:830–6.
54. Carlson LA, Böttiger LE. Ischaemic heart-disease in relation to fasting values of plasma triglycerides and cholesterol: Stockholm Prospective Study. *Lancet* 1972; **1**:865–8.
55. Hokanson JE, Austin MA. Plasma triglyceride level is a risk factor for cardiovascular disease independent of high-density lipoprotein cholesterol level: a meta-analysis of population-based prospective studies. *J Cardiovasc Risk* 1996; **3**:213–19.
56. Law MR, Wald NJ, Wu T, Hackshaw A, Bailey A. Systematic underestimations of association between serum cholesterol concentration and ischaemic heart disease in observational studies: data from the BUPA study. *Br Med J* 1994; **308**:363–6.
57. Egger M, Davey Smith G, Pfluger D, Altpeter E, Elwood PC. Triglyceride as a risk factor for ischaemic heart disease in British men: effect of adjusting for measurement error. *Atherosclerosis* 1999; **143**:275–84.
58. Durrington PN. Triglycerides are more important in atherosclerosis than

epidemiology has suggested. *Atherosclerosis* 1998; **141**(Suppl 1): S57–62.
59. Kannel WB, Wolf PA, Castelli WP, D'Agostini RB. Fibrinogen and risk of cardiovascular disease: the Framingham Study. *JAMA* 1987; **258**:1185–6.
60. Craig WY, Neveux LM, Palomaki GE, Cleveland MM, Haddow JE. Lipoprotein(a) as a risk factor for ischaemic heart disease: meta-analysis of prospective studies. *Clin Chem* 1998; **44**:2301–6.
61. Duell PB, Malinow MR. Homocyst(e)ine: an important risk factor for atherosclerotic vascular disease. *Curr Opin Lipidol* 1997; **8**:28–34.
62. Agatston AS, Janowitz WR, Hildner FJ, Zusmer NR, Viamonte NR, Detrano R. Quantitation of coronary artery calcium using ultrafast computed tomography. *J Am Coll Cardiol* 1990; **15**:827–32.
63. West of Scotland Coronary Prevention Group. Influence of pravastatin and plasma lipids on clinical events in the West of Scotland Coronary Prevention Study (WOSCOPS). *Circulation* 1998; **97**:1440–5.
64. Wallis EJ, Ramsay LE, Haq IU, Ghahramani P, Jackson PR, Rowland-Yeo K, Yeo WW. Coronary and cardiovascular risk estimation for primary prevention: validation of a new Sheffield table in the 1995 Scottish Health survey population. *Br Med J* 2000; **320**:671–6.
65. Jones AF, Walker J, Jewkes C, Game FL, Bartlett WA, Marshall T, Bayly GR. Comparative accuracy of cardiovascular risk prediction methods in primary care patients. *Heart* 2001; **85**:37–43.
66. Durrington PN, Prais H. Methods for the prediction of coronary heart disease risk. *Heart* 2001; **85**:489–90.
67. British Cardiac Society, British Hyperlipidaemia Association, British Hypertension Society, British Diabetic Association. Joint British recommendations on prevention of coronary heart disease in clinical practice. Summary. *Br Med J* 2000; **320**:705–8.
68. Isles CG, Ritchie LD, Murchie P, Norrie J. Risk assessment in primary prevention of coronary heart disease: randomized comparison of three scoring methods. *Br Med J* 2000; 320:690–1.
69. Grundy SM, Pasternak R, Greenland P, Smith S, Fuster V. Assessment of cardiovascular risk by use of multiple-risk-factor assessment equations. A statement for healthcare professionals from the American Heart Association and American College of Cardiology. *Circulation* 1999; **100**:1481–92.
70. Borsch-Johnsen K, Kreiner S. Proteinuria: value as a predictor of cardiovascular mortality in insulin dependent diabetes mellitus. *Br Med J* 1987; **294**:1651–4.
71. American Diabetes Association. Management of dyslipidaemia in adults with diabetes. *Diabetes Care* 1998; **21**(Suppl 1): S36–9.
72. Short CD, Durrington PN. Renal disorders. In: Betteridge DJ, Illingworth DR, Shepherd J (eds). *Lipoproteins in Health and Disease.* London: Arnold, 1999:943–66.
73. McKeigue PM, Miller GJ, Marmot MG. Coronary heart disease in south Asians overseas: a review. *J Clin Epidemiol* 1989; **42**:597–609.
74. Tunstall-Pedo H. The Dundee coronary risk-disk for management of change in risk factors. *Br Med J* 1991; **303**:744–7.

Part III
Diabetic dyslipidaemia

Diabetic dyslipidaemia

Introduction

Diabetes mellitus (DM) is one of the most common chronic conditions in the world, with a prevalence of between 4 and 8% in the USA and Europe |**1,2**|. The predominant clinical form of diabetes is type 2 or non-insulin-dependent DM, accounting for about 90% of all cases.

Type 2 diabetes increases the risk for all manifestations of vascular disease including coronary heart disease (CHD), cerebrovascular disease and peripheral vascular disease, with CHD accounting for the majority of type 2 diabetes related morbidity and mortality |**3**|. The incidence of CHD in type 2 diabetes is increased by a factor of 2–4 |**4**|. Furthermore the prevalence of type 2 diabetes is at least two- to threefold higher in patients with established CHD than in the general population |**2**|.

The propensity for early and diffuse macrovascular disease in type 2 diabetes also reduces the efficacy of revascularization procedures, such as coronary artery angioplasty, stenting and coronary bypass surgery, compared with results from non-diabetic patients |**5**|.

Defective glucose homeostasis alone does not appear to be the major determinant of CHD in type 2 diabetes, a concept supported by observations from the UK Prospective Diabetes Study |**6**|. Furthermore, patients with established type 2 diabetes and the pre-diabetic state of impaired glucose tolerance (IGT) have higher rates of recurrent coronary events and gain significantly greater risk reduction with risk factor modification, compared with those with normal glucose tolerance |**7–9**|.

Indeed, many of the established risk factors for atherosclerosis not only co-segregate in both type 2 diabetes and IGT but are characteristic of those conditions. The so-called metabolic syndrome |**10**| includes insulin resistance, hyperinsulinaemia, obesity, increased concentrations of plasminogen activator inhibitor |**11**| and dyslipidaemia defined by hypertriglyceridaemia, reduced high-density lipoprotein (HDL) cholesterol and abnormal postprandial lipaemia |**12–14**|. Postprandial lipaemia in type 2 diabetes is characterized by prolonged and exaggerated excursions in potentially atherogenic triglyceride (TG)-rich lipoproteins.

This leads us to the concept of a ‘common soil’ hypothesis, whereby impaired insulin physiology and the consequent metabolic effects, results in a clinical phenotype that can variably be expressed as excess macrovascular disease, IGT and/or overt type 2 diabetes |**15**|. However, hyperglycaemia is a very late stage in the sequence of events leading from insulin resistance to frank diabetes, whereas disturbances of lipid metabolism are already manifest during the largely asymptomatic diabetic prodrome |**15,16**|. A growing body of evidence thus supports a central role for diabetic dyslipidaemia, in particular hypertriglyceridaemia with elevated levels of TG-rich lipoproteins (TGRL) and exaggerated postprandial lipaemia, in the pathogenesis of atheromatous disease in type 2

diabetes. The precise mechanisms involved are unclear; however, dysfunctional vascular endothelium and enhanced oxidative stress represent potentially important factors |**17**|.

Aetiology of diabetic dyslipidaemia

Type 2 diabetes results in diverse alterations of lipid and lipoprotein metabolism in both the fasting and the postprandial states |**18–20**|, with both quantitative and qualitative changes in lipoprotein concentrations and composition. The major classical abnormality of diabetic dyslipidaemia is hypertriglyceridaemia with reduced HDL levels, which is seen in up to one-third of type 2 diabetic patients even those with good glycaemic control |**21**|. In many studies the typical dyslipidaemia of type 2 diabetes is more severe in women than in men, which is consistent with the reported excess CHD risk in diabetic women |**22,23**|. In the Framingham study prevalence of hypertriglyceridaemia in type 2 diabetic (defined by fasting plasma TG≥2.7 mmol/l) men and non-diabetic men was 19% and 9%, respectively. In type 2 diabetic and non-diabetic women the prevalence of hypertriglyceridaemia was 17% and 8%, respectively. Prevalence for low HDL cholesterol (<0.9 mmol/l) was 21% (versus 9%) in men as opposed to 25% (versus 10%) in women.

By contrast levels of total and low-density lipoprotein (LDL) cholesterol are similar to those in subjects without diabetes. Of the 347 978 men (aged 35–57 years) screened for inclusion in the Multiple Risk Factor Intervention Study (MRFIT), 5163 subjects were reported as taking medication for diabetes. Baseline cholesterol concentrations were similar for both diabetic and non-diabetic subjects (5.56 versus 5.52 mmol/l).

The aetiology of diabetic dyslipidaemia is complex and involves a variety of factors, including insulin resistance |**23**|, hyperinsulinaemia |**24**|, hyperglycaemia |**25**| and disturbed fatty acid metabolism |**26**|.

A state of insulin resistance, as exemplified in patients with type 2 diabetes, impairs the normal suppression of fatty acid release from adipose tissue |**27**|. Consequently, the flux of free fatty acids to the liver increases and overproduction of TG-enriched very low-density lipoprotein (VLDL) from these substrates occurs. Furthermore, in type 2 diabetes, the inhibitory effect of insulin on hepatic synthesis of apolipoprotein B containing particles (VLDL) is impaired |**28**|. Thus as a consequence of both increased substrate supply and defective inhibitory control, type 2 diabetes results in increased hepatic production of large TG-rich VLDL particles and thus elevated plasma TG levels. An important function of insulin is to maintain a balance between intestinally derived TG containing lipoproteins (chylomicrons) and TG containing lipoproteins (VLDL) of hepatic origin. In type 2 diabetes this regulatory mechanism fails and thus inappropriate hepatic production of VLDL occurs by the liver thus further promoting a state of hypertriglyceridaemia.

The catabolism of TGRLs is initiated by an endothelial bound enzyme lipoprotein lipase (LPL). This enzyme hydrolyses the TG moiety of both VLDL and chylomicrons releasing fatty acids for energy production in muscle and for storage in adipose tissue. There is good evidence that LPL activity is impaired in type 2 diabetic patients compared with non-diabetic subjects of similar age and adiposity |**13**|. This reduced activity of LPL

is even more striking in those type 2 diabetic patients with CHD. Thus in type 2 diabetes there is impaired catabolism of TGRL due to a combination of defective LPL function and an overproduction of TGRL, which saturates the available LPL. This has a variety of important effects with respect to the pathophysiology of diabetic dyslipidaemia.

The two components of diabetic dyslipidaemia, namely, hypertryglyceridaemia with elevated concentrations of TGRL and reduced concentrations of HDL, are closely related. Hypertriglyceridaemia contributes to reducing HDL levels in two ways. The first process involves defective transfer of surface remnants (redundant phospholipid and apolipoprotein) from TGRL to HDL particles during lipolysis. As LPL activity is impaired and lipolysis decreased in type 2 diabetes, there are fewer surface remnants to be incorporated into HDL. Secondly, the expanded pool of TGRL and their prolonged residence time in the circulation increase the exchange (mediated by cholesterol ester transfer protein) of cholesterol ester from HDL to TGRL particles and TG from TGRL to HDL particles. These mechanisms result in the excess production of TG-enriched HDL particles, which have a higher catabolic rate, thus ultimately resulting in lower levels of circulating HDL. Furthermore, TG-rich HDL particles have a higher affinity for hepatic lipase, the activity of which is upregulated in type 2 diabetes |**29**|. Hepatic lipase hydrolyses TG in the HDL core resulting in the production of smaller denser HDL particles. Thus in type 2 diabetes there is a reduction in HDL levels that is predominantly in the form of small dense particles with altered physiological function.

The hypertriglyceridaemia and excess production of TG-rich VLDL particles in type 2 diabetes also affects LDL particle metabolism. The increased neutral lipid exchange mediated by cholesterol ester transfer protein between TG-rich VLDL and HDL particles also occurs between TG-rich VLDL and LDL particles, resulting in the synthesis of TG-rich LDL particles. These particles also have an increased affinity for hepatic lipase, which has enhanced activity in type 2 diabetes. This results in excess hydrolysis of TG within the LDL particles, with the subsequent generation of smaller denser and more atherogenic LDL particles |**30**|.

Small dense LDL particles are well recognized in type 2 diabetes; however, there is no evidence that diabetes *per se* is a determinant of the small dense LDL pattern other than through hypertriglyceridaemia |**19**|.

References

1. Harris MI, Hadden WC, Knowler WC, Bennett PH. Prevalence of diabetes and impaired glucose tolerance in U.S. population aged 20–74 yr. *Diabetes* 1987; **36**:523–34.

2. Currie CJ, Morgan C, Peters JR. Patterns and costs of hospital care for coronary heart disease related and not related to diabetes. *Heart* 1997; **78**(6): 544–9.

3. Laakso M, Lehto S. Epidemiology of risk factors for cardiovascular disease in diabetes and impaired glucose tolerance. *Atherosclerosis* 1998; **137**(Suppl): S65–73.

4. Wingard DL, Barrett-Connor E. Heart disease and diabetes. In: Harris MI (ed.) *Diabetes in America*, 2nd edn. NIH Publication 95–1468. Bethesda MD: National Institutes of Health. National Institutes of Diabetes and Digestive and Kidney Disease, 1995: 429–48.

5. The By-pass Angioplasty Revascularization Investigation (BARI) investigators. Comparison of coronary by-pass surgery with angioplasty in patients with multivessel disease. *N Engl J Med* 1996; **335**:217–25.
6. UK, Prospective Diabetes Study (UKPDS) group. Intensive blood glucose control with sulphonylureas or insulin compared with conventional treatment and risk of complications in patients with type 2 diabetes (UKPDS 33). *Lancet* 1998; **352**:837–53.
7. Goldberg RB, Mellies MJ, Sacks FM, Howard BV, Howard WJ, Davis BR, Cole TG, Pfeffer MA, Braunwald E. Cardiovascular events and their reduction with pravastatin in diabetic and glucose intolerant myocardial infarction survivors with average cholesterol levels: subgroup analyses in the cholesterol and recurrent events (CARE) trial. The Care Investigators. *Circulation* 1998; **98**:2513–19.
8. Stamler J, Vaccaro O, Neaton JD, Wentworth D, for the Multiple Risk Factor Intervention Trial Research Group. Diabetes, other risk factors and 12-year cardiovascular mortality for men screened in the Multiple Risk Factor Intervention Trial. *Diabetes Care* 1993; **16**:434–44.
9. Haffner SM, Lehto S, Ronnemaa T, Pyorala K, Laakso M. Mortality from coronary heart disease in subjects with type 2 diabetes and in non-diabetic subjects with and without prior myocardial infarction. *N Engl J Med* 1998; **339**:229–34.
10. Reaven GM. Banting lecture. Role of insulin resistance in human disease. *Diabetes* 1988; 37:1595–607.
11. Vague P, Juhan Vague I, Aillaud MF. Correlation between blood fibrinolytic activity, plasminogen activator inhibitor level, plasma insulin level and relative body-weight in normal and obese subjects. *Metabolism* 1986; **35**:250–3.
12. Garg A. Insulin resistance in the pathogenesis of dyslipidaemia. *Diabetes Care* 1996; **4**: 387–9.
13. De Man FHA, Cabezas MC, Van Barlingen HHJ, Erkelens DW, De Bruin TWA. Triglyceride rich lipoproteins in non-insulin dependent diabetes mellitus: postprandial metabolism and relation to premature atherosclerosis. *Eur J Clin Invest* 1996; **26**: 89–108.
14. Briones ER, Mao SJ, Palumbo PJ, O'Fallon WM, Chenoweth W, Kottke BA. Analysis of plasma lipids and lipoproteins in insulin dependent and non-insulin dependent diabetics. *Metabolism* 1984; **33**:4–9.
15. Haffner SM, Stern MP, Hazuda HP, Mitchell BD, Patterson JK. Cardiovascular risk factors in confirmed prediabetic individuals: Does the clock for coronary heart disease begin ticking before the onset of clinical diabetes? *JAMA* 1990; **263**:2893–8.
16. Kreisberg RA. Diabetic dyslipidaemia. *Am J Cardiol* 1998; **82**(12A): 67–73U.
17. Watts GF, Playford DA. Dyslipoproteinaemia and hyperoxidative stress in the pathogenesis of endothelial dysfunction in non-insulin dependent diabetes mellitus: a hypothesis. *Atherosclerosis* 1998; **141**:17–30.
18. Taskinen M-R. Dyslipidaemia in non-insulin dependent diabetes. *Cardiovasc Risk Factors* 1995; **5**:22–9.
19. Syvanne M, Taskinen M-R. Lipids and lipoproteins as coronary risk factors in non-insulin dependent diabetes mellitus. *Lancet* 1997; **350**(Suppl 1): 20–23.
20. Syvanne M, Hilden H, Taskinen M-R. Abnormal metabolism of post-prandial lipoproteins in patients with non-insulin dependent diabetes is not related to coronary artery disease. *J Lipid Res* 1994; **35**:15–26.
21. Betteridge DJ. Diabetic dyslipidaemia—implications for vascular risk. In: Betteridge DJ (ed.) *Lipids: Current Perspectives.* London: Martin Dunitz, 1996:135–57.
22. Kannel WB, McGee DI. Diabetes and glucose intolerance as risk factors

cardiovascular disease: The Framingham study. *Diabetes Care* 1979; **2**:120–6.

23. Malmstrom R, Packard CJ, Caslake M, Bedford D, Stewart P, Yehi-Jaronian H. Defective regulation of triglyceride metabolism by insulin in the liver in non-insulin dependent diabetes mellitus. *Diabetologia* 1997; **40**:454–62.

24. Reaven GM, Greenfield HS. Diabetic hypertriglyceridaemia. Evidence for three clinical syndromes. *Diabetes* 1981; **31**(Suppl 2): 66–75.

25. Howard BV. Lipoprotein metabolism in diabetes mellitus. *J Lipid Res* 1987; **28**:613–28.

26. Lewis GF, Uffelman KD, Szeto LW, Weller B, Steiner G. Interaction between free fatty acids and insulin in the acute control of very low density lipoprotein production in humans. *J Clin Invest* 1995; **95**:158–66.

27. Frayn KN. Insulin resistance and lipid metabolism. *Curr Opin Lipidol* 1993; **4**:194–204.

28. Lewis GF, Uffelman KD, Szeto LW, Steiner G. Effects of acute hyperinsulinaemia on VLDL triglyceride and VLDL apo B production in normal weight and obese individuals. *Diabetes* 1993; **43**:833–42.

29. Harno K, Nikkila EA, Kuusi T. Plasma HDL-cholesterol and post-heparin plasma hepatic endothelial lipase (HL) activity: Relationship to obesity and non-insulin dependent diabetes (NIDDM). *Diabetologia* 1980; **19**:281.

30. Austin MA, Breslow JL, Hennekens CH, Buring JE, Willet WC, Krauss RM. Low-density lipoprotein subclass patterns and the risk of myocardial infarction. *JAMA* 1988; **260**: 1917–21.

10
Diabetic dyslipidaemia and coronary heart disease

Diabetes and coronary heart disease

Introduction

Type 2 diabetes mellitus (DM) increases the risk of coronary heart disease (CHD) by a factor of 2–4 |**1**|, with a greater increase of relative risk for CHD being shown in women, compared with men, in most studies. Moreover, the prognosis for patients with type 2 diabetes is worse both with respect to myocardial infarction (MI) rate and subsequent mortality |**2**|. Even after a first cardiac event, 50% of patients with diabetes may die within 1 year, and half of those who die, do so before they reach hospital. These data, therefore, suggest that a greater emphasis should be placed on primary prevention of CHD in patients with type 2 diabetes, as secondary prevention is clearly not an option for a significant proportion of patients. Indeed, there is a great deal of merit in the suggestion that all patients with type 2 DM should be considered as having the same absolute risk for a coronary event as non-diabetic patients who have survived their initial MI. Put another way, patients with type 2 diabetes should be considered in the same risk category as 'secondary prevention' patients whether they have overt coronary symptoms or not.

Mortality from coronary heart disease in subjects with type 2 diabetes and in non diabetic subjects with and without prior myocardial infarction.

S M Haffner, S Lehto, T Rönnemaa, K Pyörälä, M Laakso. *N Engl J Med* 1998; **339**:229–34.

B**ACKGROUND. Type 2 (non-insulin-dependent) diabetes is associated with a marked increase in the risk of CHD. It has been debated whether patients with diabetes who have not had MIs should be treated as aggressively for cardiovascular risk factors as patients who have had MIs.**

I**NTERPRETATION.** To address the issue, a comparison was made between the 7-year incidence of MI (fatal and non-fatal) among 1373 non-diabetic subjects and the incidence among 1059 diabetic subjects, all from a Finish population-based study. The

7-year incidence rates of MI in non-diabetic subjects with and without prior MI at baseline were 18.8% and 3.5%, respectively ($P<0.001$). The 7-year incidence rates of MI in diabetic subjects with and without prior MI at baseline were 45.0% and 20.2%, respectively ($P<0.001$). The hazard ratio for death from CHD for diabetic subjects without prior MI as compared with non-diabetic subjects with prior MI was not significantly different from 1.0 (hazard ratio, 1.4; 95% confidence interval, 0.7–2.6) after adjustment for age and sex, suggesting similar risks of infarction in the two groups. After further adjustment for total cholesterol, hypertension and smoking, this hazard ratio remained close to 1.0 (hazard ratio, 1.2; 95% confidence interval, 0.6–2.4).

Comment

In middle-aged patients, the 7-year incidence of MI in patients who had preexisting CHD but did not have DM, was similar to that among patients with type 2 diabetes who did not have CHD, suggesting that type 2 diabetes may confer the same degree as risk as the preceding CHD.

Conclusion

These data suggest that diabetic patients without previous MI have as high a risk of MI as non-diabetic patients with previous MI. The data provide a rationale for treating cardiovascular risk factors in diabetic patients as aggressively as in non-diabetic patients with prior MI.

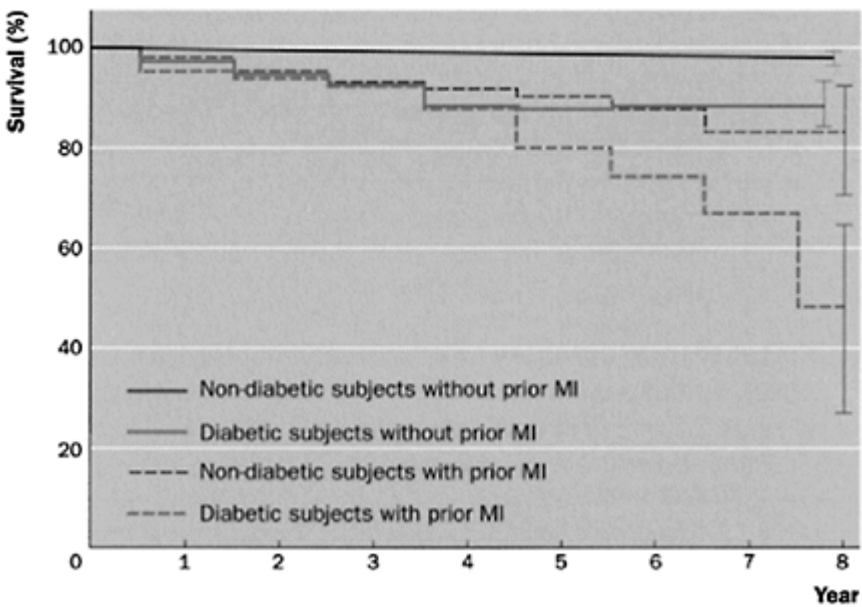

Fig. 10.1 Kaplan-Meier estimates of the probability of death from CHD in 1059 subjects with type 2 diabetes and 1378 non-diabetic subjects with and without prior MI. I bars indicate 95% confidence intervals. Source: Haffner *et al.* (1998).

Table 10.1 Incidence of cardiovascular events during a 7-year follow-up in relation to history of MI in subjects with type 2 diabetes and in non-diabetic subjects*

Event	Non-diabetic subjects			Subjects with type 2 diabetes			All subjects	
	Prior MI (*n*=69)	No prior MI (*n*=1304)	*P* value	Prior MI (*n*=169)	No prior MI (*n*=890)	*P* value	*P* value for prior MI *vs* no prior MI	*P* value for diabetes *vs* no diabetes
Fatal or non-fatal MI								
Incidence during follow-up	18.8	3.5	<0.001	45.0	20.2	<0.001	<0.001	<0.001
Events/100 person-yr	3.0	0.5		7.8	3.2			
Fatal or non-fatal stroke								
Incidence during follow-up	7.2	1.9	0.01	19.5	10.3	<0.001	<0.001	<0.001
Events/100 person-yr	1.2	0.3		3.4	1.6			
Death from cardiovascular causes								
Incidence during follow-up	15.9	2.1	<0.001	42.0	15.4	<0.001	<0.001	<0.001
Events/100 person-yr	2.6	0.3		7.3	2.5			

* *P* values were calculated with Cox proportional-hazards models. The Cox models were adjusted for age and sex. MI denotes myocardial infarction.
Source: Haffner *et al.* (1998).

Rapid rise in the incidence of type 2 diabetes from 1987 to 1996: results from the San Antonio Heart Study.
J P Burke, K Williams, S P Gaskill, *et al. Arch Intern Med* 1999; **159**: 1450–6.

B**ACKGROUND. The prevalence of type 2 diabetes has increased in the early part of the 20th century, particularly in developing countries. There is now evidence that the prevalence also continues to increase in developed countries, including the United States. However, it is not known whether this increase is due to a rise in the incidence of diabetes or to diminishing diabetic mortality, or both. Participants in the San Antonio Study, who were non-diabetic at baseline and who returned for a 7 to 8-year follow-up examination, were examined for secular trends in the incidence of type 2 diabetes. Risk factors for diabetes, such as obesity, were also examined. Patients were enrolled in the San Antonio Heart Study from 1979 to 1988, and 7 to 8-year incidence of diabetes were determined from 1987 to 1996.**

I**NTERPRETATION.** A significant secular trend in the 7 to 8-year incidence of type 2 diabetes was observed in Mexican Americans (5.7% for participants enrolled in 1979 to 15.7% for participants enrolled in 1988). In non-Hispanic whites, the incidence increased from 2.6% for participants enrolled in 1980 to 9.4% for participants enrolled in 1988 (P=0.07). After adjustments for age and sex, the secular trend remained significant in Mexican Americans and borderline significant in non-Hispanic whites. This demonstrates that between 1987 and 1996 the 7 to 8-year incidence of type 2 diabetes approximately tripled in both ethnic groups. The overall secular trend also remained significant after adjusting for additional risk factors for diabetes such as obesity. Also observed was a rising secular trend in obesity. There has been a significant increasing secular trend in the incidence of type 2 diabetes in Mexican Americans and a borderline significant trend in non-Hispanic whites participating in the San Antonio Heart Study. Unlike other cardiovascular risk factors such as lipid levels, cigarette smoking, and blood pressure, which are either declining or under progressively better medical management and control, and unlike cardiovascular mortality, which is also declining, obesity and type 2 diabetes are showing increasing trends. Obesity and diabetes could therefore easily become the pre-eminent US public health problem.

Comment

The clinical association between type 2 DM and CHD is likely to become even more of a public health issue with time. The explosion in the incidence of type 2 diabetes is occurring world-wide across all races and social class and in both high- and low-risk populations. This will have a consequent exacerbation of long-term chronic vascular risk for the world population.

Diabetes Atlas 2000. Brussels: International Diabetes Federation, July 2000.

International Diabetes Federation. World Health Organization. World Health Report 1977. Geneva: WHO, 1977.

B**ACKGROUND/INTERPRETATION. The current scale of the world-wide diabetes epidemic is described in this paper.**

Comment

It is now estimated that in the 134 countries with diabetes organizations affiliated to the International Diabetes Federation, there are currently 151 million people with clinically diagnosed diabetes in the age group 20–79 years. There is an overall prevalence of 4.6% and over 80% of these are type 2 diabetes. Of particular importance is that type 2 diabetes will become more common over the next 25 years mainly because of increasing levels in childhood, adult obesity, decreasing levels of physical activity, increasing urbanization and increasing longevity.

World Health Organization (WHO) predictions of the future prevalence of diabetes in various parts of the world suggest that it will double over the next 25 years in Africa, Asia and eastern Mediterranean countries. The most dramatic increase in these countries will affect ‘economically active’ people aged between 30 and 65 years. The main impact will be with concomitant comorbidities such as

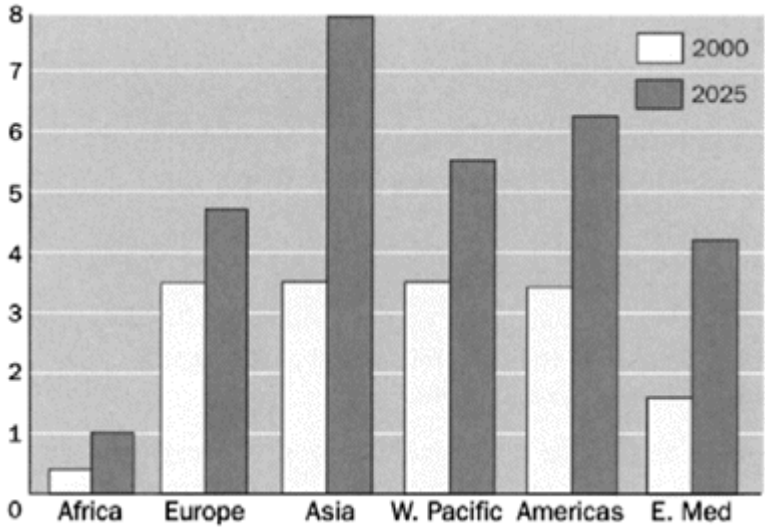

Fig. 10.2 WHO predictions of the prevalence (%) of diabetes in 2025 compared with estimates for 2000 in the six WHO regions. Adapted from WHO. Source: International Diabetes Federation. World Health Organization. World Health Report (1977).

CHD, stroke and hypertension. In Europe, the prevalence will increase from about 3.5 to 4.7% mainly affecting older adults.

Type 2 diabetes in children and adolescents.
American Diabetes Association. *Diabetes Care* 2000; **23**:381–9.

B**ACKGROUND. Type 2 diabetes is a serious and costly disease affecting more than 15 million adult Americans. The chronic complications of diabetes include accelerated development of cardiovascular disease, end-stage renal disease, loss of visual acuity and limb amputations. All these complications contribute to excess morbidity and mortality in individuals with diabetes. Moreover, the prevalence of type 2 diabetes in adults is increased. Superimposed on this disturbing picture in adults are the recent reports of the emerging problem of type 2 diabetes in children and adolescents.**

I**NTERPRETATION.** If there is an increase in the incidence and prevalence of type 2 diabetes in children and if this increase cannot be reversed, society will face major challenges. The burden of diabetes and its

complications will affect many more individuals than currently foreseen, and the cost of diabetes to society will consume enormous resources. Also, many more Americans will be taking potent medications, with attendant risks, for most of their lives. Despite the wealth of experience and knowledge concerning the epidemiology, pathophysiology and medical management of type 2 diabetes in adults, little is known about the disease in children. To assess present knowledge and understanding and to provide guidance to practitioners on medical management, the American Diabetes Association (ADA) convened a consensus development conference on type 2 diabetes in children and adolescents from 30 August 1999 to 1 September 1999.

Type 2 diabetes among North American children and adolescents: an epidemiological review and a public health perspective. A Fagot-Campagna, D J Pettitt, M M Engelgau, *et al. J Pediatr* 2000; **136**: 664–72.

B**ACKGROUND. To review the magnitude, characteristics, and public health importance of type 2 diabetes in North American youth.**

I**NTERPRETATION.** Among 15 to 19 year old North American Indians, prevalence of type 2 diabetes per 1000 was 50.9 for Pima Indians, 4.5 for all US American Indians, and 2.3 for Canadian Cree and Ojibway Indians in Manitoba. From 1967–1976 to 1987–1996, prevalence for Pima Indian adolescents increased 6-fold. Among African Americans and whites aged 10 to 19 years in Ohio, type 2 diabetes accounted for 33% of all cases of diabetes. Young people with type 2 diabetes were generally 10 to 19 years old, were obese and had a family history of type 2 diabetes, had acanthosis nigricans, belonged to minority populations and were more likely to be girls than boys. At

follow-up, glucose control was often poor, and diabetic complications could occur early. Type 2 diabetes is an important problem among American Indian and First Nation youth. Other populations have not been well studied but cases are now occurring in all population groups, especially in ethnic minorities. Type 2 diabetes among young people is an emerging public health problem, for which there is a great potential to improve primary and secondary prevention.

Comment

Type 2 diabetes in children is an emotionally charged issue and an emerging public health problem. Increasingly, type 2 diabetes is being reported in children from the United States, Canada, Japan, Hong Kong, Australia, New Zealand, Libya and Bangladesh. The emergence of the disease in young people embodies the growing problem of chronic diseases world-wide and their extension into youth.

Management of coronary heart disease in patients with diabetes mellitus

Although there has been a marked decline in mortality due to CHD in the overall population over the past two or three decades, this appears not to be the case in patients with diabetes. The possible explanations for these observations are controversial and contentious. They include inadequate control or interventions with respect to conventional risk factors, such as blood glucose control, blood pressure, dyslipidaemia and smoking. The achievement of improving glycaemic control, by the use of sulphonylurea drugs and exogenous insulin, conferred only a very modest benefit in terms of the risk of MI in the UK Prospective Diabetes Study 33 |**3**|, thus lessening concerns that sulphonylurea drugs increase mortality after MI. The case for intensive glycaemic control is more compelling in the period immediately after a MI, during which a significant reduction in mortality was observed in a 1-year randomized control trial |**4**|.

Another possibility is that patients with diabetes may not receive beta-blockers (which have been shown to reduce mortality after a MI) because of theoretical concerns about masking the symptoms of early hypoglycaemia or alternatively exacerbating the typical dyslipidaemia seen in patients with type 2 diabetes. In addition, a *post hoc* analysis of the results of the Bypass Angioplasty Revascularization Investigation (BARI) |**5**| suggested that patients with diabetes who were randomized to undergo angioplasty did significantly worse than those randomly assigned to undergo coronary artery bypass grafting. Unfortunately, no data on glycaemic control are available in the BARI study, so the effect of this putative confounding variable is impossible to evaluate. The data from BARI have, however, led to the suggestion that coronary artery bypass surgery is a better and preferable means of revascularization in patients with type 2 DM than angioplasty. Only 66% of randomized patients survived 5 years following angioplasty compared with 81% of those who had coronary artery bypass grafting. One criticism of this study was the lack of use of stents in the angioplasty group, as stenting appears to improve outcome in patients with type 2 diabetes.

Optimizing the percutaneous interventional outcomes for patients with diabetes mellitus: results of the EPISTENT diabetic substudy.

S P Marso, A M Lincoff, S G Ellis, *et al. Circulation* 1999; **100**:2477–84.

B**ACKGROUND. Stenting likely decreases the need for target-vessel**

revascularization procedures in diabetic patients compared with balloon angioplasty; however, the efficacy of stenting with platelet glycoprotein IIb/IIIa blockade has not yet been assessed in diabetics.

I**NTERPRETATION.** The outcomes of 491 diabetic patients within the multicentre Evaluation of Platelet IIb/IIIa Inhibitor for Stenting Trial (EPISTENT) were analysed. Diabetic patients were a prospectively defined subset: 173 were randomized to stentplacebo, 162 to stent-abciximab, and 156 to balloon angioplasty-abciximab. The main end-point for this analysis was combined 6-month death, MI or target-vessel revascularization. The composite end-point occurred in 25.2% of stent-placebo, 23.4% of balloon-abciximab and 13.0% of stent-abciximab patients (P=0.005). Abciximab therapy, irrespective of revascularization strategy (stent or balloon angioplasty), resulted in a significant reduction in the 6-month death or MI rate: 12.7% for stent-placebo, 7.8% for balloon angioplasty-abciximab and 6.2% for the stent-abciximab group (P=0.029). The 6-month target-vessel revascularization rate was 16.6% for stent-placebo, 18.4% for balloon-abciximab and 8.1% for stent-abciximab (P=0.021). Compared with stent-placebo, stent-abciximab therapy was associated with a significant increase in angiographic net gain (0.88 versus 0.55 mm; P=0.011) and a decrease in the late loss index (0.40 versus 0.60 mm; P=0.061). The 1-year mortality rate for diabetics was 4.1% for stent-placebo and 1.2% for stent-abciximab patients (P=0.11). The combination of stenting and abciximab therapy among diabetics resulted in a significant reduction in 6-month rates of death, MI and target-vessel revascularization compared with stent-placebo or balloon-abciximab therapy.

Comment

This trial (with a composite end-point of death within 6 months, MI and target-vessel revascularization) showed maximal benefit when abciximab (a platelet IIb/IIIa inhibitor) was combined with a coronary artery stent. In diabetic patients receiving abciximab and stenting, the risk of a clinical event was reduced to that of a patient without diabetes.

Conclusion

CHD and type 2 DM go hand in hand. There is a world-wide pandemic of type 2 diabetes that will result in considerable macrovascular comorbidities such as CHD,

Table 10.2 Six-month event rates for study groups

Stent-placebo (n=809)	**Stent-abciximab (n=794)**	P^*	**PTCA-abciximab (n=796)**	$p^\dagger$

Death, MI or TVR					
Diabetics	43 (25.2)	21 (13.0)	0.005	36 (23.4)	0.6
Nondiabetics	104 (16.5)	81 (13.0)	0.062	126 (19.9)	0.17
Death or MI					
Diabetics	22 (12.7)	10 (6.2)	0.041	12 (7.8)	0.13
Nondiabetics	70 (11.0)	34 (5.4)	<0.001	50 (7.8)	0.049
Death					
Diabetics	3 (1.7)	1 (0.6)	0.35	2 (1.3)	0.74
Nondiabetics	7 (1.1)	3 (0.5)	0.21	12 (1.9)	0.26
MI					
Diabetics	19 (11.0)	10 (6.2)	0.11	10 (6.5)	0.14
Nondiabetics	64 (10.1)	31 (4.9)	<0.001	42 (6.6)	0.024
TVR					
Diabetics	28 (16.6)	13 (8.1)	0.021	28 (18.4)	0.7
Nondiabetics	56 (9.0)	55 (8.8)	0.95	92 (14.6)	0.002

* Stent-placebo *vs* stent-abciximab;
† Stent-placebo *vs* PTCA-abciximab.
Source: Marso *et al.* (1999).

stroke and peripheral vascular disease. The prognosis for CHD in people with type 2 diabetes is considerably worse with high mortality as a significant proportion of patients die before they even reach hospital. This inexorably leads to the conclusion that a primary prevention strategy is the essential approach to CHD prevention in people with type 2 diabetes. The data suggest that people with type 2 diabetes have an absolute risk of a coronary event equivalent to non-diabetic patients who have survived their first MI; i.e. their absolute risk for CHD qualifies them for aggressive risk factor modification (lipids, blood pressure control, optimizing glycaemic control, stopping smoking, etc.) before they develop symptoms or signs of overt vascular disease. Should they survive their initial event, previous data have suggested that the choice of revascularization procedure should be coronary artery bypass grafting. More recent evidence however indicates that the judicial use of coronary artery stenting combined with a platelet IIb/IIIa inhibitor is a viable alternative to achieving successful revascularization of the coronary circulation. Hitherto, there has been a belief that coronary artery disease (CAD) in diabetes is more diffuse than that in the general population. However a recent examination of coronary angiograms undertaken for clinical reasons found that the distribution of angiographic CAD in type 2 diabetes is similar to that in matched controls who do not have diabetes |**6**|. This again suggests that coronary artery stenting and platelet IIb/IIIa inhibitors is a legitimate option for revascularization in patients with DM.

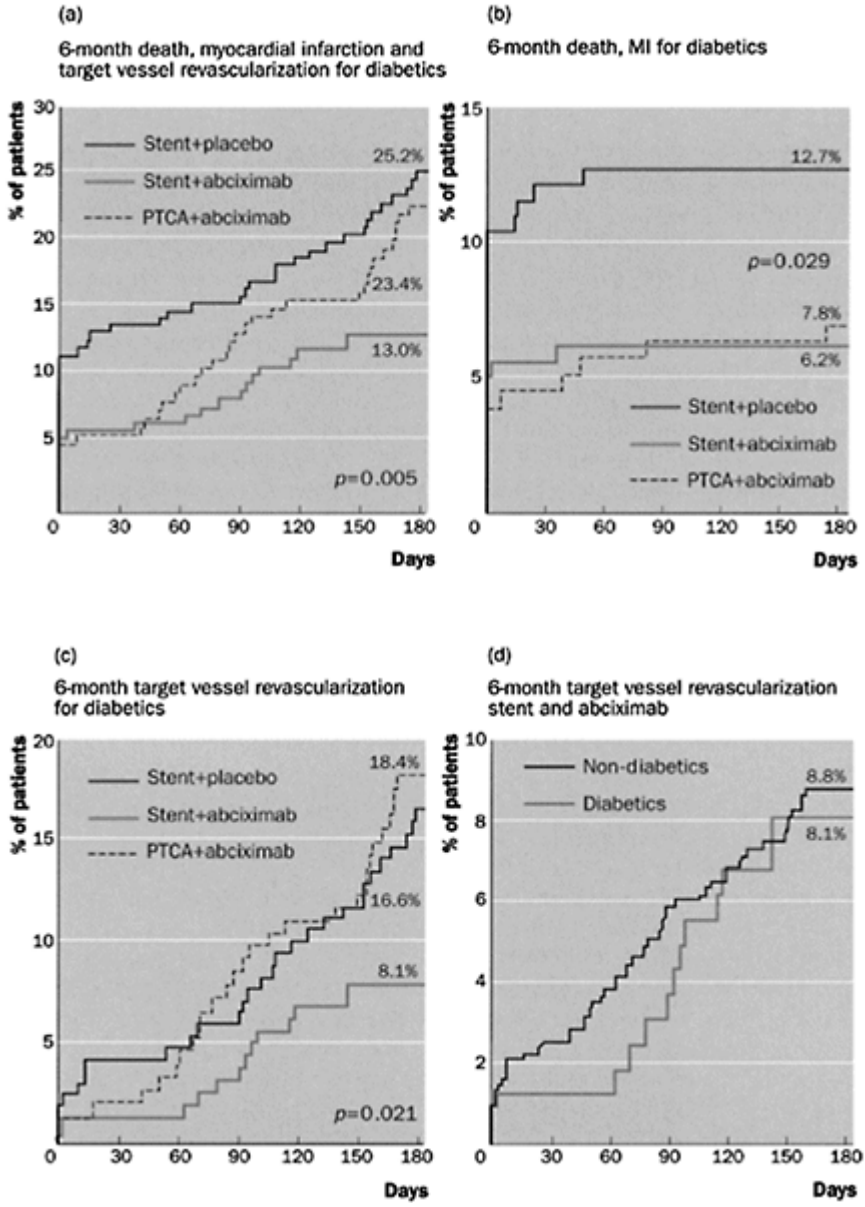

Fig. 10.3 Kaplan-Meier estimates for depicted end-points and study groups. Source: Marso *et al.* (1999).

Postprandial hyperlipidaemia, oxidative stress and endothelial dysfunction in type 2 diabetes mellitus

Type 2 diabetes also results in diverse alterations in postprandial lipid and lipoprotein metabolism. Postprandial lipaemia represents the state of lipid metabolism between food ingestion and the post-absorptive state when all components of the lipid transport system are in a state of equilibrium |**7**|. The rise in plasma triglyceride (TG) and TG-rich lipoproteins (TGRL) after a fatty meal is exaggerated in type 2 diabetic patients compared with non-diabetics |**8**|, with fasting plasma TG levels of≥1.5 mmol/l being predictive of exaggerated postprandial lipaemia |**9**|.

Much of the lipoprotein in postprandial plasma from diabetic patients is in the excess production of hepatic VLDL as a result of insulin resistance, combined with form of very low-density lipoprotein (VLDL) particles |**10**|. This is partly due to competition from chylomicrons and their remnants with endogenous VLDL for catabolism through common removal pathways |**11**|. Indeed the excess production of TG-rich VLDL during postprandial lipaemia in type 2 diabetes exacerbates the existing pro-atherogenic perturbations in lipoprotein metabolism, further lowering high-density lipoprotein (HDL) cholesterol, promoting plasma accumulation of atherogenic VLDL remnants and adding further to the pool of small dense LDL.

The atherogenic potential of disturbed postprandial lipid metabolism was first described by Zilversmit in 1979. However, this had been overlooked for some time as most epidemiological studies of CHD studied fasting subjects only, and may potentially explain the less than expected CHD absolute risk reduction observed following cholesterol reduction in type 2 diabetic subgroups of the major statin trials |**12**|.

There is convincing evidence that exaggerated post-prandial lipaemia is associated with atherosclerotic disease severity and progression in both diabetic and non-diabetic subjects |**13,14**|. In a large study |**15**|, 101 male patients were ascertained on the basis of coronary angiography as having coronary atherosclerosis or not. Postprandial plasma TG showed an accuracy of 68% in predicting the presence of coronary disease, independent of HDL levels, compared with an accuracy of 64% for fasting cholesterol levels. Furthermore, early structural changes of atherosclerosis, as detected by carotid intima thickening on ultrasound, have been shown to correlate strongly with postprandial plasma TG levels |**16**|. Thus postprandial hypertriglyceridaemia with subsequent TG enrichment of lipoproteins appears to be an important determinant of CHD risk.

There is considerable evidence both clinical and experimental supporting the role of TGRL in atherogenesis. This is illustrated by remnant removal disease or familial dysbetalipoproteinaemia, which results in massive accumulation of TGRL remnant particles and associated premature atherosclerosis |**17**|. Lipoprotein analyses from the Monitored Atherosclerosis Regression Study (MARS) |**18**| confirm the importance of TG-rich VLDL and IDL, as strong predictors of atherosclerotic disease progression. Accumulation of TGRL within the vascular endothelium leads to cholesterol accumulation and deposition in the vessel wall |**19**|, and TG-rich VLDL particles have been shown to undergo preferentially endocytosis by receptors on macrophages to form foam cells |**20**|. Additionally, TGRL have been shown to have a direct cytotoxic effect on the vessel wall |**21**| and in accordance with the response to injury hypothesis |**22**| promote atherogenesis. Furthermore, lipolytic products of TGRL and TG-rich apolipoprotein (apo)B containing particles have been isolated in excess from atherosclerotic plaques |**23**|.

Thus, there is compelling evidence of a strong association between postprandial

lipaemia with the production of TGRL and atherosclerosis. Recent evidence suggests that potential mechanisms accounting for this association, in both diabetic and non-diabetic subjects, may involve enhanced oxidative stress and endothelial dysfunction |**24,25**|.

Endothelial dysfunction in type 2 diabetes

Endothelial dysfunction, with a reduction in bioavailable nitric oxide (NO), represents a key early and potentially reversible event in the pathology of atherosclerosis |**26,27**|. As well as being a vasodilator, NO has a variety of actions considered central to the prevention of atherosclerosis. It inhibits both platelet adhesion and migration |**28**|, NO also prevents neutrophil adhesion to the endothelium, prevents vascular smooth muscle proliferation and migration and inhibits adhesion molecule expression |**16,27**|. The recognized antiatherogenic properties of intact endothelium and their attenuation with the development of detectable atherosclerotic plaques, suggests that endothelial dysfunction may not be simply a marker of early vascular disease but that it may represent the primary vascular wall injury that initiates the atherosclerotic process.

Endothelial dysfunction, in the form of impaired hyperaemic flow related vasodilatation, has been demonstrated in the presence of various known risk factors for atherosclerosis, including hypertension, hypercholesterolaemia |**29**|, passive as well as active smoking |**30**|. In addition endothelial dysfunction is well described in both type 1 and type 2 diabetes |**31,32**|. Furthermore, a correlation between the severity of atherosclerotic risk factors and the degree of endothelial dysfunction as well as interaction between risk factors similar to that seen in population outcome studies can be demonstrated |**33**|. There are no clinical trials to date, correlating the presence of endothelial dysfunction with later morbidity and mortality. However, interventions that reduce the incidence of cardiovascular events, such as cholesterol lowering and exercise, also produce improvements in endothelial function |**31,34**|, thus suggesting a link between endothelial dysfunction and later cardiovascular events.

Numerous studies have consistently demonstrated the presence of endothelial dysfunction in type 2 diabetes |**32,35**|. The pathogenesis of endothelial dysfunction in type 2 diabetes is, however, not fully understood. A growing body of evidence supports a role for diabetic dyslipidaemia, in particular hypertriglyceridaemia and exaggerated postprandial lipaemia with elevated levels of TGRLs, together with enhanced oxidative stress, in this process |**36,37**|.

Oxidative stress in type 2 diabetes

Oxidative stress represents a pathophysiological mechanism that stems from a state of disequilibrium between free radical production and natural antioxidant defences. The pathophysiology of enhanced oxidative stress in type 2 diabetes is complex and is likely to involve disturbances in glucose as well as lipid metabolism, support for the concept of increased oxidative stress in diabetes being derived principally from *in vitro* experiments |**38**,**39**|.

Hyperglycaemia may result in enhanced oxidative stress via several mechanisms, although the individual contribution of each mechanism to oxidative stress remains

undefined, as does any dose-response relationship between glucose and oxidative stress.

The term autoxidation describes the capability of glucose to enolize, thereby reducing molecular oxygen and yielding oxidizing intermediates |**40**|. The reduced oxygen products formed in the autoxidative reaction are superoxide anions, the hydroxyl radical and hydrogen peroxide. All can damage lipids, as well as proteins through cross-linking and fragmentation. Free radicals also accelerate the formation of advanced glycation end-products (AGEs), which in turn supply more free radicals; a process termed autoxidative glycosylation or glycoxidation |**41**|. Increased cellular uptake of glucose stimulates protein kinase C activity |**42**| which among other effects, activates peroxidase enzymes and the cyclo-oxygenase (COX) pathway |**42,43**|, with resultant overproduction of oxidative molecules. By elevating endothelial cell calcium hyperglycaemia also stimulates the synthesis and release of NO |**44,45**|. However, in the presence of superoxide anions, NO is converted into the highly potent oxidant molecule peroxynitrite (ONOO-), thus further contributing to a state of enhanced oxidative stress.

There is also evidence that diabetes is associated with decreased tissue concentrations of antioxidants. In general, antioxidant enzymes such as superoxide dismutase, catalase and glutathione peroxidase are reduced in non-vascular beds of diabetic animals |**46**|. In vascular beds |**47**| and cardiac tissue |**48**|, there is a selective increase in catalase but not in superoxide dismutase or glutathione peroxidase. This has been interpreted as evidence that diabetic blood vessels are chronically exposed to enhanced oxidative stress due to increased production of hydrogen peroxide.

Disturbances in lipid metabolism in type 2 diabetes also contribute significantly to a state of enhanced oxidative stress. Diabetic dyslipidaemia, characterized by hypertriglyceridaemia and reduced HDL cholesterol, also results in compositional changes in all lipoproteins, resulting in the production of smaller denser lipoprotein particles with altered physiological function. HDL cholesterol appears to have antioxidant properties inhibiting the oxidation of LDL both *in vitro* and *in vivo* |**49,50**|. The antioxidant properties of HDL relate to its protein components, which bind transition metals |**51**| and to two intrinsic enzyme systems: platelet-activating factor acetyl hydrolase |**52**| and paraoxonase that can hydrolyse lipid hydroperoxide within the LDL particle |**52**|. HDL particles may also play a part in the reverse transport of hydroperoxide species for hepatic detoxification |**53**|. Impaired removal of these species as a result of reduced HDL cholesterol concentrations could be particularly deleterious under conditions favouring a prolonged endothelial residence time and accumulation of susceptible lipoprotein particles, in particular small dense LDL. Accordingly, in type 2 diabetes, a reduced pool size of HDL with altered chemical composition and reduced paraoxonase activity |**54**|, results in enhanced oxidative stress and promotes LDL oxidation (in particles that are already more susceptible to oxidation), thus further increasing the atherogenicity of LDL.

Small dense LDL particles (particle diameter <25.5 nm) are a well-described consequence of lipid metabolism in type 2 diabetes |**55**|. These LDL particles are highly susceptible to oxidative modification |**53**|, particularly when glycated and in the presence of hyperglycaemia and enhanced oxidative stress |**56,57**|. Oxidized LDL particles can further promote enhanced oxidative stress, both directly, by stimulating free radical release from the vessel wall |**58**| and indirectly, by inhibiting NO synthesis thus reducing

antioxidant potential.

Hypertriglyceridaemia *per se* may also promote an environment of enhanced oxidative stress. Monocytes and polymorphonuclear cells release significantly more free radicals when exposed to hypertriglyceridaemic plasma |**59**| compared with plasma from hypercholesterolaemic patients or normolipidaemic plasma. In these studies leucocyte production of free radicals correlated positively with plasma TG and negatively with HDL cholesterol levels. Further studies have demonstrated that hypertriglyceridaemia particularly in the presence of hyperglycaemia as in type 2 diabetes can stimulate leucocyte activation and so promote free radical release. Additionally, plasma antioxidant levels (superoxide dismutase, β-carotene, lycopene) are reduced in hypertriglyceridaemic plasma thus further contributing to the oxidative burden imposed by hypertriglyceridaemia |**60**|.

Dyslipidaemia and atherosclerosis in type 2 diabetes: Role of enhanced oxidative stress and endothelial dysfunction

Injury to the endothelium, with reduced bioavailable NO, initiates the process of atherosclerosis |**22**|. Available evidence suggests that the extent of endothelial dysfunction may reflect the degree of oxidative stress imposed on the endothelium. This concept is supported by reports that antioxidants such as vitamins C and E and probucol ameliorate the degree of endothelial vasodilator dysfunction in both diabetic and non-diabetic subjects |**24,37,61**|. Moreover vitamin E may actually improve coronary outcome |**62,63**|. The potential role of enhanced oxidative stress in association with perturbed fasting and postprandial lipid metabolism in the pathogenesis of endothelial dysfunction in type 2 diabetes has recently become increasingly apparent.

Ciprofibrate therapy improves endothelial function and reduces postprandial lipemia and oxidative stress in type 2 diabetes mellitus.

M Evans, R A Anderson, J Graham, *et al. Circulation* 2000; **101**(15): 1173–9.

B**ACKGROUND. Exaggerated postprandial lipaemia is a factor in atherogenesis, involving endothelial dysfunction and enhanced oxidative stress. The effects of 3 months of ciprofibrate therapy on these parameters were examined in this double-blind placebo-controlled study of 20 type 2 DM patients.**

I**NTERPRETATION.** This study demonstrates that fibrate therapy improves fasting and postprandial endothelial function in type 2 diabetes. Attenuation of postprandial lipaemia and the associated oxidative stress, with increased HDL cholesterol levels, may be important. After ciprofibrate, fasting and postprandial flow-mediated endothelium-dependent vasodilatation values were significantly higher, and fasting and postprandial TG levels fell.

Fasting and postprandial HDL cholesterol was increased but total and LDL cholesterol were unchanged. Fasting and postprandial TG enrichment of all lipoproteins was attenuated, with cholesterol depletion of VLDL and enrichment of HDL. There were similar postprandial increases in oxidative stress in both groups at baseline, which was significantly attenuated by ciprofibrate.

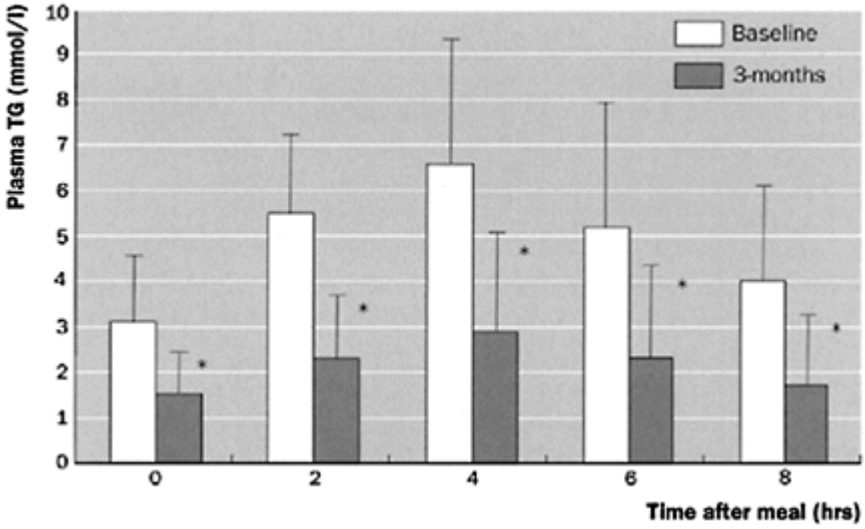

Fig. 10.4 Postprandial lipaemia in ciprofibrate group at baseline and after 3 months of treatment. **P*>0.05 for pretreatment *versus* post-treatment.
Source: Evans *et al.* (2000).

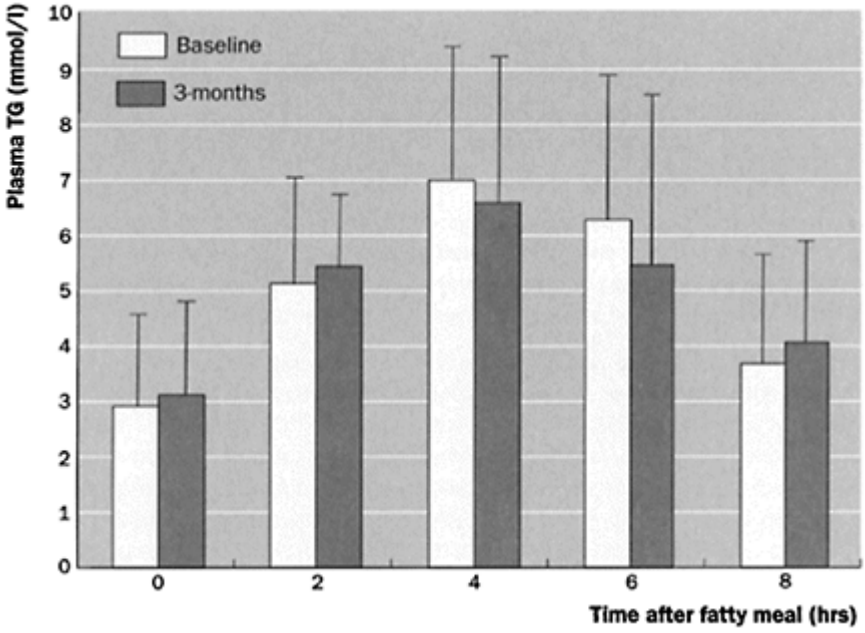

Fig. 10.5 Postprandial lipaemia in placebo group at baseline and after 3 months of treatment. Source: Evans *et al.* (2000).

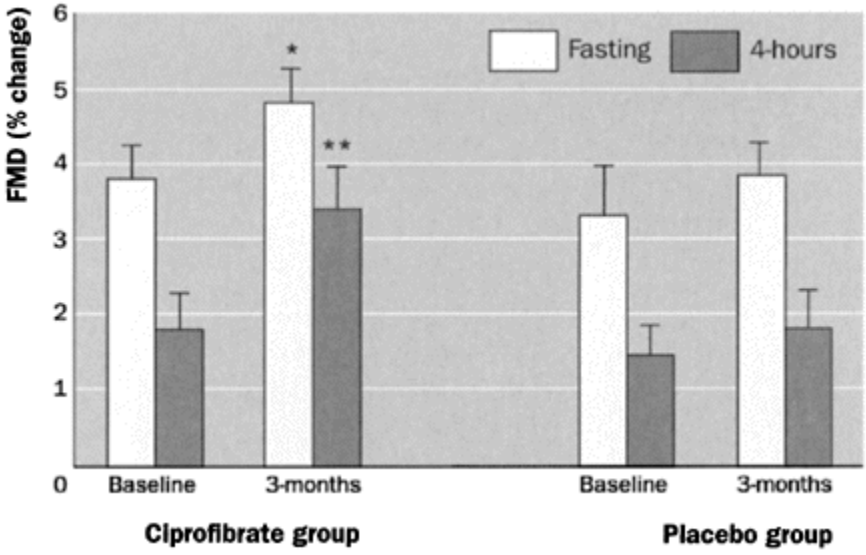

Fig. 10.6 Flow-mediated dilation in placebo and ciprofibrate groups at baseline and after treatment. **P*<0.05 for fasting post-treatment *versus* baseline; ***P*<0.05 for postprandial post-treatment *versus* baseline.

Source: Evans *et al.* (2000).

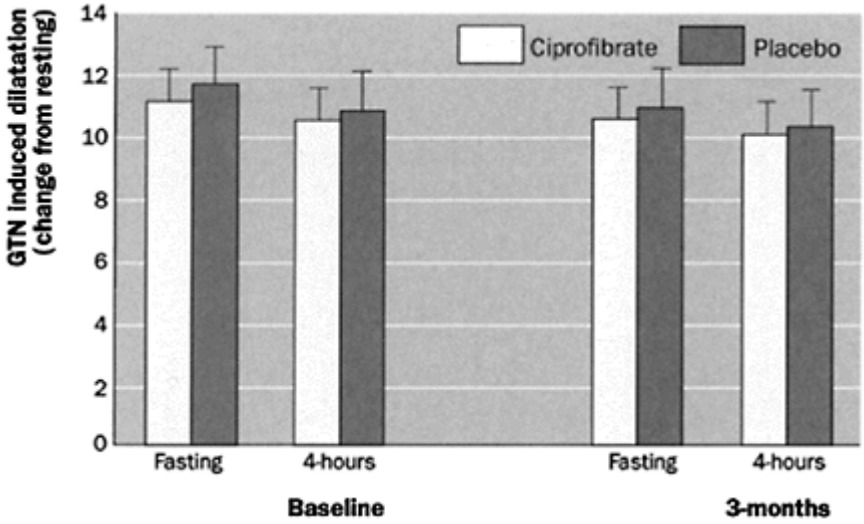

Fig. 10.7 Non-triglyceridaemia-induced endothelium-independent brachial artery dilatation at baseline and after 3 months in both groups. Source: Evans *et al.* (2000).

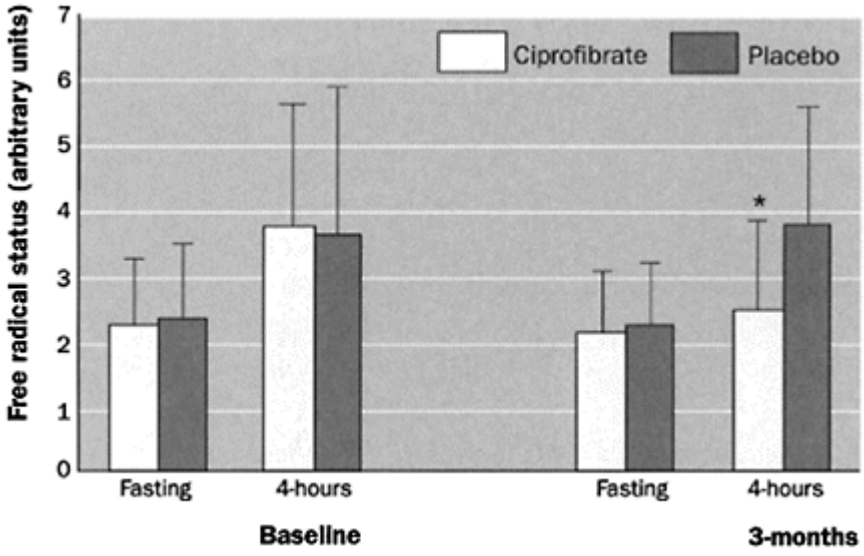

Fig. 10.8 Fasting and postprandial oxidative stress in placebo and ciprofibrate groups at baseline and after treatment. *$P<0.05$ for post-treatment *versus* baseline in ciprofibrate. Source: Evans *et al.* (2000).

Table 10.3 Fasting biochemistry in both groups at baseline and after treatment

	Baseline		3 Months	
	Ciprofibrate	**Placebo**	**Ciprofibrate**	**Placebo**
Glucose, mmol/L	11.1±3.3	10.1±4.1	10.5±3.4	11.2±4.6
Cholesterol, mmol/L	5.9±1.1	5.6±1.4	5.6±1.3	5.7±1.2
LDL-C, mmol/L	3.57±0.6	3.46±0.9	3.51±0.5	3.42±0.5
HDL-C, mmol/L	1.03±0.2	1.09±0.2	1.26±0.1*	0.98±0.3
Insulin, IU/L	32.1±17	31.9±10.8	28.9±10.1	32.4±14.7
HbAIc, %	8.3±1.3	8.01±1.5	7.9±1.5	8.15±1.6
TG, mmol/L	2.8±2.1	2.8±1.7	1.5±0.8*	3.1±2.7

Values are mean±SD.
*P<0.05 *vs* baseline.
Source: Evans *et al.* (2000).

Comment

This group from Cardiff, Wales, test the hypothesis that exaggerated postprandial excursions of TGRLs are important in atherogenesis. This study shows for the first time that a fibric acid derivative improves fasting and postprandial endothelial function in 20 patients with type 2 diabetes. They postulate that this improvement is due to attenuation of fasting and postprandial TG levels, attenuation of TG enrichment of lipoproteins across all classes, and a parallel reduction in oxidative stress. These interesting findings clearly require confirmation in larger studies. However, they emphasize that kinetic changes in lipoprotein metabolism may have an equally malign effect on the endothelium as do quantitative changes.

The relationships between postprandial lipaemia, endothelial function and oxidative stress in healthy individuals and patients with type 2 diabetes.
R A Anderson, L M Evans, G R Ellis, *et al. Atherosclerosis* 2001; **154**: 475–83.

B**ACKGROUND. Postprandial lipaemia (PPL) is a factor in atherogenesis and results in reversible endothelial dysfunction in healthy individuals. Oxidative stress and triglyceride (TG)-rich lipoproteins have been implicated. Type 2 diabetes (NIDDM) results in exaggerated PPL.**

I**NTERPRETATION.** The delineation of the mechanisms of PPL induced, endothelial dysfunction (EF) and oxidative stress in 12 NIDDM and 12 matched healthy subjects was attempted. Subjects underwent a fat tolerance

test, with endothelial function assessed by flow-mediated vasodilatation and oxidative stress measured by venous lipid-derived

free radicals *ex vivo* and lipid peroxidation products over the postprandial phase. Fasting TG, postprandial hypertriglyceridaemia and the TG enrichment of all lipoproteins was significantly greater in NIDDM. Post-prandial endothelial function inversely correlated with fasting HDL-C (r=−0.84, P=0.001) in both the control and NIDDM groups. The deterioration in EF in the NIDDM group also correlated with TG enrichment of VLDL and LDL. PPL in both groups also resulted in increased oxidative stress. The increment in free radicals correlated with TG enrichment of VLDL in both groups and was, therefore, greater in NIDDM. Thus, PPL—with the production of TG-enrichment of VLDL—results in endothelial dysfunction by an oxidative stress mechanism in both groups. The magnitude is greater in NIDDM. Fasting HDL-C appears to contribute to the protection of the endothelium against this phenomenon. Hence, exaggerated PPL associated with reduced HDL-C may be important in the pathogenesis of vascular disease, particularly in NIDDM.

Comment

An extension of the previous study to try and delineate the mechanism of post-prandial lipaemia induced endothelial dysfunction and oxidative stress in type 2 diabetes. They found that deterioration in flow-mediated dilatation in the type 2

Table 10.4 Postprandial area under the curve (AUC) for TG content (mol/ml per 8 h) of lipoproteins in both groups, following a fatty meal[a]

	AUC VLDL-TG	AUC LDL-TG	AUC HDL-TG
NIDDM	1.79±0.22*	1.22±0.35*	1.01±0.43*
Controls	1.13±0.41	0.56±0.27	0.81±0.28

[a] Data is expressed as mean±SD.
* Denotes P<0.05 NIDDM>controls, when analysed using two sample Mann-Whitney U-test.
Source: Anderson *et al.* (2001).

Table 10.5 Oxidative stress and vascular data

EPR height (arbitrary units)	TBARS (mol/I)	FMD (%)	GTN-induced vasodilatation (%)

Controls				
Baseline	2.4±0.1	3.5±2.1	6.3±1.3	11.5±1.1
4 h	3.3±0.2*	6.2±3.6*	4.77±1.4*	10.8±0.8
NIDDM				
Baseline	2.3±0.4	7.5±3.6†	2.65±0.96†	11.1±0.7
4 h	4.53±1.3*†	9.6±3.5*†	1.45±1*†	10.5±1

* Denotes $P<0.05$ versus baseline.
† Denotes $P<0.05$ versus controls.
Source: Anderson *et al.* (2001).

diabetes group correlated best with TG enrichment of VLDL and LDL. Moreover, oxidative stress correlated strongly with TG enrichment of VLDL. The augmented oxidative stress seen in patients with type 2 diabetes was measured in two ways: by electron paramagnetic resonance spectroscopy and by measuring plasma thiobarbituric acid-reacting substances, which are indicators of lipid peroxides in plasma. The authors suggest that oxidative stress results from TG enrichment of lipoproteins, particularly VLDL, and that therapeutic interventions aimed at decreasing TG-VLDL and oxidative stress postprandially may have important anti-atherogenic sequelae.

Conclusion

Exaggerated postprandial lipaemia is recognized as an important factor in atherogenesis in both diabetic and non-diabetic individuals. Recent evidence suggests that the mechanisms underlying these observations involve endothelial dysfunction and enhanced oxidative stress.

In a study by Plotnick (1997) |**124**|, a group of healthy, non-diabetic subjects demonstrated transient endothelial dysfunction in association with postprandial lipaemia following the consumption of a high fat meal. This postprandial endothelial dysfunction was abolished by concomitant consumption of antioxidant vitamins and was not observed following the consumption of a low fat meal. These observations imply a mechanistic association between hypertriglyceridaemia and the subsequent generation of TGRL with enhanced oxidative stress and endothelial dysfunction.

The relationship between postprandial lipaemia, endothelial function and oxidative stress as assessed by directly measuring lipid-derived free radicals using electron paramagnetic resonance spectroscopy has recently been investigated in both diabetic and non-diabetic subjects. In these studies postprandial hypertriglyceridaemia was associated with endothelial dysfunction and enhanced oxidative stress in both diabetic and non-diabetic subjects. The exaggerated and prolonged nature of post-prandial lipaemia in the diabetic subjects was associated with relatively greater deterioration in endothelial function and enhanced excursions in oxidative stress compared with non-diabetic subjects. Furthermore, postprandial endothelial dysfunction and oxidative stress in both diabetes and non-diabetic subjects correlated most strongly with the production of TG-

rich VLDL particles. In addition, modifying TG metabolism and attenuating postprandial lipaemia in the diabetic subjects, using fibrate therapy, resulted in improved fasting and postprandial endothelial function and reduced oxidative stress. Associated with these changes in endothelial function and oxidative stress fibrate therapy had various effects on lipid metabolism, including increased levels of HDL cholesterol and TG depletion of all lipoproteins in both fasting and postprandial states particularly VLDL. Indeed the reduction in oxidative stress and improvements in fasting and postprandial endothelial function following fibrate therapy correlated most strongly with reduction in fasting and postprandial plasma TG levels and TG depletion of VLDL and HDL. It is thus clear that perturbed postprandial lipaemia in type 2 diabetes with the excess production of TGRLs appears to be an important factor in atherogenesis in these patients, by mechanisms involving enhanced oxidative stress and endothelial dysfunction. Furthermore, attenuating postprandial lipaemia in type 2 diabetes may provide therapeutic benefit in reducing cardiovascular risk in these patients.

Endothelial function studies in type 1 diabetes

Endothelial vasodilator function is related to low-density lipoprotein particle size and low-density lipoprotein vitamin E content in type 1 diabetes.

R A Skyrme-Jones, R C O'Brien, M Luo, *et al. J Am Coll Cardiol* 2000; **35**: 292–9.

B**ACKGROUND. Impaired endothelial vasodilator function (EVF) is an early feature of diabetic vascular disease and may be related to oxidant stress. Although small, dense LDL and oxidized LDL are features of type 2 diabetes and predict the development of CAD, their role in type 1 diabetes is less clear. This study sought to determine whether EVF in patients with type 1 diabetes was related to LDL particle size (LDLPS), LDL vitamin E content (LDLVE) or the susceptibility of LDL to oxidation (OxLDL). EVF was assessed in 37 patients with type 1 DM and 45 matched controls.**

I**NTERPRETATION.** The results suggest that LDL particle size and LDL vitamin E may be determinants of conduit and resistance vessel EVF in type 1 diabetes. Total, LDL and HDL cholesterol, TGs and OxLDL did not differ in DM compared with controls, but LDLPS was significantly smaller and LDLVE was reduced. Flow-mediated vasodilation was significantly impaired in DM compared with controls, as was the vasodilator response to acetylcholine. Flow-mediated vasodilation was directly related to LDLPS and LDLVE in the entire study cohort and DM alone, but not to other parameters of the standard lipid profile. Similarly, endothelium-dependent vasodilation in

the resistance circulation was directly related to LDLPS and LDLVE, but not to OxLDL.

Comment

Another paper investigating endothelial function but this time in type 1 DM. This group, based in Melbourne, Australia, investigated 37 patients with type 1 diabetes and 45 matched controls. No differences between patients and controls were seen in the conventional lipid profile but the LDL particle size and its vitamin E content was lower in patients with type 1 diabetes and their endothelial function was significantly impaired. Further analyses suggested that there were no differences in the susceptibility of the LDL particle to oxidation and that it was LDL particle size and vitamin E content that correlated best with the degree of endothelial dysfunction. These data clearly need confirmation but raises the spectra of vitamin E supplementation (an antioxidant vitamin) as a therapeutic option for people with diabetes.

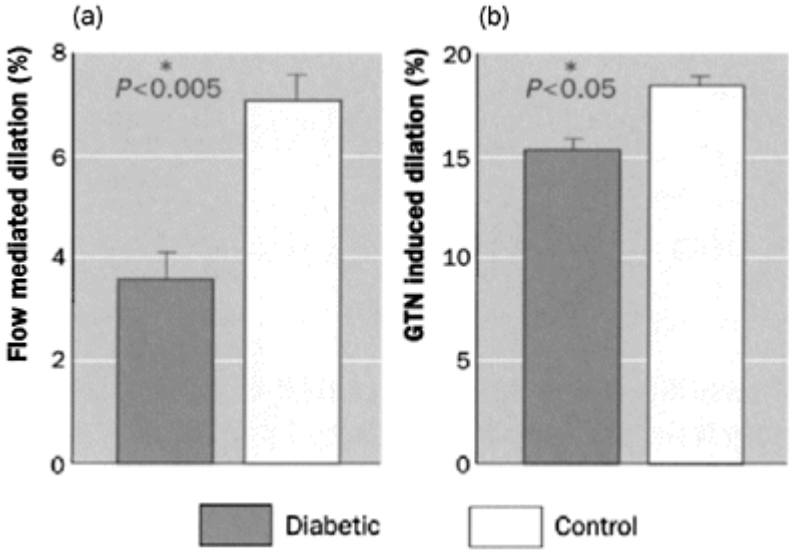

Fig. 10.9 Endothelium-dependent flow-mediated dilation (a) in diabetic (black bar) and control (white bar) subjects. Flow-mediated dilation was significantly reduced in DM compared with control subjects (3.6±0.6% versus 7.1±0.5%). Endothelium-independent nitroglycerine-induced vasodilation (b) was also reduced in DM compared with controls (15.4±1.1% versus 18.5±0.8%), albeit to a lesser extent than flow-mediated dilation. Source: Skyrme-Jones *et al.* (2000).

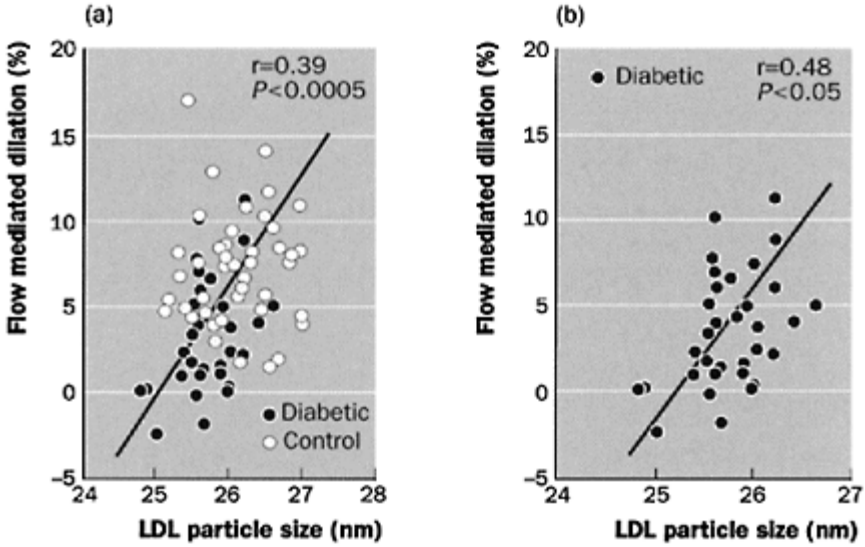

Fig. 10.10 Relationship between flow-mediated dilation in diabetic subjects (filled circles) and control (open circles) subjects with LDL particle size. There was a significant correlation between flow-mediated dilation and LDL particle size in the study cohort (a) and the diabetic group alone (b). Source: Skyrme-Jones *et al.* (2000).

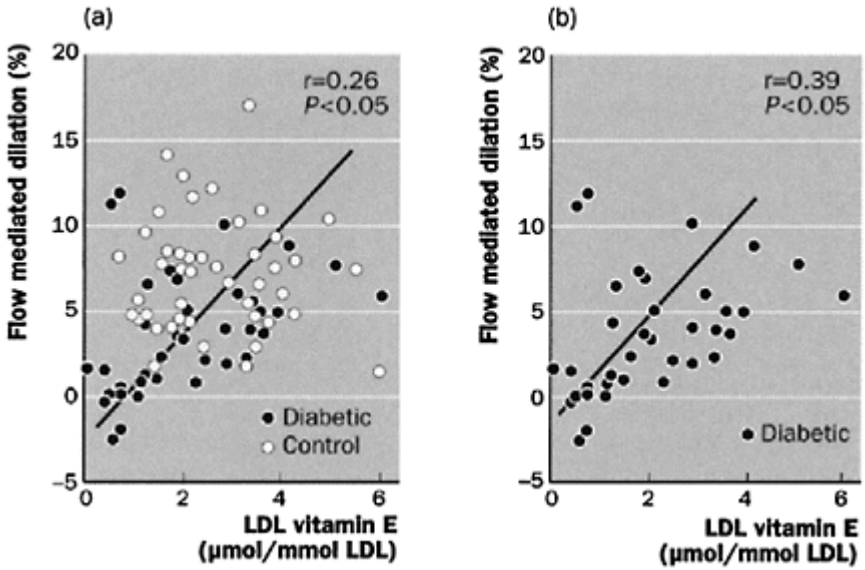

Fig. 10.11 Relationship between flow-mediated dilation in diabetic (filled circles) and control (open circles) subjects with LDL vitamin E.

There was a significant correlation between flow-mediated dilation and LDL vitamin E in the study cohort (a) and the diabetic group alone (b). Source: Skyrme-Jones *et al.* (2000).

Compositional changes of lipoproteins in diabetes mellitus

Very low density lipoprotein subfractions in type 2 diabetes mellitus: alterations in composition and susceptibility to oxidation.
J McEneny, M J O'Kane, K W Moles, *et al. Diabetologia* 2000; **43**:485–93.

B**ACKGROUND. Type 2 DM is associated with raised TGs and increased VLDL cholesterol. This study aimed to assess whether the VLDL subfraction composition and oxidation potential were altered in this condition.**

I**NTERPRETATION.** The results indicate abnormalities in VLDL subfraction composition and oxidation profile in type 2 diabetic patients that are characteristic of more atherogenic particles and may contribute to the development of cardiovascular disease in these patients.

Comment

Diabetic dyslipidaemia is characterized by both quantitative and qualitative changes in lipoprotein metabolism. The TG enrichment across the lipoprotein classes,

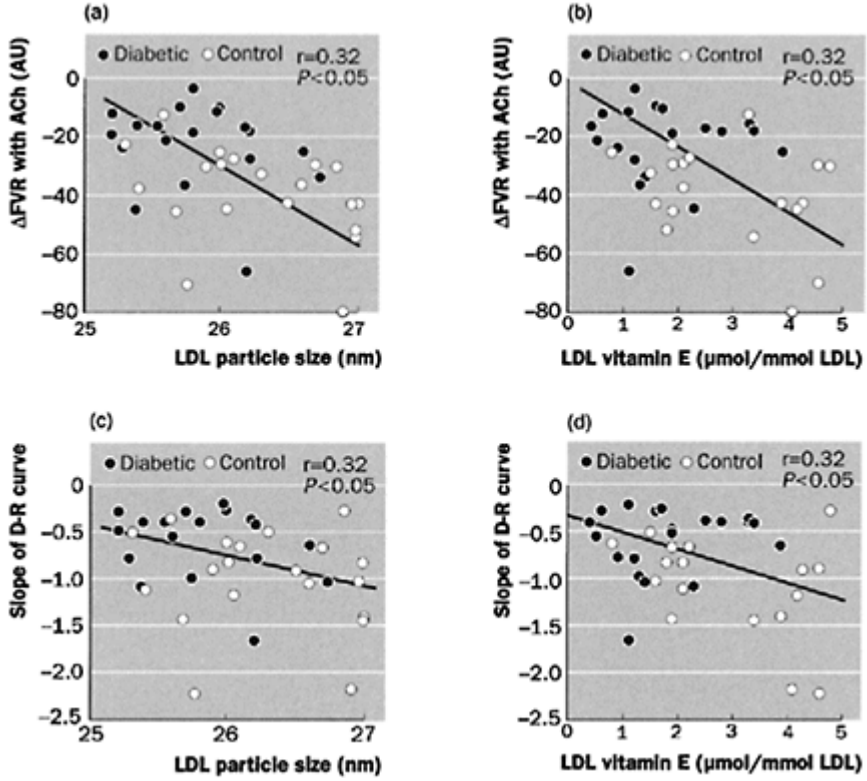

Fig. 10.12 (a, b) The relationship between the absolute change in forearm vascular resistance (ΔFVR) at peak dose of acetylcholine (ACh) and LDL particle size (a) and LDL vitamin E (b) in diabetic (filled circles) and control subjects (open circles). (c, d) The relationship between the slope of the ΔFVR ACh dose-response curve and LDL particle size (c) and LDL vitamin E (d). Source: Skyrme-Jones *et al.* (2000).

influences the susceptibility of these lipoproteins to oxidative changes with consequent increased atherogenicity. This group, from Belfast, separated the VLDL from patients with type 2 diabetes into four subfractions by a novel ultracentrifugation procedure and analysed each subfraction according to lipid and fatty acid composition. Comparing patients with controls, preformed peroxides were higher in all the subfractions from patients with type 2 diabetes and some of the patients' subfractions were more susceptible to *in vitro* oxidation. These findings are characteristic of abnormalities found in the more atherogenic lipoprotein particles. This study indicates that subjects with type 2 diabetes have much larger, lipid-rich VLDL particles and that as the VLDL subfractions decrease in size, and increase in density,

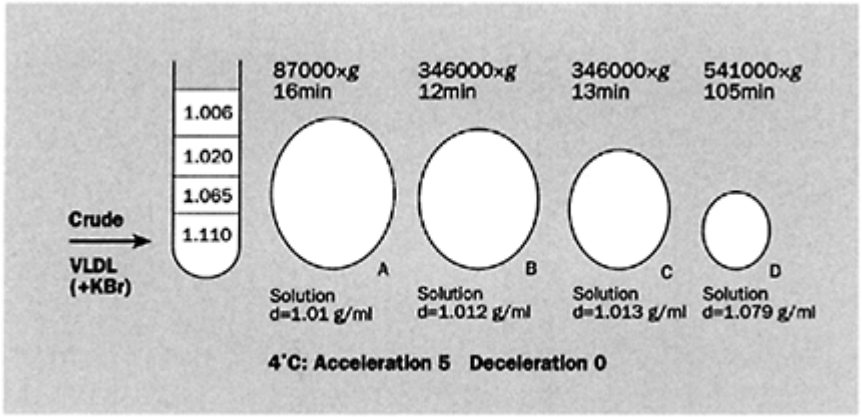

Fig. 10.13 VLDL subfractionation by sequential ultracentrifugation. Source: McEneny *et al.* (2000).

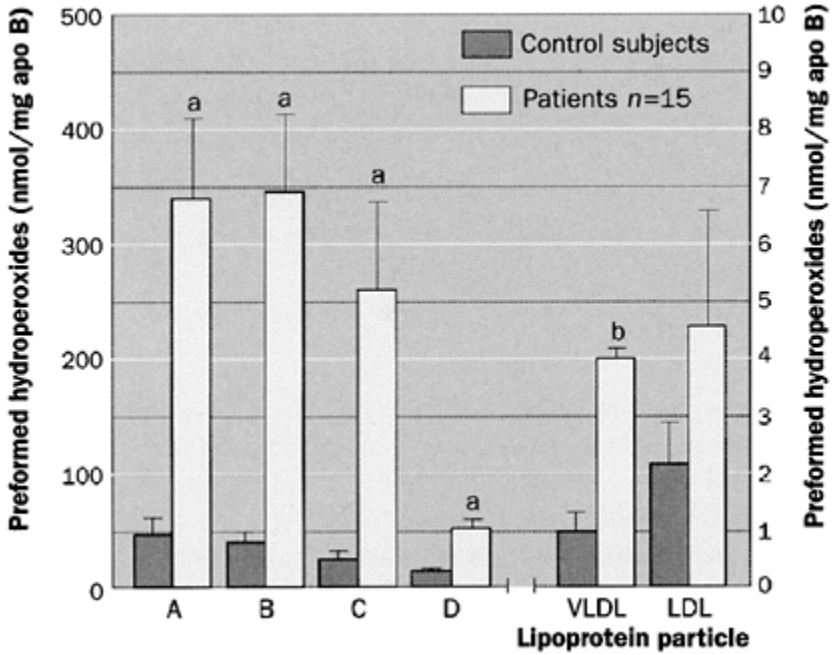

Fig. 10.14 Distribution of preformed hydroperoxides standardized for apoB found in VLDL subfractions (A→D), crude VLDL and LDL in patients with type 2 diabetes compared with matched control subjects. Data presented as mean±SD: $^{a}P<0.001$, $^{b}P<0.05$. Source: McEneny et al. (2000).

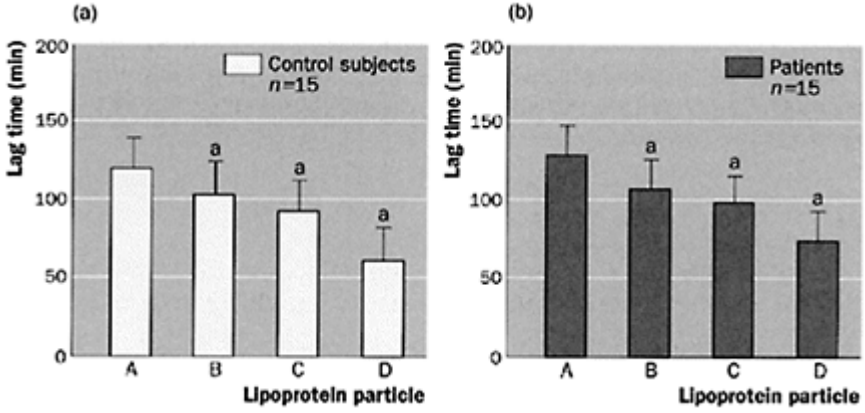

Fig. 10.15 Change in VLDL subfractions susceptibility to copper-mediated oxidation with increasing density (A→D). (a) Control. (b) Type 2 diabetes. Data presented as mean±SD: [a]compared with subfraction A; $P<0.004$. Source: McEneny *et al.* (2000).

the particles become more susceptible to copper mediated oxidation, thus making them more pro-atherogenic.

Postprandial apolipoprotein B48- and B100-containing lipoproteins in type 2 diabetes: do statins have a specific effect on triglyceride metabolism?

S B Battula, O Fitzsimons, S Moreno, *et al. Metabolism* 2000; **49**: 1049–54.

B**ACKGROUND. Little is known about the effects of modifying LDL turnover on chylomicron and VLDL metabolism, yet chylomicron remnant particles are thought to be particularly atherogenic. This study examined the effects of statin-induced inhibition of cholesterol synthesis on postprandial lipoproteins. Eight type 2 diabetic patients were examined (after a high-fat meal) before treatment with cerivastatin, after 4 weeks on active treatment, and 4 weeks after stopping treatment.**

I**NTERPRETATION.** This study suggests that major postprandial lipoprotein changes on statin therapy that may partly explain the beneficial effects of statins in the prevention of MI. During statin treatment, there was a significant reduction in postprandial apoB48 and apoB100 in the chylomicron fraction. Postprandial cholesterol, TG and phospholipid also decreased. In the VLDL fraction, postprandial cholesterol and TG were significantly reduced by statin.

Comment

This study from Dublin looked at eight patients with type 2 diabetes before and after 4 weeks of cerivastatin therapy. They found substantial reduction in the post-prandial apoB-48 area under the curve while on statin therapy and speculate that part of the beneficial effects of statin therapy on CHD prevention may be mediated by effects on postprandial lipid metabolism.

Inhibitory effects of low-density lipoproteins from men with type 2 diabetes on endothelium-dependent relaxation.
K L McNeill, L Fontana, D L Russell-Jones, *et al. Am Coll Cardiol* 2000; **35**: 1622–7.

B**ACKGROUND. Endothelium-dependent vasodilation is impaired in men with type 2 diabetes and this may result from qualitative rather than quantitative abnormalities of LDL. The present study aimed to determine whether native (n) LDL isolated from men with type 2 diabetes and abnormal endothelial function inhibits endothelium-dependent relaxation more than n-LDL isolated from non-diabetic control subjects.**

I**NTERPRETATION.** A qualitative abnormality of LDL may account for endothelial dysfunction in men with type 2 diabetes. Forearm blood-flow responses to acetylcholine but not nitroprusside were significantly impaired in diabetic men compared with controls. n-LDL from diabetic men inhibited relaxation to acetylcholine by significantly more than n-LDL from controls. Relaxation to nitroprusside was not significantly inhibited by n-LDL.

Comment

Another study in type 2 diabetes this time from London. Ten patients with type 2 diabetes and 10 controls were investigated. Significant endothelial dysfunction was demonstrated in the patients with type 2 diabetes. Native LDL was isolated from the subject by using discontinuous density gradient ultracentrifugation using ethylene-diamine tetraacetic acid to prevent oxidation. A preconstricted rabbit aortic ring bioassay was used to assess the inhibitory properties of the LDL on endothelial-dependent relaxation. They found that the LDL from patients with type 2 diabetes inhibited endothelial-dependent relaxation to a significantly greater degree than LDL from the controls. They interpret this as evidence for a qualitative abnormality in the LDL of patients with type 2 diabetes.

Postprandial hypertriglyceridaemia and insulin resistance in normoglycemic first-degree relatives of patients with type 2 diabetes.
M Axelsen, U Smith, J W Eriksson, *et al. Ann Intern Med* 1999; **131**: 27–31.

B**ACKGROUND. Impaired ability to eliminate lipids in the postprandial state is an atherogenic trait associated with insulin resistance. This study was designed to assess insulin sensitivity and postprandial TG metabolism in pre-diabetic individuals. The subjects were 13 healthy, normotriglyceridaemic men, each with two first-degree relatives with type 2 diabetes, and 13 carefully matched controls without known diabetes heredity.**

I**NTERPRETATION.** Despite having normal fasting TG levels these healthy male first-degree relatives of patients with type 2 diabetes are insulin resistant and show postprandial lipid intolerance. These characteristics, which occur in the absence of glucose intolerance, are associated with an increased risk of macroangiopathy.

Comment

A study from Gothenburg, Sweden, of 13 healthy, normotriglyceridaemic men with two first degree relatives with type 2 diabetes and 13 carefully matched controls with no family history of type 2 diabetes. Using euglycaemic clamp techniques, the men with relatives having type 2 diabetes were shown to have insulin resistance but normal glucose tolerance. They also showed significantly larger postprandial TG excursions (area under the curve 50% greater than control group) despite having normal fasting TG levels. These data suggest an increased vascular risk for these individuals.

Diabetic dyslipidaemia and coronary heart disease: new perspectives.
M Evans, N Khan, A Rees. *Curr Opin Lipidol* 1999; **10**:387–91.

B**ACKGROUND. Atherosclerotic macrovascular disease is the leading cause of both morbidity and mortality in non-insulin dependent DM (NIDDM). Endothelial dysfunction is a key, early and potentially reversible event in the pathogenesis of atherosclerosis, which occurs in NIDDM. NIDDM results in diverse abnormalities of lipid and lipoprotein metabolism, in particular hypertriglyceridaemia, low levels of HDL and**

abnormalities of postprandial lipaemia. There is evidence of enhanced oxidative stress in NIDDM, and recent data imply an association between oxidative stress, postprandial lipaemia and endothelial dysfunction in non-diabetic subjects.

I**NTERPRETATION.** Based on *in vitro* and human studies, this article develops the hypothesis that in NIDDM endothelial dysfunction results from diabetic dyslipidaemia, in

particular postprandial lipaemia, and from oxidative stress on the action of nitric oxide. The practical applications of this theory provide potential therapeutic options, which may reduce the risk of vascular disease in NIDDM.

Comment

A review describing experimental evidence linking postprandial, endothelial dysfunction and oxidative stress. The authors suggest therapeutic options which may improve endothelial function in patients with type 2 diabetes and give an update of current thinking in this field.

Diabetic dyslipidaemia in Baillière's Best Practice and Research.

P N Durrington. *Clin Endocrinol Metab* 1999; **13**:265–78.

B**ACKGROUND. The dyslipidaemia of type 2 diabetes consists of hypertriglyceridaemia and low levels of HDL cholesterol. In type I diabetes, hypertriglyceridaemia is present but, when glycaemic control is good, HDL cholesterol levels may be normal or even increased. The risk of CHD associated with serum cholesterol is, however, higher in diabetic than in non-diabetic individuals. So far, the strongest evidence that lipid-lowering drug therapy decreases the risk of CHD, particularly in secondary prevention, comes from trials of statins that lower cholesterol.**

I**NTERPRETATION.** Evidence is growing that hypertriglyceridaemia, because of its effects on cholesteryl ester transfer, leading to the formation of small LDL particles susceptible to oxidation, compounds the risk of serum cholesterol in diabetes. Both fibrates and statins can decrease cholesteryl ester transfer. Further studies of fibrates, with clinical end-points, should clarify their role in CHD prevention. Meanwhile, statins should be part of routine diabetic clinical practice, fibrates having a more limited role when

hypertriglyceridaemia is extreme.

Comment

An authoritative overview of the clinical trial data and clinical management consensus in this field. It emphasizes the need for routine use of statin therapy in the management of type 2 diabetes and assesses the need for further outcome studies with clinical end-points before the wider clinical use of fibrates can be recommended.

Clinical relevance of the oxidative stress concept.
S M Haffner. *Metabolism* 2000; **49**(2 Suppl 1):30–4.

B**ACKGROUND. Patients with type 2 diabetes have markedly increased rates of CHD that are only partly explained by increased levels of conventional risk factors such as**

total cholesterol, hypertension and smoking. Many studies have suggested a role for glycaemia in cardiovascular disease but this remains controversial. Patients with diabetes seem to have increased oxidative stress, a factor that has often been associated with cardiovascular disease, and could thus explain the high level of CHD in diabetes.

I**NTERPRETATION.** Increased levels of oxidative stress may underlie some of the increased risk of cardiovascular disease in diabetic subjects. Interventions to decrease levels of oxidative stress by methods such as improved glycaemic control, antioxidant therapy (i.e. α-tocopherol), and gliclazide are indicated.

Comment

The author reviews the possible role of oxidative stress in the aetiology of vascular disease in patients with type 2 diabetes.

Epidemiology of insulin resistance and its relation to coronary artery disease.
S M Haffner. *Am J Cardiol* 1999; **84**(1A):11–14J.

B**ACKGROUND. The relationship between insulin resistance and cardiovascular risk, particularly CAD, is well established. The clustering of insulin resistance and/or hyperinsulinaemia, hypertriglyceridaemia, hypertension and low HDL is now considered a feature of the insulin resistance syndrome. The pathways by which elevated insulin adversely affects both CAD risk factors and the risk of developing CAD have yet to be elucidated, however.**

I**NTERPRETATION.** Postprandial lipaemia may be a mechanistic link between insulin resistance and CAD. Hyperinsulinaemia appears to be a weak, but positive, independent cardiovascular risk factor. The strongest relationships are seen in middle-aged rather than older persons and at higher elevations of plasma insulin levels. The risk of MI in those with type 2 diabetes is equivalent to that among non-diabetic persons who have had a previous MI. Given the relatively weak associations between duration of diabetes and severity of hyperglycaemia, and cardiovascular disease, common factors may underlie both CAD and type 2 diabetes.

Comment

A review of the complex inter-relationship between CAD, insulin resistance and type 2 diabetes. This review emphasizes the high risk of MI in patients with type 2 diabetes and develops the so-called ‘common soil’ hypothesis that postulates that common factors underlie CHD and type 2 diabetes and the other manifestations of insulin resistance such as low levels of LDL, hypertriglyceridaemia and hypertension.

Postprandial metabolism of apolipoprotein B-48- and B-100-containing particles in type 2 diabetes mellitus: relations to angiographically verified severity of coronary artery disease.
N Mero, R Malmstrom, G Steiner, *et al. Atherosclerosis* 2000; **150**: 167–77.

B**ACKGROUND. This cross-sectional angiographic study aimed to examine whether there is a relationship between the severity of CAD and postprandial lipaemia in patients with type 2 DM. The contribution of apoB-48-containing and apoB-100-containing TG-rich particles to postprandial lipaemia and degree of CAD was determined.**

I**NTERPRETATION.** Postprandial apoB-48 and apoB-100 metabolism in TGRLs is distorted in type 2 diabetic patients, even in those with only mild CAD. The data suggest that postprandial changes in small remnant particle numbers may contribute to the severity of CAD in type 2 diabetes.

Comment

This study from Helsinki, Finland, is a cross-sectional angiograph study focusing on the relative contribution of the gut-derived apoB-48 and the liver-derived apoB-100 containing particles to the magnitude of postprandial lipaemia and degree of CHD. Patients with type 2 diabetes exhibited the typical dsylipidaemia found in insulin-resistant states with delayed clearance of hepatic and intestinally derived particles. There was no difference in fasting or postprandial B48 and B100 levels found and apoE phenotype seemed not to influence postprandial lipaemia. Multiple comparisons were made but no compelling conclusions could be drawn apart from confirming abnormalities in postprandial metabolism at both apoB-48 and apoB-100 particles.

Significant improvement of apolipoprotein B-containing lipoprotein metabolism by insulin treatment in patients with non-insulin-dependent diabetes mellitus.
L Duvillard, F Pont, E Florentin, *et al. Diabetologia* 2000; **43**:27–35.

B**ACKGROUND. Patients with type 2 DM have multiple abnormalities in apoB-containing lipoprotein metabolism, which are likely to contribute to the development of premature atherogenesis. The effect of insulin therapy for 2 months on apoB metabolism in six poorly controlled type 2 diabetic patients and five controls was studied in stable isotope kinetic experiments**

I**NTERPRETATION.** Insulin treatment in type 2 diabetes induced profound changes in lipoprotein metabolism, resulting in a significant reduction in the intravascular residence time of VLDL, intermediate density lipoprotein (IIDL) and LDL particles. These changes are likely to make these particles less harmful. Insulin treatment decreased the VLDL apoB plasma concentration and accelerated IDL apoB turnover without changing its plasma concentration. The treatment increased LDL apoB production and restored a normal LDL apoB fractional catabolic rate, resulting in a constant LDL apoB plasma concentration.

Comment

This study from Dijon, France, used a stable isotope kinetic experiment to study the effects of insulin therapy on apoB metabolism. They found a significant decrease in intravascular residence time of VLDL, IDL and LDL particles. However, whether such changes would occur in patients who had less poorly controlled type 2 diabetes remains

to be seen.

Conclusion

The concept that diabetic dyslipidaemia is in essence a quantitative abnormality of the lipid profile characterized by relatively 'normal' levels of total and LDL cholesterol level together with elevated levels of TGs and reduced levels of HDL cholesterol is an oversimplification. There are quantitative, qualitative and kinetic abnormalities of lipoprotein metabolism resulting in increased atherogenicity of the particles and increased susceptibility to oxidation and other chemical modifications. The effects of these changes on the vasculature can be investigated using surrogate markers of clinical end-points such as flow-mediated dilatation in assessing endothelial dysfunction. With time, and with luck, new treatment modalities may be developed that may benefit endothelial function and ultimately reduce morbidity and mortality in patients with type 2 diabetes.

References

1. Wingard DL, Barrett-Connor E. Heart disease and diabetes. In: Harris MI (ed.). *Diabetes in America*, 2nd edn. NIH publication 95–1468. Bethesda National Institutes of Health, National Institutes of Diabetes and Digestive and Kidney Disease, 1995:429–48.

2. Miettinen H, Lehto S, Salomaa W, Mahonen M, Niemela M, Haffner SM, Pyorala K, Tuomilehto J. Impact of diabetes on mortality after the first myocardial infarction. *Diabetes Care* 1998; **21**:69–75.

3. UK Prospective Diabetes Study Group. Intensive blood-glucose control with sulphonyl-ureas or insulin compared with conventional treatment and risk of complications in patients with type 2 diabetes (UKPDS 33). *Lancet* 1998; **352**:837–53.

4. Malmberg K. Prospective randomised study of intensive insulin treatment on long term survival after acute myocardial infarction in patients with diabetes mellitus. *BMJ* 1997; **314**:1512–15.

5. The Bypass Angioplasty Revascularization Investigation (BARI) Investigators. Comparison of coronary bypass surgery with angioplasty in patients with multivessel disease. *N Engl J Med* 1996; **335**:217–25.

6. Pajunen P, Nieminen MS, Taskinen MR, Syvanne M. Quantitative comparison of angiographic characteristics of coronary artery disease in patients with noninsulin-dependent diabetes mellitus compared with matched nondiabetic control subjects. *Am J Cardiol* 1997; **80**:550–6.

7. Kirchmair R, Ebenbichler CF, Patsch JR. Postprandial lipaemia. *Baillière's Clin Endocrinol Metab* 1995; **9**:705–15.

8. Syvanne M, Hilden H, Taskinen M-R. Abnormal metabolism of postprandial lipoproteins in patients with non-insulin dependent diabetes is not related to coronary artery disease. *J Lipid Res* 1994; **35**:15–26.

9. Lewis GFO, Mera P, Soltys JD, Blackman P, Iverius H, Pugh WL, Getz GS, Polonsky KS. Fasting hypertriglyceridaemia in non-insulin dependent diabetes mellitus is an important predictor of postprandial lipid and lipoprotein abnormalities. *J Clin Endocrinol Metab* 1991; **72**:934–44.

10. Attia N, Durlach V, Roche D, Paul JL, Soni T, Zahouani A, Landron F, Labrousse F, Leutenegger M, Girard-Globa A. Postprandial metabolism of triglyceride rich lipoproteins in non-insulin dependent diabetic patients before and after bezafibrate treatment. *Eur J Clin Invest* 1997; **27**:55–63.
11. Demant T, Packard C. In-Vivo studies of VLDL metabolism and LDL heterogenicity. *Eur Heart J* 1998; **19**(Suppl. H): H7–10.
12. Goldberg RB, Mellies MJ, Sacks FM, Howard BV, Howard WJ, Davis BR, Cole TG, Pfeffer MA, Braunwald E. Cardiovascular events and their reduction with pravastatin in diabetic and glucose intolerant myocardial infarction survivors with average cholesterol levels: subgroup analyses in the cholesterol and recurrent events (CARE) trial. The Care Investigators. *Circulation* 1998; **98**:2513–19.
13. Karpe F, Bard JM, Steiner G, Carlson LA, Fruchart JC, Hamsten A. HDL and alimentary lipaemia. Studies in men with myocardial infarction at young age. *Atheroscler Thromb* 1993; **13**:11–22.
14. Karpe F, Steiner G, Uffelman K, Olivecrona T, Hamsten A. Postprandial lipoproteins and progression of coronary atherosclerosis. *Atherosclerosis* 1994; **106**:83–97.
15. Patsch JR, Miesenbock G, Hopferweiser T, Muhlberger V, Knapp E, Dunn JK, Gotto AM, Patsch W. Relation of triglyceride metabolism and coronary artery disease. *Arterioscler Thromb* 1992; **12**:1336–45.
16. Karpe F, de Faire U, Mercuri M, Gene-Bond M, Hellenius M-L, Hamsten A. Magnitude of alimentary lipaemia is related to intima-media thickness of the common carotid artery in middle aged-men. *Atherosclerosis* 1998; **141**:307–15.
17. Havel RJ. Familial dysbetalipoproteinaemia. New aspects of pathogenesis and diagnosis. *Med Clin North Am* 1982; **66**:441–54.
18. Grenholdt MLN, Nordestgaard BG, Wiebee BM, Wilhjelm JE, Sillesen H. Echo-lucency 7of computerized ultrasound images of carotid atherosclerotic plaques are associated with increased levels of triglyceride rich lipoproteins as well as increased plaque content. *Circulation* 1998; **97**:34–40.
19. Floren CH, Albers JJ, Bierman EL. Uptake of chylomicron remnants causes cholesterol accumulation in cultured human arterial smooth muscle cells. *Biochem Biophys Acta* 1981; **663**:336–49.
20. Gianturco SH, Bradley WA, Gotto AM. Hypertriglyceridaemic very low density lipoproteins induce triglyceride synthesis and accumulation in mouse peritoneal macrophages. *J Clin Invest* 1982; **70**:168–78.
21. Chung BH, Segrest JP. Cytotoxicity of remnants of triglyceride rich lipoproteins: an atherogenic insult? *Adv Exp Med Biol* 1991; **285**:341–51.
22. Ross R. The pathogenesis of atherosclerosis: a perspective for the 1990s. *Nature* 1993; **362**:801–9.
23. Rapp JH, Lespine A, Hamilton RL, Colyvas N, Chaumeton AH, Tweedie-Hardman J, Kottie L, Kunitake ST, Havel RJ, Kane JP. Triglyceride rich lipoproteins isolated by selected affinity anti apolipoprotein B immunosorption from human atherosclerotic plaque. *Arterioscler Thromb Vasc Biol* 1994; **14**:1767–74.
24. Plotnick GD, Correti MC, Vogel RA. Effect of antioxidant vitamins on the transient impairment of endothelium dependent brachial artery vasoactivity following a single high fat meal. *JAMA* 1997; **278**:1682–6.
25. Evans M, Anderson RA, Graham J, Ellis GR, Morris K, Davies S, Jackson SK, Lewis MJ, Frenneaux MP, Rees A. Ciprofibrate therapy improves endothelial function and reduces postprandial lipaemia and oxidative stress in type 2 diabetes. *Circulation* 2000; **101**: 1773–9.

26. Healy B. Endothelial cell dysfunction: an emerging endocrinopathy linked to coronary disease. *J Am Coll Cardiol* 1990; **16**:357–8.
27. Berliner J, Navab M, Fogelman A, Dermmer LL, Edwards PA, Watson AD. Atherosclerosis: Basic mechanisms. *Circulation* 1995; **91**:2488–96.
28. Radomski MW, Palmer RMJ, Moncada S. The role of nitric oxide and cGMP in platelet adhesion to vascular endothelium. *Biochem Biophys Res Commun* 1987; **148**:1482–9.
29. Celermajer DS, Sorenson K, Gooch V, Spieglhalter D, Deanfield J. Non-invasive detection of endothelial dysfunction in children and adults at risk of atherosclerosis. *Lancet* 1992; **340**:1111–15.
30. Celermajer DS, Sorensen K, Georgakopoulos D, Robinson J, Bull C, Deanfield JE. Cigarette smoking is associated with dose related and potentially reversible impairment of endothelium-dependent dilatation in healthy young adults. *Circulation* 1993; **88**:2148–55.
31. Clarkson P, Celermajer D, Donald A, Sampson M, Sorensen KE, Adams M, Yue DK, Betteridge DJ, Deanfield JE. Impaired vascular reactivity in insulin dependent diabetes mellitus is related to disease duration and low-density lipoprotein cholesterol levels. *J Am Coll Cardiol* 1996; **28**:573–9.
32. Goodfellow J, Ramsey MW, Luddington LA, Jones CH, Coates PA, Dunstan F, Lewis M, Owens DR, Henderson AH. Endothelium and inelastic arteries: an early marker of vascular dysfunction in non-insulin dependent diabetes. *BMJ* 1996; **312**:744–5.
33. Celermajer DS, Sorensen KE, Bull C, Robinson J, Deanfield JE. Endothelium-dependent dilatation in the systemic arteries of asymptomatic subjects relates to coronary risk factors and their interaction. *J Am Coll Cardiol* 1994; **24**:1468–74.
34. Bones C, Bellamy M, Brownlee M, Goodfellow J, Ramsey MW, Gorman ST, Lewis MJ. Endothelial function in healthy young adults is rapidly improved by physical training. *Eur Heart J* 1997; **18**:265–71.
35. O'Brien SF, Watts GF, Playford DA, Burke V, O'Neal DN, Best JD. Low density lipoprotein size, high density lipoprotein concentration and endothelial dysfunction in non insulin dependent diabetes. *Diabetic Med* 1997; **14**:974–8.
36. Watts GF, Playford DA. Dyslipoproteinaemia and hyperoxidative stress in the pathogenesis of endothelial dysfunction in non-insulin dependent diabetes mellitus: a hypothesis. *Atherosclerosis* 1998; **141**:17–30.
37. Anderson RA, Evans LM, Ellis GR, Morris-Thorgood JA, Jackson SK, Lewis MJ, Rees A, Frenneaux MP. Postprandial lipaemia is associated with increased oxidative stress and endothelial dysfunction in healthy subjects and is augmented in non-insulin dependent diabetes. *Atherosclerosis*, 2001; **154**:475–83.
38. Wolf SP. Diabetes mellitus and free radicals. Free radicals, transition metals and oxidative stress in the aetiology of diabetes mellitus and complications. *Br Med J* 1993; **49**: 642–52.
39. Giugliano D, Ceriello A, Paolisso G. Oxidative stress and diabetic vascular complications. *Diabetes Care* 1996; **3**:257–67.
40. Hunt JV, Dean RT, Wolff SP. Hydroxyl radical production and autoxidative glycosylation. Glucose autoxidation as the cause of protein damage in the experimental model of diabetes mellitus and ageing. *Biochem J* 1988; **256**:205–12.
41. Baynes JW. Role of oxidative stress in development of complications in diabetes. *Diabetes* 1991; **40**:405–12.
42. Lee TS, Saltsman KA, Ohashi H, King GL. Activation of protein kinase C by

elevation of glucose concentration: Proposal for a mechanism in the development of diabetic vascular complications. *Proc Natl Acad Sci USA* 1989; **86**:5141–5.
43. Feener EP, King GL. Vascular dysfunction in diabetes mellitus. *Lancet* 1997; **i** (Suppl.): 9–13.
44. Cohen RA. Dysfunction of vascular endothelium in diabetes mellitus. *Circulation* 1993; **87**(Suppl.V):V67–76.
45. Poston L, Taylor PD. Endothelium mediated vascular function in insulin dependent diabetes mellitus. *Clin Sci* 1995; **88**:245–55.
46. Matkovics B, Varga S-I, Szabo L, Witas H. The effect of diabetes on the activities of the peroxide metabolism enzymes. *Hormone Metab Res* 1982; **14**:77–9.
47. Pieper GM, Jordan M, Dondlinger LA, Adams MB, Roza AM. Peroxidative stress in diabetic blood vessels. *Diabetes* 1995; **44**:884–9.
48. Wohaieb SA, Godin DV. Alterations in tissue antioxidant systems in spontaneously diabetic (BB Wistar) rats. *Can J Physiol Pharmacol* 1987; **65**:2191–5.
49. Mackness MI, Abbott CA, Arrol S, Durrington PN. The role of high-density lipoprotein and lipid soluble antioxidant vitamins in inhibiting low-density lipoprotein oxidation. *Biochem J* 1993; **294**:829–35.
50. Klimov AN, Gurevich VS, Nikoforava AA, Shatilina LV, Kuzmin AA, Plavinski SL. Antioxidant activity of high density lipoproteins in-vivo. *Atherosclerosis* 1993; **100**: 13–18.
51. Kwiterovich PO. The antiatherogenic role of high-density lipoprotein cholesterol. *Am J Cardiol* 1998; **82**:13–20Q.
52. Mackness MI, Durrington PN. Paraoxonase: another factor in NIDDM cardiovascular disease. *Lancet* 1995; **346**:385.
53. Tribble DL. Lipoprotein oxidation in dyslipidaemia: Insights into general mechanisms affecting lipoprotein oxidative behaviour. *Curr Opin Lipidol* 1995; **6**:196–208.
54. Abbott CA, Mackness MI, Kumar S, Boulton AJ, Durrington PN. Serum paraoxonase activity, concentration and phenotype distribution in diabetes mellitus and its relationship with serum lipids and lipoproteins. *Arterioscler Thromb Vasc Biol* 1995; **15**:1812–18.
55. Taskinen M-R. Dyslipidaemia in non-insulin dependent diabetes. *Cardiovasc Risk Factors* 1995; **5**:22–9.
56. Kawamura M, Heinecke JW, Chait A. Pathophysiological concentrations of glucose promote oxidative modification of low-density lipoprotein by a superoxide dependent pathway. *J Clin Invest* 1994; **94**:771–8.
57. Bowie A, Owens D, Collins P, Johnson A, Tomkin GH. Glycosylated low density lipoprotein is more sensitive to oxidation. Implications for diabetic patients. *Atherosclerosis* 1993; **102**:63–7.
58. Harrison DG, O'Hara Y. Pathophysiologic consequences of increased vascular oxidant stress in hypercholesterolaemia. Implications of impaired vasomotion. *Am J Cardiol* 1995; **75**:75–81B.
59. Proani L, Hiramatsu K, Saiusa Y, Nakazawa H. Low superoxide scavenging activity associated with enhanced superoxide generation by monocytes from male hypertriglyceridaemics with and without diabetes. *Atherosclerosis* 1991; **90**:39–47.
60. Armstrong AM, Chestnutt JE, Gromley MJ, Young IS. The effect of dietary treatment on lipid peroxidation and antioxidant status in newly diagnosed non-insulin dependent diabetes. *Free Rad Biol Med*, 1996; **21**:719–26.
61. Ting HH, Timimi FK, Boles KS, Creager SJ, Ganz P, Creager MA. Vitamin C

improves endothelium dependent vasodilatation in patients with non-insulin dependent diabetes mellitus. *J Clin Invest* 1996; **97**:22–8.

62. Diaz MN, Frei B, Vita JA, Keaney JF. Antioxidants and atherosclerotic heart disease. *N Engl J Med* 1997; **337**:408–16.

63. Stephens NG, Parsons A, Schofield PM, Kelly F, Cheesman K, Mitchinson MJ. Randomised control trial of vitamin E in patients with coronary heart disease: Cambridge Heart Antioxidant Study. *Lancet* 1996; **347**:781–6.

11
Lipid intervention

Lipid intervention trials in diabetes

Introduction

Most clinical trials of lipid intervention and coronary heart disease (CHD) prevention have been conducted in study populations that excluded patients with diabetes. Three trials have conducted *post hoc* analyses of subgroups of patients with type 2 diabetes mellitus (DM). One of these was a primary prevention study with Gemfibrozil (Helsinki Heart Study) |**1**|. This trial found a reduction in coronary events, but the numbers were too small to reach significance. The other two trials were the Scandinavian Simvastatin Survival Study (4S) |**2,3**| and the Cholesterol and Recurrent Events (CARE) |**4**| studies, which were secondary prevention trials using hydroxymethylglutaryl coenzyme A reductase inhibitors, simvastatin and pravastatin, respectively. These two trials present the most compelling evidence in support of lipid reduction in DM, but as these are *post hoc* subgroup analyses, some caution must be exercised in their interpretation. The gold standard for developing specific robust recommendations for lipid regulating therapy in patients with type 2 diabetes must be randomized controlled trials in patients with type 2 diabetes with clear unambiguous primary clinical end-points.

The evidence from statins

Cholesterol lowering with simvastatin improves prognosis of diabetic patients with coronary heart disease.
K Pyorala, T R Pedersen, J Kjekhus, *et al. Diabetes Care* 1997; **20**:614–20.

B**ACKGROUND. To assess in diabetic patients with coronary heart disease (CHD) the effect of cholesterol lowering with simvastatin on mortality and the risk of CHD and other atherosclerotic events.**

I**NTERPRETATION.** A *post hoc* subgroup analysis was carried out on data from 202 diabetic patients and 4242 non-diabetic patients with previous myocardial infarction or

angina pectoris, serum total cholesterol 5.5–8.0 mmol/l, and serum triglycerides<or =2.5 mmol/l who were participating in the Scandinavian Simvastatin Survival Study (4S). Participants in the 4S were randomly assigned to double-blind treatment with simvastatin, 20 mg daily, with blinded dosage titration up to 40 mg daily, according to cholesterol response during the first 6–18 weeks, or placebo. End-points were 1) total mortality, 2) major CHD events (CHD death or non-fatal myocardial infarction), 3) other acute atherosclerotic events, 4) myocardial revascularization procedures. Over the 5.4-year median follow-up period, simvastatin treatment produced mean changes in serum lipids in diabetic patients similar to those observed in non-diabetic patients. The relative risks (RRs) of main end-points in simvastatin-treated diabetic patients were as follows: total mortality 0.57 (95% CI, 0.30–1.08; *P*=0.087), major CHD events 0.45 (95% CI, 0.27–0.74; *P*=0.002), and any atherosclerotic event 0.63 (95% CI, 0.43–0.92; *P*=0.018). The corresponding RRs in non-diabetic patients were the following: 0.71 (95% CI, 0.58–0.87; *P*=0.001), 0.68 (95% CI, 0.60–0.77; *P*<0.0001), and 0.74 (95% CI, 0.68–0.82; *P*<0.0001). The results strongly suggest that cholesterol lowering with simvastatin improves the prognosis of diabetic patients with CHD. The absolute clinical benefit achieved by cholesterol lowering may be greater in diabetic than in non-diabetic patients with CHD because diabetic patients have a higher absolute risk of recurrent CHD events and other atherosclerotic events.

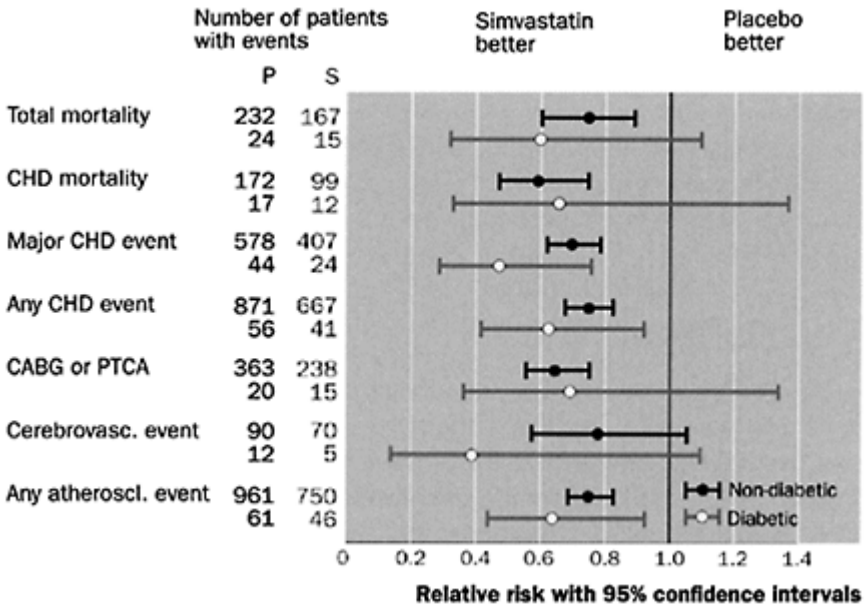

Fig. 11.1 Reduction in the risk of different end-points expressed as relative risk

(simvastatin group versus placebo group) with 95% confidence intervals in non-diabetic and diabetic patients. Source: Pyorala *et al.* (1997).

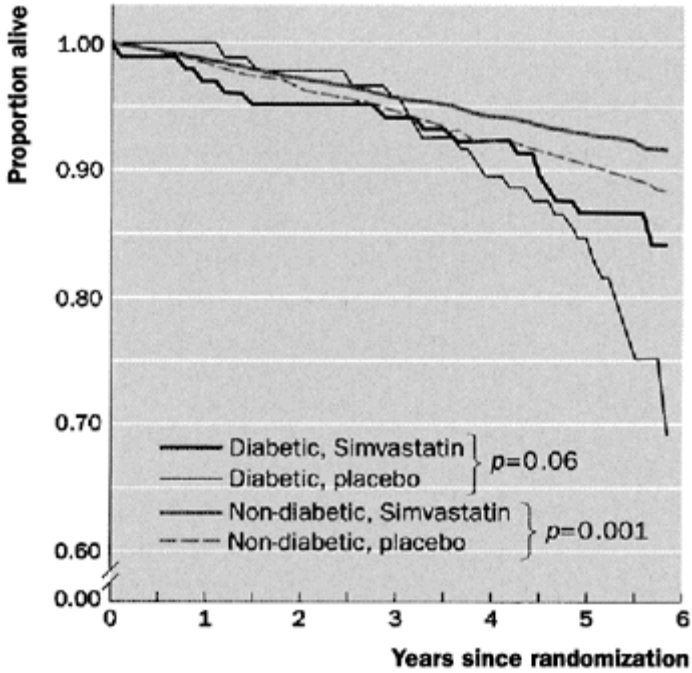

Fig. 11.2 Kaplan-Meier curves for total mortality during follow-up in non-diabetic and diabetic patients treated with placebo or simvastatin in the 4S. Source: Pyorala *et al.* (1997).

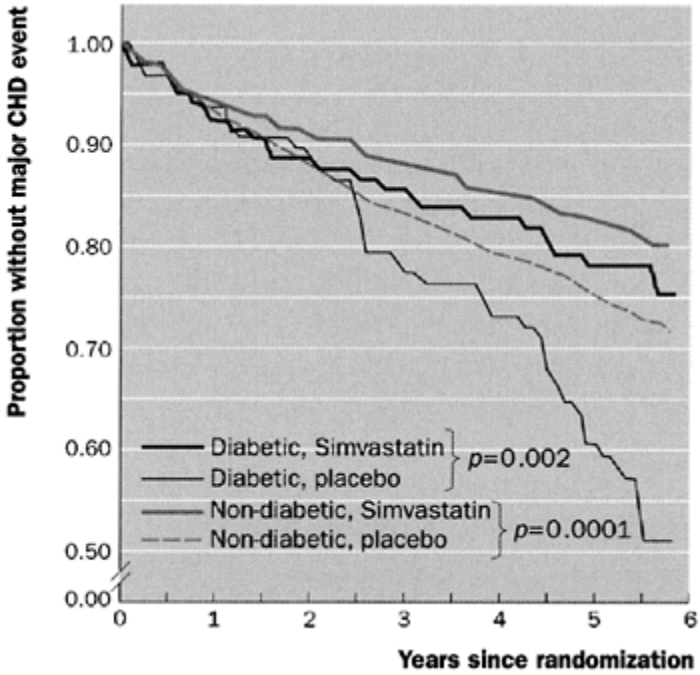

Fig. 11.3 Kaplan-Meier survival curves for the probability of remaining free of a major CHD event during follow-up in non-diabetic and diabetic patients treated with placebo or simvastatin in the 4S. Source: Pyorala *et al.* (1997).

Table 11.1 Mortality and occurrence of non-fatal atherosclerotic events during follow-up in non-diabetic and diabetic patients randomized to placebo or simvastatin

	Non-diabetic		**Diabetic**	
	Placebo	**Simvastatin**	**Placebo**	**Simvastatin**
n	2,126	2,116	97	105
Death				
Death from CHD	172 (8.1)	99 (4.7)	17 (17.5)	12 (11.4)
Death from other cardiovascular cause	15 (0.7)	24 (1.1)	3 (3.1)	0
Death from non-cardiovascular cause	45 (2.1)	44 (2.1)	4 (4.1)	3 (2.9)
Death from any cause	232 (10.9)	167 (7.9)	24 (24.7)	15 (14.3)
Non-fatal events				

Definite MI	246 (11.6)	157 (7.4)	24 (24.7)	7 (6.7)
Probable MI	191 (9.0)	135 (6.4)	11 (11.3)	4 (3.8)
Intervention-associated MI	22 (1.0)	10 (0.5)	3 (3.1)	2 (1.9)
Silent MI	97 (4.6)	85 (4.0)	12 (12.4)	5 (4.8)
Resuscitated cardiac arrest	0	1 (0.1)	0	0
Non-fatal major CHD event	466 (21.9)	339 (16.0)	35 (36.1)	14 (13.3)
Non-MI CHD event	310 (14.6)	280 (13.2)	21 (21.6)	15 (14.3)
CABG or angioplasty	363 (17.1)	237 (11.2)	20 (20.6)	15 (14.3)
Cerebrovascular disease event	85 (4.0)	56 (2.6)	10 (10.3)	5 (4.8)
Other atherosclerotic event	12 (0.6)	12 (0.6)	0	1 (1.0)

Data are number of patients (%). A patient with two or more events of different types will appear more than once in a column but only once in a row. MI, myocardial infarction. CABG, coronary artery bypass grafting.
Source: Pyorala *et al.* (1997).

Comment

The 4S study was a cholesterol lowering trial involving 4444 patients with prevalent CHD and a relatively high low-density lipoprotein (LDL) cholesterol level at baseline. Patients were randomized to simvastatin 20 mg daily versus placebo. In the subgroup with diabetes (*n*=202), a 55% reduction in CHD was seen in the simvastatin group compared with the placebo group (*P*=0.002). The percentage reduction in major coronary events in the group with diabetes was particularly impressive—55% compared with 32% in the group without diabetes.

Cardiovascular events and their reduction with pravastatin in diabetic and glucose-intolerant myocardial infarction survivors with average cholesterol levels.
R B Goldberg, J Margot, M D Mellies, *et al.* Subgroup Analyses in the Cholesterol and Recurrent Events (CARE) Trial. *Circulation* 1998; **98**: 2513–19.

B**ACKGROUND. Although diabetes is a major risk factor for CHD, little information is available on the effects of lipid lowering in diabetic patients. We determined whether**

Table 11.2 Effect of simvastatin treatment on the risk of major CHD events in diabetic

patients by baseline serum lipid levels

Lipid variable	Placebo/ simvastatin	Subjects with events		RR (95% CI) (simvastatin vs. placebo)	*P* value
		Placebo	Simvastatin		
Total cholesterol (mmol/l)					
<6.25	46/52	18 (39)	11 (21)	0.50 (0.23–1.05)	0.07
≥6.25	51/53	26 (51)	13 (25)	0.44 (0.22–0.85)	0.02
LDL cholesterol (mmol/l)					
<4.85	47/50	22 (47)	12 (24)	0.46 (0.23–0.93)	0.03
≥4.85	49/55	22 (45)	12 (22)	0.45 (0.22–0.91)	0.03
HDL cholesterol (mmol/l)					
<1.10	43/50	21 (49)	10 (20)	0.34 (0.16−0.72)	0.005
≥1.10	53/55	23 (43)	14 (26)	0.56 (0.29–1.08)	0.08
Triglycerides (mmol/l)					
<1.70	45/54	19 (42)	12 (22)	0.50 (0.24–1.02)	0.06
≥1.70	52/51	25 (48)	12 (24)	0.42 (0.21–0.84)	0.01

Data are *n* or *n* (%) unless otherwise indicated. The groups were divided into two strata by the median for each lipid variable in the whole diabetic group. Treatment-by-lipid level interactions were not significant.
Source: Pyorala *et al.* (1997).

lipid-lowering treatment with pravastatin prevents recurrent cardiovascular events in diabetic patients with CHD and average cholesterol levels.

I**NTERPRETATION.** CARE is a 5-year trial that compared the effect of pravastatin and placebo, which included 586 patients (14.1%) with clinical diagnoses of diabetes. The participants with diabetes were older, more obese and more hypertensive. The mean baseline lipid concentrations in the group with diabetes 136 mg/dl LDL cholesterol, 38 mg/dl HDL cholesterol, and 164 mg/dl triglycerides (TGs) were similar to those in the non-diabetic group. LDL cholesterol reduction by pravastatin was similar (27% and 28%) in the diabetic and non-diabetic groups, respectively. In the placebo group, the diabetic patients suffered more recurrent coronary events [CHD death, non-fatal myocardial infarction (MI), coronary artery bypass grafting and

percutaneous transluminal coronary angioplasty] than did the non-diabetic patients (37% versus 25%). Pravastatin treatment reduced the absolute risk of coronary events for the diabetic and non-diabetic patients by 8.1% and 5.2% and the relative risk by 25% (P=0.05) and 23% (P<0.001), respectively. Pravastatin reduced the relative risk for revascularization procedures by 32% (P=0.04) in the diabetic patients. In the 3553 patients who were not diagnosed as diabetic, 342 had impaired fasting glucose at entry defined by the American Diabetes Association as 110–125 mg/dl. These non-diabetic patients with impaired fasting glucose had a higher rate of recurrent coronary events than those with normal fasting glucose (e.g. 13% versus 10% for non-fatal MI). Recurrence rates tended to be lower in the pravastatin compared with placebo group (e.g. −50%, P=0.05 for non-fatal MI). Diabetic patients and non-diabetic patients with impaired fasting glucose are at high risk of recurrent coronary events that can be substantially reduced by pravastatin treatment.

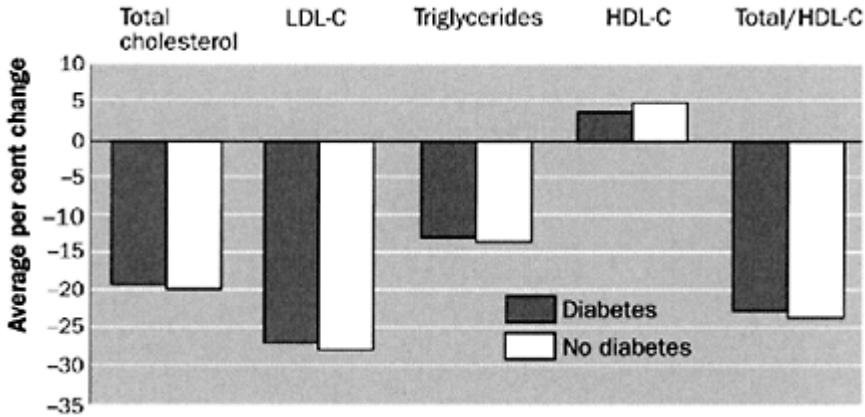

Fig. 11.4 Effect of pravastatin on cholesterol and TGs in diabetes and non-diabetes groups. Change in pravastatin group is shown relative to change in placebo group. Average for 5-year trial duration. Source: Goldberg *et al.* (1998).

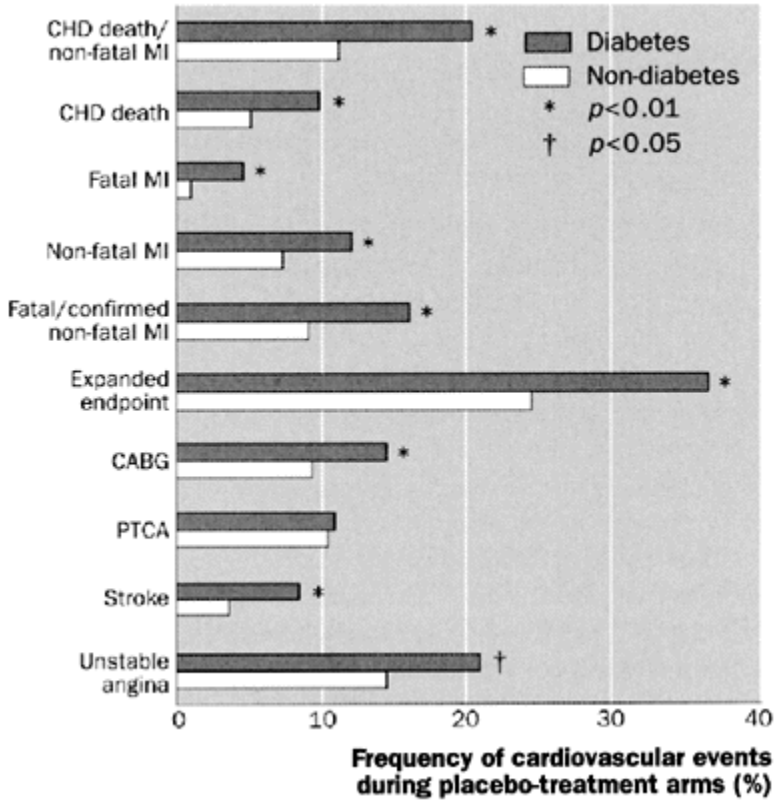

Fig. 11.5 Cardiovascular event rates in patients with or without clinically diagnosed diabetes in placebo group (n=304 and 1774, respectively). Expanded end-point was CHD death, non-fatal MI, coronary artery bypass grafting or percutaneous transluminal coronary angioplasty. **P*<0.01; †*P*<0.05. Source: Goldberg *et al.* (1998).

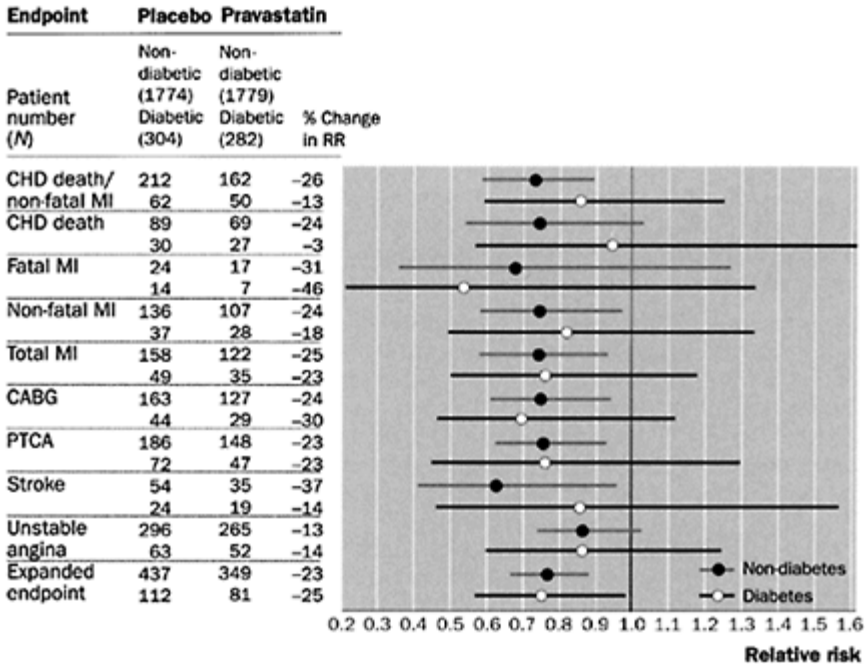

Fig. 11.6 Effect of pravastatin on risks of cardiovascular events in patients with or without clinically diagnosed diabetes. Numbers indicate patients with cardiovascular events. Relative risks (RR) for pravastatin compared with placebo group are shown with 95% confidence intervals. v—v, indicates total non-diabetes group; v—v, diabetes group. Source: Goldberg *et al.* (1998).

Comment

The CARE study was a randomized control trial that compared pravastatin 40 mg with placebo in patients with clinical CHD and lowish LDL cholesterol levels. In 586 subjects with diabetes, pravastatin was associated with a 25% reduction in major CHD events with revascularization procedures (P=0.05), which was similar to the 23% reduction seen in patients without diabetes (P<0.001). Thus the effect of LDL reductions by pravastatin in patients with diabetes was as great in CARE as in those without diabetes.

Reduced coronary events in simvastatin-treated patients with coronary heart disease and diabetes or impaired fasting glucose levels.

S M Haffner, C M A Alexander, T J Cook, *et al. Arch Intern Med* 1999; **159**: 2661–7.

B**ACKGROUND. Patients with DM have a marked increase in CHD events relative to those without DM. In a previous report from the 4S Study using a clinical case definition of DM (*n*=202), simvastatin-treated patients had significantly fewer CHD events compared with placebo-treated control subjects.**

I**NTERPRETATION.** To examine the effect of simvastatin therapy on CHD in patients with DM and impaired fasting glucose levels. Using the 1997 American Diabetes Association diagnostic criteria, we assessed the effect of simvastatin therapy *post hoc* for an average of 5.4 years in the 4S Study patients with normal fasting glucose (*n*=3237), impaired fasting glucose (*n*=678) and DM (*n*=483). Simvastatin-treated patients with DM had significantly reduced numbers of major coronary events [relative risk (RR) =0.58; *P*=0.001] and revascularizations (RR=0.52, *P*=0.005). Total (RR=0.79; *P*=0.34) and coronary (RR=0.72, *P*=0.26) mortality were also reduced in DM, but not significantly, due to small sample size. In impaired fasting glucose subjects, simvastatin use significantly reduced the number of major coronary events (RR=0.62; *P*=0.003), revascularizations (RR=0.57; *P*=0.009), and total (RR=0.57; *P*=0.02) and coronary (RR=0.45; *P*=0.007) mortality. Our results extend previous findings in patients with DM to a larger cohort, confirming the benefit of cholesterol lowering with simvastatin treatment on CHD events. In addition, significant decreases in total mortality, major coronary events and revascularizations were observed in simvastatin-treated patients with impaired fasting glucose levels. These results strongly support the concept that cholesterol lowering with simvastatin therapy improves the prognosis of patients with elevated fasting glucose levels [≥6.0 mmol/l (≥110 mg/dl)] or DM and known CHD.

Comment

This *post hoc* subgroup analysis of the 4S study extends the previous findings by Pyorala *et al.* by assessing the effect of simvastatin in a cohort of the 4S patients with normal fasting glucose (*n*=3237), impaired fasting glucose according to the 1997 American Diabetes Association (ADA) diagnostic criteria (*n*=678), and established DM (*n*=483). The results of this further analysis strongly support the concept that cholesterol lowering with simvastatin improves outcome in diabetic patients with known CHD. Data in this present study were also analysed for a number-needed-to-treat (NNT) approach. The NNT was 12 for normal fasting glucose, eight for impaired fasting glucose and seven for type 2 DM. In their 1999 position paper on the management of diabetic dyslipidaemia, the ADA recommended more aggressive management for all adults with type 2 diabetes and a target LDL cholesterol level of ≤2.59 mmol/l (≤100 mg/dl) was suggested. The evidence from this

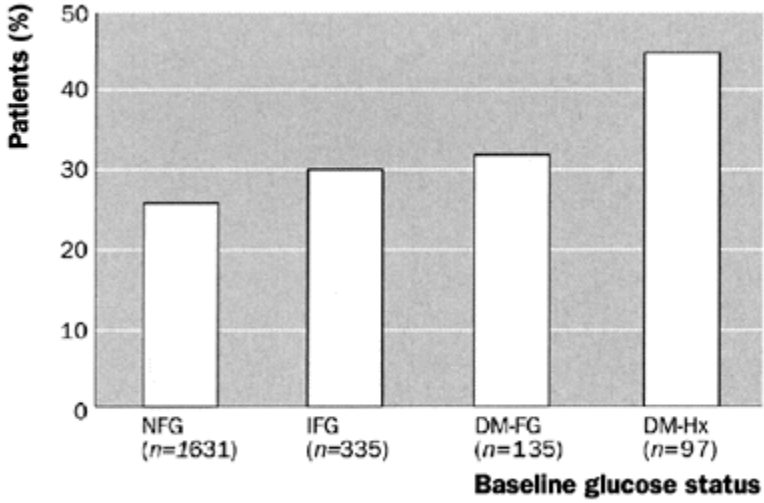

Fig. 11.7 Incidence of major CHD events by glucose status in the placebo group in the Scandinavian Simvastatin Survival Study (4S). NFG, normal fasting glucose; IFG, impaired fasting glucose; DM-FG, diabetes by elevated fasting glucose; DM-Hx, diabetes by clinical history. Source: Haffner *et al.* (1999).

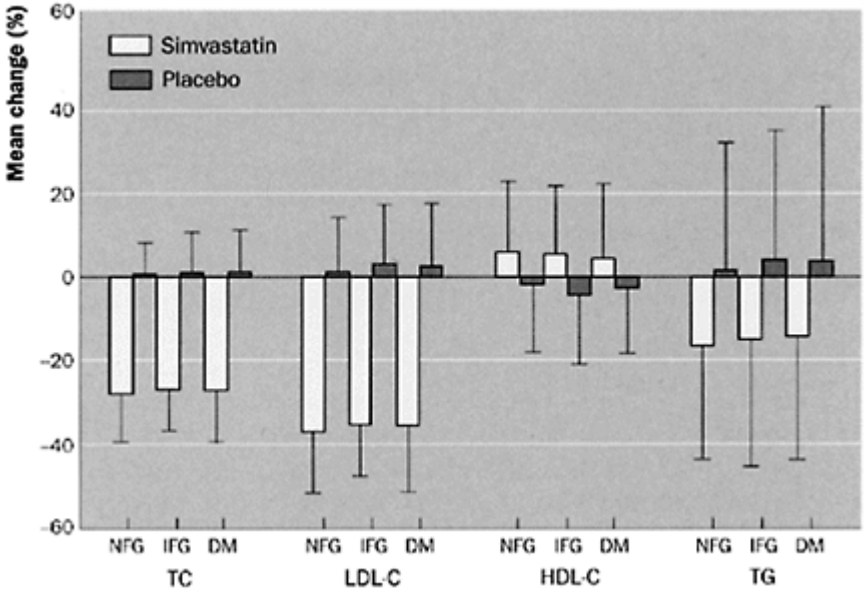

Fig. 11.8 Mean percentage change in lipid and lipoprotein levels by glucose status and treatment assignment (placebo *versus* simvastatin). NFG, normal lasting glucose, IFG, impaired fasting glucose; DM, diabetes

mellitus; TC, total cholesterol; LDL-C, LDL cholesterol; HDL-C, HDL cholesterol. Error bars indicate SEM. Source: Haffner *et al.* (1999).

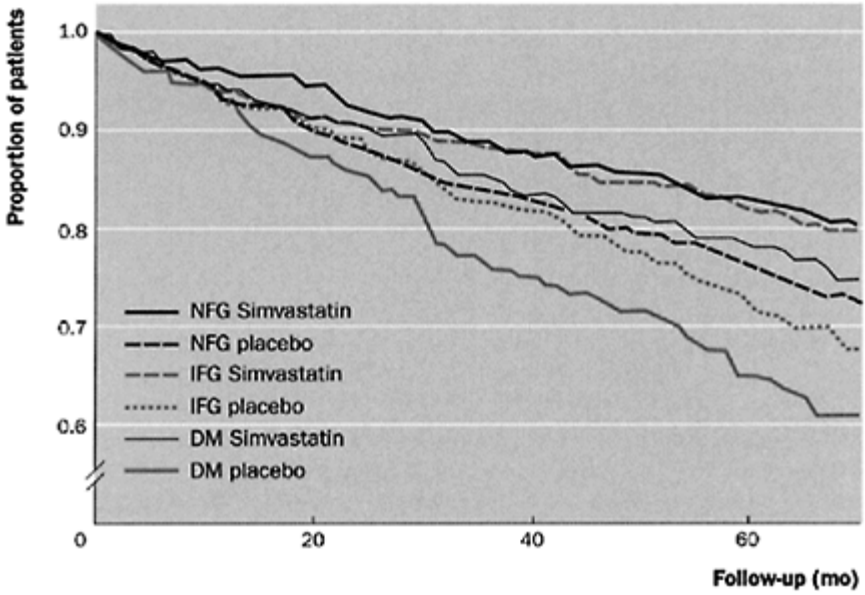

Fig. 11.9 Kaplan-Meier survival curves from the probability of remaining free of a major CHD event during follow-up in placebo- and simvastatin-treated patients with normal fasting glucose (NFG) levels, impaired fasting glucose (IFG) levels, and diabetes mellitus (DM). Source: Haffner *et al.* (1999).

particular paper supports the ADA position that all patients with DM should have the same LDL cholesterol goal as those with established CHD. Whilst again this study was a *post hoc* subgroup analysis of the 4S study, the percentage reduction of LDL cholesterol levels in patients with type 2 diabetes was similar to that seen in non-diabetic patients in the same study, and supports the inference that reduction of LDL cholesterol levels by statin therapy is likely to benefit patients with DM. These data suggest that patients with CHD and impaired fasting glucose (6–6.9 mmol/l) benefit from treatment by significant reduction in total and coronary mortality, major coronary events and revascularizations.

Cost-effectiveness of treating hyperlipidaemia in the presence of diabetes: who should be treated?
S A Grover, L Coupal, H Zowall, M Dorais. *Circulation* 2000; **102**:722–7.

B**ACKGROUND. This study estimated the long-term costs and benefits of treating hyperlipidaemia in diabetic patients with and without known cardiovascular disease after validating the Cardiovascular Life Expectancy Model. Model estimates were**

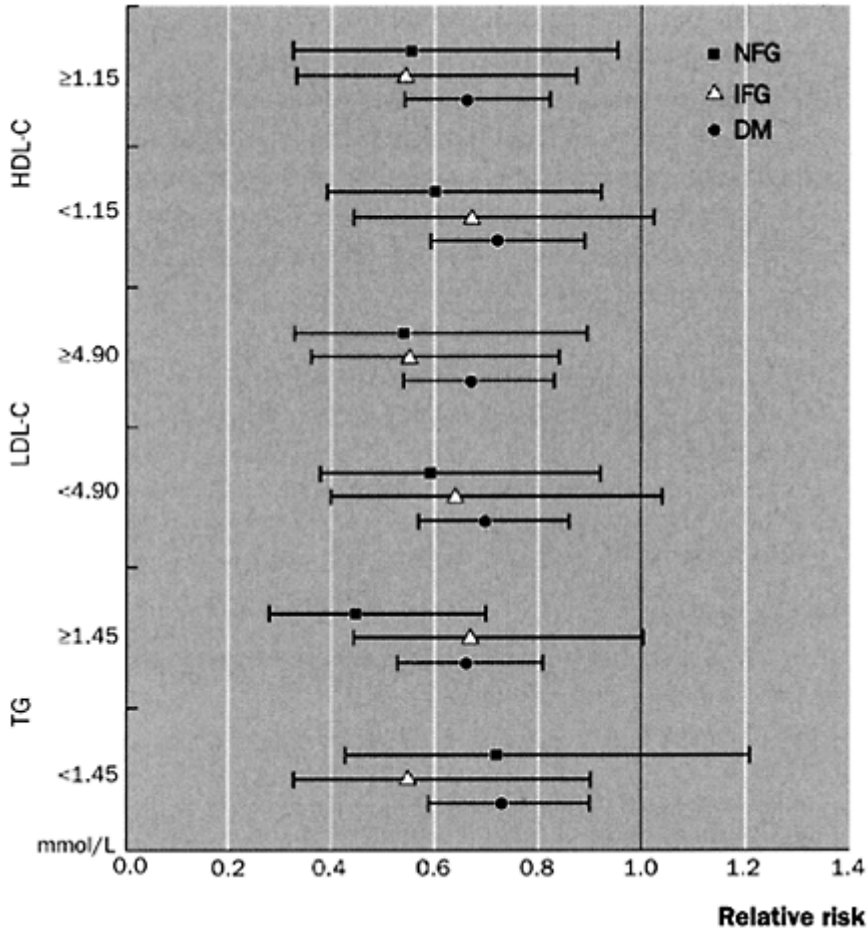

Fig. 11.10 Effects of simvastatin therapy on major coronary events by glucose status, stratified by level of lipid variables. NFG, normal fasting glucose; IFG, impaired fasting glucose; DM, diabetes mellitus, LDL-C, LDL cholesterol, HDL-C, LDL cholesterol. Error bars indicate SEM. To convert HDL-C and LDL-C from mmol/l to mg/dl, divide mmol/l by 0.02586. To convert TG from mmol/l to mg/dl, divide mmol/l by 0.01129.

Source: Haffner *et al*. (1999).

compared with the 4S study and used to estimate the long-term costs and benefits of treatment with simvastatin. Simulations were done for men and women with different pre-treatment LDL cholesterol levels.

I**NTERPRETATION.** Among adults with hyperlipidaemia, the presence of diabetes identifies men and women for whom lipid therapy is likely to be effective and cost-effective even in the absence of other risk factors or known cardiovascular disease. The model estimates showed good agreement with the results for the 4S diabetic patients. Treatment with simvastatin for patients with cardiovascular disease is cost-effective for men and women, with or without diabetes. For diabetic individuals without cardiovascular disease, primary prevention gave substantial benefits and was cost-effective.

Comment

There are many cost-effectiveness evaluations of statin therapy in the literature. In essence, the higher the initial risk of CHD in an individual patient, the greater the benefit from statin therapy. It therefore follows that the greater cost-effectiveness is in those high-risk patients—typically exemplified by a patient with type 2 diabetes. This cost-effectiveness model estimated the benefits and costs of treating patients with type 2 diabetes with and without cardiovascular disease, and showed that both primary and secondary prevention with statin therapy gives substantial benefit and was cost-effective. These conclusions were robust even among diabetics with low pre-treatment LDL levels and where small reductions of LDL were observed. The authors concluded that the presence of type 2 DM identifies men and women for whom statin therapy is likely to be cost-effective even in the absence of other risk factors or known cardiovascular disease.

LDL cholesterol as a strong predictor of coronary heart disease in diabetic individuals with insulin resistance and low LDL: the Strong Heart Study.

B V Howard, D C Robbins, M L Sievers, *et al. Arterioscler Thromb Vasc Biol* 2000; **20**:830–5.

B**ACKGROUND. The relative importance of diabetes-associated risk factors for cardiovascular disease, especially the role of lipid levels remains unclear, because LDL cholesterol often is not elevated in diabetic individuals. This analysis was designed to evaluate cardiovascular disease risk factors in diabetic individuals (4549 American Indians, 2034 with diabetes) and to compare the importance of dyslipidaemia (i.e. elevated TGs and low HDL cholesterol) and LDL cholesterol in determining cardiovascular disease risk in diabetic individuals.**

INTERPRETATION. At concentrations well below the National Cholesterol Education Program target of 130 mg/dl, LDL cholesterol is a strong independent predictor of CHD in individuals with diabetes, even when components of diabetic dyslipidaemia are present. The results support recommendations for aggressive control of LDL cholesterol in diabetic individuals, with a target level of<100 mg/dl.

Comment

In the UKPDS report on the risk factors for CHD in type 2 diabetes, LDL cholesterol was the best predictor of macrovascular disease events. In this study of American Indians, even at relatively low levels, LDL cholesterol was a strong independent predictor of CHD in patients with type 2 diabetes and supports recommendations in the USA for aggressive control of LDL cholesterol in patients with type 2 diabetes to target levels<100 mg/dl. High-density lipoprotein (HDL) levels also had a strong inverse effect in the multivariate model.

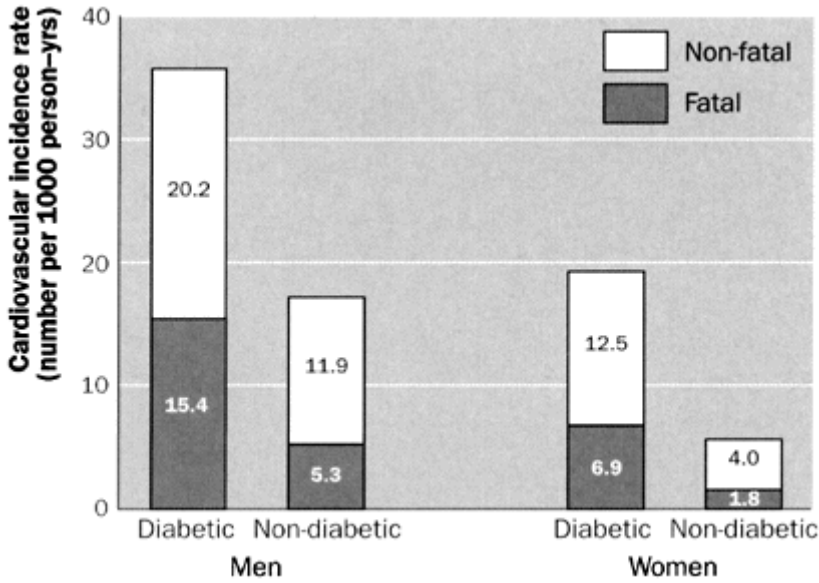

Fig. 11.11 Incidence rates of cardiovascular disease (CVD) by diabetic status: the Strong Heart Study. Overall incidence rates per 1000 person-years were as follows: for diabetic men, 31.8; for non-diabetic men, 16.4; for diabetic women, 17.9; and for non-diabetic women, 5.8. Person-years were calculated to whichever fatal or non-fatal event occurred for each individual. For fatal CVD, rates with 95% confidence intervals (CIs) were 15.4 (CI 11.2–19.6) in diabetic men, 5.3 (CI 3.0–7.6) in non-diabetic men, 6.9 (CI 4.9–8.9) in diabetic

women, and 1.8 (CI 0.6–3.1) in non-diabetic women. The relative risks for fatal CHD, stroke, and total fatal CVD were 3.1 (CI 1.8–5.3), 2.3 (CI 0.6–9.2), and 2.9 (CI 1.8–4.9) in diabetic men and 5.1 (CI 2.0–12.9), 1.5 (CI 0.4–5.9), and 3.8 (CI 1.8–8.0), respectively, in diabetic women. For non-fatal CVD, rates with 95% CIs for non-fatal CVD were 20.2 (CI 15.2–25.1) in diabetic men, 11.9 (CI 8.4–15.4) in non-diabetic men, 12.5 (CI 9.8–15.2) in diabetic women, and 4.0 (CI 2.1–5.8) in non-diabetic women. The relative risks for non-fatal CHD, non-fatal stroke, and total non-fatal CVD were 1.8 (CI 1.2–2.8), 1.7 (CI 0.7–4.2), and 1.7 (CI 1.2–2.5) in diabetic men and 3.3 (CI 1.8–6.0), 3.4 (CI 1.2–10.0), and 3.2 (CI 1.9–5.3), respectively, in diabetic women. Source: Howard *et al.* (2000).

Patients with type 2 diabetes: the case for primary prevention. S M Haffner. *Am J Med* 1999; **107**:43S–45S.

B**ACKGROUND/INTERPRETATION. A cogently argued editorial making the case that the risk of CHD is so high in those with type 2 diabetes that they qualify for treatment with statins even in the absence of clinically overt CHD.**

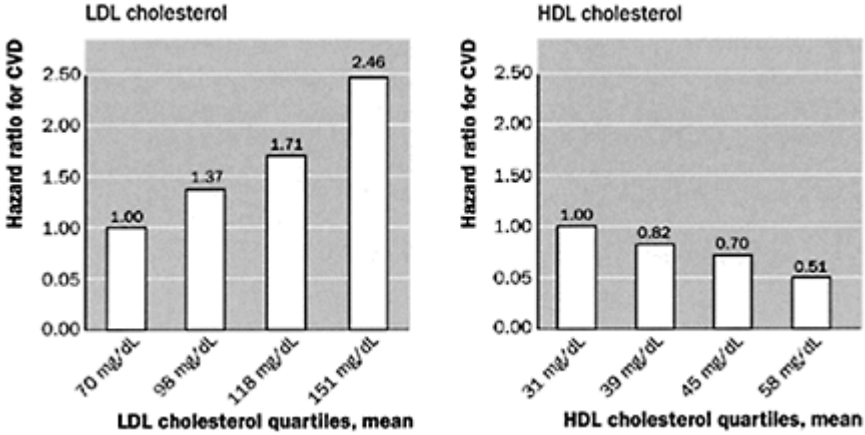

Fig. 11.12 Hazard ratios for fatal and non-fatal diabetes-associated cardiovascular disease by quartile for LDL and HDL cholesterol for diabetic men and women, calculated from results in this manuscript. The *y*-axis is logarithmic scale. Source: Howard *et al.* (2000).

Diabetes and cardiovascular disease. A statement for healthcare professionals from the American Heart Association.
S M Grundy, I J Benjamin, G L Burke, *et al. Circulation* 1999; **100**: 1134–46.

B**ACKGROUND/INTERPRETATION. A statement for healthcare professionals prepared by a committee of the American Heart Association chaired by Scott Grundy.**

Management of dyslipidaemia in adults with diabetes (supplement).
American Diabetes Association. *Diabetes Care* 1999; **22**:S56–9.

B**ACKGROUND/INTERPRETATION. A document outlining the goals of therapy and management priorities for diabetic dyslipidaemia.**

Comment

The above three important documents give an overview of the cardiovascular complications of diabetes, the opportunities for prevention, and the goals of therapy and management priorities.

Pravastatin and the development of diabetes mellitus. Evidence for a protective treatment effect in the West of Scotland Coronary Prevention Study.
D J Freeman, J Norrie, N Sattar, *et al. Circulation* 2001; **103**:357–62.

B**ACKGROUND/ INTERPRETATION. Hypertriglyceridaemia is a recognized precursor of the development of type 2 DM and, more recently, markers of low-grade inflammation have also been shown to have predictive value. Pravastatin has significant lipid regulating properties, lowering both total and LDL cholesterol levels as well as TG levels. Furthermore, it raises HDL cholesterol and has other pleiotropic effects, including antithrombotic and anti-inflammatory effects.**

Comment

This paper, a *post hoc* analysis of the West of Scotland Coronary Prevention Study, using serial fasting glucose measurements and ADA criteria, confirms the previously established predictors of glucose intolerance—namely body mass index and plasma TGs. However, it also identifies pravastatin therapy in this primary prevention trial to be associated with a reduced risk of developing type 2 diabetes. One possible explanation is that the impact of pravastatin on coronary events may lead to reduced prescription of thiazide and beta-blocking agents—both of which adversely affect glucose homeostasis. The West of Scotland Coronary Prevention Study database is unable to resolve this issue at present. An alternative explanation is that the 12% reduction in TG levels in this study could be responsible. The effects of TG lowering by fibric acid derivatives on glucose tolerance is controversial with contradictory reports in the literature. Another explanation is that pravastatin may improve insulin sensitivity by its beneficial effects on endothelial function.

This is an interesting hypothesis-generating paper that should be confirmed in appropriate prospective trials.

Conclusion

While we await appropriately designed prospective randomized control trials in patients with type 2 diabetes, to provide the incontrovertible evidence of the benefit of lipid-lowering therapy in these high-risk patients, there is already compelling evidence from a number of *post hoc* subgroup analyses of completed trials with statin therapy to justify an aggressive approach in risk factor management in patients with type 2 diabetes. As the present evidence stands, priorities should be given to the reduction of LDL cholesterol with a statin as well as tight control of blood pressure.

Hypertriglyceridaemia and the fibrate trials

The concept of statistical ‘independence’ for the effects of TGs on CHD risk is of limited value given the complex interdependence of the various lipoproteins involved in TG metabolism. These lipoproteins differ in their atherogenicity and the fact that the highest risk for CHD is linked to moderate, rather than severe hypertriglyceridaemia, is somewhat puzzling and counter-intuitive to many clinicians. Nevertheless, the meta-analysis published in 1996 by Hokanson and Austin |**5**|, which included 17 population-based studies comprising some 46 000 men and 10 000 women showed that the risk of cardiovascular disease increased by 30% in men and by 75% in women for every 1 mmol/l increase in TGs. After adjustments were made for changes in HDL levels, the figures were 14% and 37%, respectively, and were significant in both multivariate and univariate analyses. Subsequently, the 8-year follow up to the Copenhagen Male Study |**6**| showed that the risk of coronary death increased as TGs increased to 2.5 mmol/l across three levels of HDL cholesterol, supporting the concept of at least partial statistical independence from HDL.

Fibrates are ligands for the peroxisome proliferator activator receptor and lower plasma TGs and raise HDL—a lipid regulating effect that is particularly desirable in patients with type 2 diabetes. However, it is the clinical benefits that result from proper randomized control trials, which is the gold standard required before clinicians will embrace the widespread use of fibric acid derivatives in high-risk patients with a so-called atherogenic lipoprotein phenotype (ALP). A review of trials published between 1966 and 1996 includes 12 trials of therapy with fibrates or placebo in more than 21 000 patients |**7**|. Overall, these trials indicate no clinical benefit in terms of reduction in risk of coronary death. Since 1966, five additional trials have been published with fibrates compared with placebo in 6144 patients. Three will be discussed further, the VAHIT trial, the Bezafibrate Infarction Prevention (BIP) trial and the Diabetes Atherosclerosis Intervention Study (DAIS) trial.

Secondary prevention by raising HDL cholesterol and reducing triglycerides in patients with coronary artery disease: the Bezafibrate Infarction Prevention (BIP) study.

Israeli Society for Prevention of Heart Attacks. *Circulation* 2000; **102**:21–7.

B**ACKGROUND. CHD patients with low HDL cholesterol levels, high TG levels, or both, are at an increased risk of cardiovascular events. Whether raising HDL cholesterol or decreasing TGs reduces this excess risk remains to be confirmed.**

I**NTERPRETATION.** Bezafibrate, 400 mg daily for a mean 6.2 years, was safe and effective in elevating HDL cholesterol levels and lowering TGs in this double-blind trial of 3090 patients with a previous MI or stable angina. An overall trend towards reduction of the primary end-point (incidence of fatal or non-fatal MI or sudden death) was observed. The reduction in the primary end-point in patients with high baseline TGs (≥200 mg/dl) needs further confirmation.

Comment

The results from BIP were disappointing because they showed no significant effect of treatment with bezafibrate in either the primary end-point of the trial (non-fatal and fatal MI and sudden death) or on rates of coronary death (fatal MI and sudden death). In fact the rates of coronary deaths are the same in both bezafibrate and placebo groups. The rates of non-coronary death did not differ significantly between the two groups but tended to be slightly higher in the bezafibrate group (6.5% cf. 6%).

Patients in the BIP study differed to those in the VAHIT study by their level of baseline risk. Nearly 10% of patients in BIP were women and the mean age at entry was

60 years. Moreover, there were fewer patients with type 2 diabetes in BIP compared with VAHIT. The crude annual rate of the primary end-point in the placebo arm of BIP was 2.4%, which is substantially lower than the 3.7% equivalent seen in VAHIT. Moreover, a possible confounding issue is the relatively high rate of use of open label lipid-lowering drugs in the bezafibrate arm (11%) and particularly in the placebo arm (15%).

A *post hoc* subgroup analysis of the BIP trial data suggests a particular subgroup of patients with baseline TGs above 200 mg/l (approximately 2.2 mmol/l) had an accumulative probability of a primary end-point 39.5% lower (P=0.02) in the 234 patients randomized to bezafibrate compared with the 225 patients randomized to placebo. However, this is a *post hoc* analysis and needs to be tested prospectively in a proper randomized control trial.

Gemfibrozil for the secondary prevention of coronary heart disease in men with low levels of high-density lipoprotein cholesterol. Veterans Affairs High-Density Lipoprotein Cholesterol Intervention Trial Study Group.

H B Rubins, S J Robins, D Collins, *et al. N Engl J Med* 1999; **341**:410–18.

B**ACKGROUND. Although it is generally accepted that lowering elevated serum levels of low-density lipoprotein (LDL) cholesterol in patients with coronary heart disease is beneficial, few data exist to help make decisions about therapy for patients whose primary lipid abnormality is a low level of high-density lipoprotein (HDL) cholesterol.**

I**NTERPRETATION.** A double-blind trial was conducted to compare gemfibrozil (1200 mg per day) with placebo in 2531 men with coronary heart disease (CHD) and HDL cholesterol level of 40 mg per decilitre (1.0 mmol per litre) or less and an LDL cholesterol level of 140 mg per decilitre (3.6 mmol per liter) or less. The primary study

outcome was non-fatal myocardial infarction or death from coronary causes. The median follow-up was 5.1 years. At 1 year, the mean HDL cholesterol level was 6% higher, the mean triglyceride level was 31% lower, and the mean total cholesterol level was 4% lower in the gemfibrozil group than in the placebo group. There was no significant difference in LDL cholesterol levels between the groups. A primary event occurred in 275 of the 1267 patients assigned to placebo (21.7%) and in 219 of the 1264 patients assigned to gemfibrozil (17.3%). The overall reduction in the risk of an event was 4.4 percentage points, and the reduction in relative risk was 22% (95% CI, 7 to 35%; P=0.006). A 24% reduction in the combined outcome of death from CHD, non-fatal myocardial infarction, and stroke (P<0.001) was noted. There were no significant differences in the rates of coronary revascularization,

hospitalization for unstable angina, death from any cause, and cancer. Gemfibrozil therapy resulted in a significant reduction in the risk of major cardiovascular events in patients with coronary disease whose primary lipid abnormality was a low HDL cholesterol level. The findings suggest that the rate of coronary events decreased by raising HDL cholesterol levels and lowering levels of triglycerides without lowering LDL cholesterol levels

Comment

The inverse relationship between the level of HDL and the development of premature CHD has been recognized for two decades or so and is generally considered secure. In clinical practice, the ratio of total cholesterol to HDL cholesterol is more informative than HDL cholesterol alone. This is particularly shown to be so in the PROCAM study of more than 80 000 men and women |**8**|. Until recently there was little evidence for the clinical benefits of raising HDL levels from randomized control trials other than *post hoc* subgroup analyses of fibrate trials such as the Helsinki Heart Study |**9**| and secondary prevention trials in Stockholm using a combination of clofibrate/nicotinic acid |**10**|.

The recently reported VAHIT trial provides the first direct evidence that raising the concentration of HDL and lowering plasma TGs reduces the incidence of cardiovascular events. In approximately 30% of patients with CHD, the predominant dyslipidaemia is lowered levels of HDL and elevated levels of TGs. Patients with such lipid profiles were in general not included in the statin trials. Moreover, the levels of total and LDL cholesterol at entry into the VAHIT were within the limits of most recent European guidelines for coronary prevention. The primary results are shown in the Table 11.3. The numbers included in the trial were 2531. There was no change in the non-cardiac mortality or in the incidence of cancer observed in this trial. The 6% increase in HDL cholesterol in VAHIT was associated with a 22% reduction in cardiac events. This result confirms the analysis of the Helsinki Heart Study that predicts that an 8% rise in HDL will give a 23% reduction in event rate |**11**|.

Although the intention of VAHIT was to investigate the effects of raising HDL cholesterol, the benefits seen are undoubtedly due to the combined effects of a 6% increase in HDL coupled with a 31% fall in TGs. It is precisely this type of response that the clinician seeks in treating the typical dyslipidaemia or patients with type 2

Table 11.3 Primary results

1.	Fatal and non-fatal MI	−22%	*P*=0.006
2.	Deaths due to CHD	−22%	*P*=0.07
3.	Non-fatal MI	−23%	*P*=0.02
4.	Confirmed stroke	−25%	*P*=0.01

Source: Rubins *et al.* (1999).

Table 11.4 Lipid levels at 1 year

Lipid	Placebo (*n*=1267)	Gemfibrozil (*n*=1264)	*P*
Total cholesterol (mmol/l)	4.6	4.4 (−4%)	<0.01
LDL cholesterol (mmol/l)	2.9	2.9 (0%)	NS
HDL cholesterol (mmol/l)	0.8	0.9 (+6%)	<0.001
Triglycerides (mmol/l)	1.9	1.3 (−31%)	<0.001

Source: Rubins *et al.* (1999).

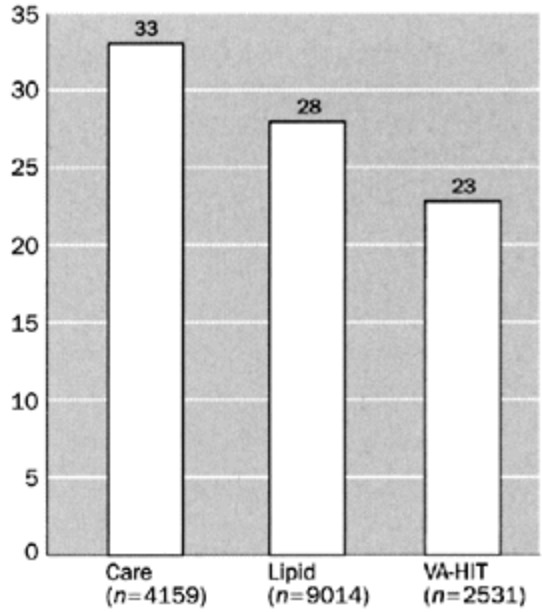

Fig. 11.13 VAHIT, CARE and the Long-term Intervention with Pravastatin in Ischaemic Disease Study. Source: Rubins *et al.* (1999).

diabetes or the metabolic syndrome. This is sometimes called the atherogenic lipoprotein phenotype.

The effect of fenofibrate on the progression of coronary heart

disease.

Diabetes Atherosclerosis Intervention Study. *Lancet* 2001; **357**:905–10.

B**ACKGROUND. Atherosclerosis is the most common complication of diabetes. Correction of hyperglycaemia helps to prevent microvascular complications but has little effect on macrovascular disease. *Post hoc* analyses of diabetic subpopulations in lipid intervention trials suggest that correction of lipoprotein abnormalities will lead to a decrease in coronary-artery disease. The Diabetes Atherosclerosis Intervention Study (DAIS) was specifically designed to assess the effects of correcting lipoprotein abnormalities on coronary atherosclerosis in type 2 diabetes.**

I**NTERPRETATION.** Metabolic and angiographic criteria were used to screen 731 men and women with type 2 diabetes. Of these, 418 were randomly assigned micronized fenofibrate (200 mg/day) or placebo for at least 3 years. They were in good glycaemic control (mean haemoglobin A16 7.5%), had mild lipoprotein abnormalities, typical of type 2 diabetes, and at least one visible coronary lesion. Half had no previous clinical coronary disease. Initial and final angiograms followed a standard protocol and were analysed by a computer-assisted quantitative approach. Missing data for the primary end-points (minimum lumen diameter, mean segment diameter, and mean percentage stenosis) were imputed. Total plasma cholesterol, HDL-cholesterol, LDL-cholesterol, and triglyceride concentrations all changed significantly more from baseline in the fenofibrate group (n=27) than in the placebo group (n=211). The fenofibrate group showed a significantly smaller increase in percentage diameter stenosis than the placebo group (mean 2:11 [SE 0.594] *vs* 3.65 [0.608]%, P=0.02), a significantly smaller decrease in minimum lumen diameter (−0.06 [0.016] *vs*−0.10 [0.016] mm, P=0.029, and a non-significantly smaller decrease in mean segment diameter (−0.06 [0.017] *vs*−0.08 [0.018] mm, P=0.171. The trial was not powered to examine clinical end-points, but there were fewer in the fenofibrate group than the placebo group (38 *vs* 50). DAIS suggests that treatment with fenofibrate reduces the angiographic progression of coronary-artery disease in type 2 diabetes. This effect is related, at least partly, to the correction of lipoprotein abnormalities, even those previously judged not to need treatment.

Comment

This is the first trial performed specifically in a cohort with type 2 diabetes and was performed in 11 centres in Canada and Europe; 418 (305 men, 113 women) patients with type 2 diabetes were screened by metabolic and angiographic criteria and randomized to micronized fenofibrate or placebo therapy for at least 3 years. Glycaemic control was good (mean HbA1c 7.5%) and the mean lipid concentrations were generally within or near those that are widely considered ‘normal’ and resemble the typical profile of type 2 diabetes. This study was underpowered to allow definitive conclusions to be drawn about clinical end-points but there was a consistently lower rate of each individual end-point

with micronized fenofibrate compared with placebo except for hospital admission with angina. The DAIS population was small with a relatively short follow-up period but the difference in the

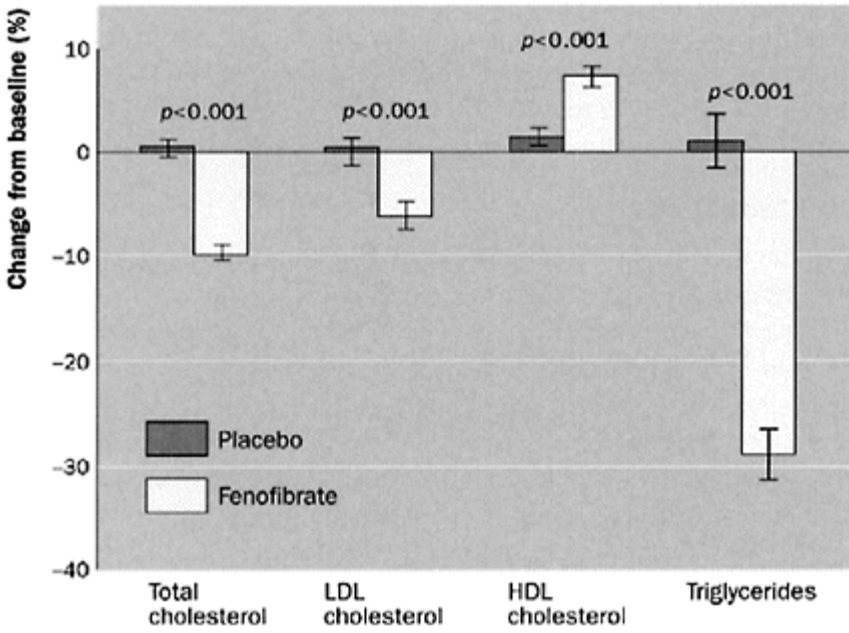

Fig. 11.14 Changes in lipid values in placebo and fenofibrate groups. Error bars=SE.

Source: Diabetes Atherosclerosis Intervention Study (2001).

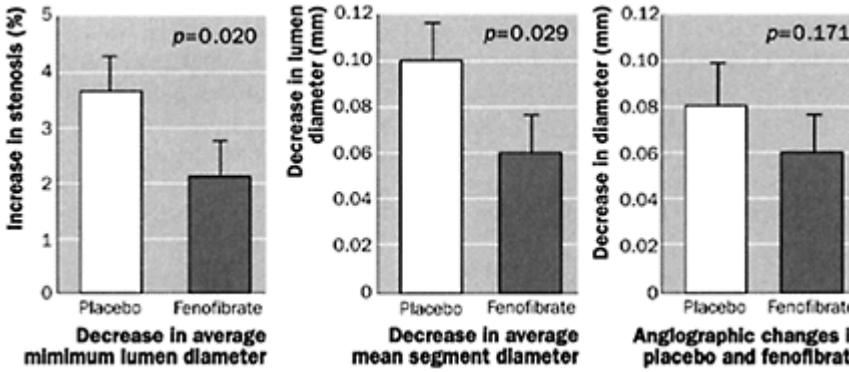

Fig. 11.15 Angiographic changes in placebo and fenofibrate groups. Error bars=SE.

Source: Diabetes Atherosclerosis Intervention Study (2001).

rate of end-points (23%) is very similar to that seen in the *post hoc* analyses of the larger and longer trials VAHIT, 4S and CARE. The study population in DAIS was recruited on metabolic and angiographic criteria and only 50% had clinically overt CHD. Thus the clinical event rate was less than in the aforementioned trials.

The angiographic changes observed in DAIS are of the same magnitude as several other angiographic trials in people without diabetes. In some of these trials, despite small populations and short duration, significant differences in clinical end-points were observed. Moreover, pooling of the angiographic trials revealed significant reduction in clinical end-points. The findings of DAIS are, therefore, encouraging and lend credence to the hypothesis that correction of the relatively mild lipid abnormalities typically found in type 2 diabetes reduces the risk of CHD.

Future and ongoing trials

Ongoing trials, either specifically in patients with type 2 diabetes (Collaborative Atorvastatin Diabetes Study, Atorvastatin as Prevention of Coronary Heart Disease in Patients with Type 2 Diabetes Mellitus, The Fenofibrate Intervention and Event Lowering in Diabetes Trial, Lipids in Diabetes Study) or trials containing substantial numbers of patients with type 2 diabetes (Heart Protection Study) will provide the clinical outcome evidence base, which will dictate specific recommendations for lipid regulating therapy in the management of type 2 diabetes. Dyslipidaemia is a major risk factor of macrovascular disease in diabetes. In the UK Prospective Diabetes Study Group (UKPDS), more intensive glycaemic control was associated with significant reduction microvascular end-points but resulted in a very modest effect on macrovascular end-points (UKPDS 33). Moreover, LDL cholesterol was the best predictor of macrovascular events (UKPDS 23).

Heart Protection Study |**12,13**|

This study, which is co-ordinated by the University of Oxford Clinical Trials Centre UK contains a large number of diabetic patients with and without vascular disease; 20 536 individuals have been recruited and are considered to be at high risk because of either established CHD, cerebrovascular risk peripheral vascular risk or hypertension. It is a double blind randomized trial of either simvastatin (40 mg/day) versus placebo and antioxidant vitamins (vitamins C, E and β-carotene) versus placebo. It is scheduled for 5 years duration and contains 5963 patients with DM, approximately 90% of which have type 2 diabetes. The primary end-points are all-cause mortality, CHD mortality and non-CHD mortality. It is expected to report in 2001.

As this trial population contains large numbers of elderly patients and patients with below average cholesterol levels, it will substantially increase the evidence base for these groups of patients.

Lipids in Diabetes Study (LDS)

Again co-ordinated by the Clinical Trials Unit at the University of Oxford, this is the first

study to examine the combination of a statin (cerivastatin 400 μg) and a fibrate (micronized fenofibrate 200 mg) in a study population of approximately 5500 men and women with type 2 diabetes but without clinical evidence of CHD. The primary end-point will be a composite of fatal and non-fatal CHD and a revascularization procedure. It is expected to be completed by 2005 and is currently recruiting.

Collaborative Atorvastatin Diabetes Study (CARDS)

This study is a randomized placebo controlled double-blind multicentre trial that is now focused on the effect of atorvastatin 10 mg versus placebo in patients with type 2 diabetes who have normal or moderately raised LDL cholesterol levels but no previous history of CHD or other macrovascular disease. In total 2750 will be recruited with a 4-year follow-up. Patients will be aged 40–75 years at entry and must have either hypertension, retinopathy, microalbuminuria or macroalbuminuria or be a current smoker. The LDL cholesterol must be 4.14 mmol/ or less (≤160 mg/dl) and the TGs 6.7 mmol/l or less (≤ 600 mg/dl). The primary end-point is the time to the first of any of the following: fatal or non-fatal MI, acute CHD death, coronary revascularization procedure or stroke. Results are expected to report in 2004.

Atorvastatin as Prevention of Coronary Heart Disease in Patients with Type 2 Diabetes Mellitus (ASPEN)

ASPEN involves 2250 patients in 70 centres in the USA, Europe and Australia. This study is ongoing and is due to complete in 2003. It has finished randomization and in many ways is similar to CARDS. It is a randomized trial of atorvastatin 10 mg versus placebo but includes patients with type 2 diabetes with a previous history of a MI as well as patients with type 2 diabetes without a history of MI. The lipid entry criteria is the same as CARDS for those without a history of previous MI. Those with a history of previous MI must have an LDL cholesterol level 3.62 mmol/l or below and TGs of 6.78 mmol/l or below. Similarities between CARDS and ASPEN will allow pooling of the data in subgroup analyses.

The Fenofibrate Intervention and Event Lowering in Diabetes Trial (FIELD)

This is a randomized controlled trial comparing micronized fenofibrate 200 mg/day versus placebo in 8000 men and women with type 2 diabetes. It is a combined primary and secondary prevention trial, including 2000 patients with established CHD and 6000 without CHD. Lipid entry criteria are a total/HDL cholesterol ratio of 4 or over or a fasting TG level of between 1 and 5 mmol/l. Total cholesterol levels are 3–6.5 mmol/l. Trial duration will be 5–7 years and it is currently recruiting in Australia, New Zealand and Finland.

Conclusion

As yet, apart from DAIS, there are no trials reported of lipid lowering specifically in

patients with type 2 diabetes. The ongoing trials will provide the evidence base for specific recommendations as well as allowing cost-benefit analyses to be performed.

The statin drugs are highly effective in lowering total and LDL cholesterol levels and have been shown to reduce significantly mortality and morbidity in *post hoc* analysis of patients with type 2 diabetes. Moreover, they also lower TGs, particularly when levels are raised, as well as increasing HDL levels. Thus, at present, the reduction of LDL cholesterol levels with a statin remains the priority in the management of diabetic dyslipidaemia. The ongoing trials will, hopefully, clarify the role of fibrates in clinical practice and in particular, the LDS study with its statin/ fibrate combination arm will allow an evaluation as to whether there is a synergistic effect on clinical events with this potentially attractive treatment modality.

Pharmacotherapy efficacy studies

Atorvastatin for the management of type 2 diabetic patients with dyslipidaemia. A mid-term (9 months) treatment experience.
M Velussi, A M Cernigoi, C Tortul, M Merni. *Diabetes Nutr Metab* 1999; **12**: 407–12.

B**ACKGROUND. Dyslipidaemia, particularly increased TGs and low HDL cholesterol, represents an important risk factor for macrovascular complications in type 2 diabetes. This study evaluated the effects of atorvastatin in a population of type 2 DM patients according to their cardiovascular risk: evidence of myocardial or coronary lesions (group A); evidence of familiar hypercholesterolaemia (group B); evidence of stable cardiovascular risk (group C).**

I**NTERPRETATION.** This preliminary study suggests that the management of hypercholesterolaemia with atorvastatin may be useful for both primary and secondary prevention of chronic complications of type 2 diabetes. Lipid profile improved significantly during atorvastatin treatment (generally 10 mg/day): total cholesterol, TGs and LDL cholesterol were reduced and HDL was increased. Atorvastatin treatment significantly reduced microalbuminuria and fibrinogen levels. In patients with hypertension, diastolic blood pressure values were reduced without modifying antihypertensive treatment.

Comment

Atorvastatin 10 mg can be an effective treatment for the typical dyslipidaemia seen in type 2 diabetes.

Efficacy and safety of micronised fenofibrate in a randomized double-blind study comparing four doses from 200 mg to 400 mg daily with placebo in patients with hypercholesterolemia.
M Krempf, V Rohmer, M Farnier, *et al. Diabetes Metab* 2000; **26**:184–91.

B**ACKGROUND. This study aimed to evaluate the efficacy on LDL cholesterol of micronized fenofibrate given for 3 months at doses of 200–400 mg once daily, compared with placebo. A double-blind, randomized, parallel group, trial was performed in 340 hypercholesterolaemic patients.**

I**NTERPRETATION.** This study showed treatment for 3 months with micronized fenofibrate at doses of up to 400 mg daily is effective and can reduce LDL cholesterol by up to 30%. Further evaluation of these doses in longer trials is needed. LDL cholesterol, total cholesterol, total TGs and apolipoprotein B were significantly decreased compared with placebo in all four fenofibrate groups. Lipid values in the placebo group remained unchanged. There were no major clinical or biological adverse events at doses of 200–400 mg micronized fenofibrate daily.

Comment

This is an efficacy trial of micronized fenofibrate showing the usual lipid-regulating effects of the fibric acid derivative. Fenofibrate appears to be a well tolerated treatment modality.

Ciprofibrate effects on carbohydrate and lipid metabolism in type 2 diabetes mellitus subjects.
A Hernandez-Mijares, I Lluch, E Vizcarra, *et al. Nutr Metab Cardiovasc Dis* 2000; **10**:1–6.

B**ACKGROUND. Atherosclerosis is the most common cause of morbidity and mortality in type 2 DM. Hypertriglyceridaemia and low HDL cholesterol, important cardiovascular risk factors, seem to be related to insulin resistance. Fibrates are effective for treating dyslipoproteinaemia in diabetes. This crossover study compared the effects of ciprofibrate 100 mg for 4 weeks with those of placebo on dyslipoproteinaemia, fibrinogen plasma concentrations, glycaemic**

control and insulin action in 13 patients with type 2 DM.

I**NTERPRETATION.** Ciprofibrate has a potent hypolipidaemic effect, especially decreasing TGs, very low-density lipoprotein and fibrinogen, and increasing HDL cholesterol, but does not influence glycaemic control or insulin action. Decreased insulin secretion may be due to peripheral use of glucose due to the drug's antilipolytic action.

Comment

This study illustrates the typical lipid regulating effect of ciprofibrate—a fibric acid derivative. This study also indicates that fibrinogen levels are lowered with ciprofibrate therapy, which may have a beneficial effect on the risk of CHD. This fibrinogen lowering effect has also been demonstrated with bezafibrate. There was no overall effect on glycaemic control in this study contributing to the contradictory data published relating as to whether improvement of the lipid profile in patients with type 2 diabetes also results in improved glycaemic control.

Pravastatin compared with bezafibrate in the treatment of dyslipidaemia in insulin-treated patients with type 2 diabetes mellitus.
C Rustemeijer, J A Schouten, H J Voerman, *et al. Metab Res Rev* 2000; **16**: 82–7.

B**ACKGROUND. Both statins and fibrates are used to treat dyslipidaemia in type 2 diabetes patients. The lipid-lowering effects of pravastatin 40 mg and bezafibrate 400 mg on serum lipids, lipoproteins and lipoprotein composition in 45 dyslipidaemic, insulin-treated type 2 diabetes patients were compared in this double-blind, cross-over study.**

I**NTERPRETATION.** Pravastatin treatment is superior for reducing cholesterol-enriched lipoprotein subpopulations and improving cardiovascular risk factors. Bezafibrate is more effective for raising HDL cholesterol and improving LDL particle composition.

Comment

This study illustrates that pravastatin is most effective in reducing LDL cholesterol levels while bezafibrate is most effective in reducing TGs and elevating HDL levels. Moreover, the lipid regulating effects of bezafibrate has a knock-on effect on LDL particle composition, which is also thought to be of benefit.

Bezafibrate reduces blood glucose in type 2 diabetes mellitus.
S Ogawa, K Takeuchi, K Sugimura, *et al. Metabolism* 2000; **49**:331–4.

B**ACKGROUND. The clinical efficacy of bezafibrate was examined with special reference to glucose metabolism in patients with type 2 DM. In protocol 1, 342 patients with type 2 DM and hyperlipidaemias were randomly divided to receive bezafibrate for 16 weeks or no bezafibrate. In protocol 2, 20 type 2 DM patients were randomly divided to receive bezafibrate for 8 weeks or no bezafibrate, and a meal tolerance test (MTT) was done.**

I**NTERPRETATION.** In patients with type 2 DM both hyperglycaemia and hyperlipidaemia can be improved by bezafibrate treatment. In protocol 1, bezafibrate

significantly reduced fasting levels of TG, total cholesterol, plasma glucose and haemoglobin A_{1c} and significantly increased HDL cholesterol. In protocol 2, fasting TG, plasma glucose and insulin levels were significantly reduced by bezafibrate. In the MTT, postprandial increments of TG were significantly blunted after bezafibrate, whereas postprandial plasma glucose and insulin levels were not significantly changed. Leptin levels were significantly decreased, while tumour necrosis factor-α levels were not changed.

Comment

The possible role of fibrates in improving insulin sensitivity is controversial. The literature has many contradictory reports. This report supports the hypothesis that improving the lipid profile in type 2 diabetes also results in improved glycaemic control. Moreover, postprandial lipaemia is also attenuated. No studies on endothelial function was done in this paper and thus any benefits from surrogate markers of CHD as a consequence of bezafibrate therapy cannot be commented on.

Effects of psyllium on glucose and serum lipid responses in men with type 2 diabetes and hypercholesterolaemia.
J W Anderson, L D Allgood, J Turner, *et al. Am J Clin Nutr* 1999; **70**: 466–

73.

B**ACKGROUND. Water-soluble dietary fibres decrease postprandial glucose and serum cholesterol concentrations. This study examined the effects of administering psyllium husk fibre to men with type 2 diabetes. and mild-to-moderate hypercholesterolaemia; 34 men were randomly assigned to receive 5.1 g psyllium or cellulose placebo twice daily for 8 weeks.**

I**NTERPRETATION.** The results indicate that adding psyllium to a traditional diet for patients with diabetes is safe and well tolerated, and improves glycaemic and lipid control in men with type 2 diabetes and hypercholesterolaemia. Serum total and LDL cholesterol concentrations were significantly lower in the psyllium group compared with the placebo group. All-day and postprandial glucose concentrations were lower in the psyllium than in the placebo group. Both products were well tolerated.

Comment

You are what you eat! A reminder that significant benefits obtain from healthy eating and water-soluble dietary fibres can have beneficial effects on both glycaemic control and postprandial glycaemic excursions. There are also beneficial changes on the lipid profile observed. Psyllium husk is a viscous, mostly water-soluble fibre prepared by the mechanical removal of the husk from blonde psyllium seed *(Plantaga ovata)*. This 8-week study showed that the addition of psyllium to a traditional diet in patients with type 2 diabetes is safe and well tolerated with improved metabolic control.

Table 11.5 Serum glycemic and lipid responses in metabolic ward and outpatient settings in subjects in the psyllium and control groups[1]

	Control (*n*=14)		**Psyllium (*n*=15)**	
	Baseline	**Percentage change**	**Baseline**	**Percentage change**
Outpatient				
Body weight (kg)	87.1±3.3	1.5±0.7	89.6±2.4	−0.3±0.4[3]
Glucose (mmol/L)	10.74±0.56	2.8±4.6	10.02±0.41	−6.1±4.5
Hb A_{1c}	0.075±0.002	−0.8±4.3	0.073±0.003	−6.3±3.1
Glycated albumin	0.0222±0.0010	−5.6±6.2	0.020±0.001	−3.1±4.1
Total cholesterol (mmol/L)	5.89±0.15	2.8±2.3	6.08±0.18	−2.3±2.2
LDL cholesterol (mmol/L)	3.80±0.17	2.8±3.4	4.00±0.23	−4.9±2.4

HDL cholesterol (mmol/L)	0.94±0.05	8.8±2.3	0.97±0.07	−0.9±3.0[2]
Triacylglycerols (mmol/L)	2.50±0.20	−0.4±5.3	2.71±0.35	−7.0±13.3
Metabolic ward				
Glucose (mmol/L)				
Postbreakfast	13.54±0.95	3.8±4.7	13.44±0.82	−3.0±4.6
Postlunch	10.43±0.83	12.7±5.6	10.75±0.69	−6.5±4.2[3]
Postdinner	10.89±0.61	2.2±3.9	11.80±0.75	−5.7±4.5
All day	11.53±0.76	6.8±3.9	11.90±0.70	−4.2±3.3[2]
Total cholesterol (mmol/L)	5.39±0.17	6.9±2.4	5.69±0.20	−2.1±2.3[2]
LDL cholesterol (mmol/L)	3.39±0.17	8.3±5.3	3.81±0.19	−4.7±4.3
HDL cholesterol (mmol/L)	0.85±0.05	2.0±2.2	0.88±0.07	0.6±3.1
Triacylglycerols (mmol/L)	2.50±0.23	13.7±7.3	2.54±0.31	6.5±6.8
VAP lipoprotein cholesterol (mmol/L)				
LDL	3.39±0.14	2.7±3.5	3.70±0.20	−7.0±3.7
HDL	0.86±0.05	−2.6±3.6	0.85±0.07	−0.1±4.2
HDL_2	0.14±0.02	−19.2±9.2	0.12±0.04	8.7±15.9
HDL_3	0.72±0.03	1.4±4.3	0.72±0.05	−3.1±4.2
Apolipoprotein B (g/L)	1.45±0.08	5.7±2.3	1.46±0.08	−1.5±3.7
Apolipoprotein A (g/L)	1.064±0.032	2.5±3.0	1.047±0.079	2.2±4.4

1 ±SEM. Baseline values were measured at weeks −2, −1, and 0; final values were measured at week 8. VAP, vehicle auto profile; Hb A1c, glycated hemoglobin.
2,3 Significantly different from control group: [2] $P<0.05$, [3] $P=0.01$.
Source: Anderson *et al.* (1999).

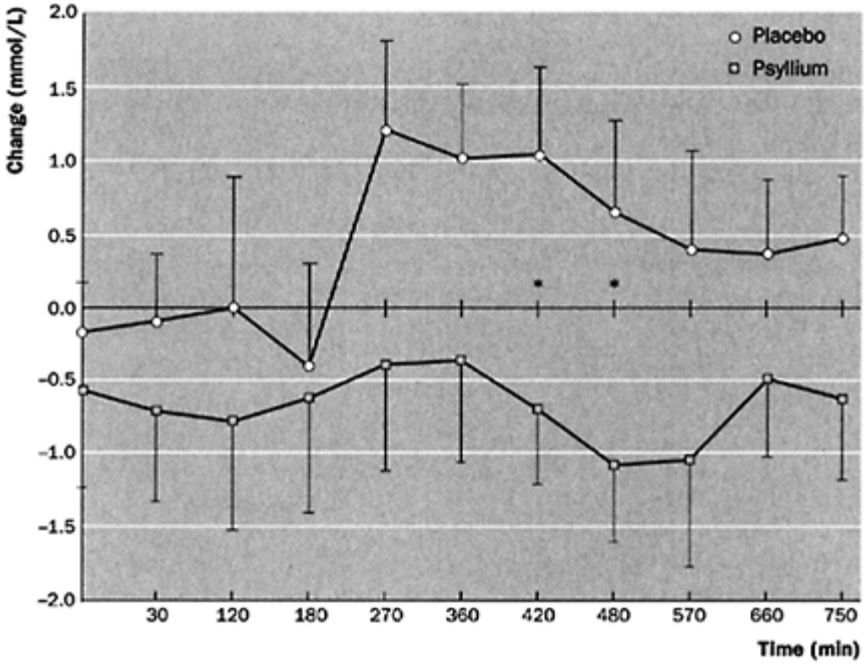

Fig. 11.16 Mean (±SEM) changes (final values at week 8—initial values at week 0) in serum glucose concentrations before and after meals for subjects in the placebo (○) and psyllium ( ) groups. *Significant difference between groups, $P<0.05$.

Source: Anderson *et al.* (1999).

Efficacy and safety of cerivastatin for type 2 diabetes and hypercholesterolaemia. Hyperlipidaemia in diabetes mellitus investigators.

A Rubinstein, F J Maritz, S G Soule, *et al. Cardiovasc Risk* 1999; **6**: 399–403.

B**ACKGROUND. CHD is much more prevalent in diabetic patients compared with non-diabetic individuals, and its prognosis is poorer. Serum total and LDL cholesterol concentrations are powerful predictors of CHD morbidity and mortality in patients with type 2 diabetes. The available data suggest that the target cholesterol concentration in patients with diabetes should be similar to that in non-diabetic individuals with a previous MI. This double-blind, randomized study**

assesses a new, highly potent statin, cerivastatin in type 2 diabetic patients with hypercholesterolaemia.

Table 11.6 Changes in efficacy variables in the patient population valid for efficacy analysis

	Control (*n*=45)		**Cerivastatin 0.1 mg (*n*=101)**		**Cerivastatin 0.3 mg (*n*=106)**	
	Baseline	**Endpoint**	**Baseline**	**Endpoint**	**Baseline**	**Endpoint**
Cholesterol (mmol/I)**	6.39	6.46 (+1.5%)	6.45	5.55 (−13.7%)	6.34	4.84 (−23.5%)
LDL cholesterol (mmol/I)**	4.29	4.29 (+0.6%)	4.34	3.45 (−20.2%)	4.24	2.80 (−33.8%)
HDL cholesterol (mmol/I)*	1.14	1.17 (+3.1%)	1.13	1.19 (+5.7%)	1.15	1.21 (+6.2%)
Triglycerides (mmol/l)U	2.09	2.17 (+4.5%)	2.12	1.97 (−3.9%)	2.07	1.79 (−12.3%)

Values in parentheses are % changes from baseline to endpoint. ** $P<0.001$ for difference between the three groups.

* $P<0.05$ for 0.3 mg compared with 0.1 mg and for 0.1 mg compared with placebo; no significant difference between the three groups.

$P=0.0004$ for difference between the three groups, $P<0.05$ for 0.3 mg compared with placebo; no significant difference between 0.1 mg and placebo. LDL, low-density lipoprotein; HDL, high-density lipoprotein.

Source: Rubinstein *et al.* (1999).

Table 11.7 Reduction in LDL cholesterol in the study population

		Cerivastatin	
	Placebo (*n*=45)	**0.1 mg (*n*=101)**	**0.3 mg (*n*=106)**
Baseline LDL cholesterol (mmol/l)	4.29	4.34	4.24
Endpoint LDL cholesterol			
<3.35 mmol/l	4 (8.8)	56 (55.4)	87 (82)
<2.58 mmol/l	0(0)	13 (12.3)	47 (44.3)
<2.58 mmol/l with coronary disease	1/17 (6)	6/25 (24)	13/36 (36)
Endpoint LDL cholesterol in patients with	4.37	3.53	2.85

coronary disease (mmol/l)

Values are number (%) of patients. LDL, low-density lipoprotein.
Source: Rubinstein *et al.* (1999).

INTERPRETATION. Hypercholesterolaemic patients with type 2 diabetes showed significant reductions in LDL and total cholesterol concentrations after cerivastatin treatment. The dose of 0.3 mg cerivastatin is effective in diabetic hypercholesterolaemia, with co-reduction of TG concentrations. Current clinical trials are assessing the effects of cerivastatin on coronary morbidity and mortality.

Comment

Another efficacy and safety trial of the HMG CoA reductase inhibitor cerivastatin using a dosage of 0.3 mg. It shows the predictable effect on lipoprotein levels seen in all the other statin trials and cerivastatin appears to be safe and well tolerated in patients with type 2 diabetes. The potential for cerivastatin to be effective in reducing coronary artery disease morbidity and mortality in patients with type 2 diabetes is currently being investigated in clinical trials.

Effect of niacin on lipid and lipoprotein levels and glycemic control in patients with diabetes and peripheral arterial disease: the ADMIT study: A randomized trial. Arterial Disease Multiple Intervention Trial.

M B Elam, D B Hunninghake, K B Davis, *et al. JAMA* 2000; **284**:1263–70.

B**ACKGROUND. Niacin increases low levels of HDL cholesterol, which often accompany diabetes; however, niacin use in patients with diabetes is not currently recommended because of concerns about adverse effects on glycaemic control, although this concern is based on limited data. This prospective, randomized placebo-controlled trial aimed to determine the efficacy and safety of lipid-modifying dosages**

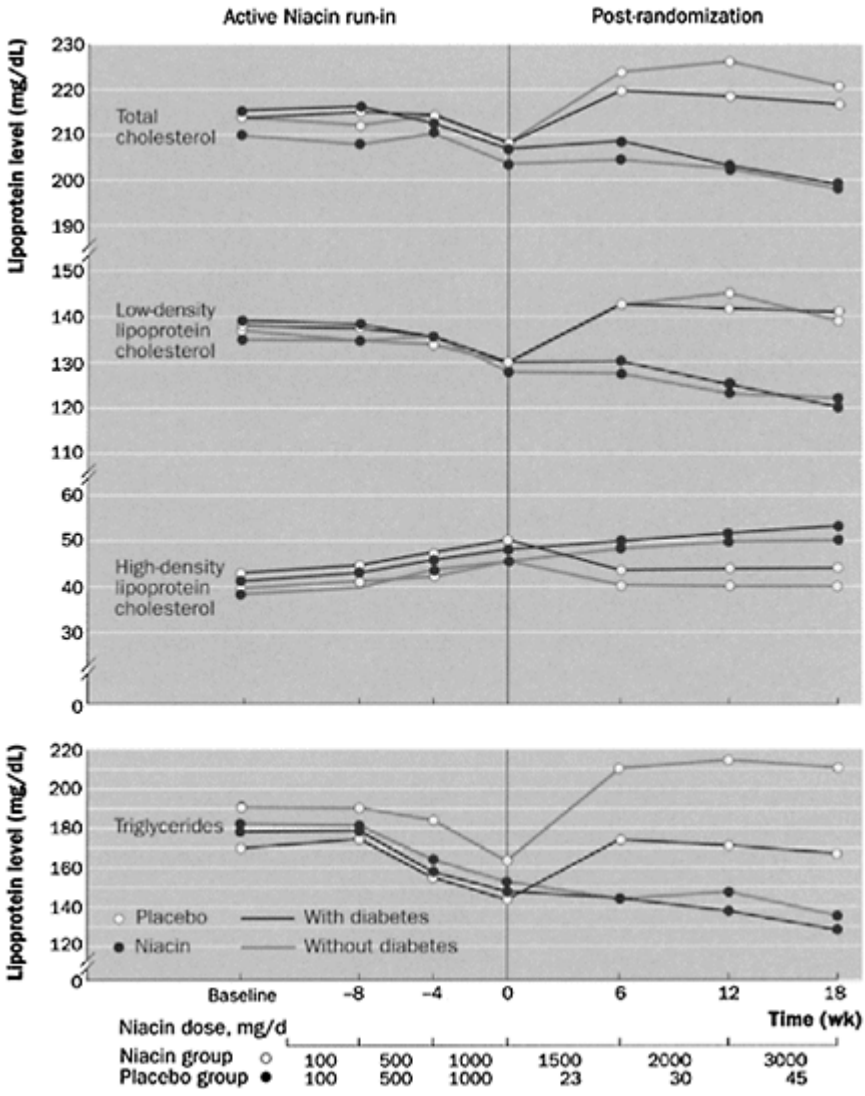

Fig. 11.17 Data are expressed as mean plasma lipoprotein levels in mg/dl. During the 12-week active niacin run-in, all participants received niacin (maximum dose, 1000 mg/day). At week 0, participants were randomly assigned in double-blind fashion to receive niacin (closed circles, maximum dose, 3000 mg/day) or its placebo (open circles). Niacin treatment resulted in significant and equal reductions in plasma TGs, cholesterol, LDL cholesterol and equivalent increase in HDL cholesterol in participants with and without diabetes (P<0.001). To convert total LDL and HDL cholesterol from mg/dl to mmol/l, multiply by 0.0259. To convert TGs from mg/dl to mmol/l, multiply

by 0.0113. Source: Elam *et al.* (2000).

of niacin in patients with diabetes. Patients with diagnosed peripheral arterial disease (*n*=468), including 125 with diabetes, were randomized to receive niacin (crystalline nicotinic acid), 3000 mg/day or the maximum tolerated dose, or placebo for up to 60 weeks.

I**NTERPRETATION.** The results suggest that lipid-modifying dosages of niacin can be safely used in patients with diabetes and that niacin may be considered as an alternative to statins or fibrates for patients with diabetes in whom these agents are not tolerated or not effective. Niacin use significantly increased HDL cholesterol, and decreased TGs and LDL cholesterol in all participants. Glucose levels were modestly increased by niacin in all participants. Levels of HbA(1c) were unchanged in patients with diabetes treated with

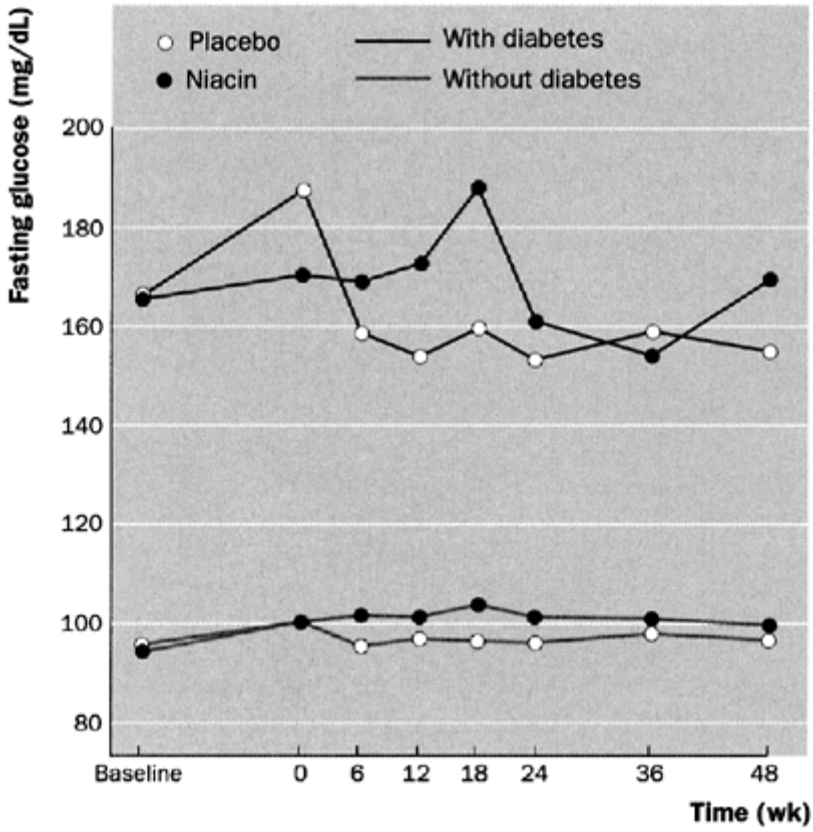

Fig. 11.18 Effect of niacin treatment on fasting glucose in participants with and without diabetes. All participants received niacin in doses up to 1000

:

mg/day during the initial 12-week active niacin run-in (baseline to week 0). At the end of the active niacin run-in period (week 0), participants were randomly assigned to receive continued niacin (titrated up to 3000 mg/day by week 18) or its placebo. Data are mg/dl of fasting glucose at baseline and subsequent follow-up visits by niacin treatment group. Niacin treatment was associated with a transient but statistically significant increase in glucose in participants with and without diabetes during follow-up (P=0.05). The effect of niacin onplasma glucose was more pronounced in participants with versus without diabetes (P=0.04). To convert fasting glucose from mg/dl to mmol/l, multiply by 0.0555.

Source: Elam *et al.* (2000).

Table 11.8 Effect of niacin on average plasma glucose, hemoglobin A_{1c}, and uric acid in participants with or without diabetes*

	With diabetes					**Without diabetes**		
	No. of participants	**Baseline**	**Follow-up†**	**Change‡**	***P* Value**	**No. of participants**	**Baseline**	**Fo up**
Glucose, mg/dL §								
Placebo	59	165(54)	157(47)	−8.7	0.04	161	96(20)	
Niacin	61	165(63)	173(61)	8.1		169	95(13)	1
Hemoglobin A_{1c} %								
Placebo	60	7.7(1.4)	7.4(1.2)	−0.3	0.05	164	5.3(0.6)	5
Niacin	61	7.8(1.5)	7.8(1.3)	0		167	5.3(0.6)	5
Uric acid, µmol/L								
Placebo	56	333(89)	333(71)	0	<0.001	153	380(83)	3
Niacin	58	339(71)	386(95)	47		162	375(87)	4

* Values are expressed as mean (SD).

† Postrandomization values for glucose and uric acid were taken at weeks 6, 12, 18, 24, 36, and 48;

‡ Difference between the baseline value and the mean follow-up value.

§ To convert to mmol/L, multiply by 0.0555.

Source: Elam *et al.* (2000).

niacin but decreased significantly in patients with diabetes treated with placebo. There were no significant differences in niacin discontinuation, niacin dosage or hypoglycaemic therapy in patients with diabetes assigned to niacin versus placebo.

Comment

Nicotinic acid, in the doses usually required to treat significant dyslipidaemia (2–4 g/day) is thought to increase insulin resistance and even precipitate type 2 diabetes in susceptible patients. From this study, however, it seems to suggest that doses of 3 g/day does not significantly alter overall glycaemic control in patients with type 2 diabetes as estimated by HbA1c levels, although patients receiving placebo had significantly reduced HbA1c. Nevertheless, statins remain the first line drugs of choice in patients with type 2 diabetes with fibrates reserved as a second line adjunctive therapy in certain circumstances.

Effects of fluvastatin on prothrombotic and fibrinolytic factors in type 2 diabetes mellitus.

K C Tan, E D Janus, K S Lam. *Am J Cardiol* 1999; **84**:934–7 (A7).

B**ACKGROUND. The effects of fluvastatin therapy on parameters of coagulation and fibrinolysis were assessed in patients with diabetic dyslipidaemia in a randomized, placebo-controlled study.**

I**NTERPRETATION.** Fluvastatin therapy resulted in small reductions in factor VII coagulant activity, von Willebrand factor, plasminogen activator inhibitor 1 and tissue plasminogen activator antigens. The effects of fluvastatin on haemostatic factors were, however, much less marked than its effects on plasma lipids.

Comment

There is currently much interest in the so-called pleiotropic effects of statins; i.e. their possible effects on aspects other than LDL cholesterol reduction. This paper suggests that fluvastatin has only very modest effects on thrombotic and haemostatic factors compared with changes to plasma lipids.

Lack of effect of simvastatin on insulin sensitivity in Type 2 diabetic patients with hypercholesterolaemia: results from a double-blind, randomized, placebo-controlled crossover study.

C M Hwu, C F Kwok, H S Chen, *et al. Diabet Med* 1999; **16**:749–54.

B**ACKGROUND. This study aimed to evaluate the effects of simvastatin on serum lipids and insulin sensitivity in type 2 diabetic**

patients with hypercholesterolaemia.

Table 11.9 Fasting lipid profiles and glycemic control

	Placebo Group (*n*=20)			**Fluvastatin Group (*n*=37)**		
	Week 0	**Week 6**	**Week 12**	**Week 0**	**Week 6 (20 mg)**	**Week 12 (40 mg)**
Total cholesterol						
mg/dl	225±24	239±34	252±21	260±31	213‡±31†	208‡±34†
mmol/L	6.61±0.61	6.20±0.89	6.52±0.55	6.74±0.80	5.53‡±0.81†	5.40‡±0.88†
Triglyceride¶	132	158	153	148	142	134
mg/dl	(107–156)	(114–202)	(119–187)	(129 −167)	(116–166)	(114–153)
mmol/L	1.49	1.79	1.73	1.67	1.60	1.51
	(1.21–1.76)	(1.29–2.28)	(1.34–2.11)	(1.46–1.89)	(1.31–1.88)	(1.29–1.73)
LDL						
mg/dl	1.86±20	166±29	178±22	187±25	140‡±25†	135§±26†
mmol/L	4.83±0.53	4.30±0.76	4.61±0.57	4.84±0.65	3.62‡±0.64†	3.51§±0.68†
HDL						
mg/dl	42±8	41±8	43±9	44±12	46±12	46±14*
mmol/L	1.10±0.20	1.07±0.21	1.11±0.24	1.13±0.30	1.18±0.32	1.19±0.35*
Glycated hemoglobin A1c (%)	7.6±1.0	7.7±1.0	7.7±1.2	7.7±1.0	7.6±0.9	7.6±1.0
Fasting glucose (mmol/L)	7.8±2.5	7.6±2.1	8.7±3.0	8.3±2.2	8.4±2.2	8.5±1.9
Creatinine (μmol/L)	85.2±12.6	84.2±10.4	83.7±12.7	81.1±11.2	81.0±12.9	81.1±13.3

Values are expressed as mean±SD.

¶ Expressed as geometric means (95% confidence intervals).
Within group: *p<0.05; †p<0.001 *versus* week 0.
Between group: ‡p<0.01; §p<0.001 *versus* placebo.
Source: Tan *et al.* (1999).

Table 11.10 Hemostatic variables

	Placebo Group (*n*=20)			Fluvastatin Group (*n*=37)		
	Week 0	**Week 6**	**Week 12**	**Week 0**	**Week 6 (20 mg)**	**Week 12 (40 mg)**
Fibrinogen (g/L)‡	3.09	3.17	2.83	2.97	2.82	3.02
	(2.86–3.32)	2.94–3.39)	(2.55–3.10)	(2.76–3.18)	(2.63–3.00)	(2.84–3.21)
Factor VIIc (%)‡	135	134	136	130	128	123*
	(122–148)	(120–149)	(124–147)	(122–139)	(119–136)	(115–131)
von Willebrand factor antigen (U/ml)	1.33±0.28	1.39±0.30	1.29±0.30	1.37±0.22	1.34±0.22	1.28±0.25†
Plasminogen activator inhibitor 1 antigen (U/ml)	36.77±26.60	39.68±28.97	42.33±17.18	52.91±30.22	49.97±27.79	41.48±21.30*
Plasminogen activator inhibitor 1 activity (U/ml)	12.61±9.48	16.44±8.87	17.18±10.08	17.64±7.97	19.61±9.93	18.41±9.75
Tissue plasminogen activator antigen (U/ml)	12.58±3.80	12.78±3.50	12.03±3.85	13.43±2.57	12.39±2.91*	12.51±2.69*
Tissue plasminogen activator activity (U/ml)	0.60±0.20	0.60±0.23	0.62±0.20	0.55±0.18	0.55±0.21	0.61±0.19

‡ Expressed as geometric means (95% confidence intervals).
Within group: *p<0.05; †p<0.01 *versus* week 0.
Values are expressed as mean±SD.
Source: Tan *et al.* (1999).

This was a double-blind, randomized, placebo-controlled cross-over study in which 19 type 2 diabetic patients with hypercholesterolaemia

were randomized to receive either simvastatin or placebo for 3 months.
I**NTERPRETATION.** Simvastatin significantly reduced the serum total and LDL cholesterol levels without altering glycaemic control or insulin sensitivity in type 2 diabetic patients with hypercholesterolaemia.

Comment

In this small study, no effect on glycaemic control was seen with short-term treatment with simvastatin. As stated previously, the literature is full of contradictory reports. Thus, lipid regulating drugs should only be prescribed on the basis of assessment of absolute risk of CHD in an individual patient. Any possible knock-on effect on improving insulin sensitivity should be considered a bonus.

Table 11.11 Effects of simvastatin on lipid profiles in type 2 diabetic patients with hypercholesterolaemia

	Placebo period*	**Simvastatin period***	**Difference†**	***P*-value‡**
Subjects (n)	17	17	16	
TC (mmol/l)	6.35±1.05	4.68±1.10	−1.59±1.27	0.0002
TG (mmol/l)	3.28±1.56	2.55±1.42	−0.71±2.00	0.18
HDL-C (mmol/l)	0.84±0.22	0.93±0.18	0.08±0.17	0.08
LDL-C (mmol/l)	4.06±1.19	2.58±0.86	−1.40±1.18	0.0003
NEFA (mol/l)	0.833±0.135	0.757±0.169	−0.076±0.216	0.18

* There were no period and carryover effects in the study. Data of the same (placebo or simvastatin) period were pooled and the difference tested by a matched pairs Student's *t*-test. The data shown are the means±SD of measurements at the end of each treatment period. † The difference was calculated from the difference between the simvastatin period and the placebo period. † The statistical significance of difference by a matched pairs Student's *t*-test.
Source: Hwu *et al.* (1999).

Table 11.12 Effects of simvastatin on fasting plasma glucose and glycated haemoglobin in type 2 diabetic patients with hypercholesterolaemia

	Placebo period*	**Simvastatin period***	**Difference†**	***P*-value‡**
Subjects (n)	17	17	16	
Fasting PG (mmol/l)	7.49±1.45	8.26±2.09	0.85±2.18	0.14

HbAtc (%)	7.9±1.5	8.2±1.8	0.3±0.9	0.24

Data were expressed as means±SD. For explanation of symbols please refer to the footnotes of Table 11.11.
Source: Hwu *et al.* (1999).

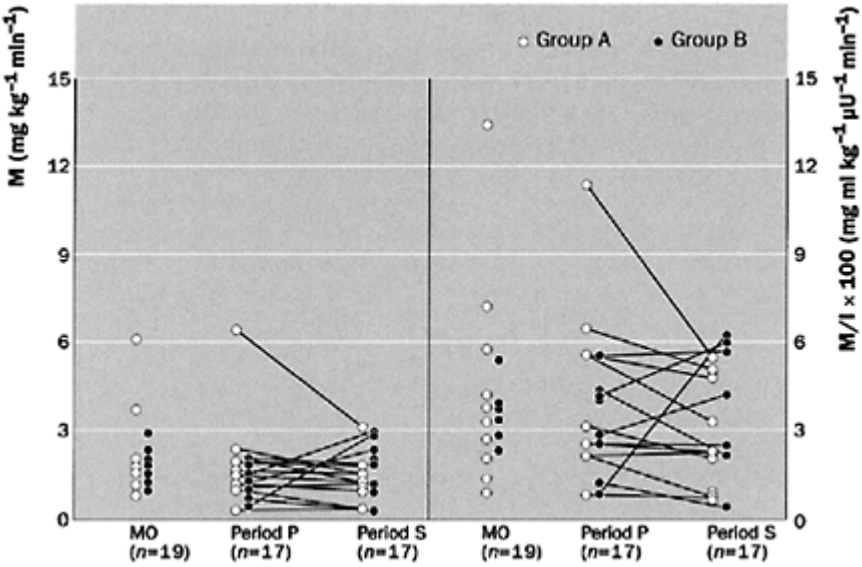

Fig. 11.19 *In vivo* insulin sensitivity as measured by modified euglycaemic insulin clamp during the administration of placebo or simvastatin in type 2 diabetic patients with hypercholesterolaemia. The M-values (mean±SD) in month 0 (M0) were 2.05±1.64 in group A and 1.83±0.57 mg/kg per min in group B. The M/I ratios in M0 were 4.46±3.68 in group A and 3.45±0.94×100 mg/ml per kg per μU per min in group B. The M-values (1.65±1.36 *versus* 1.42±0.94) and M/I ratios (3.84±2.59 *versus* 3.21±2.03) were similar in the placebo and the simvastatin periods, and no drug effect on insulin sensitivity could be identified. Source: Hwu *et al.* (1999).

Bezafibrate and simvastatin combination therapy for diabetic dyslipidaemia: efficacy and safety.
D Gavish, E Leibovitz, I Shapira, *et al. J Intern Med* 2000; **247**:563–9.

B**ACKGROUND. This open study in 148 patients was designed to determine the efficacy and safety of a statin-fibrate combination in**

patients with type 2 DM. Each patient received either bezafibrate slow release 400 mg daily or simvastatin 20 mg daily for 6 months and then a combination of the two for 1 year.

I**NTERPRETATION.** The statin and fibrate combination was more efficacious than either of the single medications for the treatment of diabetic dyslipidaemia, as shown by improvements in the lipoprotein profile and reductions in lipoprotein(a), fibrinogen and the cardiovascular event rate. The cardiovascular event rate was significantly reduced, from 9.5% during the first 6 months of the study to less than 2% during the last year while patients were on the combination treatment.

Comment

This is an interesting study showing the synergistic effect of a statin/fibrate combination on diabetic dyslipidaemia. This was designed as an efficacy and safety study that showed encouraging reduction in the cardiovascular event rate. Definitive data on statin-fibrate combinations will, however, be available when the LDS trial is completed in 2005—when over 5500 men and women with type 2 diabetes will

Table 11.13 Number and percentage of patients suffering from side-effects according to treatment group

Group	Mild CPK elevation	>×2 CPK elevation	Liver function abnormality	Clinical myopathy
All	16 (11%)	5 (3.4%)	4 (3%)	3 (2%)
Statins only	3 (3%)	1 (1%)	1 (1%)	1 (1%)
Fibrates only	1 (2%)	1 (2%)	1 (2%)	–
Combination	12 (8%)	3 (2%)	2 (1.4%)	2 (1.4%)

Source: Gavish *et al.* (2000).

Table 11.14 All events (cardiovascular and non-cardiovascular) occurring during the 18 months of follow-up

	Statin (*n*=100)	Fibrate (*n*=48)	Combination (*n*=148)	All
Cardiovascular events				
All	6 (6%)	6 (12%)	2 (1.4%)	14 (9.5%)
Death	1 (1%)	1 (2.1%)	0	2 (1.4%)

Acute MI	0	1 (2.1%)	1 (0.7%)	2 (1.4%)
Unstable AP	2 (2%)	1 (2.1%)	1 (07%)	4 (2.8%)
Angiography	2 (2%)	2 (4.2%)	0	4 (2.8%)[a]
Angioplasty	2 (2%)	–	–	2 (1.4%)[a]
CABG	–	–	–	1 (0.7%)[a]
TIA	1 (1%)	1 (2.1%)	–	2 (1.4%)
Non-cardiovascular events				
All	1 (1%)	1 (2.1%)	2 (1.4%)	4 (2.8%)
Pneumonia	1 (1%)	–	1 (0.7%)	2 (1.4%)
Urosepsis	–	–	1 (0.7%)	1 (0.7%)
Acute cholecystitis	–	1 (2.1%)	–	1 (0.7%)

MI, myocardial infarction; AP, angina pectoris; CABG, coronary artery bypass graft; TIA, transient ischaemic attack.

[a] The four patients who needed angiography during follow-up include two patients who had undergone angioplasty and one patient who underwent CABG.

Source: Gavish *et al.* (2000).

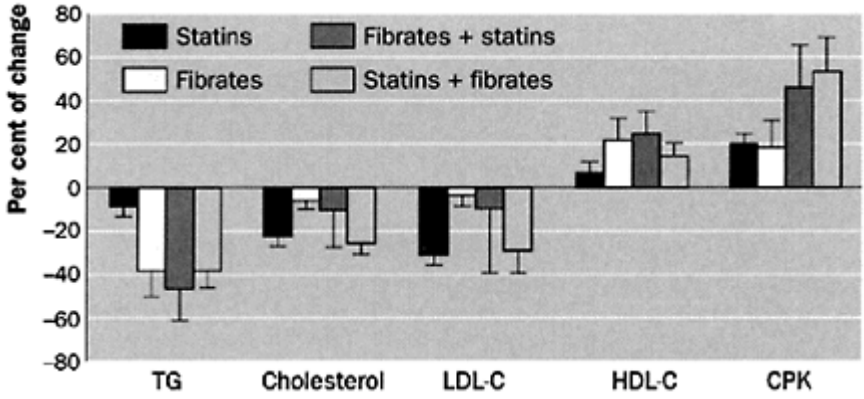

Fig. 11.20 Percentage change from baseline in risk factors: TGs, total cholesterol (Chol), LDL cholesterol (LDL-C), HDL cholesterol (HDL-C) and creatinine phosphokinase (CPK). Source: Gavish *et al.* (2000).

have participated in a 2×2 factorial designed trial for at least 5 years with the primary end-point being a composite of fatal and non-fatal CHD together with a coronary or peripheral revascularization procedure. The design provides the opportunity to assess whether there are additional benefits (or hazards) from the combination of cerivastatin and fenofibrate over either alone or neither.

Conclusion

A review of the currently licensed statins and fibric acid derivatives show that in general they are well tolerated, and result in significant improvement in the dyslipidaemia of type 2 diabetes. Currently, outcome data are only available from *post hoc* subgroup analyses in trials using simvastatin and pravastatin. The role of statin/ fibrate combination therapy is an appealing treatment strategy from first principles but requires randomized control trials with clinical outcome, primary end-points and a full safety evaluation before they can be generally recommended or incorporated into treatment guidelines.

References

1. Koskinen P, Manttäri M, Manninen V, Huttunen JK, Heinonen OP, Frick MH. Coronary heart disease incidence in NIDDM patients in the Helsinki Heart Study. *Didbetes Care* 1992; **15**:820–5.
2. Scandinavian Simvastatin Survival Study Group. Randomized trial of cholesterol lowering in 4444 patients with coronary heart disease. Scandinavian Simvastatin Survival study. *Lancet* 1994; **344**:1383–9.
3. Pyörälä K, Pedersen TR, Kjeksus J, Faergerman O, Olsson AG, Thorgeirsson G. Cholesterol lowering with simvastatin improves prognosis of diabetic patients with coronary heart disease: a subgroup analyses of the Scandinavian Simvastatin Survival Study (4S). *Diabetes Care* 1997; **20**:614–20.
4. Sacks FM, Pfeffer MA, Moye LA, Rouleau JL, Rutherford JO, Cole TG, Brown L, Warnica JW, Arnold JM, Wun CC, Davis BR, Braunwald E, for the Cholesterol and Recurrent Events Trial Investigators. The effect of pravastatin on coronary events after myocardial infarction in patients with average cholesterol levels: Cholesterol and Recurrent Events Trials Investigators. *N Engl J Med* 1996; **335**:1001–9.
5. Hokanson JE, Austin MA. Plasma triglyceride level is a risk factor for cardiovascular disease independent of high-density lipoprotein cholesterol level: a meta-analysis of population-based prospective studies. *J Cardiovasc Risk* 1996; **3**:213–19.
6. Jeppesen J, Hein HO, Suadicani P, Gyntelberg F. Triglyceride concentration and ischaemic heart disease: an eight-year follow-up in the Copenhagen Male Study. *Circulation* 1998; **97**:1029–36.
7. Bucher HC, Griffith LE, Guyatt GH. Systematic review on the risk and benefit of different cholesterol-lowering interventions. *Arterioscler Thromb Vasc Biol* 1999; **19**:187–95. (A review of results of cholesterol-lowering with statins, fibrates, resins, niacin hormones, n-3 fatty acids and diet in 59 trials involving 173 000 patients published between 1966 and 1996.)
8. Assmann G, Betteridge DJ, Gotto Jr AM, Steiner G. Management of hypertriglyceridaemic patients. A treatment classification goal. *Am J Cardiol* 1991; **68**:30–4A.
9. Frick MH, Elo O, Haapa K, Heinonen OP, Heinsalmi P, Helo P, Huttunen JK, Kaitaniemi P, Koskinen P, Manninen V, *et al.* Helsinki Heart Study. Primary prevention trial with gemfibrozil in middle-aged men with dyslipidaemia. Safety of treatment, changes in risk factors and incidence of coronary heart disease. *N Engl J Med* 1987; **317**: 1237–45.

10. Carlson LA, Rosenhamer G. Reduction of mortality in the Stockholm ischaemic disease secondary prevention study by combined treatment with clofibrate and nicotinic acid. *Acta Med Scand* 1988; **223**:405–18.

11. Manninen V, Tenkanen L, Koskinen P, Huttunen JK, Manttari M, Heinonen OP, Frick MH. Joint effect of serum triglyceride and LDL cholesterol and HDL cholesterol concentrations on coronary heart disease risk in the Helsinki Heart Study: implications for treatment. *Circulation* 1992; **85**:37–45.

12. MRC/BHF Heart Protection Study Collaborative Group. Study of cholesterol-lowering therapy and of antioxidant vitamin supplementation in a wide range of patients at increased risk of coronary heart disease death: early safety and efficacy experience. *Eur Heart J* 1999; **20**:725–41.

13. Armitage J, Collins R. Need for large scale randomized evidence about lowering LDL cholesterol in people with diabetes mellitus: MRC/BHF heart protection study and other major trials. *Heart* 2000; **84**(4): 357–60.

12
Hypertension in patients with diabetes

Introduction

Hypertension and diabetes mellitus (DM) are independent risk factors for the development of coronary heart disease (CHD) and frequently coexist in the same patient. In fact hypertension is twice as prevalent in patients with diabetes compared with the general population. Several factors contribute to the high prevalence of hypertension. They include sodium retention and vascular hyper-reactivity. These abnormal vascular responses are best explained by early changes in vascular structure and endothelial dysfunction, mediated by the metabolic disorders in DM. Blood pressure and plasma glucose levels are positively correlated |**1**|. Tight glucose control decreases blood pressure in diabetes, despite increases in plasma volume and exchangeable sodium |**2**| and blood pressure increases with worsening of metabolic control. Controversy surrounds the issue as to whether insulin or glucose levels or indeed both, participate in the microvascular and macrovascular complications of diabetes. Glucose has direct cellular effects on blood vessels. Elevated glucose levels increase endothelin-1 production |**3**| and decrease nitric oxide (NO) formation |**4**|. Glucose has direct toxic effects on endothelial cells, delaying cell replication and accelerating cell death |**4**|. The glucose molecule also links to proteins through a non-enzymatic reaction leading to the formation of highly reactive late addition products called advance glycosylation end-products (AGEs). These products remain irreversibly bound to proteins at multiple sites, particularly to subendothelial basement membrane proteins and their deposition takes place at an accelerated rate in diabetes. The AGEs elicit multiple effects that may promote vascular changes and hypertension. They induce growth-promoting cytokines such as interleukin-1 |**5**| and transendothelial migration of monocytes to subendothelial spaces. AGEs can also interfere with NO action, and this partly accounts for the defective NO-dependent vasodilatation seen in diabetes |**6**|.

In numerous studies, blood pressure has correlated with insulin levels and insulin resistance in obese patients with type 2 diabetes, obese patients without type 2 diabetes and patients with essential hypertension |**7,8**|. In experimental models of hypertension, agents that reverse insulin resistance such as metformin can reduce blood pressure |**9**|. The combined disorders of glucose and insulin, therefore, have major independent but perhaps additive effects that contribute to abnormal blood pressure regulation, particularly in patients with type 2 diabetes. Although circulating plasma renin activity (PRA) levels are low in patients with type 2 diabetes, it is thought that the tissue rather than the circulating renin-angiotensin system may have the more important role in vascular autoregulation, and thus be of central importance in the aetiology of hypertension in type 2 diabetes |**10**|.

Hypertension, when present, almost always exacerbates the prognosis of

macrovascular and microvascular complications in diabetes. According to the results of the UK Prospective Diabetes Study (UKPDS), tight blood pressure control offers better protection against cardiovascular end-points and death than does tight glycaemic control |**11**|. These findings are consistent with those of the other large trials in both the management of dyslipidaemia and hypertension that have included diabetic patients. Until relatively recently, DM has been generally considered to be an abnormality of carbohydrate metabolism with an overfocused approach on glycaemic control in the management of the patient. DM is best considered part of a chronic vascular risk syndrome leading to the suggestion that it should be redefined as a condition of premature vascular disease. While the goal of achieving optimum glycaemic control remains a central part of the management strategy in patients with diabetes, recent outcome trials in both hypertension and dyslipidaemia have resulted in a significant shift in therapeutic emphasis in current clinical practice. Overall risk factor management is the name of the game, with particular emphasis on the control of blood pressure, dyslipidaemia, diet, obesity, life-style and quitting smoking. Recent trends in the management of blood pressure, which have included significant cohorts of patients with both hypertension and DM, will now be reviewed.

UK Prospective Diabetes Study

The UKPDS was a clinical outcome trial designed to assess whether intensive treatment with either oral hypoglycaemic agents or insulin reduced the risk of micro-vascular and macrovascular disease relative to less intensive conventional measures such as diet. It was also a comparison as to whether there were benefits or indeed hazards from any of the treatments studied.

In 1987 a subgroup was identified for the Hypertension in Diabetes Study and this was subsequently incorporated into the main UKPDS study via a factorial design to compare relatively tight blood pressure control compared with ‘less tight’ blood pressure control; 1148 patients were randomized for this study with subsequent subdivision in the tight control group to a treatment arm based either on an angiotensin-converting enzyme (ACE) inhibitor (captopril) or a beta-blocker (atenolol).

UKPDS 33

Intensive blood-glucose control with sulphonylureas or insulin compared with conventional treatment and risk of complications in patients with type II diabetes: UKPDS 33.
United Kingdom Prospective Diabetes Study Group. *Lancet* 1998; **352**: 837–53.

B**ACKGROUND. Improved blood glucose control slows the progression of diabetic microvascular disease. However, the effect on macrovascular complications is unknown. There is concern that sulphonylureas may increase cardiovascular mortality in patients with type 2 diabetes and that high insulin concentrations may enhance atheroma formation. The effects of intensive blood glucose control were compared with either sulphonylurea or insulin and conventional treatment on the risk of microvascular and macrovascular complications in patients with type 2 diabetes in a randomized controlled trial.**

I**NTERPRETATION.** Controlling intensive blood glucose by either sulphonylureas or insulin substantially lowers the risk of microvascular complications (but not macrovascular disease) in patients with type 2 diabetes. There were no adverse effects on cardiovascular outcomes by any of the individual drugs. All intensive treatment increased the risk of hypoglycaemia.

Comment

Intensive treatment with sulphonylureas or insulin resulted in a mean HbA1c of 7% over 10 years compared with 7.9% for the diet group. However, there was no difference between the different intensive treatment subgroups. Improved glycaemic control resulted in 12% reduction for any diabetes related end-point mainly attributed to a 25% reduction in microvascular end-points ($P<0.005$). A modest reduction of 16% in fatal and non-fatal myocardial infarction (non-significant) was observed. Moreover, sudden death was significantly reduced in the intensive treatment subgroup ($P<0.05$). No differences between treatment modalities, chlorpropamide, glibenclamide and insulin were observed. As a consequence, these results have lessened previous concerns that sulphonylurea drugs may in fact increase mortality and/or morbidity after a myocardial infarction.

UKPDS 34

Effect of intensive blood glucose control with metformin on complications in overweight patients with type 2 diabetes: UKPDS 34. United Kingdom Prospective Diabetes Study Group. *Lancet* 1998; **352**: 854–65.

B**ACKGROUND. In patients with type 2 diabetes, controlling intensive blood glucose with insulin or sulphonylurea therapy slows the progression of microvascular disease and may also reduce the risk of heart attacks. This study researched whether intensive glucose control**

with metformin has specific advantages or disadvantages.

I**NTERPRETATION.** Because intensive glucose control with metformin appears to reduce the risk of diabetes-related end-points in overweight diabetic patients, and is associated with less weight gain and fewer hypoglycaemic attacks than control with insulin and sulphonylureas, it could be the first-line pharmacological therapy of choice for these patients.

Comment

Metformin resulted in a significant reduction in risk for all-cause mortality, diabetes related death and all diabetes-related end-points. The metformin arm has a significantly lower $HbA1_c$ (7.4%) compared with 8% in the diet group. For all macrovascular events and myocardial infarction, a significant risk reduction of 30% was observed with metformin treatment compared with conventional dietary treatment.

UKPDS 38

Tight blood pressure control and risk of macrovascular and microvascular complications in type 2 diabetes: UKPDS 38.
United Kingdom Prospective Diabetes Study Group. *BMJ* 1998; **317**: 703–13.

B**ACKGROUND. The goal of this research was to establish whether tight control of blood pressure (BP) prevents macrovascular and microvascular complications in patients with type 2 diabetes.**

I**NTERPRETATION.** Tight BP control in patients with hypertension and type 2 diabetes attains a clinically important decrease in the risk of deaths related to diabetes, complications related to diabetes, progression of diabetic retinopathy and deterioration in visual acuity.

Table 12.1 Tight blood pressure (BP) control in type 2 diabetes: events per 1000 patient-years

	BP control		**Relative risk**	
	Tight	**Less**	**Reduction**	
Any diabetes-related endpoint	50.9	67.4	−24%	$P<0.005$
All-cause mortality	22.4	27.2	−18%	
Myocardial infarction	18.6	23.5	−21%	

Stroke	6.5	11.6	−44%	$P<0.02$
Microvascular disease	12.0	19.2	−37%	$P<0.01$

Source: UKPDS 38 (1998).

Comment

In 1987 the Hypertension in Diabetes Study was incorporated into the main UKPDS study. The objective was to compare 'tight' blood pressure control (target <150/85 mmHg) with 'less tight' control of blood pressure (<180/105 mmHg); 1148 patients were randomized with the 'tight control' arm further randomly allocated to either a beta-blocker (atenolol) or an ACE inhibitor (captopril) based regimen. The mean blood pressure achieved in the 'tight' control arm was 144/82 which was significantly lower than in the less tight' control arm—154/87, $P<0.001$. This difference resulted in significant reductions in both microvascular and macrovascular events (see Table 12.1). The risk in microvascular complications was mainly attributable to a reduction in risk and progression of retinopathy.

UKPDS 39

Efficacy of atenolol and captopril in reducing risk of macrovascular and microvascular complications in type 2 diabetes: UKPDS 39.

United Kingdom Prospective Diabetes Study Group. *BMJ* 1998; **317**: 713–20.

B**ACKGROUND. The goal of this research was to establish whether tight control of BP with either a beta-blocker or an angiotensin-converting enzyme inhibitor had specific advantages or disadvantages in preventing the macrovascular and microvascular complications in patients with type 2 diabetes.**

I**NTERPRETATION.** Using captopril or atenolol to lower BP was similarly effective in diminishing the incidence of diabetic complications. This research provided no evidence that either drug had any specific beneficial or deleterious effect. This suggests that BP reduction in itself may be more important than the treatment used.

Table 12.2 Captopril versus atenolol diabetes-related endpoints

Clinical endpoint	Patients with aggregate endpoints		*P*	Relative risk for capropril
	Captopril (*n*=400)	Atenolol (*n*=358)		
Any endpoint	141	118	0.43	1.10
Deaths	48	34	0.28	1.27
Acute myocardial infarction	61	46	0.35	1.20
Stroke	21	17	0.74	1.12
Peripheral vascular disease	5	3	0.59	1.48
Microvascular disease	40	28	0.30	1.29

Source: UKPDS 39 (1998).

Comment

Captopril and atenolol were equally effective in reducing the incidence of diabetic complications in the 'tight' control of blood pressure arm of the Hypertension in Diabetes Study of the UKPDS. However, this study did not have the statistical power to address the controversial issue as to whether ACE inhibition or beta blockade have additional benefits or disbenefits.

For the clinician, an important point to take away from the UKPDS 38 and the UKPDS 39 reports is that single drug therapy resulted in 'tight' control of blood pressure in <50% of patients and that three or more agents were required in nearly 30% of patients. Moreover, after 9 years of treatment, only 56% of the 'tight' control group had attained a target blood pressure of <150/85 mmHg. The practical implications of these observations are self-evident.

Captopril prevention project (CAPPP)

Effect of angiotensin-converting enzyme inhibition compared with conventional therapy on cardiovascular morbidity and mortality in hypertension: the Captopril Prevention Project (CAPPP) randomized

trial.
L Hansson, L H Lindholm, L Niskanen, *et al.* for the Captopril Prevention Project (CAPPP) Study Group. *Lancet* 1999; **353**:611–16.

B**ACKGROUND. Angiotensin-converting enzyme (ACE) inhibitors have been used for more than a decade to treat high BP, although there has been little data from randomized intervention trials demonstrating that such treatment affects cardiovascular morbidity and mortality. The Captopril Prevention Project is a**

randomized intervention trial comparing the effects of ACE inhibition and conventional therapy on cardiovascular morbidity and mortality in patients with hypertension.

I**NTERPRETATION.** The efficacy of captopril and conventional treatment did not differ in preventing cardiovascular morbidity and mortality. The difference in stroke risk is probably due to the lower levels of BP obtained initially in previously treated patients by conventional therapy.

Comment

Two observations were made from the subgroup analysis of patients with hypertension and DM:

- The treatment regimen utilizing thiazides or beta-blockers was more likely to result in new cases of diabetes with 43 cases being identified during 30 000 patient-years of treatment. This is conventionally interpreted as being secondary to the effect of these agents on intermediary metabolism. Another interpretation is that ACE inhibition may protect against the development of type 2 diabetes [see Heart Outcomes Prevention Evaluation (HOPE) Trial and commentary].
- As only 572 patients with DM were diagnosed prior to entry into the study it is, therefore, completely underpowered to draw any conclusions as to whether any additional benefits accrue from captopril therapy compared with conventional therapy. Moreover, about half of the patients in the captopril arm were administered captopril once daily—a regimen unlikely to provide adequate 24-h blood pressure control.

SYST-EUR: Diabetic Cohort

Effects of calcium channel blockade in older patients with diabetes and systolic hypertension.

J Tuomilehto, D Rastenyte, W H Berkenhäger, *et al.* for the Systolic Hypertension in Europe Trial Investigators. *N Engl J Med* 1999; **340**: 677–84.

B**ACKGROUND. It has been recently suggested that calcium channel blockers may be harmful in patients with diabetes and hypertension. These authors previously reported that antihypertensive treatment with the calcium channel blocker nitrendipine lowered the risk of cardiovascular events. This *post hoc* analysis compared the outcome of treatment with nitrendipine in diabetic and non-diabetic patients.**

I**NTERPRETATION.** Nitrendipine-based antihypertensive therapy is particularly beneficial in older patients with diabetes and isolated systolic hypertension. Thus, the findings do not support the hypothesis that the use of long-acting calcium channel blockers may be harmful in diabetic patients.

Table 12.3 Reduction in mortality and cardiovascular events in diabetics *vs* non-diabetics

	Diabetic patients (*n*=492)	**Non-diabetic patients (*n*=4203)**
All strokes	73%	38%
All cardiac end-points	63%	21%
All cardiovascular end-points	69%*	26%
Total mortality	55%	6%
Cardiovascular mortality	76%*	13%

**P*<0.05 for diabetic *vs* non-diabetic patients.
Source: Tuomilehto *et al.* (1999).

Comment

This *post hoc* analysis of a calcium channel blocker nitrendipine in both diabetic and non-diabetic patients revealed significantly greater benefits in the diabetic compared to non-diabetic patients for all cardiovascular events and for cardiovascular mortality ($P<0.05$) (see Table 12.3). This was after adjusting for confounding factors. The diabetic subgroup in this study comprised 492 patients and systolic and diastolic blood pressures were reduced by 8.6/3.9 mmHg in the treatment group. While the treatment group received nitrendipine (a dihydropyridine calcium channel blocker) more than half the patients ultimately required multiple drug combination to produce this reduction, reinforcing the observations made in UKPDS 38. This study also emphasizes the benefits of risk reduction in high-risk patients (older patients with diabetes and isolated systolic hypertension).

Hypertension Optimal Treatment (HOT) Study: diabetic cohort

Effects of intensive blood pressure lowering and low-dose aspirin in patients with hypertension: principal results of the Hypertension Optimal Treatment (HOT) randomized trial.
L Hansson, A Zanchetti, S G Carruthers, *et al*. for the HOT Study Group. *Lancet* 1998; **351**:1755–62.

B**ACKGROUND. Despite treatment, there is often a higher incidence of cardiovascular complications in patients with hypertension than in normotensive individuals. Inadequate reduction of their BP is a likely cause, but the optimum target BP is not known. The impact of acetylsalicylic acid (aspirin) has never been investigated in patients with hypertension. This study aimed to assess the optimum target diastolic BP and the potential benefit of a low dose of acetylsalicylic acid in the treatment of hypertension.**

I**NTERPRETATION.** Intensive lowering of BP in patients with hypertension was associated with a low rate of cardiovascular events. The HOT study shows the benefits of lowering the diastolic blood pressure down to 82.6 mmHg. Acetylsalicylic acid significantly reduced major cardiovascular events, with the greatest benefit seen in myocardial infarction. There was no effect on the incidence of stroke or fatal bleeds, but non-fatal major bleeds were twice as common.

Comment

There were 18790 patients in the HOT Study, with treatment based on the dihydropyridine calcium channel blocker felodipine. A cohort of 1501 patients with DM were identified and in the group randomized to a target diastolic of <80 mmHg, the risk of major cardiovascular events was halved in comparison with that of the group randomized to <90 mmHg (see Fig. 12.1). The cardiovascular event rates in both groups was similarly reduced, but not always consistently reaching conventional levels of statistical significance. In relation to the clinical point made earlier with respect to the UKPDS 38 and the SYST-EUR trials, only 26% of patients targeted to achieve <80 mmHg diastolic blood pressure achieved this with single drug treatment. Once again, multiple drug regimens were required.

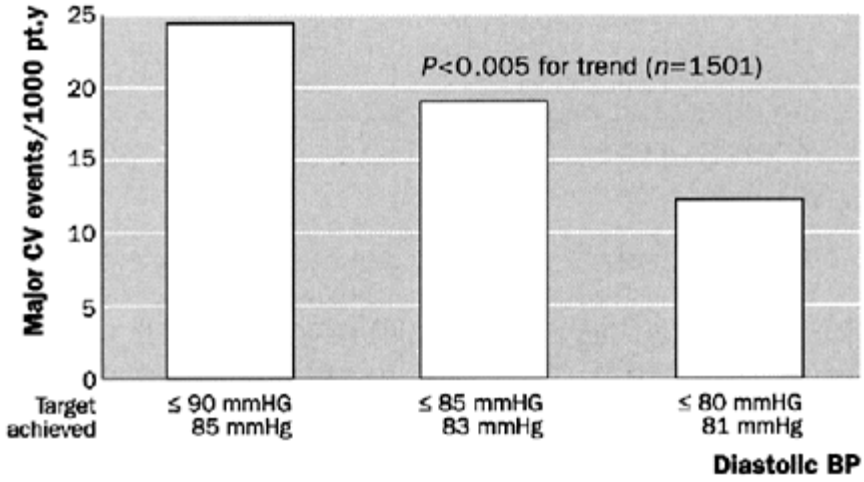

Fig. 12.1 The HOT Study: cardiovascular (CV) risk reduction in diabetics. Major cardiovascular event rates according to the target blood pressure (BP) category in the HOT Study; pt.y=patient-year. Source: Hansson *et al.* (1998).

Heart Outcomes Prevention Evaluation Study

Effects of an angiotensin-converting enzyme inhibitor, ramipril, on cardiovascular events in high-risk patients.
HOPE Study Investigators. *N Engl J Med* 2000; **342**:145–53.

B**ACKGROUND. ACE inhibitors improve the outcome among patients with left ventricular dysfunction, whether or not they have heart failure. HOPE assessed the role of an ACE inhibitor, ramipril, in patients who were at high risk for cardiovascular events but who did not have left ventricular dysfunction or heart failure.**

I**NTERPRETATION.** Ramipril significantly reduced the rates of death, myocardial infarction, and stroke in a broad range of high-risk patients who are not known to have a low ejection fraction or heart failure.

Effects of ramipril on cardiovascular and microvascular outcomes in people with diabetes mellitus: results of the HOPE study and MICRO-HOPE substudy.
HOPE Study Investigators. *Lancet* 2000; **355**:253–9.

B**ACKGROUND. Diabetes mellitus is a strong risk factor for cardiovascular and renal disease. HOPE investigated whether the ACE inhibitor ramipril can lower these risks in patients with diabetes.**

I**NTERPRETATION.** Ramipril was beneficial for cardiovascular events and overt nephropathy in people with diabetes. The cardiovascular benefit appeared greater than that attributable to the decrease in blood pressure.

Comment

This landmark trial was initiated in the early 1990s; an important objective being to clarify the controversial issue as to whether the antioxidant vitamin E, would benefit in a reduction of cardiovascular events in high-risk patients with vascular disease. A 2×2 factorial study design was employed evaluating both vitamin E and the ACE inhibitor ramipril in these patients. An a priori substudy included the evaluation of patients with DM. The study was stopped 6 months early (after 4½ years) by the independent data safety and monitoring board, because of both a lack of effect of vitamin E and a consistent benefit of ramipril compared with placebo. The results of the diabetic study for the ramipril-treated subjects was consistent with those of the overall study, showing relative risk of approximately 0.8 for all major end-points. Importantly, the relative risk of overt nephropathy was

Table 12.4 Incidence of the primary outcome and of deaths from any cause

Outcome	**Ramipril group (*n*=4645)**	**Placebo group (*n*=4652)**	**Relative risk (95% CI)***	**z Statistic**	***P* Value†**
Myocardial infarction, stroke, or death	no. (%)				
from cardiovascular causes‡	651 (14.0)	826 (17.8)	0.78 (0.70–0.86)	−4.87	<0.001
Death from cardiovascular causes§	282(6.1)	377(8.1)	0.74(0.64–0.87)	−3.78	<0.001
Myocardial infarction§	459 (9.9)	570 (12.3)	0.80 (0.70–0.90)	−3.63	<0.001

Stroke§	156(3.4)	226(4.9)	0.68(0.56–0.84)	−3.69	<0.001
Death from non-cardiovascular causes	200(4.3)	192(4.1)	1.03(0.85–1.26)	0.33	0.74
Death from any cause	482(10.4)	569(12.2)	0.84(0.75–0.95)	-2.79	0.005

* CI denotes confidence interval.
† *P* values were calculated with use of the log-rank test.
‡ In the substudy, 34 of 244 patients (13.9 per cent) assigned to take a low dose of ramipril (2.5 mg per day) reached the composite endpoint, as compared with 31 of 224 assigned to take 10 mg of ramipril per day (12.7 per cent) and 41 of 244 assigned to placebo (16.8). The inclusion of the data from the low-dose group did not change the overall results (relative risk of the primary outcome, 0.78; 95 per cent confidence interval, 0.70 to 0.86).
§ All patients with this outcome are included.
Source: HOPE Study Investigators (2000).

reduced in the ramipril-treated group (0.76, $P<0.03$). All studies showed no significant increase in most side-effects or adverse events in the treated groups.

The study population of 9297 high-risk patients (≥55 years age) had a history of coronary artery disease, stroke, peripheral vascular disease or DM plus one other risk factor such as hypertension, elevated total cholesterol, low high-density lipoprotein cholesterol, documented microalbuminuria or a history of smoking. Thus, while a large proportion of the patients had DM (n=3500), many of the non-diabetic patients included had features of the ‘metabolic syndrome’ or ‘insulin resistance syndrome’. However, this was not specifically examined in the HOPE

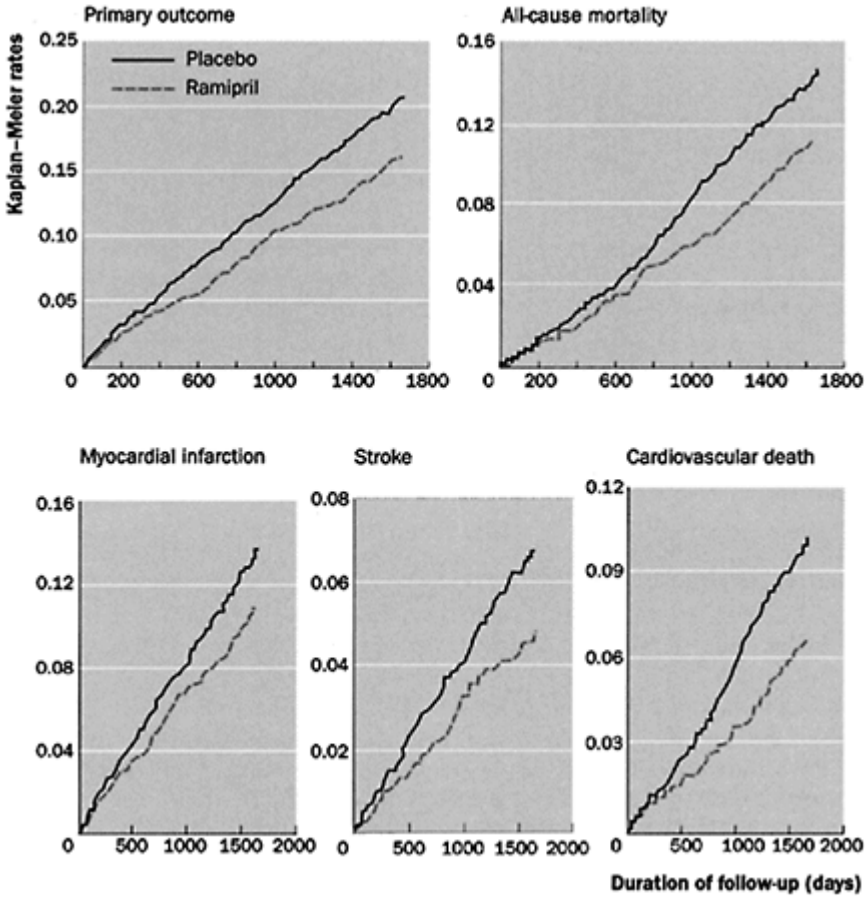

Fig. 12.2 Kaplan-Meier survival curves for participants with diabetes. Source: HOPE Study Investigators (2000).

Study. Of major interest was the reduction in the diagnosis of new diabetes (105 versus 154; relative risk 0.68, $P=0.002$) in the ramipril arm. The study did not specify which type of diabetes, but it is reasonable to assume that in this older population (≥55 years) that it is type 2 diabetes and insulin resistance, which is reduced. The mechanism for this 31% reduction in newly diagnosed diabetes is uncertain, but ACE inhibitors have been shown to increase insulin sensitivity.

There has been particular interest in the role of ACE inhibitor drugs as first-line drugs in the treatment of hypertension in patients with type 2 diabetes. There is increasing evidence that ACE inhibitor drugs have a specific effect on intraglomerular pressures in the renal microcirculation and may have particular reno-protective effects in early nephropathy in patients with both type 1 and type 2 DM. The results of the HOPE Study extend the evidence in favour of ACE inhibitors particularly in the subgroup of 3500 patients with DM. The primary efficacy endpoint was a combination of myocardial

infarction, stroke or cardiovascular death.

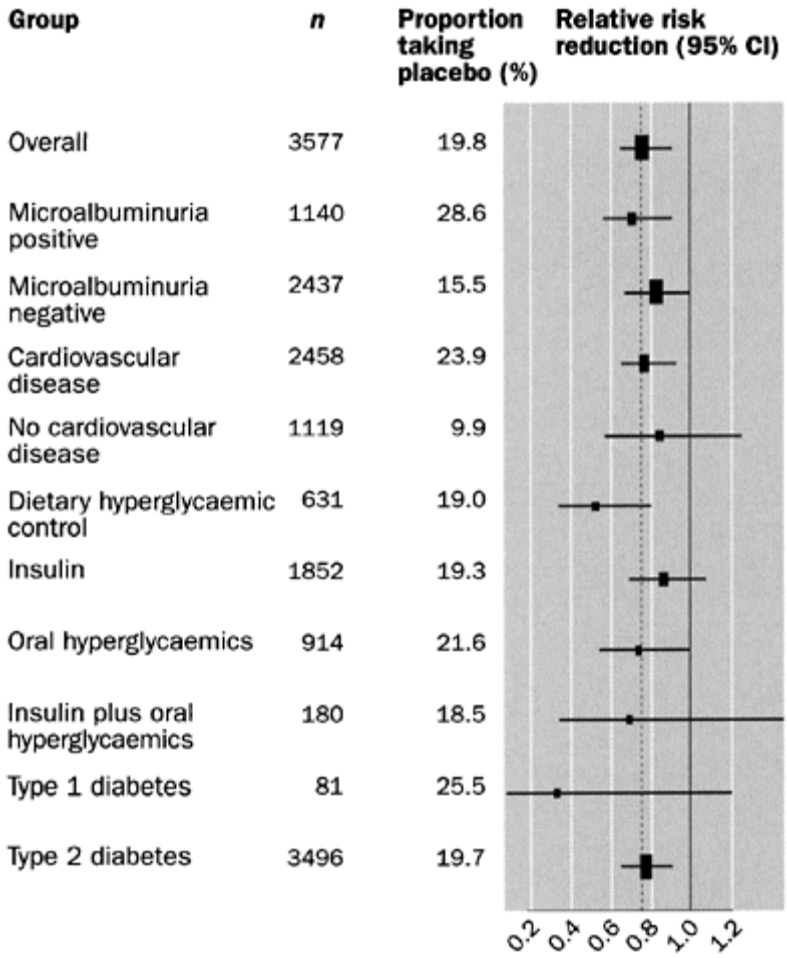

Fig. 12.3 Effect of ramipril on combined primary outcome in subgroups. Size of symbol is proportional to number of participants in subgroup; broken line=overall relative risk.
Source: HOPE Study Investigators (2000).

In the whole group, ACE inhibition was associated with a 22% reduction of risk. Total mortality was also reduced by 22% in the general population and 24% in the diabetic population. The difference in blood pressure in the active group was only 2 mmHg systolic and 1 mmHg diastolic at the end of the study. It is unlikely that these small differences can account for the large differences in cardiovascular end-

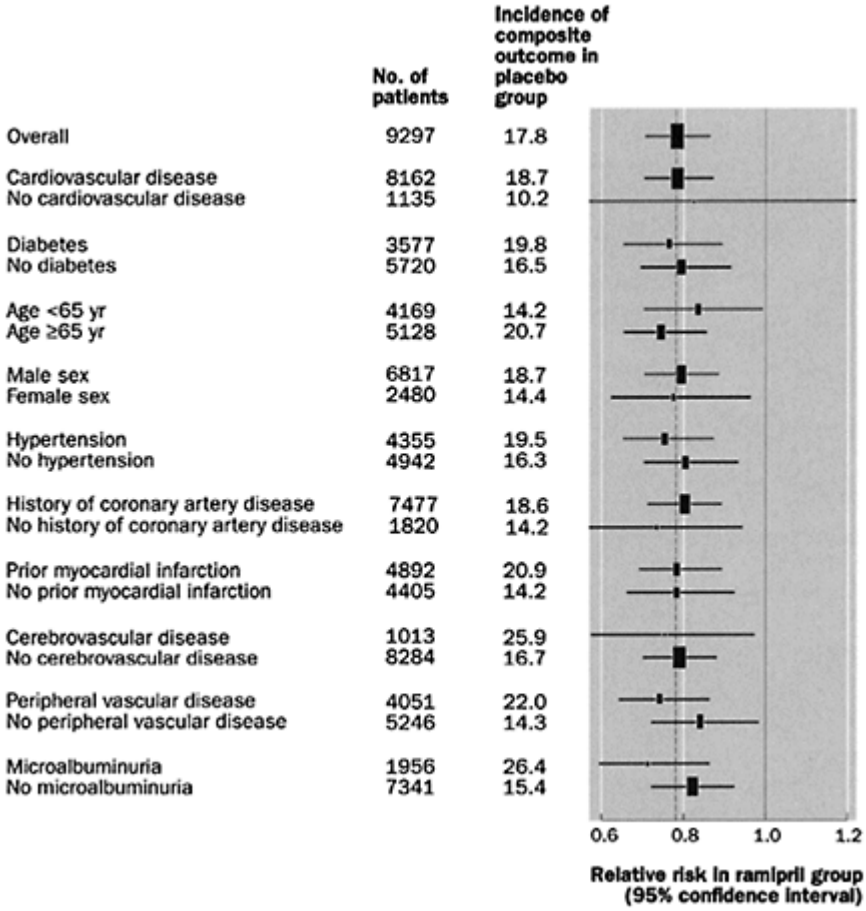

Fig. 12.4 The beneficial effect of treatment with ramipril on the composite outcome of myocardial infarction, stroke or death from cardiovascular causes overall and in various pre-defined subgroups. Cerebrovascular disease was defined as stroke or transient ischaemic attacks. The size of each symbol is proportional to the number of patients in each group. The dashed line indicates overall relative risk. Source: HOPE Study Investigators (2000).

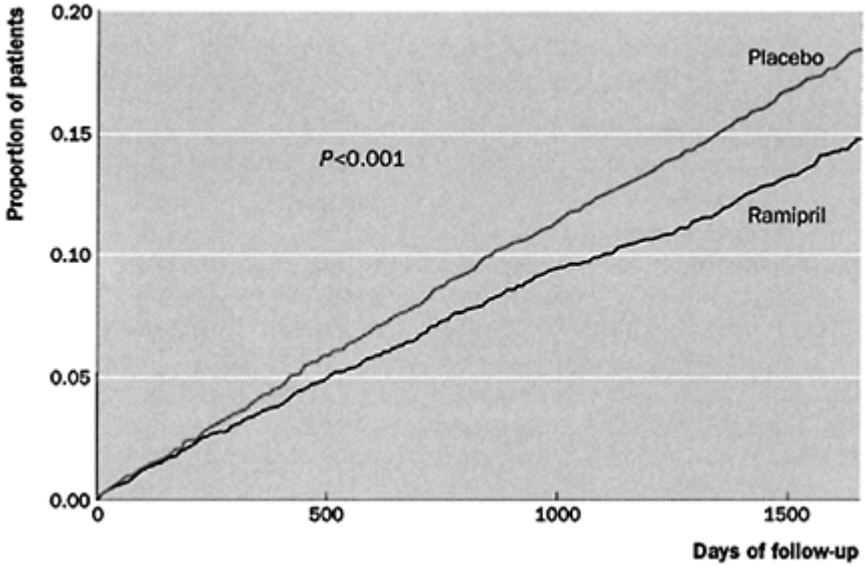

Fig. 12.5 Kaplan-Meier estimates of the composite outcome of myocardial infarction, stroke, or death from cardiovascular causes in the ramipril group and the placebo group. The relative risk of the composite outcome in the ramipril group as compared with the placebo group was 0.78 (95% confidence interval, 0.70–0.86). Source: HOPE Study Investigators (2000).

points and mortality, thus giving credence to the hypothesis that inhibition of the renin-angiotensin system by an ACE inhibitor may have benefits over and above the effects on blood pressure. These benefits seem to apply to patients at high risk for CHD, particularly if they have DM and/or features of the metabolic syndrome. A clear understanding of the basic mechanism that results in these clinical benefits is impossible to obtain from a trial such as HOPE, or to whether the results obtain to a class effect of ACE inhibition. This is a major shortcoming for theoretically minded individuals as it is precisely the definition of these mechanisms that will provide targets for new intervention strategies with additional significant effects on cardiovascular disease in the future. In the meantime, for the clinician, managing cardiovascular risk in the individual patient with DM, there is encouraging evidence for the use of ACE inhibitors as first-line therapy in CHD prevention.

Conclusion

There is a general philosophical shift from identifying individual risk factors and treating when values exceed a certain threshold (e.g. blood pressure, lipids, etc.) to the estimation of absolute risk for the development of either cardiovascular disease or CHD over time, and intervening when the threshold of absolute risk is crossed. In general, the higher the absolute risk for an individual the greater the benefit that accrues from a therapeutic intervention. The British Cardiac Society, the British Hyperlipidaemia Association and the British Hypertension Society have co-operated in preparing national recommendations, which have been endorsed by the British Diabetic Association |**12**|. To calculate absolute risk a computer program or coronary risk chart is provided to enable practitioners to stratify and prioritize patients according to risk and advocate a staged approach to management. Other guidelines include the Recommendations of the Second Joint Task Force of European and other Societies on Coronary Prevention |**13**| and the American Heart Association/American College of Cardiology Statement on Risk Assessment |**14**|. All these current guidelines emphasize the importance of risk assessment in order to identify patients who benefit from treatment. It is the overlap rather than the differences between the various guidelines that is striking.

In general there is a convergence of views based on conclusive evidence from randomized control trials (RCT). However, the various national and international guidelines will still reflect different interpretations, views and even different resources of the health-care system in which clinicians practice.

The guidelines for the Management of Hypertension Report of the 3rd Working Party of the British Hypertension Society, have updated their previous report in 1989 and 1993. They recommend a formal estimation of 10-year CHD risk using the charts of the Joint British Recommendations on prevention of CHD |**12,15,16**|. As a consequence of the RCT evidence that has accrued, the advice for the management of hypertension in people with diabetes is separate and succinct.

In type 1 and type 2 DM, the threshold for starting antihypertensive therapy is ≥140/90 mmHg and the target blood pressure is <140/80 mmHg. In type 1 diabetes with nephropathy, the target blood pressure is 130/80 mmHg; or lower if there is proteinuria >1 g/24 h. In such cases, the target is <125/75 mmHg. An ACE inhibitor titrated to the maximum dose recommended and tolerated is preferred first-line therapy. It is acknowledged that in both forms of diabetes, rigorous control of blood pressure will result in multiple drug regimens. ACE inhibitors, low-dose diuretics, calcium channel blockers, and both alpha- and beta-blockers are all suitable agents.

References

1. Kelleher C, Kingston SM, Barry DG, Cole MM, Ferriss JB, Grealy G, Joyce C, O'Sullivan DJ. Hypertension in diabetic clinic patients and their siblings. *Diabetologia* 1998; **31**: 76–81.
2. Ferriss JB, O'Hare JA, Kelleher CC, Sullivan PA, Cole MM, Ross HF, O'Sullivan DJ.

Diabetic control and the renin-angiotensin system, catecholamines and blood pressure. *Hypertension* 1985; **7**(Suppl 2): 58–63.
3. Yamauchi T, Ohnaka K, Takayanagi E, Umeda F, Nawata H. Enhanced secretion of endothelin-1 by elevated glucose levels from cultured bovine aortic endothelial cells. *FEBS Lett* 1990; **267**:16–18.
4. Kamal K, Du W, Mills I, Sumpio BE. Antiproliferative effect on elevated glucose in human microvascular endothelial cells. *J Cell Biochem* 1998; **71**:449–501.
5. Vlassara H, Brownlee M, Manogue KR, Dinarello CA, Pasagian A. Cachectin/TNF, and IL-I induced by glucose-modified proteins: role in normal tissue remodelling. *Science* 1998; **240**:1546–8.
6. Bucola R, Tracey KJ, Cerami A. Advanced glycosylation products quench nitric oxide and mediate defective endothelium-dependent vasodilatation in experimental diabetes. *J Clin Invest* 1991; **87**:432–8.
7. Tuck ML, Corry DB. Pathophysiology and management of hypertension in diabetes. *Annu Rev Med* 1991; **42**:533–48.
8. Corry DB, Tuck ML. Hypertension and diabetes. *Semin Nephrol* 1991; **5**:561–70.
9. Muntzel MS, Hamidou I, Barrett S. Metformin attenuates salt-induced hypertension in spontaneously hypertensive rats. *Hypertension* 1999; **33**:1135–40.
10. Stern N, Tuck ML. Diabetes and hypertension, in LeRoith D, Olefsky JM, Taylor S (eds). *Diabetes Mellitus: A Fundamental and Clinical Text.* New York: Lippincott-Raven, 1996: 357–72.
11. UK, Prospective Diabetes Study Group. Tight blood pressure control and risk of macrovascular complication in type 2 diabetes. UKPDS 38. *BMJ* 1998; **317**:703–13.
12. Wood D, Durrington P, McInnes G, Pulter N, Rees A, Wray A. Joint British recommendations on prevention of coronary heart disease in clinic practice. *Heart* 1998; **80**(Suppl 2): S1–9.
13. Wood D, De Backer G, Faergeman O, Graham I, Mancia G, Pyörälä K. Prevention of coronary heart disease in clinical practice. Summary of recommendations of the Second Joint Task Force of European and other Societies on Coronary Prevention. *J Hypertens* 1998; **16**:1407–14.
14. Grundy SM, Pasternak R, Greenland P, Smith S, Fuster V. Assessment of cardiovascular risk by use of multiple risk factor assessment equations. A statement for healthcare professionals from the American Heart Association and the American College of Cardiology. *Circulation* 1999; **100**:1481–92.
15. Ramsay LE, Williams B, Johnston GD, MacGregor GA, Poston L, Potter JF, Poulter NR, Russell G. Guidelines for management of hypertension: report of the third working party of the British Hypertension Society. *J Hum Hypertens* 1999; **13**:569–92.
16. Ramsay LE, Williams B, Johnston GD, MacGregor GA, Poston L, Potter JF, Poulter NR, Russell G. British Hypertension Society guidelines for hypertension management. 1999; Summary.*BMJ* 1999; **319**:630–5.

Part IV
Lipids and atherosclerosis

Lipids and atherosclerosis

Introduction

In this section of the first *Year in Dyslipidaemia* the opportunity has been taken to include reviews published in the year 2000 in the field of lipids and atherosclerosis. These reviews provide excellent essential up-to-date information for the non-expert and those new to the field and are intended to supplement and enhance the understanding of the highlighted original research papers also included in this section.

13
Non-lipid lowering effects of vastatins

Introduction

Vastatins (statins) inhibit the enzyme 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase which is an essential enzyme in cellular cholesterol biosynthesis. In order to obtain the cholesterol they need, cells upregulate the low-density lipoprotein (LDL) receptor, thereby obtaining cholesterol from LDL and lowering circulating LDL and therefore total cholesterol concentrations. The large primary and secondary prevention studies using statins, i.e. Scandinavian Simvastatin Survival Study (4S), West of Scotland Coronary Prevention Study (WOSCOPS), Cholesterol and Recurrent Events (CARE), Air Force/Texas Coronary Atherosclerosis Prevention Study (AFCAPS/TexCAPS) have all shown approximately 30% improvement in cardiovascular end-points (see Part II Lipid-lowering trials by Professor Durrington). However, this improvement is not fully explained by the LDL lowering effects of the statins. The following two papers illustrate current research into the direct antiatherogenic effects of statins.

Effects of statins on vascular wall: vasomotor function, inflammation, and plaque stability.
K K Koh. *Cardiovasc Res* 2000; **47**:648–57.

B**ACKGROUND. Statins improve cardiovascular end-points and coronary stenosis, but the improvements are incompletely explained by improvements in LDL cholesterol levels. The clinical benefits of statins may involve non-lipid mechanisms that modify endothelial function, smooth muscle cells and monocyte-macrophage: vasomotor function, inflammatory responses and plaque stability. Statin therapy might enhance endothelium-dependent vasodilation by augmenting the bioactivity of nitric oxide. Studies show that statins decrease smooth muscle cell migration and proliferation, independently of their ability to reduce plasma cholesterol. They also reduce *in vitro* cholesterol accumulation in macrophages and expression of matrix metalloproteinase, resulting in plaque stability. These effects were prevented completely by adding mevalonate and partially by all-*trans* farnesol and all-*trans* geranylgeraniol, confirming the regulatory role of isoprenoid metabolites. Statins prevent the activation of monocytes into**

macrophages, inhibit the production of pro-inflammatory cytokines,

C-reactive protein and cellular adhesion molecules, and decrease the adhesion of monocytes to endothelial cells.

I**NTERPRETATION.** Statins exert their cardiovascular benefits through direct antiatherogenic properties in the arterial wall, beyond their effects on plasma lipids.

Comment

The manuscript reviews some of the direct antiatherogenic effects of statins on the arterial wall and provides excellent basic information on the so-called 'pleiotrophic effects' of statins.

Statins as a newly recognized type of immunomodulator.
B Kwak, F Mulhaupt, S Myit, F Mach. *Nat Med* 2000; **6**(12):1399–402.

B**ACKGROUND. Statins are effective lipid-lowering agents. They have never been shown to be involved in the immune response, although a report indicated a better outcome of cardiac transplantation in patients treated with pravastatin. Major histocompatibility complex (MHC) class II molecules are directly involved in the activation of T lymphocytes and control of the immune response. Only a few specialized cell types express MHC II constitutively, but numerous others become MHC II positive on induction by interferon-γ. This complex regulation is controlled by the transactivator CIITA.**

I**NTERPRETATION.** This report shows that statins directly inhibit the induction of MHC II expression by interferon-γ and thus repress MHC II-mediated T-cell activation. This effect of statins is due to inhibition of the inducible promoter IV of the transactivator CIITA. This inhibition is specific for inducible MHC II expression. In repressing induction of MHC II, and subsequent T-lymphocyte activation, statins provide a new type of immunomodulation. This provides a rationale for using statins as immunosuppressors in organ transplantation and in many other pathologies.

Comment

In addition to inhibiting the inflammatory immune response in atherosclerosis this work indicates a wider role for statins as immunosuppressive agents that may be useful in the

treatment of transplant rejection and of various autoimmune diseases such as rheumatoid arthritis.

Conclusion

These aspects of statin function are very much a ‘new field’ and require much more *in vitro* and *in vivo* experimentation to confirm their clinical usefulness in addition to cholesterol lowering. However, it is a potentially exciting development with implications both for the treatment of atherosclerosis in those patients who are at risk without the presence of high total or LDL cholesterol concentrations. In addition immunosuppression by statins has important implications for the treatment of organ transplantation recipients and for those diseases with an autoimmune component such as rheumatoid arthritis, insulin-dependent diabetes mellitus and multiple sclerosis. It will be interesting to watch developments in these fields in the future.

14
High-density lipoproteins

Introduction

Among the many independent risk factors for coronary heart disease (CHD) identified by epidemiological studies, low plasma high-density lipoprotein (HDL) concentration is one of the strongest |**1**|. The Framingham Heart Study demonstrated that for any given low-density lipoprotein (LDL) concentration, HDL cholesterol concentration is inversely correlated with CHD risk |**2**|. A review of 19 prospective risk factor studies for CHD by a National Institutes of Health consensus panel in 1992 indicated that 15 of the studies reported a significant association between low HDL and CHD while three of the other studies showed a non-significant trend towards this association |**3**|. Prospective studies conducted since this time have continued to show the strong association between low HDL and CHD |**4,5**|.

Similarly, studies in animal models susceptible to the development of athero-sclerosis, such as the apolipoprotein (apo)E-deficient mouse, have shown that expression of the human apo A1 transgene increases HDL concentration and greatly diminishes fatty streak formation |**6–8**|.

One explanation for the inverse association between HDL and the development of CHD is that HDL has a protective role against atherogenesis. How might such protection be achieved? Several possible potential mechanisms have been reported. Most research in this area has focused on the central role of HDL in reverse-cholesterol transport, which has been the subject of an excellent recent review |**9**| and will not be dealt with here. Other potentially antiatherogenic properties of HDL, such as reducing blood viscosity, regulation of prostaglandin and thromboxane synthesis and the activation of fibrinolysis have also been reported (reviewed in reference 10). However, the greatest interest in recent years has been the capacity of HDL to protect LDL against lipid peroxidation. This section reviews some of the latest developments in the field of HDL research.

Epidemiology

Building on previous epidemiological studies of HDL and CHD, two large studies described below are of interest because of the additional information they provide.

HDL cholesterol as a marker of coronary heart disease risk: the Quebec cardiovascular study.

J Despres, I Lemieux, G Dagenais, B Cantin, B Lamarche. *Atherosclerosis* 2000; **153**:263–72.

B**ACKGROUND. Primary as well as secondary prevention trials have shown the relevance of lowering LDL-cholesterol to reduce CHD risk. However, although the association between LDL-cholesterol and CHD is well recognized, there is a considerable overlap in the distribution of plasma LDL-cholesterol levels between CHD patients and healthy subjects. The objective of the present review article is to use data from the Quebec cardiovascular study to demonstrate that in men, a low HDL-cholesterol (HDL-C) may be even more of a risk factor and a target for therapy than a high LDL-cholesterol. Results of the Quebec cardiovascular study, a prospective study of 2103 middle-aged men followed for a period of 5 years, have confirmed results of previous studies in showing that plasma HDL-C concentration was an independent predictor of a first ischaemic heart disease (IHD) event which included typical effort angina, coronary insufficiency, non-fatal myocardial infarction and coronary death. In addition, a reduced plasma HDL-C concentration was found to have a greater impact than raised LDL-cholesterol on the atherogenic index (total cholesterol/HDL-C ratio), this ratio being the best variable of the traditional lipid profile for the prediction of IHD events in the Quebec cardiovascular study (Fig. 14.1). However, a low HDL-cholesterol**

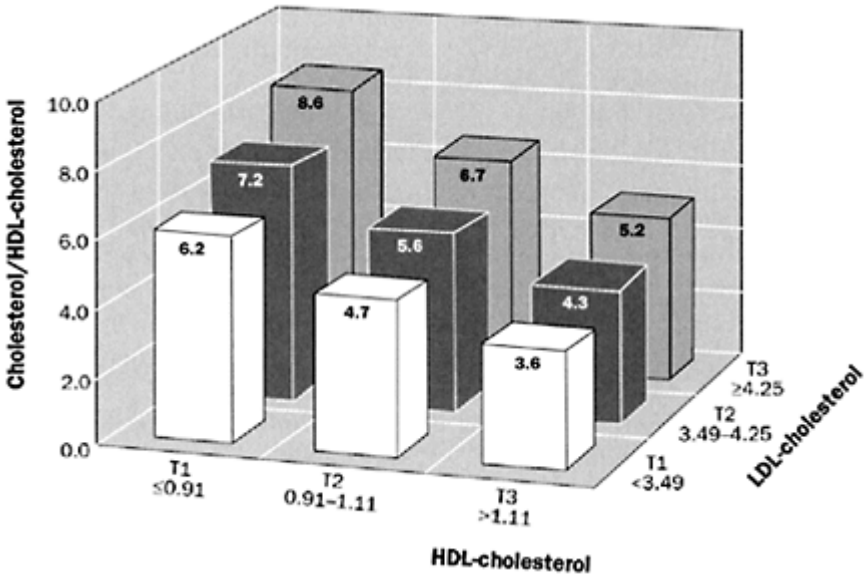

Fig. 14.1 Cholesterol/HDL cholesterol ratio values among men of the Quebec cardiovascular study stratified into tertiles of LDL cholesterol and HDL cholesterol. Numbers within bars represent total/HDL cholesterol values. Source: Despres *et al.* (2000).

concentration is not often observed as an isolated disorder but also includes hypertriglyceridemia, elevated apo B concentration, and an increased proportion of small, dense LDL particles. These abnormalities are features of an insulin resistant-hyperinsulinemic state resulting from abdominal obesity.

I**NTERPRETATION.** It is therefore recommended that we need to go beyond LDL-cholesterol measurement lowering therapy for the optimal management of CHD risk. Raising plasma HDL-C through weight loss and a healthy diet, by an increased physical activity and, if required, by proper pharmacotherapy is therefore a legitimate therapeutic target for the optimal prevention of CHD in a large proportion of high-risk patients.

Comment

The authors comment on the need to look beyond LDL cholesterol lowering in the treatment of atherosclerosis to therapy designed to raise HDL-C concentrations. This is a laudable goal; however, the prevention of atherosclerosis may be better served by targeting specific functions of HDL to enhance, for example, reverse cholesterol transport or the antioxidant activity of HDL.

HDL cholesterol level predicts survival in men after coronary artery bypass graft surgery: 20-year experience from the Cleveland clinic foundation.

J M Foody, F D Ferdinand, G L Pearce, *et al. Circulation (Online)* 2000; **102** (Suppl 3):11190–4.

B**ACKGROUND. HDL-C is an important independent predictor of atherosclerosis, yet the role that HDL-C may play in the prediction of long-term survival after coronary artery bypass grafting (CABG) remains unclear. The risk associated with a low HDL-C level in post-CABG men has not been delineated in relation to traditional surgical variables such as the use of arterial conduits, left ventricular function, and extent of disease. We performed a prospective, observational study of 432 men who underwent CABG between 1978 and 1979 in whom preoperative HDL-C values were available. Baseline lipid and lipoprotein values, history of diabetes mellitus and hypertension, left ventricular ejection fraction, extent of disease, and use of internal thoracic arteries were recorded. Hazard ratios (HRs) were determined in the patients with and without a low HDL-C level, which was defined as the lowest HDL-C quartile (HDL-C</=35 mg/dL). After adjustment for age, as well as for baseline metabolic parameters and surgical variables just noted, HDL-C corresponded to both overall (HR 0.40, CI 0.20–0.83, *P*=0.01) and event-free (HR 0.41, CI 0.24–0.70, *P*=0.001) survival. Patients with a high HDL-C level (>35 mg/dL) were 50% more likely to survive at 15 years than were patients with low HDL-C level (</=35 mg/dL) (74% *versus* 57% adjusted survival, respectively; HR 1.72, *P*=0.005). In addition, HDL-C showed a strong effect on time-to-event survival such that patients with an HDL-C level of>35 mg/dL were 50% more likely to survive without a subsequent myocardial infarction or revascularization (HR 1.42, *P*=0.02).**

INTERPRETATION. HDL-C is an important predictor of survival in post-CABG patients. In this study of >8500 patient-years of follow-up, HDL-C was the most important metabolic predictor of post-CABG survival. One-third fewer patients survive at 15 years if their HDL-C levels are </=35 mg/dL at the time of CABG. The measurement of HDL-C provides a compelling strategy for the identification of high-risk subsets of patients who undergo CABG (Fig. 14.2).

Comment

These findings reinforce the importance of high HDL in protecting against CHD and should provide impetus to the discovery of pharmaceutical agents to modulate HDL concentration.

ABC1 receptor

Tangier disease is an inherited HDL deficiency state. Cells from patients with Tangier disease lack the ability to efflux cholesterol and phospholipid to lipid poor apoA1 (pre-β HDL) and thus form mature HDL |**11**|. Therefore, these patients lack reverse cholesterol transport resulting in a build-up of cellular cholesterol in many cell types. Recent studies have shown that the genetic defect in Tangier disease is a

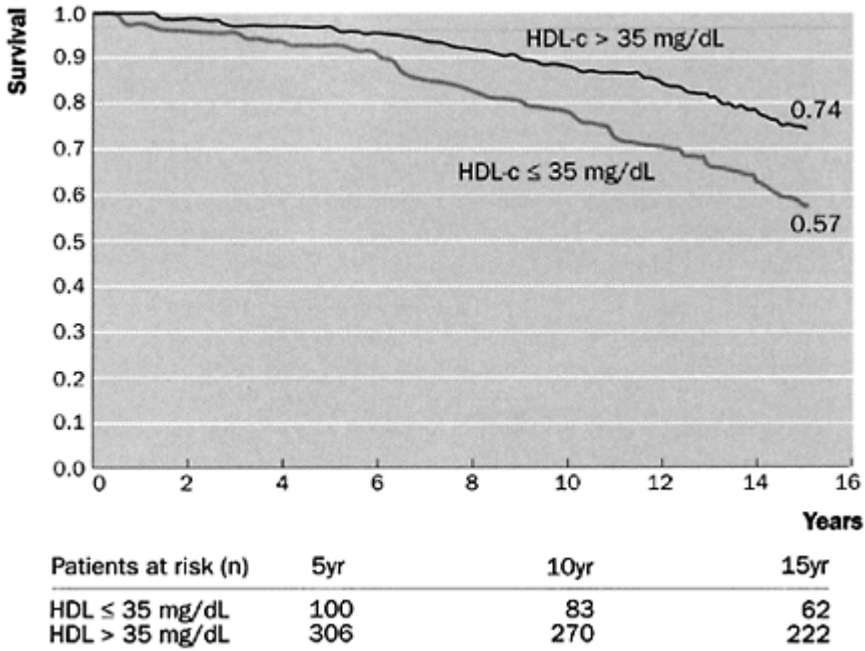

Fig. 14.2 Survival and HDL cholesterol (HDL-C). Adjusted for age, hypertension, diabetes mellitus, body mass index, total cholesterol, triglycerides, left ventricular ejection fraction, disease extent and inferior temporal artery use. Source: Foody *et al.* (2000).

mutation of the adenosine triphosphate (ATP)-binding cassette 1 (ABC 1) |**12–14**|. Further studies have implicated ABC1 as being central to the process of reverse cholesterol transport being the protein that facilitates the efflux of excess cellular cholesterol |**15**|. The following two papers have added to our knowledge the mechanism and function of ABC1.

The correlation of ATP-binding cassette 1 mRNA levels with cholesterol efflux from various cell lines.

A E Bortnick, G H Rothblat, G Stoudt, *et al. J Biol Chem* 2000; **275**: 28634–40.

B**ACKGROUND. Studies show that lipid-free apoA-I stimulates the release of cholesterol and phospholipid from fibroblasts and macrophages. ABC1 is implicated in this release and there is evidence that ABC1 is critical to the biogenesis of HDL. The present study aimed to determine whether increased efflux of cholesterol was correlated to the upregulation of ABC1 mRNA and protein.**

I**NTERPRETATION.** Studies in J774 and other cell types revealed a close correlation between ABC1 message levels and increased efflux of cholesterol. The relationship was confirmed by studies of elicited macrophages from the *Abc1*$^{-/-}$ mouse. The stimulation of efflux is specific for apoA-I, HDL and other apolipoproteins as cholesterol acceptors.

Comment

The amount of ABC1 mRNA and protein is absolutely related to the ability of a cell to efflux cholesterol (see Fig. 14.3) providing further evidence of the importance of ABC1 in reverse cholesterol transport.

Functional loss of ABCA1 in mice causes severe placental malformation, aberrant lipid distribution, and kidney glomerulonephritis as well as high-density lipoprotein cholesterol deficiency.

T A Christiansen-Weber, J R Voland, Y Wu, *et al. Am J Pathol* 2000; **157**: 1017–29.

B**ACKGROUND. Tangier disease and familial HDL deficiency have recently been linked to mutations in the human ATP-binding cassette transporter 1 (hABCA1). Both diseases are characterized by the low levels or absence of HDL-C and low serum cholesterol. The murine ABCA1**$^{-/-}$ **phenotype mimics the human Tangier disease linkage to ABCA1; HDL-C is virtually absent and serum cholesterol and tissue lipid deposition are reduced.**

I**NTERPRETATION.** This murine model of Tangier disease will assist

studies of lipid metabolism, renal inflammation and cardiovascular disease and their possible

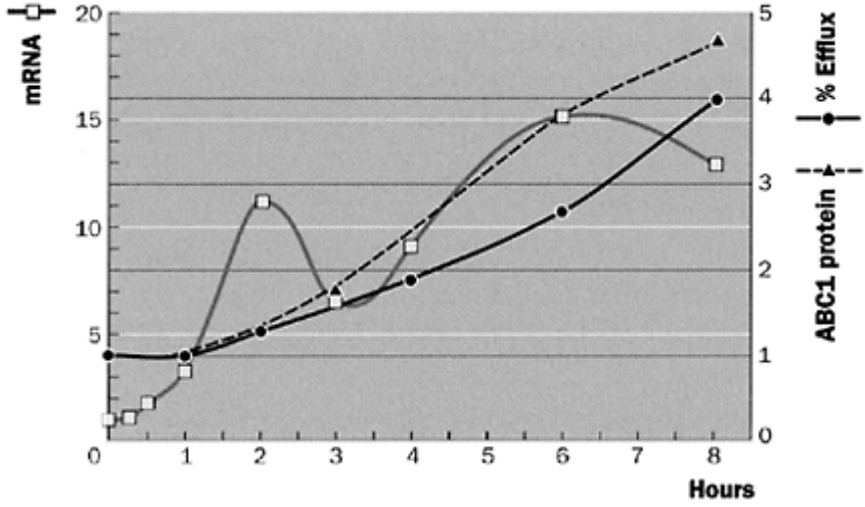

Fig. 14.3 Time course of changes in ABC1 mRNA, protein and cholesterol efflux upon cpt-cyclic adenosine monophosphate (cpt-cAMP) treatment of J774 macrophages. J774 cells were seeded at 5×10^6/100-mm dish, grown for 2 days to confluence, and incubated with acylcholesterol acyltransferase (ACAT) inhibitor. At the beginning of the experiment, a set of 100-mm plates was harvested for baseline ABC1 mRNA and protein and parallel monolayers were treated with 0.3 mM cpt-cAMP. Monolayers were sequentially harvested over the course of 8 h. In a separate experiment with triplicate wells, a radiolabelled set of cells was used to determine cholesterol efflux to lipid-free apoA-I (20 μg/ml) in the presence of 0.3 mM cpt-cAMP. , fold increase in ABC1 mRNA; ▲, fold increase in ABC1 protein; ●, absolute % cholesterol efflux to apoA-I.

Source: Bortnick *et al.* (2000).

interrelationships. The lack of HDL-C in these mice leads to altered steroidogenesis resulting in malformation of the placenta, embryo growth retardation, fetal loss and neonatal death. Surviving $ABCA1^{-/-}$ animals develop membranoproliferative glomerulonephritis, cardiomegaly and congestive heart failure.

Comment

The central role of ABC1 in reverse cholesterol transport makes it an obvious target for

future research into pharmaceutical modulation of the protein and therefore of HDL concentration.

Antioxidative function of high-density lipoprotein

It is now generally accepted that LDL oxidation is central to the pathogenesis of atherosclerosis |**16,17**|. The oxidation of LDL results in the formation of a variety of peroxidized lipids derived from the polyunsaturated fatty acids associated with phospholipids and cholesteryl esters (CE) and their breakdown products. The pro-atherogenic effects of oxidized LDL and the individual peroxidized lipid components it contains have been the subject of two excellent recent reviews |**18**,**19**| and will not be dealt with here.

The following three papers explore the possible mechanisms by which HDL is able to prevent the oxidation of LDL and acts as an antioxidant.

How high-density lipoprotein protects against the effects of lipid peroxidation.

M I Mackness, P N Durrington, B Mackness. *Curr Opin Lipidol* 2000; **11**: 383–8.

B**ACKGROUND. The capacity of HDL to protect against the development of atherosclerosis appears to involve a number of mechanisms. One of the major mechanisms is the ability of HDL to decrease, directly or indirectly, the lipid peroxidation of LDL. The hydrolysis of lipid peroxides by paraoxonase (PON1) makes a major contribution to this effect of HDL. Evidence is accumulating that the PON1 activity of human serum can be modulated by various natural compounds, which may increase or decrease the protectiveness of PON1 and, therefore, of HDL on which it is exclusively located.**

I**NTERPRETATION.** Modulations of PON1 that enhance its activity may help to delay the atherosclerotic process.

Comment

Although there is evidence that a number of mechanisms could explain the ability of HDL to prevent LDL lipid peroxidation (see Fig. 14.4), such as the transfer of phospholipid hydroperoxides from LDL to HDL, followed by transfer of the oxidized fatty acid to cholesterol by lecithin:cholesterol acyltransferase and the selective disposal of the oxidized CE by the liver. This mechanism appears to be relatively unimportant compared with the direct metabolism of lipid peroxides by the HDL-associated enzyme PON1. The manuscript provides a comprehensive review of the properties of PON1 in

relation to the development of atherosclerosis, making the very important point regarding the need to study PON1 activity in epidemiological studies of CHD. A point reinforced by later findings by Jarvik *et al.* |**20**|.

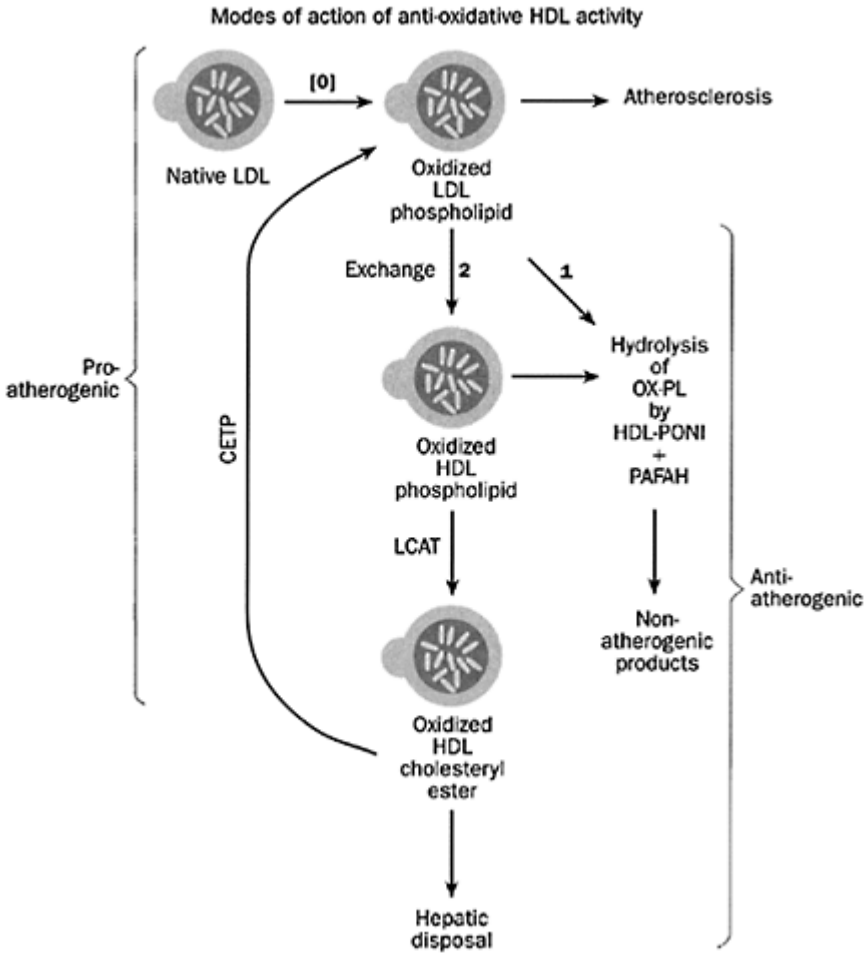

Fig. 14.4 Possible modes of action to explain the anti-oxidative activity of high-density lipoprotein. 1, direct metabolism; 2, transfer. Source: Mackness *et al.* (2000).

The next two studies explore the mechanisms by which mildly oxidized LDL is formed and how these mechanisms are prevented by HDL.

Normal high density lipoprotein inhibits three steps in the formation of mildly oxidized low density lipoprotein. Step 1.
M Navab, S Y Hama, C J Cooke, *et al. J Lipid Res* 2000; **41**:1481–94.

B**ACKGROUND. Apolipoprotein A-I (apoA-I) and an apoA-I peptide mimetic removed seeding molecules from human LDL and rendered the LDL resistant to oxidation by human artery wall cells. The apoA-I-associated seeding molecules include hydroperoxyoctadecadienoic acid (HPODE) and hydroperoxyeicosatetraenoic acid (HPETE). LDL from mice genetically susceptible to the formation of fatty streak lesions was highly susceptible to oxidation by artery wall cells and was rendered resistant to oxidation after incubation with apoA-I *in vitro.* Injection of apoA-I into mice and humans rendered their LDL resistant to oxidation within 3 h and 6 h, respectively.**

I**NTERPRETATION.** This study indicates that (a) oxidation of LDL by artery wall cells requires seeding molecules that include HPODE and HPETE; (b) LDL from mice genetically susceptible to atherogenesis is more readily oxidized by artery wall cells; and (c) normal HDL and its components can remove or inhibit the activity of lipids in freshly isolated LDL that are needed for oxidation by human artery wall cells.

Normal high density lipoprotein inhibits three steps in the formation of mildly oxidized low density lipoprotein. Steps 2 and 3.
M Navab, S Y Hama, G M Anantharamaiah, *et al. J Lipid Res* 2000; **41**: 1495–508.

B**ACKGROUND. Human artery wall cells treated with apoA-I, but not apoA-II, with an apoA-I peptide mimetic, or with HDL, or PON1, are unable to oxidize LDL. These cells contain 12-lipoxygenase (12-LO). Transfection of the cells with anti-sense to 12-LO eliminated the 12-LO protein and prevented LDL-induced monocyte chemotactic activity. Addition of 13(S)-hydroperoxyoctadecadienoic acid and 15(S)-hydroperoxyeicosatetraenoic acid greatly enhanced the non-enzymatic oxidation of both PAPC and cholesteryl linoleate. Purified PON1 inhibited the biological activity of these oxidized phospholipids. HDL from normolipidaemic patients with coronary artery disease, who were neither diabetic nor receiving hypolipidaemic medication, inhibited**

neither LDL oxidation by artery wall cells nor the biological activity of oxidized PAPC, whereas HDL from control subjects did.

I**NTERPRETATION.** Mildly oxidized LDL is formed in three steps, one of which involves 12-LO protein and each of which can be inhibited by normal HDL. HDL from at least some coronary artery disease patients with normal blood lipid levels lacks the capacity to

prevent LDL oxidation by artery wall cells and to inhibit the biological activity of oxidized PAPC (1-palmitoyl-2-arachidonoyl-sn-glycero-3-phosphocholine).

Comment

LDL is oxidized by a three-step mechanism (see Fig. 14.5). In the first step, LDL, either in the circulation or the sub-endothelial space, is 'seeded' with reactive oxygen species (oxidized lipids) probably by the action of 12-LO in the artery wall cells. Secondly, LDL is trapped in the extra-cellular matrix of the sub-endothelial space where it receives more seeding molecules. Lastly, when a critical level of seeding molecules has been reached (and, probably, LDL antioxidants depleted) LDL undergoes auto-oxidation. Normal HDL and PON1 can inhibit all three steps in the formation of oxidized LDL. Interestingly, HDL from patients with CHD is deficient in PON1 and cannot inhibit the oxidation of LDL. The antioxidative function of HDL is very important in the anti-atherosclerotic action of HDL. This antioxidative function is largely due to the action of PON1. Studies into this enzyme will obviously continue apace.

Cholesteryl-ester transfer protein (CETP)

CETP catalyses the transfer of CE from HDL to VLDL/HDL in exchange for triglyceride (TG). However, controversy has raged as to whether CETP is a pro-atherosclerotic or antiatherosclerotic agent |**21**|. On the one hand as CE transferred to VLDL/LDL can be delivered to the liver for disposal, CETP may enhance the rate of reverse cholesterol transport. On the other hand the transfer of CE from non-atherogenic HDL to pro-atherogenic VLDL/LDL may be an atherogenic mechanism. The reverse transfer of TG from LDL to HDL may also result in the formation of small dense LDL, the most atherogenic LDL species |**22**|. Although this year has seen some resolution of the controversy it remains an interesting subject for research.

Common cholesteryl ester transfer protein mutations, decreased HDL cholesterol, and possible decreased risk of ischemic heart disease: the Copenhagen city heart study.

B Agerholm-Larsen, A Tybjaerg-Hansen, P Schnohr, R Steffensen, B G Nordestgaard. *Circulation (Online)* 2000; **102**(18):2197–203.

B**ACKGROUND. CETP mediates the transfer of CE from HDL in exchange for TGs in apoB-containing lipoproteins. We studied two common mutations in CETP, A373P and R451Q, in 8467 healthy women and men from the Danish general population, and in 1636 Danish women and men with ischaemic heart disease. The prevalence of 373P and 451Q was 0.10 and 0.07, respectively, for heterozygous carriers and 0.003 and**

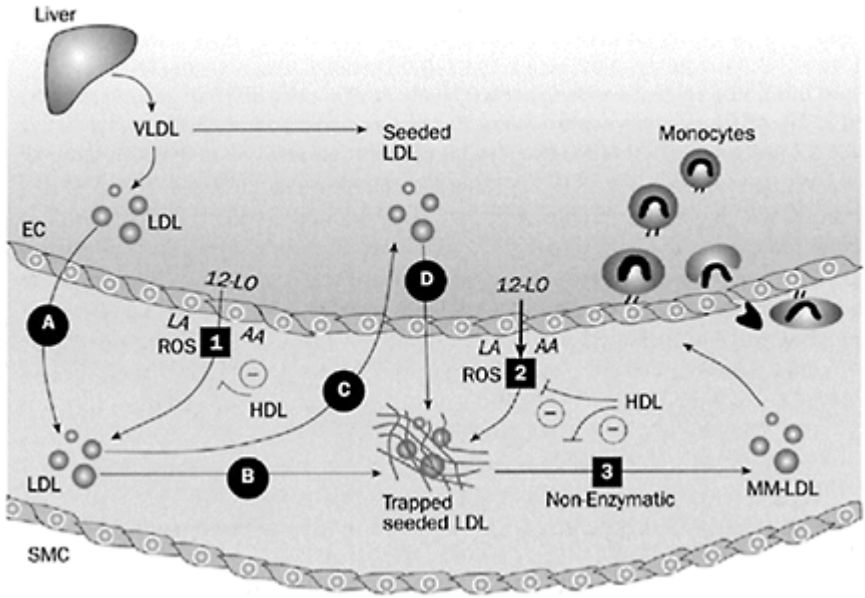

Fig. 14.5 A three-step model for LDL oxidation by artery wall cells. Step 1: LDL is seeded. Step 2: LDL is trapped in the artery wall and receives further seeding molecules. Step 3: When a critical level of seeding molecules relative to phospholipids is reached in the LDL, a non-enzymatic oxidation process generates POVPC, PGPC, and PEIPC. LDL that is formed from the hydrolysis of VLDL in the circulation may contain seeding molecules. Alternatively, LDL may enter the subendothelial space (A), where it is seeded with reactive oxygen

species (ROS) delivered from the artery wall cells [likely by the action of 12-lipoxygenase (12-L0) on linoleic (LA) and arachidonic (AA) acids (step 1)]. While the diagram depicts this as occurring in the subendothelial space, step 1 might actually occur in the microcirculation. If the LDL is seeded in the subendothelial space it might remain there, becoming trapped in the extracellular matrix (B), or the seeded LDL could exit into the circulation (C) and re-enter the subendothelial space at another site, where it would become trapped in the extracellular matrix (D). In step 2 the artery wall cells generate and transfer additional or different ROS to the trapped seeded LDL. This transfer could occur within the cell, at the cell surface, or in an adjacent protected microdomain. After this transfer of ROS to the seeded and trapped LDL, a non-enzymatic propagation of lipid oxidation occurs (step 3). This results in the formation of specific oxidized phospholipids that induce NF-κB activation, monocyte binding, MCP-I production, and macrophage colony stimulating factor production and that are present in mildly oxidized LDL (minimally modified LDL; MM-LDL). As indicated, normal HDL is capable of blocking each and every step in the formation of MM-LDL. SMC, smooth muscle cells; EC, endothelial cells. Source: Navab *et al.* (2000).

0.002, respectively, for homozygous carriers. All carriers of the 451Q allele also carried the 373P allele. HDL-C in female non-carriers, heterozygotes, and homozygotes of 373P was 1.74+/−0.01 (mean+/−SE), 1.62+/−0.02, and 1.38+/−0.09 mmol/L, respectively (ANOVA, P<0.001). In men, equivalent values were 1.40+/−0.01, 1.26+/−0.02, and 1.19+/−0.09 mmol/L, respectively (ANOVA, P: <0.001). HDL cholesterol decreased similarly as a function of 451Q genotypes and all 373P/451Q genotype combinations. Furthermore, apolipoprotein AI and the HDL-C/apolipoprotein AI ratio were also lower in carriers of either of these mutations for both sexes. Finally, the CETP genotype was not associated with risk of ischaemic heart disease unless we adjusted for HDL cholesterol: female heterozygous and homozygous carriers *versus* non-carriers had 36% lower risk of ischaemic heart disease (95% CI 4% to 57%); in male carriers, we observed a similar trend.

I**NTERPRETATION.** The A373P/R451Q polymorphism in CETP is associated with decreases in HDL-C of 0.12 to 0.36 mmol/L in women and 0.14 to 0.21 mmol/L in men (see Fig. 14.6) and possibly with a paradoxical 36% decrease in the risk of ischaemic heart disease in women.

Comment

These data amply illustrate why CETP is subject to such controversy as to whether it is pro- or antiatherogenic, and also the danger of genetic studies, which often provide conflicting results.

Vaccine-induced antibodies inhibit CETP activity in vivo and reduce aortic lesions in a rabbit model of atherosclerosis.
C W Rittershaus, D P Miller, L J Thomas, *et al. Arterioscler Thromb Vasc Biol* 2000; **20**:2106–12.

B**ACKGROUND. Using a vaccine approach, New Zealand White rabbits were immunized with a peptide containing a region of CETP known to be required for neutral lipid transfer function.**

I**NTERPRETATION.** The results demonstrate that CETP activity can be reduced *in vivo* by vaccination with a peptide derived from CETP and support the concept that inhibition of CETP activity *in vivo* can be antiatherogenic. Vaccination against a self antigen may be a viable therapeutic strategy for disease management. Vaccinated rabbits had significantly reduced plasma CETP activity and an altered lipoprotein profile. In a cholesterol-fed rabbit model of atherosclerosis, the plasma cholesterol in HDL was 42% higher and the plasma cholesterol in LDL was 24% lower in the CETP-vaccinated group than in controls. The area of the aorta surface with atherosclerotic lesions was 40% smaller in CETP-vaccinated rabbits than in controls. Vaccination against CETP, in rabbits, resulted in raised HDL cholesterol (Fig. 14.7) and reduced atherosclerosis (Fig. 14.8).

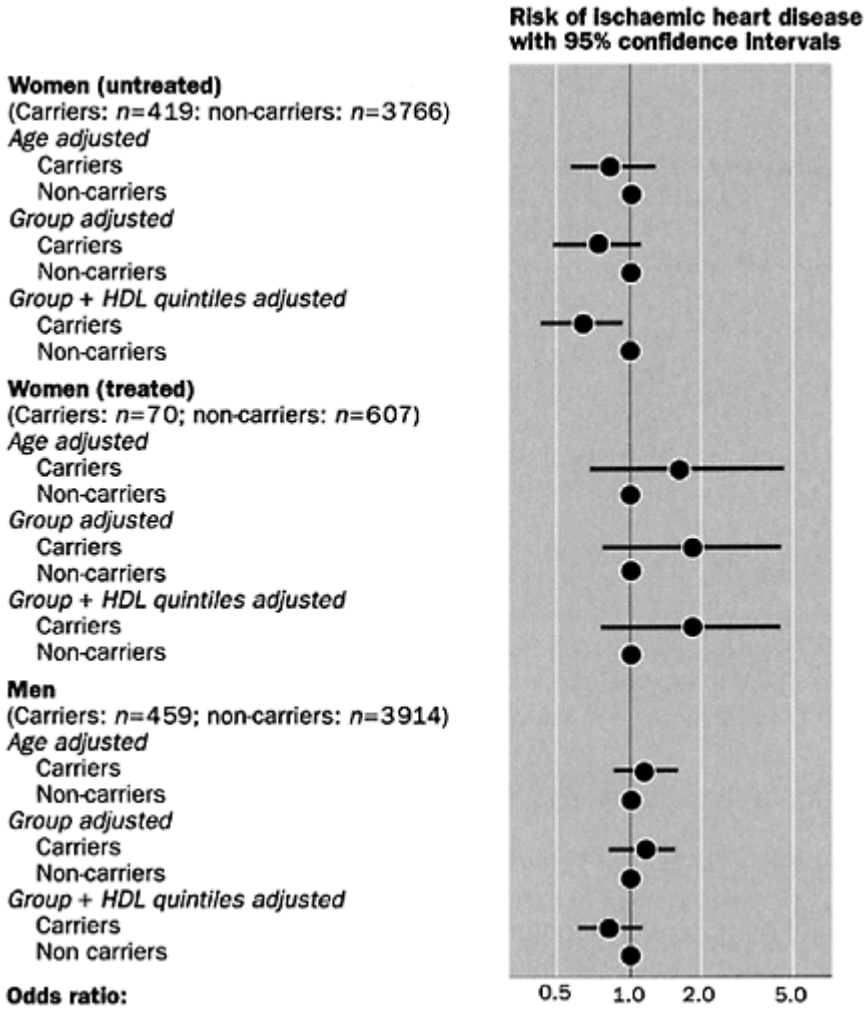

Fig. 14.6 Risk of ischaemic heart disease in heterozygous+homozygous carriers of 373P and 451Q versus non-carriers by logistic regression analysis. Models allowed either for (a) age, (b) a group of known cardiovascular risk factors (age, cholesterol, body mass index, triglycerides, lipid-lowering medication, hypertension, diabetes mellitus and smoking), or (c) the group of known cardiovascular risk factors mentioned above plus HDL cholesterol in quintiles. Source: Agerholm-Larsen *et al.* (2000).

Comment

This is a very novel approach to reducing atherosclerosis. However, whether this approach will work in humans, and have long-term benefits remains to be seen. This

approach may be more appropriate to the prevention of restenosis after percutaneous transluminal coronary angioplasty or CABG as a more short-term benefit.

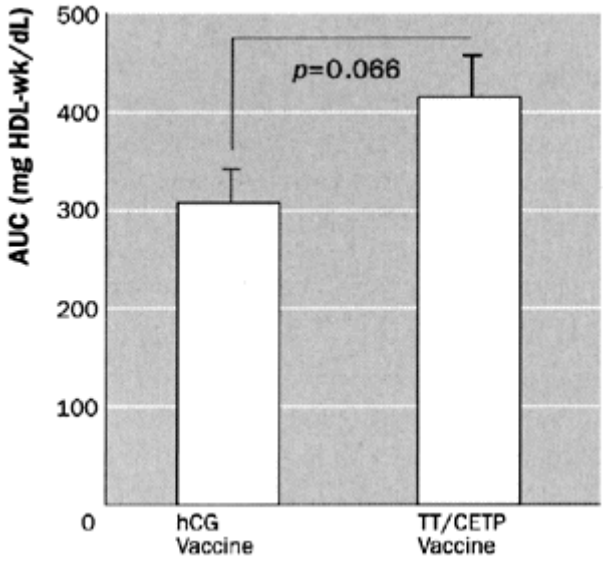

Fig. 14.7 Plasma HDL cholesterol during hypercholesterolaemia. The plasma HDL cholesterol concentration for each rabbit was determined for time points during the cholesterol-feeding portion of the experiment (weeks 19–32). The group mean HDL cholesterol concentrations for the TT/CETP vaccine group (n=12) and the hCG vaccine group (n=12) were plotted versus time, and the area under the curve (AUC) was derived for each group and is represented with standard errors. Source: Rittershaus *et al.* (2000).

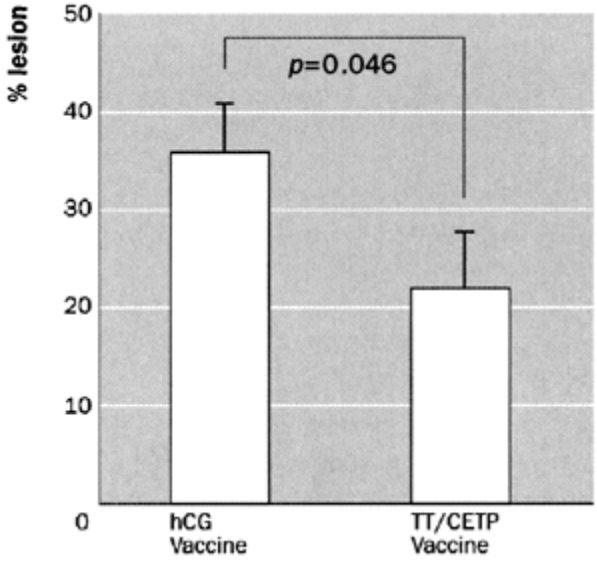

Fig. 14.8 Per cent of aortic area covered with lesion. The per cent aortic area that had lesion was calculated for each rabbit. The bar graphs represent the average (n=12) for each group with standard errors. Source: Rittershaus *et al.* (2000).

A cholesteryl ester transfer protein inhibitor attenuates atherosclerosis in rabbits.

H Okamoto, F Yonemori, K Wakitani, *et al. Nature* 2000; **406**:203–7.

B**ACKGROUND. The plasma protein CETP mediates the exchange of CE in HDL for TG in very low density lipoprotein (VLDL). This process decreases antiatherogenic HDL cholesterol and increases pro-atherogenic VLDL and LDL cholesterol. Thus, CETP is potentially atherogenic, but it could also be antiatherogenic because it participates in reverse cholesterol transport (from peripheral cells through the plasma to the liver). To clarify the role of CETP in atherosclerosis the development of a potent and specific CETP inhibitor was attempted.**

I**NTERPRETATION.** CETP inhibitors that form a disulphide bond with CETP are described, and one such inhibitor (JTT-705) that increases HDL cholesterol, decreases non-HDL cholesterol and inhibits the progression of atherosclerosis in rabbits is presented. The findings indicate that CETP may be atherogenic *in vivo* and that JTT-705 may be a potential antiatherogenic drug. Pharmacological inhibition of CETP by the compound JTT-705 (Fig.

14.9). Results in an increase in HDL cholesterol and a reduction in atherosclerosis (see Table 14.1).

Comment

These data again illustrate inhibition of CETP to be antiatherosclerotic. Safety issues aside, this approach may be more relevant to the pharmaceutical treatment of atherosclerosis in humans.

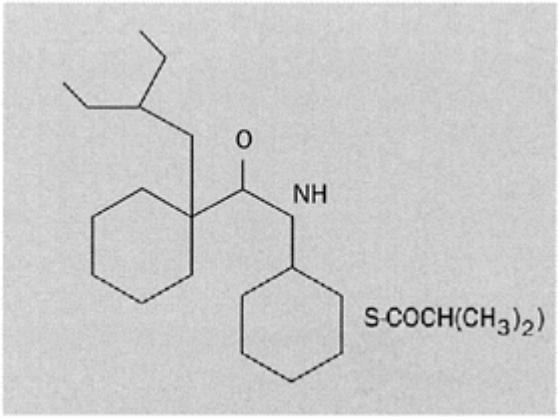

Fig. 14.9 Chemical structure of JTT-705. Source: Okamoto *et al.* (2000).

Table 14.1 Effects of JTT-705 and simvastatin on plasma CETP activity, plasma lipids, serum HDL subfractions, serum apoA-1 and aortic atheroma in rabbits fed a 0.2% cholesterol diet

Group	N	Time (months)	Body weight (kg)	CETP activity* (% of initial activity)	Total cholesterol (mg dl^{-1})	Plasma† HDL-C (mg dl^{-1})	Atherogenic index	HDL$_2$ (mg dl^{-1})
Control	10	0	2.40± 0.04	100±0	274±30	17.5±1.7	16.6±2.7	
		3	3.00±0.06	77±5	153±25	17.0±1.5	8.3±1.4	
		6	3.24±0.07	96±8	129±50	15.2±1.8	7.3±2.8	6.5±0
JTT-705	1	0	2.41±0.04	100±0	263±29	17.5±1.3	14.4±1.8	
		3	2.92±0.07	31±4¶	103±14	33.0±4.2¶	2.4±0.6¶	

		6	3.23±0.07	2.9±7.7¶	98±10	30.0±3.6¶	2.6±0.5	17.6±2
Simvastatin	10	0	2.37±0.02	100±0	269±34	16.8±1.6	16.1±2.5	
		3	2.95±0.03	94±7	57±10‖	18.1±1.6	2.1±0.3¶	
		6	3.26±0.04	68±13	77±16	19.5±2.8	2.7±0.5	8.8±1

JTT-705 (0.75%) or simvastatin (0.0075%) was added to the diet for six months after pre-loading w
* Plasma samples were taken before administration and 7 h after administration at both three and si
the per cent CETP activity was calculated as [(activity at the time of assessment/initial activity)×10
† Plasma samples were taken before administration and 16 h after administration at the three and si
were measured using a commercial available kit (Boehinger Mannheim) and the atherogenic index
C)].
‡ Serum samples were taken 16 h after administration at six months. The cholesterol content in the
and the HDL, fraction (1.125 g ml^{-1} to1.21 g ml^{-1}) was measured using the above-mentioned kit.
competitive immunoassay using goat anti-rabbit apoA-1 antiserum.
§ At the end of the experimental period, the aortas were removed and stained with 0.5% sudan IV.
evaluated as the per cent lesion area relative to the intima surface area.
Values represent the mean ±SEM. Significantly different from the control group by the Tukey-Krai
Whitney U-test ($^{a}P<0.05$).
Source: Okamoto *et al.* (2000).

Conclusion

All of the data presented in this chapter further enhance previous data on the antiatherosclerotic functions of HDL. In this era, the pharmacological regulation of HDL to enhance the prevention of atherosclerosis would seem to be an imperative |**23**|. The approach can be extremely varied, including agents that could increase the concentration of total HDL, for example by increasing ABC1 or inhibiting CETP or by increasing specific antiatherosclerotic proteins of HDL, e.g. PON1. It is obvious, however, that whatever approach is taken, is of potential benefit to the treatment of atherosclerosis.

References

1. Miller GJ, Miller NE. Plasma high-density lipoprotein concentration and the development of ischaemic heart disease. *Lancet* 1975; **1**:16–19.
2. Castelli WP, Garrison RJ, Wilson PW, Abbot RD, Kalousdiou S, Kammel WB. Incidence of coronary heart disease and lipoprotein cholesterol levels. The Framingham Study. *JAMA* 1986; **256**:2835–8.
3. Consensus Statement. Triglyceride, high-density lipoprotein and coronary heart disease 1992; **10**:1–28.
4. Assmann G, Schulte H, von Eckardstein A, Huang Y. High density lipoprotein cholesterol as a predictor of coronary heart disease risk. The PROCAM experience and patho-physiological implications for reverse cholesterol transport. *Atherosclerosis* 1996; **124**: S11–20.
5. Tanne D, Yaari S, Goldbourt U. High-density lipoprotein cholesterol and risk of

ischaemic stroke mortality. A 21 year follow-up of 8586 men from the Israeli Ischaemic Heart Disease Study. *Stroke* 1997; **21**:83–7.

6. Benoit P, Emmanuel F, Caillund JM, Bassinet L, Castro G, Gallix P, Fruchart JC, BranellecD, Denèfle P, Duverger N. Somatic gene transfer of human apo A1 inhibits atherosclerosis progression in mouse models. *Circulation* 1999; **99**:105–10.

7. Tangirala RK, Tsukamoto K, Chun SH, Usher D, Pure E, Radar DJ. Regression of atherosclerosis induced by liver-directed gene transfer of apolipoprotein AI in mice. *Circulation* 1999; **100**:1816–22.

8. Dansky HM, Charlton SA, Barlow CB, Tamminen M, Smith JD, Frank JS, Breslow JL Apo AI inhibits foam cell formation in apo E-deficient mice after monocyte adherence to the endothelium. *J Clin Invest* 1999; **104**:31–9.

9. von Eckardstein A, Nofer J-R, Assmann G. High density lipoproteins and arteriosclerosis—role of cholesterol efflux and reverse cholesterol transport. *Arterioscler Thromb Vasc Biol* 2001; **21**:13–27.

10. Mackness MI, Mackness B. HDL: Are there any benefits to raising it? In: Bettridge DJ (ed.). *Lipids and Vascular Disease: current issues.* London: Martin Dunitz, 2000:15–26.

11. Oram JF, Mendez AJ, Lymp J, Kavanagh TJ, Halbert CL. Reduction in apolipoprotein-mediated removal of cellular lipids by immortalisation of human fibroblasts and its reversion by cAMP: lack of effect with Tangier disease cells. *J Lipid Res* 1999; **40**: 1769–81.

12. Bodzioch M, Orso E, Klucken J, Langmann T, Bottcher A, Diederich W, Drobnik W, Barlage S, Buchler C, Porsch-Ozcurumez M, Kaminski WE, Hahmann HW, Oette K, Rothe G, Aslanidis C, Lackner KJ, Schmitz G. The gene encoding ATP-binding cassette transport 1 is mutated in Tangier disease. *Nat Genet* 1999; **22**:347–51.

13. Brooks-Wilson A, Marcil M, Clee SM, Zang LH, Roomp K, van Dam M, Yu L, Brewer C, Collins JA, Molhuizen HO, Loubser O, Ouelette BF, Fichter K, Ashbourne-Excoffon KJ, Sensen CW, Scherer S, Mott S, Denis M, Martindale D, Frohlich J, Morgan K, Koop B, Pimstone S, Kastelein JJ, Hayden MR. Mutations in ABC1 in Tangier disease and familial high-density lipoprotein deficiency. *Nature Genet* 1999; **22**:336–45.

14. Rust S, Rosier M, Funke H, Real J, Amoura Z, Piette JC, Deleuze JF, Brewer HB, Duverger N, Denefle P, Assmann G. Tangier disease is caused by mutations in the gene encoding ATP-binding cassette transporter 1. *Nat Genet* 1999; **22**:352–5.

15. Orsó E, Broccardo G, Kaminski WE, Böttcher A, Liebisch G, Drobnik W, Götz A, Chambenoit O, Diederich W, Langmann T, Spruss T, Luciani M-F, Rothe G, Lachner KJ, Chimini G, Schmitz G. Transport of lipids from golgi to plasma is defective in Tangier disease patients and ABC1-deficient mice. *Nat Genet* 2000*;* **24**:192–6.

16. Steinberg D, Parthasarathy S, Carew TE, Khoo JC, Witztum JL. Beyond cholesterol modifications of low-density lipoprotein that increase its atherogenicity. *N Engl J Med* 1989; **320**:915–24.

17. Parthasarathy S, Santanam N, Auge N. Oxidised low-density lipoprotein: a two-faced Janus in coronary artery disease? *Biochem Pharmacol* 1998; **56**:279–84.

18. Parthasarathy S, Santanam N, Ramachandran S, Meilhac O. Oxidants and antioxidants in atherogenesis: an appraisal. *J Lipid Res* 1999; **40**:2143–57.

19. McIntyre TM, Zimmerman GA, Prescott SM. Biologically active oxidised phospholipids. *J Biol Chem* 1999; **274**:25189–92.

20. Jarvik GP, Rozek LS, Brophy VH, Hatsukami TS, Richter RJ, Schellenberg GD, Furlong CE. Paraoxonase (PON1) phenotype is a better predictor of vascular disease

than is $PON1_{192}$ or $PON1_{55}$ genotype. *Arterioscler Thromb Vasc Biol* 2000; **20**:2441–7.

21. Barter P. CETP and atherosclerosis. *Arterioscler Thromb Vasc Biol* 2000; **20**:2029–31.

22. Hirano K-I, Yamashita S, Matsuzawa Y. Pros and cons of inhibiting cholesteryl ester transfer protein. *Curr Opin Lipidol* 2000; **11**:589–96.

23. Pearson TA, Boden WE (eds). The imperative to raise low levels of high-density lipoprotein cholesterol—A better clinical strategy in the prevention and treatment of coronary artery disease. *Am J Cardiol* 2000; **86**:12A.

15
Antioxidants and the Mediterranean diet

Introduction

Given that the oxidation of low-density lipoprotein (LDL) is central to the development of atherosclerosis, one would expect that antioxidant supplementation would be beneficial. *Ad hoc* studies have shown that the Mediterranean diet, which is high in antioxidants is beneficial in preventing atherosclerosis |**1**|. Again, a high intake of antioxidant supplements has been linked to reduced atherosclerosis |**2,3**|. Many animal studies have shown that antioxidant supplementation reduces the incidence of atherosclerosis; however, controlled clinical trials of antioxidant supplementation have been very disappointing |**4,5**|. This chapter explores recent findings in this field.

High plasma levels of alpha- and beta-carotene are associated with a lower risk of atherosclerosis. Results from the Bruneck study.
A D'Odorico, D Martines, S Kiechl, *et al*. *Atherosclerosis* 2000; **153**:231–9.

B**ACKGROUND. There is considerable evidence that carotenoids, vitamins A and E protect against atherosclerosis by antioxidant action. This study assessed the relationship between plasma levels of carotenoids and of vitamins A and E, and atherosclerosis in the carotid and femoral arteries. This was a prospective cross-sectional study of 392 randomly selected men and women.**

I**NTERPRETATION.** This study provides further epidemiological evidence of a protective role of high α- and β-carotene in early atherogenesis. No associations were found between plasma levels of vitamin A and E and atherosclerosis. Plasma levels of α- and β-carotene were significantly inversely associated with the prevalence of atherosclerosis in the carotid and femoral arteries and with the 5-year incidence of atherosclerotic lesions in the carotid arteries. Atherosclerosis risk gradually decreased with increasing plasma α- and β-carotene concentrations. Atherosclerosis risk was significantly decreased in the population as the plasma concentration of both α- and β-carotenes increased (see Fig. 15.1). This finding held true after adjustment for many other atherosclerosis risk factors. None of the other dietary antioxidants measured, including vitamin E were related to the risk of atherosclerosis.

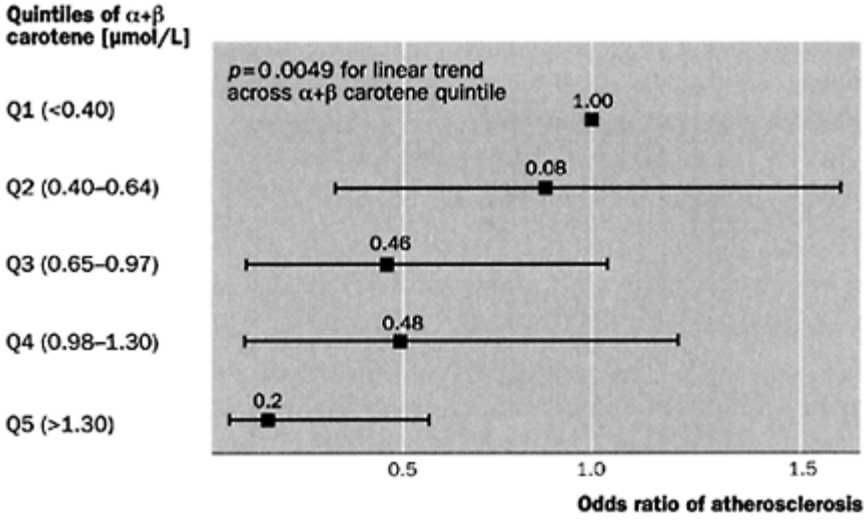

Fig. 15.1 Risk of carotid and femoral artery atherosclerosis according to quintiles of α- and β-carotene (*n*=392). Odds ratios of carotid and femoral atherosclerosis decreased with increasing plasma α+β-carotene concentrations. The horizontal bars represent 95% confidence intervals. Source: D'Odorico *et al.* (2000).

Comment

This study, as many others before it, has shown an association between selected dietary antioxidants and protection against atherosclerosis. The major weakness with this study (as with several of its predecessors) is that no details are given of dietary intake of antioxidant vitamins or whether the Bruneck population favours carotenoid supplementation or foods rich in carotenoids. Therefore, a random effect cannot be discounted.

Antioxidant Supplementation in Atherosclerosis Prevention (ASAP) study: a randomized trial of the effect of vitamins E and C on 3-year progression of carotid atherosclerosis.
J T Salonen, K Nyyssonen, R Salonen, *et al. J Intern Med* 2000; **248**: 377–86.

B**ACKGROUND. This study assessed the efficacy of vitamin E and C supplements for inhibiting the progression of carotid atherosclerosis, and tested the hypotheses: (a) that their inhibitory effects would be enhanced in men and in smokers, and (b) that these vitamins act synergistically. A**

randomized sample of 520 smoking and

Table 15.1 The mean adjusted 3-year change* of the mean carotid artery intima-media thickness in participants who received vitamins E and C supplements in a multivariate general linear model

Supplement	**Men (*n*=225)**					**Women (*n*=233)**				
	Yes		**No**			**Yes**		**No**		
	Mean (*n*)	**SE**	**Mean (*n*)**	**SE**	***P***	**Mean (*n*)**	**SE**	**Mean (*n*)**	**SE**	***P***
Vitamin E (*n*=115)	0.0118 (56)	0.005	0.0143 (169)	0.002	0.571	0.0165 (59)	0.005	0.0170 (174)	0.002	0.904
Vitamin C (*n*=120)	0.0119 (59)	0.005	0.0142 (166)	0.002	0.600	0.0174 (61)	0.005	0.0160 (172)	0.002	0.732
Both vitamins (*n*=113)	0.0086 (58)	0.005	0.0175 (167)	0.002	0.049	0.0170 (55)	0.005	0.0164 (178)	0.002	0.895

* Change estimated as the linear slope over 6-monthly assessments of mean IMT (mm year^{-1}). Covariates in the model for both men and women are serum cholesterol and ferritin concentrations, and three indicator variables for baseline examination months.
Source: Salonen *et al.* (2000)

non-smoking men and postmenopausal women with serum cholesterol of 5.0 mmol/l or more were randomized to receive 91 mg D-α-tocopherol, 250 mg slow-release vitamin C, a combination of these, or placebo for 3 years in this double-masked two-by-two factorial trial.

I**NTERPRETATION.** The results show that a combined supplementation with reasonable doses of both vitamin E and slow-release vitamin C can retard the progression of common carotid atherosclerosis in men. This may imply benefits with regard to other atherosclerosis-based events. The average annual increase in mean intima-media thickness was reduced in men randomized to either of the individual vitamins and significantly reduced in those on the vitamin combination. The proportion of men with progression was significantly reduced (by 74%) by the combined vitamin formulation, compared with placebo. Supplementation with combined vitamin E (136 IU) and vitamin C (250 mg) but not by either vitamin alone, reduced the progression of carotid atherosclerosis over a 3-year period as judged by a decrease in intima-medical thickness, in men but not women (see Table 15.1) regardless of smoking status.

Comment

These findings suggest a synergist effect of vitamins C and E in the prevention of atherosclerosis. However, the results were inconsistent between men and women, possibly due to higher baseline levels of these vitamins in women, but unlikely because supplementation increased plasma vitamin levels in both men and women. The reasons for these findings require much further investigation.

Effects of long-term supplementation with moderate pharmacologic doses of vitamin E are saturable and reversible in patients with type 1 diabetes.

W Engelen, B M Keenoy, J Vertommen, I De Leeuw. *Am J Clin Nutr* 2000; **72**:1142–9.

B**ACKGROUND. Vitamin E supplementation has been proposed as adjunctive therapy to counteract the increased LDL oxidation in diabetes and thus prevent or delay cardiovascular complications. This study investigated the effect of a moderate pharmacological dose of vitamin E for 1 year or less in patients with type 1 diabetes. Patients were randomized to receive RRR-α-tocopherol 250 IU three times daily for 1 year or placebo for 6 months followed by RRR-α-tocopherol for 6 months.**

I**NTERPRETATION.** Life-long supplementation with vitamin E should be considered in patients with type 1 diabetes because the improvement in lipoprotein peroxidizability is saturable and reversible. Serum vitamin E doubled after 3 months of supplementation. Lipid profiles, glycated haemoglobin and blood biochemistry values did not change significantly, but copper-induced *in vitro* peroxidizability of LDL and very low-density lipoprotein decreased after 3 months of supplementation. Vitamin E supplementation for a further 3–9 months resulted in no further changes in serum vitamin E and lipoprotein peroxidizability. Values returned to baseline after supplementation ended. Increased

lipoprotein oxidation is reported to be a risk factor for atherosclerosis in diabetes (although this in itself remains controversial). After 3 months supplementation with 250 IU of vitamin E, lipoprotein oxidation was significantly reduced. However, there was no further reduction with an extra 6 months supplementation. Lipid peroxidation parameters quickly returned to baseline levels when vitamin E supplementation was ceased (see Fig. 15.2).

Comment

Diabetes is a condition characterized by increased oxidative stress |**6**|. Therefore, it is one situation where antioxidant supplementation may be beneficial. However, this study has provided one surprise, that is how quickly after ceasing supplementation vitamin E returned to pre-supplementation levels and oxidative stress increased. This indicates that nothing less than life-long supplementation with vitamin E would be beneficial. If these findings are applicable to the general population it will have major consequences for health care world-wide.

Protective effect of olive oil and its phenolic compounds against low density lipoprotein oxidation.
M Fito, M I Covas, R M Lamuela-Raventos, *et al. Lipids* 2000; **35**:633–8.

B**ACKGROUND. The protective effect of phenolic compounds from an olive oil extract, and olive oils with (extra virgin) and without (refined) phenolic components, on the oxidation of LDL isolated from plasma was investigated.**

I**NTERPRETATION.** The results indicate that olive oil phenolic compounds protect LDL against peroxyl radical dependent and metal-induced oxidation *in vitro* and may associate with LDL after their incubation with plasma. Both types of olive oil protect LDL from oxidation but olive oil containing phenolics has a greater antioxidant effect than refined olive oil. The benefits of the Mediterranean diet in the prevention of cardiovascular disease are well documented |**1**|. In this *in vitro* study it is shown that olive oil, the major source of fat in the Mediterranean diet can inhibit the oxidation of LDL and that this is dependent on the phenolic (natural antioxidant) content of the olive oil (see Fig. 15.3).

Comment

This paper provides more *in vitro* data to support the antiatherogenic effects of the Mediterranean diet. Although it is undoubtedly true that this diet has many benefits it is not yet proven what mechanism(s) are involved *in vivo*.

Conclusion

In humans, the effects of antioxidant supplementation continue to provide conflicting and often controversial results. Major intervention studies, including many subjects will be required in the future to provide a definitive answer.

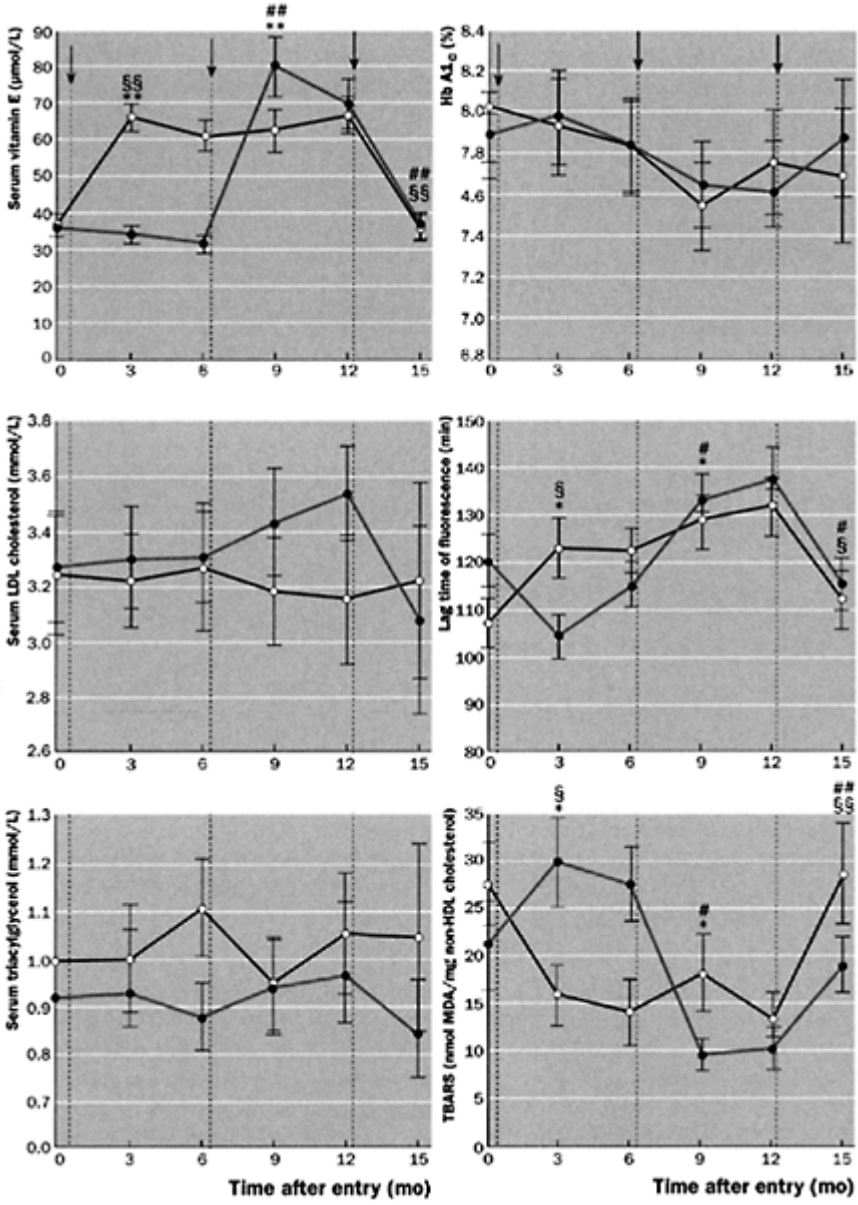

Fig. 15.2 Mean (±SEM) changes in serum vitamin E, serum LDL cholesterol, serum triacylglycerols, serum glycated haemoglobin (HbA_{1c}), lag time of fluorescence, and production of thiobarbituric acid-reactive substances (TBARS) 90 min after incubation with copper in the group receiving vitamin E [750 IU (503 mg) RRR-a-tocopherol/day] for I year (group S; solid line) and in the group receiving placebo for

the first 6 months and then vitamin E for the following 6 months (group P; dashed line). After 12 months both groups stopped taking vitamin E for 3 months and variables were then monitored for the last time (15 months after the start of the study). Significantly different from other group: **P*<0.01, ***P*<0.0005. Significant change since the previous visit in group S: §*P*<0.01, §§*P*<0.0005. Significant change since the previous visit in group P: $^{\#}P$<0.01, $^{\#\#}P$<0.0005. Source: Engelen et al. (2000).

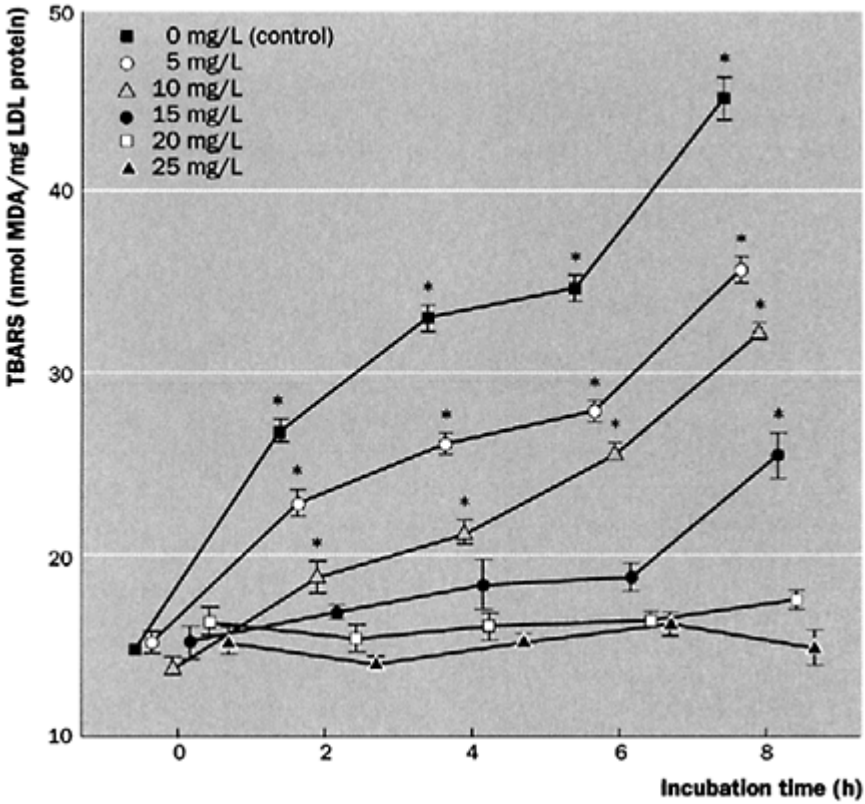

Fig. 15.3 Effect of extra-virgin olive oil phenolics on thiobarbituric acid-reactive substances (TBARS) generation following AAPH-initiated LDL oxidation. LDL (0.10 g protein/I) was incubated with azo-compound (10 mM) at 35°C for up to 8 h in the presence of the indicated amounts of phenolic compounds expressed as caffeic acid equivalents. TBARS formation was measured at intervals of 2 h. Each point represents the mean±SD of three experiments. **P*<0.01 compared with 0 h. MDA, malondialdehyde. Source: Fito *et al.* (2000).

References

1. De Lorgeril M, Salen P, Martin J-L, Moniaud I, Delaye J, Mamelle N. Mediterranean diet, traditional risk factors, and the rate of cardiovascular complications after myocardial infarction: final report of the Lyon Diet Heart Study. *Circulation* 1999; **99**:779–85.
2. Stampfer MJ, Hennekens CH, Manson JE, Colditz GA, Rosner B, Willett WC. Vitamin E consumption and the risk of coronary disease in Women. *N Engl J Med* 1993; **328**: 1444–9.
3. Rimm EB, Stampfer MJ, Ascherio A, Giovannucci E, Colditz GA, Willett WC. Vitamin E consumption and risk of coronary heart disease in men. *N Engl J Med* 1993; **328**:1450–6.
4. Stocker R. Dietary and pharmacological antioxidants in atherosclerosis. *Curr Opin Lipidol* 1999; **10**:589–97.
5. Stocker R. The ambivalence of vitamin E in atherogenesis. *TIBS* 1999; **24**:219–23.
6. Lyons TJ. Oxidised low density lipoproteins: a role in the pathogenesis of atherosclerosis in diabetes? *Diabetic Med* 1991; **8**:411–19.

16
Further issues

Introduction

The following section contains a number of disparate original manuscripts and reviews that do not conform with any particular theme but are nonetheless very important; in particular, to those new to the field, in that they provide comprehensive basic information necessary to understanding various aspects of lipids and atherosclerosis.

Atherosclerosis.
A J Lusis. *Nature* 2000; **407**:233–41.

B**ACKGROUND. Atherosclerosis is the primary cause of heart disease and stroke. In westernized societies, it is the underlying cause of about 50% of all deaths. Several important environmental and genetic risk factors are known to be associated with atherosclerosis. Progress in defining the cellular and molecular interactions involved has been hindered by the aetiological complexity of the disease. New investigative tools, including genetically modified mouse models, have clarified our understanding of the molecular mechanisms involved in the development of atherosclerotic plaque.**

I**NTERPRETATION.** It is now clear that atherosclerosis is not simply an inevitable degenerative consequence of ageing, but a chronic inflammatory condition that can be converted into an acute clinical event by plaque rupture and thrombosis.

Comment

As amply demonstrated by the figures and their accompanying legends (Figs. 16.1 and 16.2), the process of atherosclerosis is a very complex one with several stages. This review, however, provides an excellent tutorial for newcomers and ‘old hands’ alike.

Roles of PPARs in health and disease.
S Kersten, B Desvergne, W Wahli. *Nature* 2000; **405**:421–4.

B**ACKGROUND. In developed societies, chronic diseases such as diabetes, obesity, atherosclerosis and cancer are responsible for most deaths. Genetic, environmental and nutritional factors contribute to the causation of these diseases. There is evidence that a group of closely related nuclear receptors, the peroxisome proliferator-activated receptors (PPARs), may be involved in these diseases. This, and the fact that PPAR activity can be modulated by drugs such as thiazolidinediones and fibrates, has instigated a huge research effort into PPARs.**

I**NTERPRETATION.** This report presents the latest developments in the PPAR field, emphasizing the physiological function of PPARs during various nutritional states, and their possible role in chronic diseases.

Comment

PPARs are nuclear transcription factors that may be the link between lipoprotein metabolism, obesity and insulin resistance via the metabolic processes they control

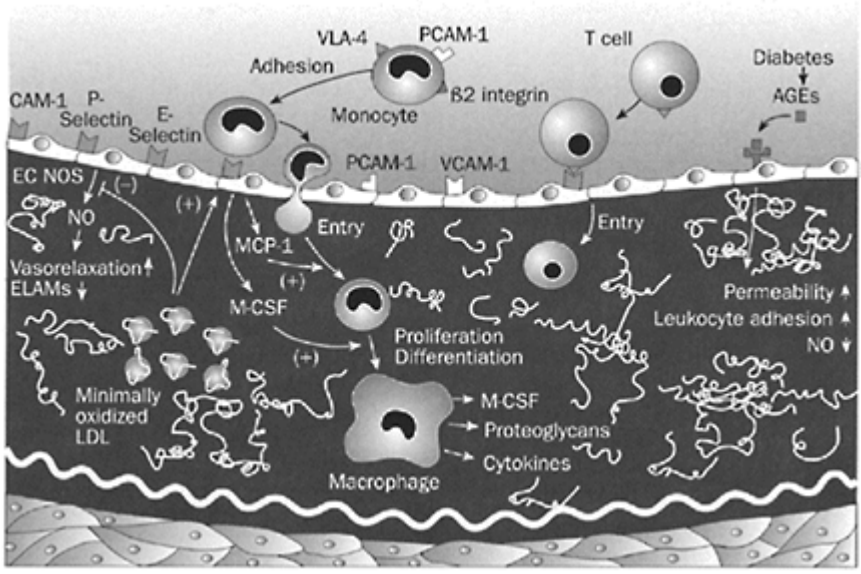

Fig. 16.1 Inflammation. Minimally oxidized LDL stimulates the overlying endothelial cells to produce adhesion molecules, chemotactic proteins such as monocyte chemotactic protein-1 (MCP-1), and growth factors such as macrophage colony-stimulating factor (M-CSF), resulting in the recruitment of monocytes to the vessel wall. Oxidized LDL has other effects, such as inhibiting the production of NO, an important mediator of vasodilation and expression of endothelial leukocyte adhesion molecules (ELAMs). Among endothelial cell adhesion molecules likely to be important in the recruitment of leukocytes are ICAM-1, P-selectin, E-selectin, PCAM-1 and VCAM-1. Important adhesion molecules on monocytes include β2 integrin, VLA-4, and PCAM-1. Advanced glycosylation endproducts (AGEs) are formed in diabetes and these promote inflammation via specific receptors on endothelial cells. Source: Lusis (2000).

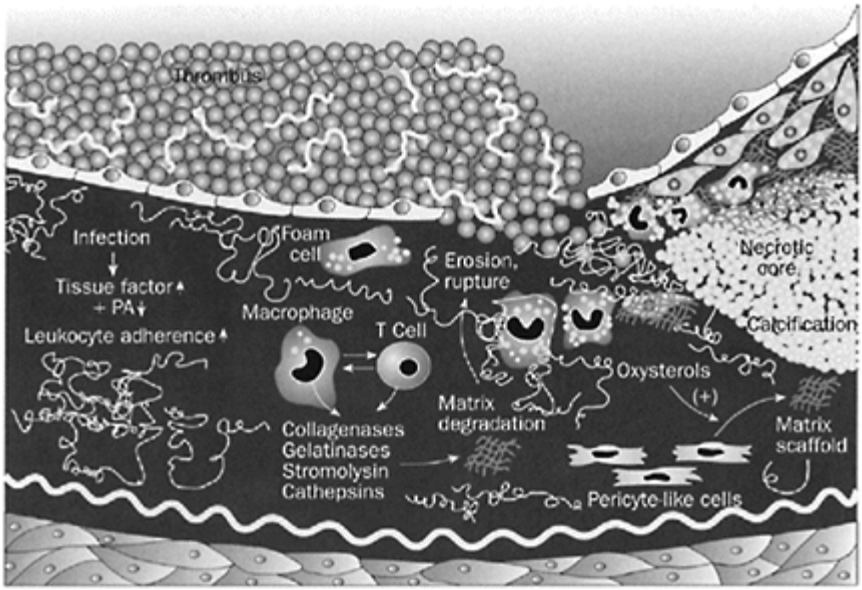

Fig. 16.2 Complex lesions and thrombosis. Vulnerable plaques with thin fibrous caps result from degradation of matrix by various proteinases such as collagenases, gelatinases, stromolysin and cathepsins and by inhibition of matrix secretion. Among various factors that may destabilize plaques and promote thrombosis are infection, which may have systemic effects such as induction of acute phase proteins and local effects such as increased expression of tissue factor and decreased expression of plasminogen activator (PA). The calcification of lesions appears to be an active, regulated process involving the secretion by pericyte-like cells in the intima of a scaffold for calcium phosphate deposition. The formation of a thrombus, consisting of adherent platelets and fibrin crosslinks,

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usually results from plaque rupture, exposing tissue factor in the necrotic core.

Source: Lusis (2000).

(Fig. 16.3). The fact that PPAR activity can be modulated by a number of drug classes has resulted in an explosion of research in this area. Again this review provides an excellent tutorial for all in the field.

Circulating oxidized low density lipoprotein levels: a biochemical risk marker for coronary heart disease.

S Toshima, A Hasegawa, M Kurabayashi, *et al. Arterioscler Thromb Vasc Biol* 2000; **20**:2243–7.

B**ACKGROUND. Recent studies have established oxidative modification of low-density lipoprotein (LDL) as an important atherogenic factor. This study examined the clinical**

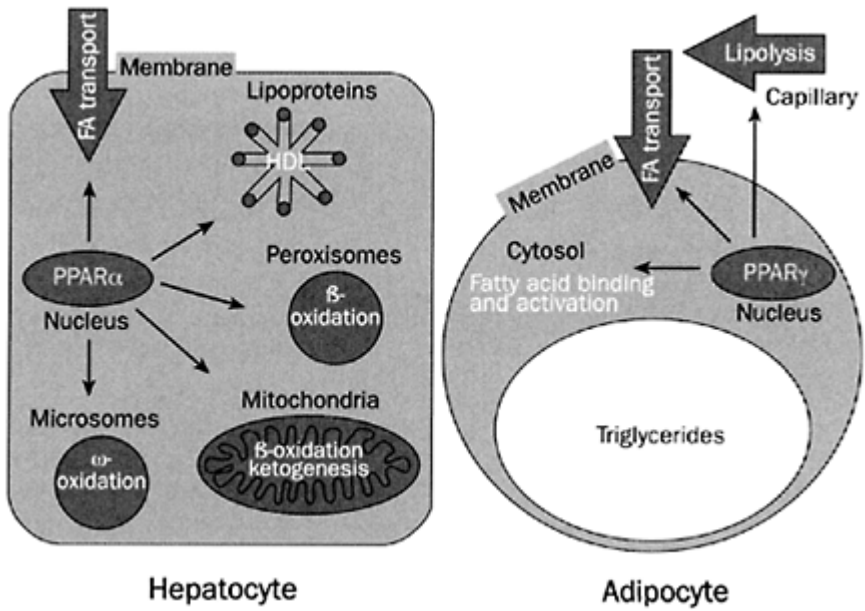

Fig. 16.3 Action of PPARα and PPARγ at the cellular level. PPARα stimulates oxidation of fatty acids in various organelles, such as mitochondria, peroxisomes and microsomes. It also stimulates uptake of fatty acids and synthesis of lipoproteins. PPARγ stimulates lipolysis of circulating triglycerides and the subsequent uptake of fatty acids into the adipose cell. It also stimulates binding and activation of fatty

acids in the cytosol, events that are required for synthesis of triglycerides. FA, fatty acid; HDL, high density lipoprotein. Source: Kersten *et al.* (2000).

relevance of circulating oxidized LDL (OxLDL) levels in atherosclerotic disease by an enzyme immunoassay using specific antibodies against OxLDL and apolipoprotein (apo)B.

I**NTERPRETATION.** The results suggest that circulating OxLDL may be a biochemical risk marker for coronary heart disease (CHD). Plasma OxLDL levels were significantly higher in patients with CHD than in controls but were not associated with age, sex, total cholesterol or apoB levels in normal control subjects. Circulating levels of OxLDL, as measured by an enzyme immunoassay using a monoclonal antibody directed against modified apoB are higher in patients with CHD than in controls or diabetics without CHD (see Fig. 16.4).

Comment

The oxidation theory of atherosclerosis is in need of *in vivo* evidence, especially with the relative failure of antioxidants to prevent atherosclerosis in large-scale clinical trials (see previous chapter). It has proven very difficult to develop a method to measure oxidative stress *in vivo* and most studies have to rely on indirect methods

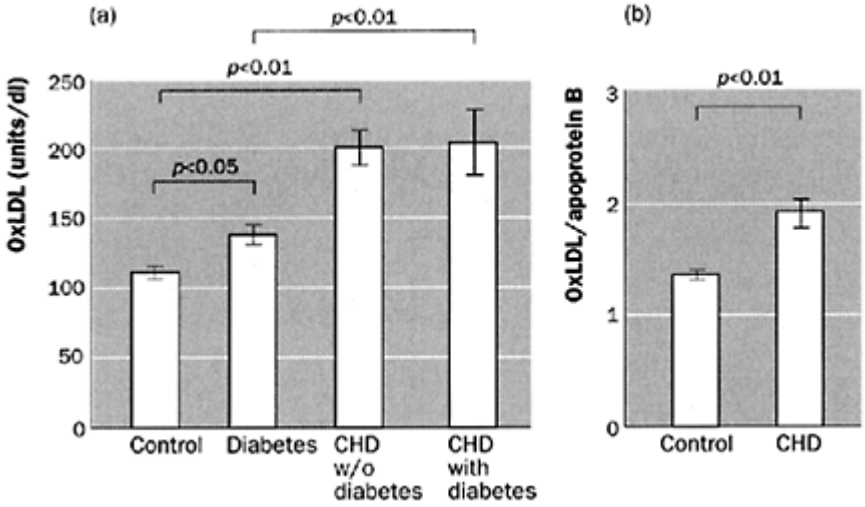

Fig. 16.4 Plasma OxLDL levels in patients. (a) Plasma OxLDL levels in control subjects, in patients with diabetes rnellitus, and in patients with CHD. OxLDL levels were significantly higher in patients with diabetes mellitus than in control subjects. OxLDL levels were significantly higher in patients with CHD without diabetes mellitus

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than in control subjects. OxLDL levels were significantly higher in patients with both CHD and diabetes than in patients with diabetes mellitus alone. (b) OxLDL/apoB ratios were higher in patients with CHD than in control subjects. Data are presented as mean±SEM. The OxLDL level of plasma samples having equivalent reactivity to 1 μg/dl of 9-CHO PC-LDL in each assay was defined as 1 U/dl. Source: Toshima *et al.* (2000).

such as the presence of antibodies to OxLDL. These, however, have provided inconsistent results |**1**|. Several groups have been active in developing methods to detect OxLDL by raising antibodies to modified apoB |**2–4**|. Although these are also indirect methods, they may be more consistent than measuring OxLDL auto-antibodies. Time will tell.

Genetics of coronary heart disease: current knowledge and research principles.
B R Winkelmann, J Hager, W E Kraus, *et al. Am Heart J* 2000; **140**: 11–26.

B**ACKGROUND. CHD is the leading cause of death in Western industrialised countries. It is often viewed as a disease of the elderly, yet the age of onset for its clinical manifestations is younger than 65 years in nearly half the persons with CHD, and approximately 5% of these are younger than 40 years. Although modifiable risk factors (hypercholesterolemia, smoking, hypertension, diabetes, sedentary lifestyle)**

Table 16.1 Genetic variation in lipid metabolism

Characteristics of study population		**Study subjects (case/control)**	**Study type**	**Frequency of variant (%) (case/control)** *	**Assoociation of intermediat clinical phenotype**
Cases	**Control group**				
Apo E polymorphism†					
Fatal CHD+ non-fatal MI	Matched controls	Men, 35–57 y (207/412)	Prospective; nested case-control (MRFIT study subset)	apo ε4/3+4/4 (32/23)	Yes/yes
Non-fatal MI	Age-	Men and	Single center	apo ε4/3+4/4	Yes/yes

(MI survivors scheduled for angiography)	matched controls (healthy factory employees from PROCAM)	women (570/624)	case-control	(26/22)	
CHD	Healthy population	Men and women 40–77 y (189/1761)	Case-control Framingham OffspringSpouse study	apo ε4/3+4/4 (29/21)	Yes/yes
LPL polymorphism‡					
CHD (population based)	No CHD (population based)	Men (418/844)	Population-based cohort (Caerphilly)	(15/19)	Yes/yes (protective)
LPL polymorphisms§					
CHD (CHD patients scheduled for angiography)	Population registry (Copenhagen City Heart Study)	Men and women, 20–80 y (948/9214)	Single-center case-control	291 Ser (4.7/5.2, men; 8.5/4.7, women)	Yes/no (men) yes/yes (women)
Parental history of MI<55 y	No parental history of MI	Men and women 18–26 y (students) (508/925; 553/1055)	Multicenter case-control (EARS I/II)	291 Ser (2.5/3.4), 9Asn (4.5/4.1)	Yes/no yes/no

Data applied with permission from Winkelmann BR, Hager J. Genetic variation in coronary heart disease and myocardial infarction: methodological overview and clinical evidence. *Pharmacogenomics* 2000; **1**:73–94 Limitations of space do not permit all studies discussed in this overview to be listed in the tables. Apo, Apoliprotem.

*Prevalence of mutation carrier defined as carrier of the less frequent allele (heterozygous+homozygous) in case of apo ε (E4/3+E4/4 phenotype).

†Apo E polymorphism: Cys112Arg and Arg158Cys; E2:112Cys158Cys; E3:112Cys158Arg; E4:112Arg158Arg.

‡LPL polymorphism: LPL Ser447Ter. §LPL polymorphisms: LPL Asn291Ser; LPL Asp9Asn. Source: Winkelmann *et al.* (2000)

and non-modifiable risk factors (age, male sex, family history of CHD), have been identified in epidemiologic studies since 1948, the relative contribution of both types of risk factors in causing coronary atherosclerosis remains poorly understood. Like most other common

chronic diseases, CHD is multifactorial and has an important hereditary component. Its complex pathophysiologic characteristics likely involve an interplay of genetic variations of molecular and biochemical pathways and their interactions with environmental factors. On a genetic level, functional allelic variations in human beings likely contribute to an individual's susceptibility to CHD, the manifestation of the disease and its prognosis.

I**NTERPRETATION.** In this review, genes that may play a role in the development and progression of athersclerosis are categorised according to the presumed major mechanisms of atherosclerosis, lipid metabolism, the coagulation cascade, smooth muscle proliferation and vascular growth, inflammatory phenomena in the arterial wall, oxidative/antioxidative balance in the vasculature, glucose/insulin metabolism and insulin resistance, other metabolic factors (ie homocysteine), and hypertension.

This overview summarizes the current knowledge of the genetic associations with CHD. It critically appraises the methods used in determining these associations and suggests guidelines or future studies. Although a comprehensive catalogue of studies and genes identified to date cannot be provided in this article, it presents findings from representative studies that have examined potentially important genes in populations sufficiently large to have possible relevance to our understanding of CHD.

Comment

For those thinking of embarking on genetic studies this review provides an excellent update of current knowledge (see Table 16.1). For these people and for those of us who think we can conduct genetic studies, it provides (too much) evidence of where we can go wrong (see Tables 16.2 and 16.3). From the available evidence in the literature, it would appear that many laboratories conducting research into the genetics of lipids and atherosclerosis, should read this review (written by a worldwide expert panel) and take its warnings to heart.

Table 16.2 Reasons for unreliable findings in case-control allelic association studies

High rate of false-positive findings related to multiple comparisons and positive study publication bias
High rate of false-negative findings related to low power, small sample size, bad phenotype
Failure to identify candidate genes involved based on incomplete pathophysiologic understanding
Heterogeneity among study populations (population stratification/admixture)
Selection bias (bias in study subject inclusion)
Deviation from Hardy-Weinberg equilibrium in the control poulation

Quality of genotypic data (e.g., error rate of genotyping >1%)

Quality of phenotypic and end point assessment (e.g., inaccurate classification of disease/control populations or clinical end points)

Source: Winkelmann *et al.* (2000).

Table 16.3 Evidence supporting a significant finding in a case-control allelic association study

Polymorphism located in a coding region and leads to an amino acid replacement of coded protein

Biologic plausibility: functional significance of polymorphism (e.g., affects activity or level of gene product)

Strength of the association (large sample size, high power)

Internal consistency (e.g., positive family history for CHD in carriers)

External consistency of association among different populations (replication of findings)

Specificity of association both in family- and population-based studies
Gene dosing effect

Source: Winkelmann *et al.* (2000).

References

1. Ylä-Herttuala S. Is oxidised low-density lipoprotein present *in vivo. Curr Opin Lipidol* 1998; **9**:337–44.
2. Holvoet P, Vanlaecke J, Janssen S, Van de Werf F, Collen D. Oxidised LDL and malondialdehyde-modified LDL in patients with acute coronary symptoms and stable coronary artery disease. *Circulation* 1998; **98**:1487–94.
3. Palinski W, Horkko S, Miller E, Steinbrecher UP, Powell HC, Curtiss LK, Witztum JL. Cloning of monoclonal auto-antibodies to epitopes of oxidised lipoproteins from apolipoprotein E-deficient mice: demonstration of epitopes of oxidised low-density lipoprotein in human plasma. *J Clin Invest* 1996; **98**:800–14.
4. Itabe H, Takeshima E, Iwasaki H, Kimura J, Yoshida Y, Imanaka T, Takano T. A monoclonal antibody against oxidised lipoprotein recognizes foam cells in atherosclerotic lesions. *J Biol Chem* 1994; **269**:15274–9.

Overall discussion

Lipids and atherosclerosis is a very wide field as (hopefully) illustrated in this chapter. Research into the role of OxLDL and atherosclerosis will continue apace. Research into the genetic determinants of atherosclerosis will increase, due to the relative ease of measuring genetic polymorphisms in most laboratories. However, the majority of studies

will simply confuse the issue because they will fail to take into account the effect of genotype on phenotype, be too small or have any of the other flaws detailed in the last paper. It is my view that this field is too diverse to produce a critical discussion. Therefore, the papers chosen were those that reflect my interests or those that excited me. However, I will go so far as to make some predictions: three of the chapters in this section, i.e. non-lipid lowering effects of statins, how HDL protects against atherosclerosis and pharmacological regulation of HDL and the role of dietary antioxidants, will continue to take centre stage. Some of these developments will be driven by the pharmaceutical industry while others, e.g. ‘eating more tomatoes and olive oil is good for you’, will be driven entirely by basic research. These aspects of research will be driven independently when a fusion of the two approaches may better serve those patients at risk of developing CHD.

Part V
Current and future trends

17
Current and future trends in lipid-regulating therapy

Summary

Evidence of clinical benefit from lowering low-density lipoprotein (LDL) cholesterol has been the main driving force behind the development of new lipid-regulating drugs during the past two decades. This conclusion is supported by the fact that most of the compounds discussed in this review are either bile acid sequestrants, statins or inhibitors of cholesterol absorption, all of which primarily lower LDL cholesterol. However, the potential benefit of raising high-density lipoprotein (HDL) cholesterol has gained support both from experimental studies with cholesterol ester transfer protein (CETP) inhibitors and from the results of recent fibrate trials. It seems likely that in future the emphasis will gradually shift from the development of LDL-lowering drugs to compounds that promote the formation and turnover of HDL.

Introduction

In the light of the positive results of five major statin trials, the therapeutic emphasis continues to be on LDL lowering. This is reflected in current guidelines on the prevention of coronary heart disease (CHD), which advocate drug therapy to lower LDL cholesterol in patients with CHD or at high risk who remain hypercholesterolaemic despite dietary intervention. Target levels of LDL cholesterol to be achieved are lower in the latest US guidelines |**1**| than in their most recent British and European counterparts |**2,3**|.

Data from the statin trials suggest that reductions in CHD events correlate better with the percentage decrease in LDL cholesterol than with its absolute level on treatment. The greatest reduction in CHD was observed in the Scandinavian Simvastatin Survival Study (4S) |**4**|, reflecting the fact that simvastatin decreased LDL cholesterol more than did either pravastatin in the West of Scotland Coronary Prevention Study (WOSCOPS), Cholesterol and Recurrent Events (CARE) and Long-term Intervention with Pravastatin in Ischaemic Disease Study (LIPID) |**5–7**| or lovastatin in the Air Force/Texas Coronary Atherosclerosis Prevention Study (AFCAPS/TexCAPS) |**8**|. Trials aimed at testing ‘the lower the LDL, the better’ hypothesis include TNT (Treating to New Targets), which will compare the effects of atorvastatin 10 mg with 80 mg daily, and IDEAL (Incremental Decrease in Endpoints through Aggressive Lipid-lowering), which compares atorvastatin 80 mg with simvastatin 20–40 mg daily; the primary end-point in both trials is CHD death and non-fatal myocardial infarction. Their outcome may help explain whether the fact that the majority of treated patients in the statin trials continued to sustain CHD

events reflects the need to achieve greater decreases in LDL cholesterol or whether it indicates an additional requirement to correct other risk factors, such as raised triglycerides (TGs) and low HDL cholesterol.

The importance of dyslipidaemia as a risk factor for macrovascular disease in diabetes is increasingly recognized. *Post hoc* analysis of the statin trials showed significant decreases in CHD among the subgroups with type 2 diabetes, ranging from 25% in CARE |**9**| to 42% in 4S |**10**|. Interestingly, a 24% decrease in CHD events was seen also in diabetics in the Veterans Affairs High Density Lipoprotein Cholesterol Intervention Trial (VAHIT) |**11**|, where the drug used was the fibrate, gemfibrozil. This illustrates that LDL lowering is not an essential prerequisite for reducing the risk of CHD in that LDL cholesterol levels were unaffected by gemfibrozil, which instead caused a 30% decrease in TG and 6% increase in HDL cholesterol. The results of the Diabetes Atherosclerosis Intervention Study (DAIS) suggest that the beneficial effects of fibrates are mediated mainly via decreased progression of coronary atherosclerosis |**12**|, like those of statins.

The results of these trials imply that the ideal lipid-regulating drug would reduce both LDL cholesterol and TGs, as well as raise HDL cholesterol. Usually, these objectives can only be achieved by combining two or more lipid-regulating agents, which may increase the risk of side-effects.

New additions to existing classes of lipid-regulating drugs

Nicotinic acid (niacin)

Nicotinic acid has been in use for many years and comes closest to achieving the objectives referred to above, but, unfortunately, its usefulness is limited by side-effects. Goldberg *et al.* |**13**| recently assessed the efficacy and safety of an extended-release form of nicotinic acid (Niaspan) in hyperlipidaemic patients. At the maximum recommended dose of 2 g daily, decreases in LDL cholesterol, TGs and lipoprotein(a) averaged 17%, 35% and 24%, respectively, whereas HDL cholesterol increased by 26%. Although 30% of those randomized to Niaspan dropped out because of side-effects, the frequency of abnormal liver function tests was similar to that on placebo. Thus for those who can tolerate it, extended-release nicotinic acid offers an equally effective but safer means of treating dyslipidaemia than sustained-release preparations, which may cause liver damage.

Bile acid sequestrants

Originally known as anion-exchange resins these compounds have been used to treat hypercholesterolaemia for the past 35 years. However, new compounds continue to be developed with the aim of improving LDL-lowering efficacy and compliance while retaining the safety conferred by non-absorbability.

Colestimide (MCI-196) has been undergoing clinical trials for several years and was recently licensed in Japan. It is administered in the form of granules or 500 mg tablets and a recent report showed that 3 g daily reduced LDL cholesterol by 22% and increased

HDL cholesterol by 8% |**14**|. Side-effects occurred in 23% of patients, mainly gastrointestinal. Thus despite its lower dose colestimide has similar disadvantages to existing compounds, such as colestyramine and colestipol.

Colesevelam hydrochloride (cholestagel) is a non-absorbed cross-linked and alkylated polyamine. Doses of ≥1.5g twice daily administered in capsules to hypercholesterolaemic patients resulted in a reduction in LDL cholesterol of 19% |**15**|. Gastrointestinal side-effects occurred in about 25% of patients but were relatively mild and compliance was good. However, this was only a short-term study and longer ones are needed before the therapeutic potential of this compound can be properly assessed.

3-hydroxy-3-methylglutaryl coenzyme A (HMG CoA) reductase inhibitors

Popularly known as statins, these compounds continue to dominate the lipid-regulating drug market. This reflects not only their proven efficacy in lowering LDL cholesterol and reducing CHD events but also their good tolerability and excellent safety record.

NK-104 is a new, synthetic HMG CoA reductase inhibitor. Preliminary data suggest that it has the advantage of undergoing little if any metabolism via the cytochrome P450 pathway, which should minimize drug interactions. A study of the efficacy of NK-104 was undertaken recently in 30 patients with familial hypercholesterolaemia treated with 2 mg/day for 8 weeks and then 4 mg/day for a further 8 weeks |**16**|. LDL cholesterol decreased by 40% and 48%, respectively, and there was a 23% decrease in TG on the higher dose. HDL cholesterol did not change but there was a significant increase in apolipoprotein (apo)A-I at both dose levels. No adverse events were recorded during the study nor were there any significant rises in liver or muscle enzymes.

Rosuvastatin (ZD-4522), the latest addition to the statin range, is a synthetic compound that may prove to be even more effective in lowering LDL than ator-vastatin. The results of a randomized, placebo-controlled dose ranging trial suggest that comparable reductions in LDL cholesterol were achieved by rosuvastatin 2.5–5 mg daily and open label atorvastatin 10 mg daily |**17**|. When given in a dose of 80 mg daily rosuvastatin reduced LDL cholesterol by 65%. In addition to lowering LDL, it decreases TGs by 10–35% and raises HDL cholesterol by 9–14%. The drug was well tolerated and there was no increase in adverse events or biochemical abnormalities compared with placebo. Preclinical and other studies indicate a high degree of hepatic selectivity and a low propensity for metabolism via the cyto-chrome P450 3A4 pathway. An application for a licence is pending.

Therapeutic potential of functional foods containing plant sterols and stanols

It has long been known that plant sterols inhibit the absorption of cholesterol, with which they are closely related structurally, and more recently plant stanols have been shown to do the same. The main plant sterols, sitosterol and campesterol, can be readily converted to their stanol counterparts, sitostanol and campestanol, by hydrogenation. All these compounds compete with cholesterol for incorporation into mixed micelles but the

limited lipid solubility of free sterols and stanols makes it difficult to dissolve them in fat spreads in high enough concentrations to be effective. This can be overcome by esterifying them with long chain fatty acids, which increases their lipid solubility and facilitates their incorporation into foods. The ester bond subsequently undergoes enzymatic hydrolysis within the intestinal lumen, releasing free sterol or stanol.

Numerous studies have shown that plant stanol ester intakes in the region of 2 g/day (expressed as free stanol) achieve reductions in LDL cholesterol of 10–15%. In the North Karelia study moderately hypercholesterolaemic subjects were randomized to receive margarine without or with added sitostanol ester 2.6 g daily |**18**|. After 1 year, total and LDL cholesterol decreased by 10% and 13%, respectively, compared with placebo margarine: this would be expected to reduce the risk of CHD by 25% |**19**|.

Plant stanol esters are marketed as Benecol in various forms of food, including margarine, cream cheese, yoghurt and cereal bars. As stanols are virtually unabsorbed side-effects are minimal, apart from a moderate decrease in plasma levels of β-carotene without any decrease in vitamin A. Similar decreases in LDL cholesterol and β-carotene occur with comparable doses of margarine containing plant sterol esters (Flora Proactiv); however, plant sterols are absorbed to some extent, which raises their plasma levels. The comparable LDL-lowering ability of equivalent amounts of plant sterol and stanol esters reflects the fact that they induce similar decreases in cholesterol absorption and compensatory increases in cholesterol synthesis.

Plant stanol esters have been shown to be an effective and safe means of lowering LDL in several categories of subjects, including children and adults with familial hypercholesterolaemia, diabetics and postmenopausal women with CHD. Any decrease in LDL cholesterol is additional to that achieved by standard lipid-lowering diet and drug therapy. For example, a further 10% decrease in LDL cholesterol was observed when plant stanol ester was given in conjunction with statins |**20**|.

A subgroup analysis of the Finnish participants in the 4S trial showed that those with the highest baseline levels of plasma cholestanol, an index of cholesterol absorption, responded to simvastatin significantly less well than subjects with the lowest levels of cholestanol |**21**|. As poor responders to statins not only have higher rates of cholesterol absorption but also lower rates of synthesis than good re-ponders they make an ideal target for stanol esters, which block cholesterol absorption and thereby upregulate cholesterol synthesis |**22**|. This non-pharmacological approach to treating dyslipidaemia provides a useful adjunct to conventional diet and drug therapy, especially in the primary prevention of CHD.

Products in the pipeline

Under this heading come drugs in various stages of development that have a novel mechanism of action.

Cholesterol absorption inhibitors

A number of naturally-occurring plant derivatives are known to inhibit cholesterol

absorption by impeding the incorporation of cholesterol into mixed micelles, as discussed above. Recently, it has been shown that certain synthetic azetidinones selectively block cholesterol absorption by a mechanism that does not impair micellar solubilization of cholesterol but which appears to inhibit the uptake of cholesterol from micelles by the brush border membrane of enterocytes.

Ezetimibe is one such compound and a randomized, double-blind, placebo-controlled trial in hypercholesterolaemic subjects compared the effect of doses ranging from 0.25 mg to 10 mg once daily over a period of 12 weeks |**23**|. Reductions in LDL cholesterol ranged from 10 to 19% and were accompanied by small but significant increases in HDL cholesterol without any change in TG; the safety profile was similar to placebo. When given together the effects of simvastatin 10 mg and ezetimibe 10 mg daily were additive, resulting in a decrease in LDL cholesterol of 52% |**24**|. The latter effect is comparable with what can be achieved by simvastatin 80 mg daily given alone and combination therapy with a statin may prove to be the most effective way in which this drug will eventually be used. Such a combination may be especially useful in poor responders to statins in whom HMG CoA reductase is downregulated secondary to excessive absorption of cholesterol; as is reported to occur in subjects with an apoE4 allele |**25**|. Inhibition of cholesterol absorption should help counteract this phenomenon and upregulate HMG CoA reductase, thereby rendering it more sensitive to pharmacological inhibition by a statin |**22**|.

Acylcholesterol:acyltransferase (ACAT) inhibitors

Strictly speaking inhibitors of ACAT are not a new class of lipid-regulating drug and until recently it was considered that these compounds acted either by inhibiting intestinal ACAT, thereby reducing cholesterol absorption |**26**|, or by inhibiting hepatic ACAT, thereby decreasing secretion of apoB-containing lipoproteins by the liver |**27**|. However, a new mechanism of action has now emerged that seems independent of any lipid-lowering effect, as described below.

Avasimibe (CI-1011) has been shown to inhibit ACAT in macrophages by Bocan *et al.* |**28**| who made New Zealand white rabbits hypercholesterolaemic and then fed them on a cholesterol-free, high fat diet, which decreased the degree of hypercholesterolaemia. After 17 weeks some rabbits were killed and necropsied, the remainder continuing on the cholesterol-free, high fat diet with or without added avasimibe 25 mg/kg for a further 5 weeks, after which they too were killed and necropsied. At the start of the study chronic endothelial damage had been surgically induced in the aorta but despite the fact that serum cholesterol levels were similar to rabbits not on avasimibe, the extent of atherosclerosis in the aorta was significantly reduced in animals receiving the compound. *In vitro* studies showed that avasimibe inhibited ACAT in monocyte-macrophages and decreased their cholesterol ester content. This study suggests that avasimibe exerts an antiatherosclerotic effect, which is independent of any cholesterol-lowering properties it may possess |**29**|. Clinical trials are now underway.

Cholesterol ester transfer protein inhibitors

The rationale for inhibiting CETP is that this will result in an increase in HDL cholesterol levels in plasma. However, there is evidence that genetic deficiency of CETP predisposes to atherosclerosis rather than protecting against it, as occurs when HDL is raised by other means. Whether pharmacological inhibition of CETP would be beneficial has now been clarified by a recent study of a novel compound which forms disulphide bonds with CETP and inhibits its activity in plasma.

JTT-705 is an orally bioavailable thioester which produces 95% inhibition of CETP in rabbits |**30**|. Oral administration of 255 mg/kg to rabbits with diet-induced hypercholesterolaemia resulted in a 90% increase in HDL cholesterol and a 40–50% decrease in non-HDL cholesterol. Simvastatin given in one hundredth the dose of JTT-705 increased HDL cholesterol by 28% and decreased non-HDL cholesterol by up to 70%. After 6 months both drugs resulted in 70–80% decreases in the extent of aortic atherosclerosis as compared with control rabbits. Preliminary data in humans showed that a dose of JTT-705 of 900 mg/day for 2 weeks resulted in a 40–45% increase in HDL cholesterol and a 15–20% decrease in LDL cholesterol. These findings suggest a new approach to the management of subjects with decreased levels of HDL cholesterol.

Future developments

A diagrammatic representation of the known metabolic targets of lipid-regulating agents is shown in Fig. 17.1. These include all the major enzymes, receptors and transporters involved in the absorption, synthesis and esterification of cholesterol, as well as its secretion into and uptake from plasma and also those involved in the

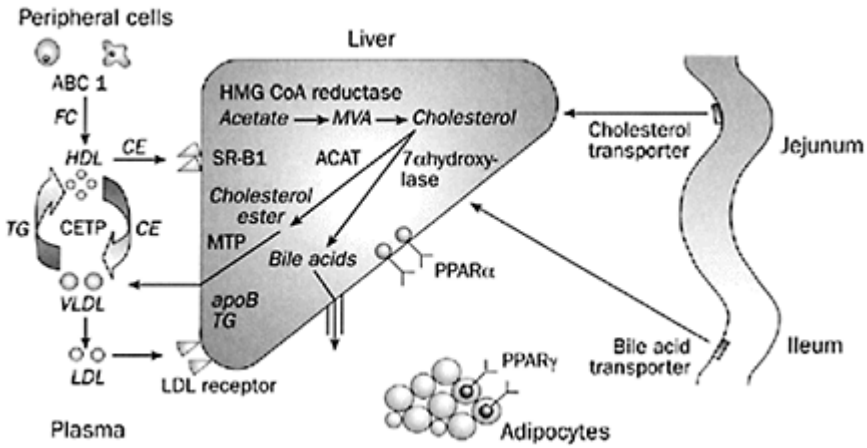

Fig. 17.1 Metabolic targets of lipid-regulating drugs. ABC1, adenosine triphosphate-binding cassette transporter; CE, cholesterol ester; FC, free cholesterol; MTP, microsomal triglyceride transfer protein; MVA, mevalonate; PPAR, peroxisome proliferator activated receptor; SR-B1, type 1, class B scavenger receptor; VLDL, very low density lipoprotein.

synthesis and reabsorption of bile acids. Compounds that influence these pathways mainly affect LDL levels, whereas those that inhibit CETP increase HDL, as has already been discussed. Ligands of the nuclear receptors, peroxisome proliferator activated receptor-α and -γ, which include fibrates and thiazolidenediones, have considerable therapeutic potential |**31**| as too have microsomal TG transfer protein inhibitors |**32**|, although the latter may ultimately prove to be too toxic for clinical use.

Two recent developments, illustrated in Fig. 17.1, provide major insights into the pathophysiology of HDL. The first is the discovery that SR-B1 mediates the uptake of cholesterol ester from HDL into the liver and thereby regulates HDL cholesterol levels in plasma |**33**|. Overexpression of SR-B1 in the liver of genetically modified mice has been shown to reverse atherosclerosis induced by cholesterol feeding |**34**|. The second is the discovery that Tangier disease is due to mutations of the ABC 1 gene, leading to the realization that ABC 1 normally plays a crucial part in the efflux of free cholesterol from cells into plasma where it is incorporated into HDL |**35**|. These discoveries will undoubtedly stimulate the development of agonists aimed at promoting efflux of cholesterol from peripheral cells to HDL via the ABC 1 pathway, as described recently |**36**|, and enhancing its subsequent removal from plasma via SR-B1. Hence, it seems that David Kritchevsky's exhorta-tion that we turn our attention from atherogenesis to atheroexodus, uttered over 30 years ago |**37**|, is proving to be prophetic.

References

1. Expert Panel on Detection, Evaluation and Treatment of High Cholesterol in Adults. Executive Summary of the Third Report of the National Cholesterol Education Program (NCEP) Expert Panel on detection, evaluation, and treatment of high blood cholesterol in adults (Adult Treatment Panel III). *JAMA* 2001; **285**:2486–97.

2. British Cardiac Society, British Hyperlipidaemia Association And British Hypertension Society. Joint British recommendations on prevention of coronary heart disease in clinical practice. *Heart* 1998; **80**(Suppl. 2): S1–29.

3. Wood D, De Backer G, Faergeman O, Graham I, Mancia G, Pyorala K. Prevention of coronary heart disease in clinical practice: Recommendations of the Second Joint Task Force of European and other Societies on Coronary Prevention. *Atherosclerosis* 1998; **140**:199–270.

4. Scandinavian Simvastatin Survival Study Group. Randomised trial of cholesterol lowering in 4444 patients with coronary heart disease: the Scandinavian Simvastatin Survival Study (4S). *Lancet* 1994; **344**:1383–9.

5. Shepherd J, Cobbe SM, Ford I, Isles CG, Lorimer AR, MacFarlane PW, McKillop JH, Packard CJ. Prevention of coronary heart disease with pravastatin in men with hypercholesterolemia. *N Engl J Med* 1995; **333**:1301–7.

6. Sacks FM, Pfeffer MA, Moye LA, Rouleau JL, Rutherford JD, Cole TG, Brown L, Warnica JW, Arnold JM, Wun CC, Davis BR, Braunwald E. The effect of pravastatin on coronary events after myocardial infarction in patients with average cholesterol levels. *N Engl J Med* 1996; **335**:1001–9.

7. The Long-Term Intervention With Pravastatin In Ischaemic Disease (Lipid) Study Group. Prevention of cardiovascular events and death with pravastatin in patients with coronary heart disease and a broad range of initial cholesterol levels. *N Engl J Med*

1998; **339**:1349–57.

8. Downs JR, Clearfield M, Weis S, Whitney E, Shapiro DR, Beere PA, Langendorfer A, Stein EA, Kruyer W, Gotto Jr AM. Primary prevention of acute coronary events with lovastatin in men and women with average cholesterol levels. *JAMA* 1998; **279**:1615–22.

9. Goldberg RB, Mellies MJ, Sacks FM, Moye LA, Howard BV, Howard WJ, Davis BR, Cole TG, Pfeffer MA, Braunwald E. Cardiovascular events and their reduction with pravastatin in diabetic and glucose-intolerant myocardial infarction survivors with average cholesterol levels: subgroup analyses in the cholesterol and recurrent events (CARE) trial. The Care Investigators. *Circulation* 1998; **98**:2513–19.

10. Haffner SM, Alexander CM, Cook TJ, Boccuzzi SJ, Musliner TA, Pedersen TR, Kjekshus J, Pyorala K. Reduced coronary events in simvastatin-treated patients with coronary heart disease and diabetes or impaired fasting glucose levels: subgroup analyses in the Scandinavian Simvastatin Survival Study. *Arch Intern Med* 1999; **159**:2661–7.

11. Rubins HB, Robins SJ, Collins D, Fye CL, Anderson JW, Elam MB, Faas FH, Linares E, Schaefer EJ, Schectman G, Wilt TJ, Wittes J. Gemfibrozil for the secondary prevention of coronary heart disease in men with low levels of high density lipoprotein cholesterol. *N Engl J Med* 1999; **341**:410–18.

12. Diabetes Atherosclerosis Intervention Study Investigators. Effect of fenofibrate on progression of coronary artery disease in type 2 diabetes: the Diabetes Atherosclerosis Intervention Study, a randomised trial. *Lancet* 2001; **357**:905–10.

13. Goldberg A, Alagona Jr P, Capuzzi DM, Guyton J, Morgan JM, Rodgers J, Sachson R, Samuel P. Multiple-dose efficacy and safety of an extended-release form of niacin in the management of hyperlipidemia. *Am J Cardiol* 2000; **85**:1100–5.

14. Nakaya N, Goto Y. Colestilan: A new bile acid sequestrant resin. A review of its clinical study in hypercholesterolaemia in Japan. *Atherosclerosis* 2000; **151**:134–5.

15. Davidson MH, Dillon MA, Gordon B. Colesevelam hydrochloride (cholestagel): a new, potent bile acid sequestrant associated with a low incidence of gastrointestinal side-effects. *Arch Intern Med* 1999; **159**:1893–900.

16. Kajinami K, Koizumi J, Ueda K, Miyamoto S, Takegoshi T, Mabuchi H. Effects of NK-104, a new hydroxymethylglutaryl-coenzyme reductase inhibitor, on low density lipoprotein cholesterol in heterozygous familial hypercholesterolemia. *Am J Cardiol* 2000; **85**:178–83.

17. Olsson AG, Pears JS, McKellar J, Caplan RJ, Raza A. Pharmacodynamics of new HMG-CoA reductase inhibitor ZD4522 in patients with primary hypercholesterolaemia. *Atherosclerosis* 2000; **151**:39.

18. Miettinen TA, Puska P, Gylling H, Vanhanen H, Vartiainen E. Reduction of serum cholesterol with sitostanol-ester margarine in a mildly hypercholesterolemic population. *N Engl J Med* 1995; **333**:1308–12.

19. Law M. Plant sterol and stanol margarines and health. *BMJ*, 2000; **320**:861–4.

20. Blair SN, Capuzzi DM, Gottlieb SO, Nguyen T, Morgan JM, Cater NB. Incremental reduction of serum total cholesterol and low-density lipoprotein cholesterol with the addition of plant stanol ester-containing spread to statin therapy. *Am J Cardiol* 2000; **86**: 46–52.

21. Miettinen TA, Strandberg TE, Gylling H, for the Finnish Investigators of the Scandinavian Simvastatin Survival Study Group. Noncholesterol sterols and cholesterol lowering by long-term simvastatin treatment in coronary patients. *Arterioscler Thromb Vasc Biol* 2000; **20**:1340–6.

22. Thompson GR. Poor responders to statins: a potential target for stanol esters. *Eur Heart J Suppl* 1999; **1**(Suppl. S): S114–17.
23. Bays H, Drehobl M, Rosenblatt S, *et al.* Low-density lipoprotein cholesterol reduction by SCH 58235 (ezetimibe), a novel inhibitor of intestinal cholesterol absorption in 243 hypercholesterolemic subjects: Results of a dose-response study. *Atherosclerosis* 2000; **151**:135.
24. Kosoglou T, Meyer I, Musiol B, *et al.* Pharmacodynamic interaction between the new selective cholesterol absorption inhibitor ezetimibe and simvastatin. *Atherosclerosis* 2000; **151**:135.
25. Kesaniemi YA, Enholm C, Miettinen TA. Intestinal cholesterol absorption efficiency in man is related to apoprotein E phenotype. *J Clin Invest* 1987; **80**:578–81.
26. Matsubara K, Matsuzawa Y, Jiao S, Kihara S, Takama T, Nakamura T, Tokunaga K, Kubom M, Tarui S. Cholesterol-lowering effect of N-(alpha-methylbenzyl)linoleamide (Melinamide) in cholesterol-fed diabetic rats. *Atherosclerosis* 1988; **72**:199–204.
27. Burnett JR, Wilcox LJ, Telford DE, Kleinstiver SJ, Barrett PH, Huff MW. Inhibition of cholesterol esterification by DuP 128 decreases hepatic apolipoprotein B secretion *in vivo:* effect of dietary fat and cholesterol. *Biochim Biophys Acta* 1998; **1393**:63–79.
28. Bocan TMA, Krause BR, Rosebury WS, Mueller SB, Lu X, Dagle C, Major T, Lathia C, Lee H. The ACAT inhibitor Avasimibe reduces macrophages and matrix metalloproteinase expression in atherosclerotic lesions of hypercholesterolemic rabbits. *Arterioscler Thromb Vasc Biol* 2000; **20**:70–9.
29. Burnett JR, Wilcox LJ, Telford DE, Kleinstiver SJ, Barrett PH, Newton RS, Huff MW. Inhibition of ACAT by avasimibe decreases both VLDL and LDL apolipoprotein B production in miniature pigs. *J Lipid Res* 1999; **40**:1317–27.
30. Okamoto H, Yonemori F, Wakitani K, Maeda K, Shinkai H. A cholesteryl ester transfer protein inhibitor attenuates atherosclerosis in rabbits. *Nature* 2000; **406**:203–7.
31. Li AC, Brown KK, Silvestre MJ, Willson TM, Palinski W, Glass CK. Peroxisome proliferator-activated receptor γ ligands inhibit development of atherosclerosis in LDL receptor-defident mice. *J Clin Invest* 2000; **106**:523–31.
32. Wetterau JR, Gregg RE, Harrity TW, Arbeeny C, Cap M, Connolly F, Chu CH, George RJ, Gordon DA, Jamil H, Jolibois KG, Kunselman LK, Lan SJ, Maccagnan TJ, Ricci B, Yan M, Young D, Chen Y, Fryszman OM, Logan JV, Musial CL, Poss MA, Robl JA, Simpkins LM, Biller SA, *et al.* An MTP inhibitor that normalizes atherogenic lipoprotein levels in WHHL rabbits. *Science* 1998; **282**:751–4.
33. Krieger M: The 'best' of cholesterols, the 'worst' of cholesterols: A tale of two receptors. *Proc Natl Acad Sci USA* 1998; **95**:4077–80.
34. Kozarsky KF, Donahee MH, Glick JM, Krieger M, Rader DJ. Gene transfer and hepatic overexpression of the HDL receptor SR-B1 reduces atherosclerosis in the cholesterolfed LDL receptor-deficient mouse. *Arterioscler Thromb Vasc Biol* 2000; **20** (3): 721–7.
35. Young SG, Fielding CJ. The ABCs of cholesterol efflux. *Nat Genet* 1999; **22**:316–18.
36. Repa JJ, Turley SD, Lobacarro JA, Medina J, Li L, Lustig K, Shan B, Heyman RA, Dietschy JM, Mangelsdorf DJ. Regulation of absorption and ABC1-mediated efflux of cholesterol by RXR heterodimers. *Science* 2000; **289**:1524–9.
37. Kritchevsky D. Current concepts in the genesis of the atherosclerotic plaque. In: Brest AN and Moyer JH (eds). *Atherosclerotic Vascular Disease*. London: Butterworths, 1967: 1–7.

Abbreviations

4S	Scandinavian Simvastatin Survival Study
A	alanine
ABCA1	ATP binding cassette A1
ABL	abetalipoproteinaemia
Ach	acetylcholine
ADA	American Diabetes Association
ADH	autosomal dominant type IIa hypercholesterolaemia
ADMIT	Arterial Disease Multiple Intervention Trial
AFCAPS/TexCAPS	Air Force/Texas Coronary Atherosclerosis Prevention Study
AG2R	angiotensin-II type I receptor
AGE	advanced glycation end-products
ALP	atherogenic lipoprotein phenotype
ANOVA	analysis of variance
AOT	antioxidant therapy
Apo	apolipoprotein
ASPEN	Atorvastatin as Prevention of Coronary Heart Disease in Patients with Type 2 Diabetes Mellitus
ATP	adenosine triphosphate
AUC	Areas under the curve
BARI	Bypass Angioplasty Revascularization Investigation
BIP	Bezafibrate Infarction Prevention
CAD	coronary artery disease
CARDS	Collaborative Atorvastatin Diabetes Study
CARE	Cholesterol and Recurrent Events
CETA	cholesteryl ester transfer activity
CETP	cholesteryl ester transfer protein
CHD	coronary heart disease
CI	confidence interval
CK	creatine kinase
COX	cyclo-oxygenase
CPK	creatinine phosphokinase
CRP	C-reactive protein

CVD	cardiovascular disease
DAIS	Diabetes Atherosclerosis Intervention Study
DALI	direct adsorption of lipoproteins
DART	Diet and Reinfarction Trial
DHA	docosahexaenoic acid
DM	diabetes mellitus
DM-FG	diabetes by elevated fasting glucose
DM-Hx	diabetes by clinical history
DSA	dextrane sulphate adsorption
EAS	European Atherosclerosis Society
ECG	electrocardiogram
EF	endothelial dysfunction
EPA	eicosapentaenoic acid
EPISTENT	Evaluation of Platelet IIb/IIIa Inhibitor for Stenting Trial
EVF	endothelial vasodilator function
FATS	Familial Atherosclerosis Treatment Study
FCH	familial combined hyperlipidaemia
FDB	familial defective apoB
FFA	free fatty acids
FH	Familial hypercholesterolaemia
FHBL	familial hypobetalipoproteinaemia
FHD	familial HDL deficiency
FH-NK	North Karelia variant of FH
FHTG	familial hypertriglyceridaemia
FIELD	Fenofibrate Intervention and Event Lowering in Diabetes Trial
FO	fish oil
GISSI	Gruppo Italiano per lo Studio della Sopravvivenza nell'Infarto Miocardico
HAART	highly active antiretroviral therapy
HDL	high-density lipoprotein
HDL-C	HDL cholesterol
HIV	human immimodeficiency virus
HL	hepatic lipase
HMG-CoA	hydroxymethly glutarate coenzyme A
HOPE	Heart Outcomes Prevention Evaluation
HRT	hormone replacement therapy
HTG	hypertriglyceridaemic

IDDM	insulin-dependent diabetes mellitus
IDL	intermediate density lipoprotein
IFG	impaired fasting glucose
IGT	impaired glucose tolerance
IHD	ischaemic heart disease
IMAL	immunoadsorption
LCAT	lecithin: cholesterol acyltransferase
LDL	low-density lipoprotein
LDL-C	LDL cholesterol
LDLPS	LDL particle size
LDL-R	LDL receptor
LDLVE	LDL vitamin E content
LDS	Lipids in Diabetes Study
LFT	liver function tests
LIPID	Long Term Intervention with Pravastatin in Ischaemic Disease
Lpa	lipoproteina
LPL	lipoprotein lipase
MARS	Monitored Atherosclerosis Regression Study
MI	myocardial infarction
MTHFR	methylenetetrahydrofolate reductase
MTP	microsomal triglyceride transfer protein
MTT	meal tolerance test
MVA	mevalonic acid
NA	Nicotinic acid
NASH	non-alcoholic steatohepatitis
NBD	nucleotide binding domain
NCEP	National Cholesterol Education Program
NFG	normal fasting glucose
NHS	National Health Service
NICE	National Institute for Clinical Excellence
NIDDM	non-insulin dependent DM type 2 diabetes
NNT	number-needed-to-treat
oo	olive oil
Ox-LDL	Oxidized LDL
PAI	plasminogen activator inhibitor
PI	protease inhibitors

PON1	paraoxonase
PPD	peak particle diameter
PPL	Post-prandial lipaemia
PROCAM	Prospective Cardiovascular Münster
PUFA	polyunsaturated fatty acids
PV	pravastatin
RD	regulatory domain
RECIFE	Reduction of Cholesterol in Ischaemia and Function of the Endothelium
RH	relative hazard
RNA	ribonucleic acid
RR	relative risk
SMR	standardized mortality ratio
T CHOL	total cholesterol
TD	Tangier disease
TG	triglyceride
TGRL	TG-rich lipoproteins
UK	United Kingdom
UKPDS	UK Prospective Diabetes Study Group
USA	United States of America
V	valine
VAHIT	Veterans Affairs HDL Intervention Trial
VLDL	very low-density lipoprotein
WHO	World Health Organization
WOSCOPS	West of Scotland Coronary Prevention Study

Index of Papers Reviewed

Part I Lipid disorders

nephrotic-range proteinuria have apolipoprotein C and E deficient VLDL1. *Kidney Int* 2000; **58**:1238–46. 82

D Demetriou, A Shabpar, G Bohmig, S Schmaldienst, WH Hörl, B Watschinger. Beneficial effects of atorvastatin in the treatment of hyperlipidemia after renal transplantation. *Wien Klin Wochenschr* 2000; **212**:358–61. 88

J Goodfellow, MF Bellamy, MW Ramsey, CJ Jones, MJ Lewis. Dietary supplementation with marine omega-3 fatty acids improve systemic large artery endothelial function in subjects with hypercholesterolemia. *J Am Coll Cardiol* 2000; **35**:265–70. 68

Gruppo Italiano per lo Studio della Sopravvivenza nell'Infarto Miocardico. Dietary supplementation with n-3 polyunsaturated fatty acids and vitamin E after myocardial infarction: results of the GISSI-Prevenzione trial. *Lancet* 1999; **354**: 447–55. 64

AB Hameed, PP Tummala, TM Goodwin, I Nuno, OR Wani, IS Karaalp, U Elkayam. Unstable angina during pregnancy in two patients with premature coronary atherosclerosis and aortic stenosis in association with familial hypercholesterolemia. *Am J Obstet Gynecol* 2000; **182**:1152–5. 23

AP Heaney, N Sharer, B Rameh, JM Braganza, PN Durrington. Prevention of recurrent pancreatitis in familial lipoprotein lipase deficiency with high-dose antioxidant therapy. *J Clin Endocrinol Metab* 1999; **84**:1203–5. 50

G Hufnagel, C Michel, F Vrtovsnik, G Queffeulou, N Kossari, F Mignon. Effects of atorvastatin on dyslipidaemia in uraemic patients on peritoneal dialysis. *Nephrol Dial Transplant* 2000; **15**:684–8. 79

HC Hsu, YT Lee, MF Chen. Effect of n-3 fatty acids on the composition and binding properties of lipoproteins in hypertriglyceridemic patients. *Am J Clin Nutr* 2000; **71**:28–35. 68

JM Jensen, LU Gerdes, HK Jensen, TM Christiansen, JU Brorholt-Petersen, O Faergeman. Association of coronary heart disease with age-adjusted aortocoronary calcification in patients with familial hypercholesterolaemia. *J Intern Med* 2000; **247**:479–84. 12

M Kawashiri, K Kajinami, A Nohara, K Yagi, A Inazu, J Koizumi, H Mabuchi. Effect of common methylenetetra-hydrofolate reductase gene mutation on coronary artery disease in familial hypercholesterolemia. *Am J Cardiol* 2000; **86**:840–5. 15

H Knobler, A Schattner, T Zhornicki, SD Malnick, D Keter, N Sokolovskaya, Y Lurie, DD Bass. Fatty liver—an additional and treatable feature of the insulin resistance syndrome. *QJM* 1999; **92**:73–9. 61

HG Kraft, A Lingenhel, FJ Raal, M Hohenegger, G Utermann. Lipoprotein(a) in homozygous familial hypercholesterolemia. *Arterioscler Thromb Vasc Biol* 2000; **20**:522–8. 8

A Lennertz, KG Parhofer, W Samtleben, T Bosch. Therapeutic plasma exchange in patients with chylomicronemia syndrome complicated by acute pancreatitis. *Ther Apher* 1999; **3**:227–33. 51

F Lepre, R Rigby, C Hawley, D Saltissi, A Brown, Z Walsh. A double-blind placebo-controlled trial of simvastatin for the treatment of dyslipidemia in renal allograft recipients. *Clin Transplant* 1999; **13**:520–5. 86

A Levin, L Duncan, O Djurdjev, RJ Shapiro, J Frohlich, A Belanger, R Dumas, S Ross. A randomized placebo-controlled doubleblind trial of lipid-lowering strategies in patients with renal insufftciency: diet modification with or without fenofibrate. *Clin Nephrol* 2000; **53**:140–6. 80

M Lopez-Santamaria, L Migliazza, M Gamez, J Murcia, M Diaz-Gonzalez, C Camarena. Liver transplantation in patients with homozygotic familial hypercholesterolemia previously treated by end-to-side portocaval shunt and ileal bypass. *J Pediatr Surg* 2000; **35**:630–3. 25

B Lundahl, TP Leren, L Ose, A Hamsten, F Karpe. A functional polymorphism in the promoter region of the microsomal triglyceride transfer protein (MTP -493G/T) influences lipoprotein phenotype in familial hypercholesterolemia. *Arterioscler Thromb Vasc Biol* 2000; **20**:1784–8. 11

hyperlipoproteinemia. *J Lipid Res* 2000; **41**: 727–33. 28
S Schmaldienst, S Banyai, TM Stulnig, G Heinz, M Jansen, WH Horl. Prospective randomised cross-over comparison of three LDL-apheresis systems in statin pretreated patients with familial hypercholesterolaemia. *Atherosclerosis* 2000; **151**:493–9. 25
Scientific Steering Committee on behalf of the Simon Broome Register Group. Mortality in treated heterozygous familial hypercholesterolaemia: implications for clinical management. *Atherosclerosis* 1999; **142**:105–12. 4
EJ Sijbrands, RG Westendorp, M Paola Lombardi, LM Havekes, RR Frants, JJ Kastelein, AH Smelt. Additional risk factors influence excess mortality in heterozygous familial hypercholesterolaemia. *Atherosclerosis* 2000; **149**:421–5. 6
TJ Smilde, FW van den Berkmortel, H Wollersheim, TM Christiansen, JU Brorholt-Petersen, O Faergeman. The effect of cholesterol lowering on carotid and femoral artery wall stiffness and thickness in patients with familial hypercholesterolaemia. *Eur J Clin Invest* 2000; **30**:473–80. 13
AF Stalenhoef, J de Graaf, ME Wittekoek, SJ Bredie, PN Demacker, JJ Kastelein. The effect of concentrated n-3 fatty acids versus gemfibrozil on plasma lipoproteins, low density lipoprotein heterogeneity and oxidizability in patients with hypertriglyceridemia. *Atherosclerosis* 2000; **153**:129–38. 66
P Stenvinkel, A Alvestrand, B Angelin, M Eriksson. LDL-apheresis in patients with nephrotic syndrome: effects on serum albumin and urinary albumin excretion. *Eur J Clin Invest* 2000; **30**:866–70. 84
P Tarugi, A Lonardo, G Ballarini, L Erspamer, E Tondelli, S Bertolini, S Calandra. A study of fatty liver disease and plasma lipoproteins in a kindred with familial hypobetalipoproteinemia due to a novel truncated form of apolipoprotein B (APO B-54.5). *J Hepatol* 2000; **33**:361–70. 94
JC Thomas, MF Lopes-Virella, VE Del Bene, JD Cerveny, KB Taylor, LS McWhorter, NC Buttemeier. Use of fenofibrate in the management of protease inhibitor-associated lipid abnormalities. *Pharmacotherapy* 2000; **20**:727–34. 58
S Tonstad, I Hjermann. Cardiovascular risk factors and testing of relatives amongst patients with familial hyperlipidaemia one decade after a clinical trial. *J Intern Med* 2000; **248**:111–18. 2
RD Toto, SM Grundy, GL Vega. Pravastatin treatment of very low density, intermediate density and low density lipoproteins in hypercholesterolemia and combined hyperlipidemia secondary to the nephrotic syndrome. *Am J Nephrol* 2000; **20**:12–17. 83
AJ van Oostrom, M Castro Cabezas, J Ribalta, L Masana, TB Twickler, TA Remijnse, DW Erkelens. Diurnal triglyceride profiles in healthy normolipidemic male subjects are associated to insulin sensitivity, body composition and diet. *Eur J Clin Invest* 2000; **30**:964–71. 35
MC Verhaar, RM Wever, JJ Kastelein, D van Loon, S Milstien, HA Koomans, TJ Rabelink. Effects of oral folic acid supplementation on endothelial function in familial hypercholesterolemia. A randomized placebo-controlled trial. *Circulation* 1999; **100**:335–8. 17
AF Vuorio, H Gylling, H Turtola, K Kontula, P Ketonen, TA Miettinen. Stanol ester margarine alone and with simvastatin lowers serum cholesterol in families with familial hypercholesterolemia caused by the FH-North Karelia mutation. *Arterioscler Thromb Vasc Biol* 2000; **20**: 500–6. 20
AS Wierzbicki, M Lambert-Hammill, PJ Lumb, MA Crook. Renin-angiotensin system polymorphisms and coronary events in familial hypercholesterolemia. *Hypertension* 2000; **36**:808–12. 7
AS Wierzbicki, PJ Lumb, G Chik, MA Crook. Comparison of therapy with simvastatin 80 mg and atorvastatin 80 mg in patients with familial hypercholesterolaemia. *Int J Clin Pract* 1999; **53**: 609–11. 19
A Zambon, JE Hokanson, BG Brown, JD Brunzell. Evidence for a new pathophysiological mechanism for coronary artery disease regression: hepatic lipase-mediated changes in LDL density.

results of the GISSI-Prevenzione trial. *Lancet* 1999; **354**: 447–55. 125
AP Heaney, N Sharer, B Rameh, JM Braganza, PN Durrington. Prevention of recurrent pancreatitis in familial lipoprotein lipase deficiency with high-dose antioxidant therapy. *J Clin Endocrinol Metab* 1999; **84**:1203–5. 128
The Heart Outcomes Prevention Evaluation Study Investigators. Vitamin E supplementation and cardiovascular events in high-risk patients. *N Engl J Med* 2000; **342**:154–60. 125
G Hufnagel, C Michel, F Vrtovsnik, G Queffeulou, N Kossari, F Mignon. Effects of atorvastatin on dyslipidaemia in uraemic patients on peritoneal dialysis. *Nephrol Dial Transplant* 2000; **15**:684–8. 136
Israeli Society for Prevention of Heart Attacks. Secondary prevention by raising HDL cholesterol and reducing triglycerides in patients with coronary artery disease: the Bezafibrate Infarction Prevention (BIP) study. *Circulation* 2000; **102**:21–7. 122
M Jansen, S Banyai, S Schmaldienst, A Goldammer, M Rohac, WH Hörl, K Derfler. Direct adsorption of lipoproteins (DALI) from whole blood: first long-term clinical experience with a new LDL-apheresis system for the treatment of familial hypercholesterolaemia. *Wien Klin Wochenschr* 2000; **112**:61–9. 132
M Kawasuji, N Sakakibara, S Fujii, T Yasuda, Y Watanabe. Coronary artery bypass surgery with arterial grafts in familial hypercholesterolemia. *J Thorac Cardiovasc Surg* 2000; **119**:1008–14. 129
A Keogh, P Macdonald, A Kaan, C Aboyoun, P Spratt, J Mundy. Efficacy and safety of pravastatin *vs* simvastatin after cardiac transplantation. *J Heart Lung Transplant* 2000; **19**:529–37. 117
KR Kulkarni, JH Markovitz, NC Nanda, JP Segrest. Increased prevalence of smaller and denser LDL particles in Asian Indians. *Arterioscler Thromb Vasc Biol* 1999; **19(11)**: 2749–55. 151
B Lamarche, I Lemieux, JP Despres. The small, dense LDL phenotype and the risk of coronary heart disease: epidemiology, patho-physiology and therapeutic aspects. *Diabetes Metab* 1999; **25**:199–211. 152
D Marks, D Wonderling, M Thorogood, H Lambert, SE Humphries, HA Neil. Screening for hypercholesterolaemia versus case finding for familial hypercholesterolaemia: a systematic review and cost-effectiveness analysis. *Health Technol Assess* 2000; **4**:1–123. 149
TA Mori, V Burke, IB Puddey, GF Watts, DN O'Neal, JD Best, LJ Beilin. Purified eicosapentaenoic and docosahexaenoic acids have differential effects on serum lipids and lipoproteins, LDL particle size, glucose, and insulin in mildly hyperlipidemic men. *Am J Clin Nutr* 2000; **71**: 1085–94. 139
A Nordoy, KH Bonaa, PM Sandset, JB Hansen, H Nilsen. Effect of omega-3 fatty acids and simvastatin on hemostatic risk factors and postprandial hyperlipemia in patients with combined hyperlipemia. *Arterioscler Thromb Vasc Biol* 2000; **20**: 259–65. 140
H Okamoto, F Yonemori, K Wakitani, T Minowa, K Maeda, H Shinkai. A cholesteryl ester transfer protein inhibitor attenuates atherosclerosis in rabbits. *Nature* 2000; **406(6792)**: 203–7. 127
S Parthasarathy, N Santanam, S Ramachandran, O Meilhac. Oxidants and antioxidants in atherogenesis. An appraisal. *J Lipid Res* 1999; **40**(12): 2143–57. 126
M Pignone, C Phillips, C Mulrow. Use of lipid lowering drugs for primary prevention of coronary heart disease: meta-analysis of randomised trials. *Br Med J* 2000; **321**: 983–6. 118
PM Ridker, N Rifai, MA Pfeffer, F Sacks, E Braunwald, for the Cholesterol and Recurrent Events (CARE) Investigators. Long-term effects of pravastatin on plasma concentration of C-reactive protein. *Circulation* 1999; **100**:230–5. 157
HB Rubins, SJ Robins, D Collins, CL Fye, JW Anderson, MB Elam, FH Faas, E Linacres, EJ Schaefer, G Schectman, TJ Wilt, J Wittes. Gemfibrozil for the secondary prevention of coronary

American Diabetes Association. Management of dyslipidaemia in adults with diabetes (supplement). *Diabetes Care* 1999; **22**:S56–9. 242

JW Anderson, LD Allgood, J Turner, PR Oeltgen, BP Daggy. Effects of psyllium on glucose and serum lipid responses in men with type 2 diabetes and hypercholesterolaemia. *Am J Clin Nutr* 1999; **70**:466–73. 255

RA Anderson, LM Evans, GR Ellis, J Graham, K Morris, SK Jackson, MJ Lewis, A Rees, MP Frenneaux. The relationships between postprandial lipaemia, endothelial function and oxidative stress in healthy individuals and patients with type 2 diabetes. *Atherosclerosis* 2001; **154**:475–83. 210

M Axelsen, U Smith, JW Eriksson, MR Taskinen, PA Jansson. Postprandial hypertriglyceridaemia and insulin resistance in normoglycemic first-degree relatives of patients with type 2 diabetes. *Ann Intern Med* 1999; **131**:27–31. 220

SB Battula, O Fitzsimons, S Moreno, D Owens, P Collins, A Johnson. Postprandial apolipoprotein B48- and B100-containing lipoproteins in type 2 diabetes: do statins have a specific effect on triglyceride metabolism? *Metabolism* 2000; **49:**1049–54. 218

JP Burke, K Williams, SP Gaskill, HP Hazuda, SM Haffher, MP Stern. Rapid rise in the incidence of type 2 diabetes from 1987 to 1996: results from the San Antonio Heart Study. *Arch Intern Med* 1999; **159**: 1450–6. 196

Diabetes Atherosclerosis Intervention Study. The effect of fenofibrate on the progression of coronary heart disease. *Lancet* 2001; **357**:905–10. 248

PN Durrington. Diabetic dyslipidaemia in Baillière's Best Practice and Research. *Clin Endocrinol Metab* 1999; **13**:265–78. 211

L Duvillard, F Pont, E Florentin, P Gambert, B Verges. Significant improvement of apolipoprotein B-containing lipoprotein metabolism by insulin treatment in patients with non-insulin-dependent diabetes mellitus. *Diabetologia* 2000; **43**:27–35. 223

MB Elam, DB Hunninghake, KB Davis, R Garg, C Johnson, D Egan, JB Kostis, DS Sheps, EA Brinton, for the ADMIT Investigators. Effect of niacin on lipid and lipoprotein levels and glycemic control in patients with diabetes and peripheral arterial disease: the ADMIT study: A randomized trial. Arterial Disease Multiple Intervention Trial. *JAMA* 2000; **284**: 1263–70. 259

M Evans, RA Anderson, J Graham, GR Ellis, K Morris, S Davies. Ciprofibrate therapy improves endothelial function and reduces postprandial lipemia and oxidative stress in type 2 diabetes mellitus. *Circulation* 2000; **101**(15):1173–9. 207

M Evans, N Khan, A Rees. Diabetic dyslipidaemia and coronary heart disease: new perspectives. *Curr Opin Lipidol* 1999; **10**:387–91. 220

A Fagot-Campagna, DJ Pettitt, MM Engelgau, NR Burrows, LS Geiss, R Valdez, GLA Beckles, J Saaddine, EW Gregg, DF Williamson, KM Venkat Narayan. Type 2 diabetes among North American children and adolescents: an epidemiological review and a public health perspective. *J Pediatr* 2000; **136**: 664–72. 198

DJ Freeman, J Norrie, N Sattar, RD Neely, SM Cobbe, I Ford, C Isles, AR Lorimer, PW MacFarlane, JH McKillop, CJ Packard, J Shepherd, A Gaw. Pravastatin and the development of diabetes mellitus. Evidence for a protective treatment effect in the West of Scotland Coronary Prevention Study. *Circulation* 2001; **103**:357–62. 243

D Gavish, E Leibovitz, I Shapira, A Rubinstein. Bezafibrate and simvastatin combination therapy for diabetic dyslipidaemia: efficacy and safety. *J Intern Med* 2000; **247**:563–9. 267

RB Goldberg, J Margot, MD Mellies, FM Sacks, LA Moye, BV Howard, WJ Howard, BR Davis, TG Cole, MA Pfeffer, E Braunwald. Cardiovascular events and their reduction with pravastatin in diabetic and glucose-intolerant myocardial infarction survivors with average cholesterol levels. Subgroup Analyses in the Cholesterol and Recurrent Events (CARE) Trial.

Geneva: WHO 1977 197
M Krempf, V Rohmer, M Farnier, M Issa-Sayegh, C Corda, I Sirugue. Efficacy and safety of micronised fenofibrate in a randomised double-blind study comparing four doses from 200 mg to 400 mg daily with placebo in patients with hypercholesterolemia. *Diabetes Metab* 2000; **26**: 184–91. 253
SP Marso, AM Lincoff, SG Ellis, DL Bhatt, JF Tanguay, NS Kleiman, T Hammoud, JE Booth, SK Sapp, EJ Topol, for the EPISTENT Investigators. Optimising the percutaneous interventional outcomes for patients with diabetes mellitus. Results of the EPISTENT Diabetes substudy. *Circulation* 1999; **100**:2477–84. 200
J McEneny, MJ O'Kane, KW Moles, C McMaster, D McMaster, C Mercer. Very low density lipoprotein subfractions in type II diabetes mellitus: alterations in composition and susceptibility to oxidation. *Diabetologia* 2000; **43**:485–93. 215
KL McNeill, L Fontana, DL Russell-Jones, I Rajman, JM Ritter, PJ Chowienczyk. Inhibitory effects of low-density lipoproteins from men with type II diabetes on endothelium-dependent relaxation. *Am Coll Cardiol* 2000; **35**:1622–7. 219
N Mero, R Malmstrom, G Steiner, MR Taskinen, M Syvanne. Postprandial metabolism of apolipoprotein B-48- and B-100-containing particles in type 2 diabetes mellitus: relations to angiographically verified severity of coronary artery disease. *Atherosclerosis* 2000; **150**:167–77. 223
S Ogawa, K Takeuchi, K Sugimura, M Fukuda, R Lee, S Ito. Bezafibrate reduces blood glucose in type 2 diabetes mellitus. *Metabolism* 2000; **49**:331–4. 254
K Pyorala, TR Pedersen, J Kjekhus, O Faergeman, AG Olsson, G Thorgeirsson. Cholesterol lowering with simvastatin improves prognosis of diabetic patients with coronary heart disease. *Diabetes Care* 1997; **20**:614–20. 229
HB Rubins, S J Robins, D Collins, CL Fye, JW Anderson, MB Elam, FH Faas, E Linares, EJ Schaefer, G Schectman, TJ Witt, J Wittes. Gemfibrozil for the secondary prevention of coronary heart disease in men with low levels of high-density lipoprotein cholesterol. Veterans Affairs High-Density Lipoprotein Cholesterol Intervention Trial Study Group. *N Engl J Med* 1999; **341**:410–18. 245
A Rubinstein, FJ Maritz, SG Soule, A Markel, T Chajek-Shaul, M Maislos, S Tal and D Stolero on behalf of the Hyperlipidaemia In Diabetes Mellitus Investigators. Efficacy and safety of cerivastatin for type 2 diabetes and hypercholesterolaemia. Hyperlipidaemia in diabetes mellitus investigators. *Cardiovasc Risk* 1999; **6**:399–403. 257
C Rustemeijer, JA Schouten, HJ Voerman, HE Hensgens, AJ Donker, RJ Heine. Pravastatin compared with bezafibrate in the treatment of dyslipidaemia in insulin-treated patients with type 2 diabetes mellitus. *Metab Res Rev* 2000; **16**:82–7. 254
RA Skyrme-Jones, RC O'Brien, M Luo, IT Meredith. Endothelial vasodilator function is related to low-density lipoprotein particle size and low-density lipoprotein vitamin E content in type 1 diabetes. *J Am Coll Cardiol* 2000; **35**:292–9. 213
KC Tan, ED Janus, KS Lam. Effects of fluvastatin on prothrombotic and fibrinolytic factors in type 2 diabetes mellitus. *Am J Cardiol* 1999; **84**:934–7 (A7). 263
J Tuomilehto, D Rastenyte, WH Berkenhäger, L Thijs, R Antikainen, CJ Bulpitt, AE Fletcher, F Forette, A Goldhaber, P Palatini, C Sarti, R Fagard, for the Systolic Hypertension in Europe Trial Investigators. Effects of calcium channel blockade in older patients with diabetes and systolic hypertension. *N Engl J Med* 1999; **340**:677–84. 277
UKPDS 33. United Kingdom Prospective Diabetes Study Group. Intensive blood-glucose control with sulphonylureas or insulin compared with conventional treatment and risk of complications in patients with type II diabetes: *Lancet* 1998; **352**:837–53. 273

effects of lipid peroxidation. *Curr Opin Lipidol* 2000; **11**:383–8. 303

M Navab, SY Hama, GM Anantharamaiah, K Hassan, GP Hough, AD Watson, ST Reddy, A Sevanian, GC Fonarow, AM Fogelman. Normal high density lipoprotein inhibits three steps in the formation of mildly oxidized low density lipoprotein. Steps 2 and 3. *J Lipid Res* 2000; **41**:1495–508. 305

M Navab, SY Hama, CJ Cooke, GM Anantharamaiah, M Chaddha, L Jin, G Subbanagounder, KF Faull, ST Reddy, NE Miller, AM Fogelman. Normal high density lipoprotein inhibits three steps in the formation of mildly oxidized low density lipoprotein. Step 1. *J Lipid Res* 2000; **41**:1481–94. 305

H Okamoto, F Yonemori, K Wakitani, T Minowa, K Maeda, H Shinkai. A cholesteryl ester transfer protein inhibitor attenuates atherosclerosis in rabbits. *Nature* 2000; **406**:203–7. 311

CW Rittershaus, DP Miller, LJ Thomas, MD Picard, CM Honan, CD Emmett, CL Pettey, H Adari, RA Hammond, DT Beattie, AD Callow, HC Marsh, US Ryan. Vaccine-induced antibodies inhibit CETP activity in vivo and reduce aortic lesions in a rabbit model of atherosclerosis. *Arterioscler Thromb Vasc Biol* 2000; **20**:2106–12. 308

JT Salonen, K Nyyssonen, R Salonen, HM Lakka, J Kaikkonen, E Porkkala-Sarataho, S Voutilainen, TA Lakka, T Rissanen, L Leskinen, TP Tuomainen, VP Valkonen, U Ristonmaa, HE Poulsen. Antioxidant Supplementation in Atherosclerosis Prevention (ASAP) study: a randomized trial of the effect of vitamins E and C on 3-year progression of carotid atherosclerosis. *J Intern Med* 2000; **248**: 377–86. 316

S Toshima, A Hasegawa, M Kurabayashi, H Itabe, T Takano, J Sugano, K Shimamura, J Kimura, I Michishita, T Suzuki, R Nagai. Circulating oxidized low density lipoprotein levels: a biochemical risk marker for coronary heart disease. *Arterioscler Thromb Vasc Biol* 2000; **20**: 2243–7. 325

BR Winkelmann, J Hager, WE Kraus, P Merlini, B Keavney, PJ Grant, JB Muhlestein, CB Granger. Genetics of coronary heart disease: current knowledge and research principles. *Am Heart J* 2000; **140**:11–26. 327

General Index

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