4. Water sampling and analysis

Ideally, a laboratory infrastructure should be established which will enable all samples to be returned to a central or regional laboratory within a few hours of being taken. However, this depends on the availability of a good road system and of reliable motorized transport for all sampling officers, and these are not available in many countries. Thus, although it may be possible to establish well-equipped central and even regional laboratories for water analysis, at the provincial and district levels it may be necessary to rely on a relatively small number of simple tests. As noted in Chapter 1, this approach is sometimes called critical-parameter water testing.

The most important factor to take into account is that, in most communities, the principal risk to human health derives from faecal contamination. In some countries there may also be hazards associated with specific chemical contaminants such as fluoride or arsenic, but the levels of these substances are unlikely to change significantly with time. Thus, if a full range of chemical analyses is undertaken on new water sources and repeated thereafter at fairly long intervals, chemical contaminants are unlikely to present an unrecognized hazard. In contrast, the potential for faecal contamination in untreated or inadequately treated community supplies is always present. The minimum level of analysis should therefore include testing for indicators of faecal pollution (thermotolerant (faecal) coliforms), turbidity, and chlorine residual and pH (if the water is disinfected with chlorine).

Even in developing countries poorly served by roads and transportation, it is usually possible to devise a rational sampling and analytical strategy. This should incorporate carefully selected critical-parameter tests in remote (usually rural) locations using simple methods and portable water-testing equipment (see pp. 65–66) where appropriate. Wherever possible the community should be involved in the sampling process. Where water is disinfected, primary health workers, schoolteachers, and sometimes community members can be trained to carry out simple chlorine residual testing. The same people could also collect samples for physicochemical analysis and arrange for their delivery to the regional laboratory. The use of community members in this way has significant implications for training and supervision but would be one way of ensuring more complete surveillance coverage.

4.1 Sampling

The guidelines provided here take into account experience in surveillance programmes in remote, typically rural, areas and in periurban communities. More general advice on sampling is given in Volume 1 and in ISO standards (see the Bibliography).

4.1.1 Location of sampling points

One objective of surveillance is to assess the quality of the water supplied by the supply agency and of that at the point of use, so that samples of both should be taken. Any significant difference between the two has important implications for remedial strategies.

Samples must be taken from locations that are representative of the water source, treatment plant, storage facilities, distribution network, points at which water is delivered to the consumer, and points of use. In selecting sampling points, each locality should be considered individually; however, the following general criteria are usually applicable:

- Sampling points should be selected such that the samples taken are representative of the different sources from which water is obtained by the public or enters the system.
- These points should include those that yield samples representative of the conditions at the most unfavourable sources or places in the supply system, particularly points of possible contamination such as unprotected sources, loops, reservoirs, low-pressure zones, ends of the system, etc.
- Sampling points should be uniformly distributed throughout a piped distribution system, taking population distribution into account; the number of sampling points should be proportional to the number of links or branches.
- The points chosen should generally yield samples that are representative of the system as a whole and of its main components.
- Sampling points should be located in such a way that water can be sampled from reserve tanks and reservoirs, etc.
- In systems with more than one water source, the locations of the sampling points should take account of the number of inhabitants served by each source.
- There should be at least one sampling point directly after the clean-water outlet from each treatment plant.

Sampling sites in a piped distribution network may be classified as:

- fixed and agreed with the supply agency;
- fixed, but not agreed with the supply agency; or
- random or variable.

Each type of sampling site has certain advantages and disadvantages. Fixed sites agreed with the supplier are essential when legal action is to be used as a

means of ensuring improvement; otherwise, the supply agency may object to a sample result on the grounds that water quality may have deteriorated in the household, beyond the area of responsibility of the supplier. Nevertheless, fixed sample points are rare or unknown in some countries.

Fixed sites that are not necessarily recognized by the supply agency are used frequently in investigations, including surveillance. They are especially useful when results have to be compared over time, but they limit the possibility of identifying local problems such as cross-connections and contamination from leaking distribution networks.

Sampling regimes using variable or random sites have the advantage of being more likely to detect local problems but are less useful for analysing changes over time.

4.1.2 Sampling frequency

The most important tests used in water-quality surveillance or quality control in small communities are those for microbiological quality (by the measurement of indicator bacteria) and turbidity, and for free chlorine residual and pH where chlorination is used. These tests should be carried out whenever a sample is taken, regardless of how many other physical or chemical variables are to be measured. The recommended minimum frequencies for these critical measurements in unpiped water supplies are summarized in Table 4.1 and minimum sample numbers for piped drinking-water in the distribution system are shown in Table 4.2.

4.1.3 Sampling methods for microbiological analysis

Detailed methods for sampling for microbiological analysis are given in Annex 4. All samples should be accompanied by an appropriate collection form; a model sample collection form is illustrated in Fig. 4.1.

4.1.4 Storage of samples for microbiological analysis

Although recommendations vary, the time between sample collection and analysis should, in general, not exceed 6 hours, and 24 hours is considered the absolute maximum. It is assumed that the samples are immediately placed in a lightproof insulated box containing melting ice or ice-packs with water to ensure rapid cooling. If ice is not available, the transportation time must not exceed 2 hours. It is imperative that samples are kept in the dark and that cooling is rapid. If these conditions are not met, the samples should be discarded. When water that contains or may contain even traces of chlorine is sampled, the chlorine must be inactivated. If it is not, microbes may be killed during transit and an erroneous result will be obtained. The bottles in which the samples are placed should therefore contain sodium thiosulfate to neutralize any chlorine present, as de-

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Source and mode of supply	Minimum frequency of	sampling and analysis	Remarks
	Bacteriological	Physical/chemical	
Open wells for community supply	Sanitary protection measures; bacteriological testing only if situation demands	Once initially for community wells	Pollution usually expected to occur
Covered dug wells and shallow tubewells with hand-pumps	Sanitary protection measures; bacteriological testing only if situation demands	Once initially, thereafter as situation demands	Situations requiring testing: change in environmental conditions, outbreak of waterborne disease, or increase in incidence of waterborne diseases
Deep tubewells with hand-pumps	Once initially, thereafter as situation demands	Once initially, thereafter as situation demands	Situations requiring testing: change in environmental conditions, outbreak of waterborne disease, or increase in incidence of waterborne diseases
Protected springs	Once initially, thereafter as situation demands	Periodically for residual chlorine if water is chlorinated	Situations requiring testing: change in environmental conditions, outbreak of waterborne disease, or increase in incidence of waterborne diseases
Community rainwater collection systems	Sanitary protection measures; bacteriological testing only if situation demands	Not needed	I

Table 4.1 Minimum frequency of sampling and analysis of unpiped water supplies

		1
WATER-QUALITY CONTROL PROGRAMME [Name of body responsible]	SAMPLE DATA Locality Sample site Place Source Residual chlorine Date of sampling Time of sampling
Place		
SourceSender Date collected Time collected Date of analysis Time of analysis Residual chlorine Results: TOTAL COLIFORMS FAECAL COLIFORMS (OTHER)	. mg/litre . /100 ml . /100 ml	Section detached and stuck to the sample bottle Analytical results entered by laboratory; copies of this section sent by laboratory to local surveillance agency or water- supply agency and person responsible for sampling
Laboratory Sample No WATER BACTERIOLOGICALLY GOOD – BAD <u>ACTION TAKEN</u>	- -	
(signed)	WH0 96543

Fig. 4.1 Model sample collection form

uisilibulion system				
Population served	No. of monthly samples			
<5000 5000-100000 >100000	1 1 per 5000 population 1 per 10000 population, plus 10 additional samples			

Table 4.2 Minimum sample numbers for
piped drinking-water in the
distribution system

scribed in Annex 4. The box used to carry samples (see Fig. 4.2) should be cleaned and disinfected after each use to avoid contaminating the surfaces of the bottles and the sampler's hands.

4.1.5 Sampling methods for physicochemical analysis

Results of physicochemical analysis are of no value if the samples tested are not properly collected and stored. This has important consequences for sampling regimes, sampling procedures, and methods of sample preservation and storage. In general, the time between sampling and analysis should be kept to a minimum. Storage in glass or polyethylene bottles at a low temperature (e.g. 4 °C) in the dark is recommended. Sample bottles must be clean but need not be sterile. Special preservatives may be required for some analytes. Residual chlorine, pH, and turbidity should be tested immediately after sampling as they will change during storage and transport.

4.2 Bacteriological analysis

The principal risk associated with water in small-community supplies is that of infectious disease related to faecal contamination. Hence, as described in Chapter 1, the microbiological examination of drinking-water emphasizes assessment of the hygienic quality of the supply. This requires the isolation and enumeration of organisms that indicate the presence of faecal contamination. In certain circumstances, the same indicator organisms may also be used to assess the efficiency of drinking-water treatment plants, which is an important element of quality control. Other microbiological indicators, not necessarily associated with faecal pollution, may also be used for this purpose.

The isolation of specific pathogens in water should be undertaken only by reference laboratories for purposes of investigating and controlling outbreaks of disease. Routine isolation in other circumstances is not practical.

Detailed methods for use in bacteriological analysis are described in Annex 5 (multiple-tube method), Annex 6 (membrane-filtration method), Annex 7 (onsite testing method), and Annex 8 (presence-absence test).



Fig. 4.2 Transport box for samples for microbiological analysis

Ice-pack or freezing mixture

4.2.1 Indicator organisms

The properties and significance of the commonly used faecal indicator bacteria are described in detail in Volume 1; a summary is provided here.

Escherichia coli is a member of the family Enterobacteriaceae, and is characterized by possession of the enzymes β -galactosidase and β -glucuronidase. It grows at 44–45 °C on complex media, ferments lactose and mannitol with the production of acid and gas, and produces indole from tryptophan. However, some strains can grow at 37 °C but not at 44–45 °C, and some do not produce gas. *E. coli* does not produce oxidase or hydrolyse urea. Complete identification of the organism is too complicated for routine use, but a number of tests have been developed for rapid and reliable identification. Some of these methods have been standardized at international and national levels and accepted for routine use; others are still being developed or evaluated.

Escherichia coli is abundant in human and animal faeces; in fresh faeces it may attain concentrations of 10^9 per gram. It is found in sewage, treated effluents, and all natural waters and soils subject to recent faecal contamination, whether from humans, wild animals, or agricultural activity. Recently, it has been suggested that *E. coli* may be present or even multiply in tropical waters not subject to human faecal pollution. However, even in the remotest regions, faecal contamination by wild animals, including birds, can never be excluded. Because animals can transmit pathogens that are infective in humans, the presence of *E. coli* or thermotolerant coliform bacteria must not be ignored, because the presumption remains that the water has been faecally contaminated and that treatment has been ineffective.

Thermotolerant coliform bacteria

Thermotolerant coliform bacteria are the coliform organisms that are able to ferment lactose at 44-45 °C; the group includes the genus *Escherichia* and some species of *Klebsiella*, *Enterobacter*, and *Citrobacter*. Thermotolerant coliforms other than *E. coli* may also originate from organically enriched water such as industrial effluents or from decaying plant materials and soils. For this reason, the term "faecal" coliforms, although frequently employed, is not correct, and its use should be discontinued.

Regrowth of thermotolerant coliform organisms in the distribution system is unlikely unless sufficient bacterial nutrients are present, unsuitable materials are in contact with the treated water, the water temperature is above 13 °C, and there is no free residual chlorine.

In most circumstances, concentrations of thermotolerant coliforms are directly related to that of *E. coli*. Their use in assessing water quality is therefore considered acceptable for routine purposes, but the limitations with regard to specificity should always be borne in mind when the data are interpreted. If high counts of thermotolerant coliforms are found in the absence of detectable sanitary hazards, additional confirmatory tests specific for *E. coli* should be carried out. National reference laboratories developing national standard methods are advised to examine the specificity of the thermotolerant coliform test for *E. coli* under local conditions.

Because thermotolerant coliform organisms are readily detected, they have an important secondary role as indicators of the efficiency of water-treatment processes in removing faecal bacteria. They may therefore be used in assessing the degree of treatment necessary for waters of different quality and for defining performance targets for removal of bacteria.

Coliform organisms (total coliforms)

Coliform organisms have long been recognized as a suitable microbial indicator of drinking-water quality, largely because they are easy to detect and enumerate in water. The term "coliform organisms" refers to Gram-negative, rod-shaped bacteria capable of growth in the presence of bile salts or other surface-active agents with similar growth-inhibiting properties and able to ferment lactose at 35-37 °C with the production of acid, gas, and aldehyde within 24–48 hours. They are also oxidase-negative and non-spore-forming and display β -galactosidase activity.

Traditionally, coliform bacteria were regarded as belonging to the genera *Escherichia*, *Citrobacter*, *Enterobacter*, and *Klebsiella*. However, as defined by modern taxonomical methods, the group is heterogeneous. It includes lactose-fermenting bacteria, such as *Enterobacter cloacae* and *Citrobacter freundii*, which can be found in both faeces and the environment (nutrient-rich waters, soil, decaying plant material) as well as in drinking-water containing relatively high concentrations of nutrients, as well as species that are rarely, if ever, found in faeces and may multiply in relatively good-quality drinking-water, e.g. *Serratia fonticola, Rabnella aquatilis*, and *Buttiauxella agrestis*.

The existence both of non-faecal bacteria that fit the definitions of coliform bacteria and of lactose-negative coliform bacteria limits the applicability of this group as an indicator of faecal pollution. Coliform bacteria should not be detectable in treated water supplies and, if found, suggest inadequate treatment, post-treatment contamination, or excessive nutrients. The coliform test can therefore be used as an indicator both of treatment efficiency and of the integrity of the distribution system. Although coliform organisms may not always be directly related to the presence of faecal contamination or pathogens in drinking-water, the coliform test is still useful for monitoring the microbial quality of treated piped water supplies. If there is any doubt, especially when coliform organisms are found in the absence of thermotolerant coliforms and *E. coli*, identification to the species level or analyses for other indicator organisms may be undertaken to investigate the nature of the contamination. Sanitary inspections will also be needed.

Faecal streptococci

Faecal streptococci are those streptococci generally present in the faeces of humans and animals. All possess the Lancefield group D antigen. Taxonomically, they belong to the genera *Enterococcus* and *Streptococcus*. The taxonomy of enterococci has recently undergone important changes, and detailed knowledge of the ecology of many of the new species is lacking; the genus *Enterococcus* now includes all streptococci that share certain biochemical properties and have a wide tolerance of adverse growth conditions—*E. avium, E. casseliflavus, E. cecorum, E. durans, E. faecalis, E. faecium, E. gallinarum, E. hirae, E. malodoratus, E. mundtii, and <i>E. solitarius*. Most of these species are of faecal origin and can generally be regarded as specific indicators of human faecal pollution for most practical purposes. They may, however, be isolated from the faeces of animals, and certain species and subspecies, such as *E. casseliflavus, E. faecalis* var. *liquefaciens, E. malodoratus*, and *E. solitarius*, occur primarily on plant material.

In the genus *Streptococcus*, only *S. bovis* and *S. equinus* possess the group D antigen and therefore belong to the faecal streptococcus group. They derive mainly from animal faeces. Faecal streptococci rarely multiply in polluted water, and they are more persistent than *E. coli* and coliform bacteria. Their primary value in water-quality examination is therefore as additional indicators of treatment efficiency. Moreover, streptococci are highly resistant to drying and may be valuable for routine control after new mains are laid or distribution systems are repaired, or for detecting pollution of groundwaters or surface waters by surface run-off.

4.2.2 Principal analytical techniques

The standardization of methods and laboratory procedures is important. International standard methods should be evaluated under local conditions before they are formally adopted by national surveillance programmes. A list of ISO standard methods is given in the Bibliography. The methods described in the annexes to this publication are based on these ISO standard methods, modified where appropriate in the light of experience in the surveillance of community water supplies.

The principal methods used in the isolation of indicator organisms from water are the membrane-filtration (MF) method, the multiple-tube (MT) or most probable number (MPN) method and presence-absence tests.

Membrane-filtration method

In the membrane-filtration (MF) method, a minimum volume of 10 ml of the sample (or dilution of the sample) is introduced aseptically into a sterile or properly disinfected filtration assembly containing a sterile membrane filter (nominal pore size 0.2 or $0.45 \,\mu$ m). A vacuum is applied and the sample is drawn

Sample type	Sample volume (ml)
Treated drinking-water	100
Protected source water or groundwater	10-100
Surface water and water from open wells	0. I-100 ^a

 Table 4.3 Typical sample volumes for membrane-filtration analysis

^a Volumes less than 10ml should be added to the filtration apparatus after addition of at least 10ml of sterile diluent to ensure adequate dispersal across the surface of the membrane filter.

through the membrane filter. All indicator organisms are retained on or within the filter, which is then transferred to a suitable selective culture medium in a Petri dish. Following a period of resuscitation, during which the bacteria become acclimatized to the new conditions, the Petri dish is transferred to an incubator at the appropriate selective temperature where it is incubated for a suitable time to allow the replication of the indicator organisms. Visually identifiable colonies are formed and counted, and the results are expressed in numbers of "colonyforming units" (CFU) per 100 ml of original sample.

This technique is inappropriate for waters with a level of turbidity that would cause the filter to become blocked before an adequate volume of water had passed through. When it is necessary to process low sample volumes (less than 10 ml), an adequate volume of sterile diluent must be used to disperse the sample before filtration and ensure that it passes evenly across the entire surface of the membrane filter. Membrane filters may be expensive in some countries.

Typical sample volumes for different water types are shown in Table 4.3. Where the quality of the water is totally unknown, it may be advisable to test two or more volumes in order to ensure that the number of colonies on the membrane is in the optimal range for counting (20–80 colonies per membrane).

Multiple-tube method

The multiple-tube method is also referred to as the most probable number (MPN) method because—unlike the MF method—it is based on an indirect assessment of microbial density in the water sample by reference to statistical tables to determine the most probable number of microorganisms present in the original sample. It is essential for highly turbid samples that cannot be analysed by membrane filtration. The technique is used extensively for drinking-water analysis, but it is time-consuming to perform and requires more equipment, glassware, and consumables than membrane filtration. However, the multiple-tube method may be more sensitive than membrane filtration.

Sample type	Number of tubes for sample volume:							
	50 ml	10 ml	1 ml	0.1 ml	0.01 ml			
Treated drinking-water	1	5		_	_			
Partially treated drinking-water		5	5	5	—			
Protected source water or groundwater	_	5	5	5	_			
Surface water or water from open wells		_	5	5	5			

Table 4.4	Typical sample	volumes	and	numbers	of	tubes	for	the
	multiple-tube m	ethod						

^a Volumes of 0.1 and 0.01 ml are tested by the addition of 1 ml of a 1/10 and 1/100 dilution sample, respectively, to 10 ml of single-strength culture medium.

The multiple-tube method depends on the separate analysis of a number of volumes of the same sample. Each volume is mixed with culture medium and incubated. The concentration of microorganisms in the original sample can then be estimated from the pattern of positive results (the number of tubes showing growth in each volume series) by means of statistical tables that give the "most probable number" per 100 ml of the original sample.

The combination of sample volumes for processing is selected according to the type of water sample or known degree of contamination. Various configurations and tables may be used; typical volumes and dilutions are summarized in Table 4.4.

Appropriate volumes of water are added aseptically to tubes or other vessels containing sterile nutrient medium of a concentration that will ensure the mixture corresponds to single-strength medium. For example, 10 ml of sample would typically be added to 10 ml of double-strength medium or 1 ml of sample to 10 ml of single-strength medium and so on. The tube must also contain a small inverted glass tube (Durham tube) to facilitate the detection of gas production. Growth in the medium is confirmed by visible turbidity and/or a colour change. Tubes are incubated without resuscitation, and the number of positive reactions is recorded after 24 and/or 48 hours, depending on the type of analysis.

Presence-absence tests

Presence–absence tests may be appropriate for monitoring good-quality drinking-water where positive results are known to be rare. They are not quantitative and, as their name suggests, they indicate only the presence or absence of the indicator sought. Such results are of very little use in countries or situations where contamination is common; the purpose of analysis is then to determine the degree of contamination rather than indicate whether or not contamination is present. Thus, presence–absence tests are not recommended for use in the analysis of surface waters, untreated small-community supplies, or larger water supplies that may experience occasional operational and maintenance difficulties.

4.2.3 Choice of methods

Very often the choice between the multiple-tube and the membrane-filtration methods will depend on national or local factors, e.g. the equipment already available or the cost of certain consumables. The advantages and disadvantages of each method should be considered when a choice has to be made; these are summarized in Table 4.5. The schematic decision-making network shown in Fig. 4.3 will aid the selection of procedure and method. The purpose of this diagram is merely to provide suggestions for the approach to be used; local or other circumstances will also affect the final decision.

4.2.4 Minimizing the cost of analysis

It is sometimes clear that faecal contamination exists (e.g. immediately downstream of a sewage discharge) or that contamination is very unlikely (e.g. in a distribution network with a free chlorine residual greater than 0.5 mg/litre, median turbidity less than 1 NTU, and pH less than 8.0). Microbiological analysis may then be deemed unnecessary. This is not appropriate, however, under certain conditions, e.g. where there is a legal requirement to conduct analysis, or where legal action that may be taken would depend on the results of a microbiological analysis of the water.

Omission of microbiological analysis under the appropriate conditions mentioned above may contribute to minimizing costs. It may also ensure that adequate numbers of samples are investigated overall where the resources available

Most probable number method	Membrane-filtration method
Slower: requires 48 hours for a negative or presumptive positive result	Quicker: quantitative results in about 18 hours
More labour-intensive	Less labour-intensive
Requires more culture medium	Requires less culture medium
Requires more glassware	Requires less glassware
More sensitive	Less sensitive
Result obtained indirectly by statistical approximation (low precision)	Result obtained directly by colony count (high precison)
Not readily adaptable for use in the field	Readily adaptable for use in the field
Applicable to all types of water	Not applicable to turbid waters
Consumables readily available in most countries	Consumables costly in many countries
May give better recovery of stressed or damaged organisms under some circumstances	

Table 4.5 Comparison of methods for analysis of coliform bacteria

Fig. 4.3 Decision-making network for selection of method of analysis

Note: Analysis may sometimes be necessary because of specific local circumstances, e.g. where legislation demands that such analysis should be undertaken, or where legal action may be taken on the basis of analytical results.



for analysis are inadequate to undertake the recommended number of microbiological analyses.

4.2.5 Laboratory-based versus on-site testing

Water-quality testing in communities may be subject to the following problems, especially when the communities or the sampling sites are remote or inaccessible:

- deterioration of samples during transport to centralized laboratory facilities;
- high cost of transporting samples;
- inadequate techniques for sample storage and preservation during prolonged transport, thus limiting the sampling range;
- increased personnel costs because of the need for repeat sampling journeys;
- the need for reporting, which may necessitate further return journeys.

If there are delays in sample transport and analysis—and therefore in reporting—remedial action is also likely to be delayed. For these reasons, on-site water testing using portable equipment is appropriate in many remote areas. Portable equipment is used in many developing countries, and does help to overcome a number of logistic and financial constraints. However, it varies widely in technical specifications, including the range of analyses that can be performed, the range of methods employed, its robustness, the degree of independence from central laboratory facilities, its portability, and requirements for consumables.

Portable testing equipment may also be favoured by agencies that undertake project monitoring in more than one area on a non-routine basis and therefore prefer portability to the establishment of a conventional laboratory. For reasons that include the following, portable equipment may also be used in conventional laboratories in place of normal laboratory equipment, especially when the number of analyses to be performed per day is relatively low.

- Independence from (unreliable) power supplies. Several types of portable equipment either incorporate a rechargeable battery or may be connected to an external battery. Where energy supplies are unreliable (because of either voltage fluctuation or intermittent supply), battery operation may be advantageous.
- Cost. Comparison of the costs of the equipment required, even after allowing for that needed for back-up, may show that it is more economical to provide portable testing equipment to peripheral or decentralized laboratories than conventional laboratory equipment.
- Ease of use. Because portable equipment is often designed for use by personnel who are not fully qualified in laboratory techniques, its use is usually straightforward. However, this does not obviate the need for proper training of personnel, particularly since some portable equipment may not be accompanied by clear, well-illustrated manuals in the language of the users.

Use of portable equipment in conventional laboratories also carries a number of disadvantages, including limitations in technical specifications. Although not invariably true, the requirement for portability may mean that portable equipment is of lower precision and sensitivity than conventional equipment. Moreover, while some types of portable equipment help to reduce dependence on expensive consumables that may be difficult to obtain in many countries (e.g. by employing reusable aluminium Petri dishes, rather than dishes made of disposable plastic or fragile glass), others actually increase dependence on non-standard glassware and, particularly, consumables (such as microbiological culture media in ampoules and preweighed reagents for chemical tests). These items are invariably more expensive than ordinary laboratory consumables and may be available only from the manufacturer of the portable equipment. Independence of special consumables is of particular importance for some reagents and microbiological culture media; ready-prepared liquid media in ampoules eliminate errors in media preparation but they have only limited shelf-life. This is an especially relevant consideration in developing countries, where delays in importation, variability of demand, and problems with transport may seriously reduce the remaining shelf-life of media. Under these conditions, it is preferable to supply dehydrated media-ideally in preweighed quantities-with a relatively long shelf-life.

The use of portable testing equipment may be the result of a commitment to the decentralization of testing facilities. Whether or not this is the case, it generally means that small numbers of analyses are undertaken at a larger number of sites, which has important implications for training:

- The number of personnel carrying out analyses will be greater so that the need for training will be greater.
- The personnel who are to use the equipment (and who are therefore to be trained) will not be working in the capital city, but in relatively remote areas far from training centres.
- These personnel are less likely to have received good initial training in laboratory techniques.

Thus there is actually a greater need for training when decentralized waterquality testing is contemplated, which is in contrast to the popular perception of "simplified" portable testing equipment for which little additional training is required. Many of the benefits expected from decentralized water-quality testing and/or on-site analysis are unlikely to be realized unless adequate resources are devoted to training.

4.2.6 Single-application (disposable) test kits

Disposable test kits are both widely marketed and increasingly used in developed countries. Their reliability may vary widely and they should be properly assessed by a reference laboratory. In developing countries, there are other drawbacks to the use of disposable kits: unit costs, which are high in developed countries, may be still higher, and the trade-off against personnel and staff costs is thus less favourable in developing countries.

4.3 Physicochemical analysis

4.3.1 Chlorine residual

The disinfection of drinking-water supplies constitutes an important barrier against waterborne diseases. Although various disinfectants may be used, chlorine in one form or another is the principal disinfecting agent employed in small communities in most countries.

Chlorine has a number of advantages as a disinfectant, including its relative cheapness, efficacy, and ease of measurement, both in laboratories and in the field. An important additional advantage over some other disinfectants is that chlorine leaves a disinfectant residual that assists in preventing recontamination during distribution, transport, and household storage of water. The absence of a chlorine residual in the distribution system may, in certain circumstances, indicate the possibility of post-treatment contamination.

Three types of chlorine residual may be measured: *free chlorine* (the most reactive species, i.e. hypochlorous acid and the hypochlorite ion); *combined chlorine* (less reactive but more persistent species formed by the reaction of free chlorine species with organic material and ammonia); and *total chlorine* (the sum of the free and combined chlorine residuals). Free chlorine is unstable in aqueous solution, and the chlorine content of water samples may decrease rapidly, particularly at warm temperatures. Exposure to strong light or agitation will accelerate the rate of loss of free chlorine. Water samples should therefore be analysed for free chlorine immediately on sampling and not stored for later testing.

The method recommended for the analysis of chlorine residual in drinkingwater employs *N*,*N*-diethyl-*p*-phenylenediamine, more commonly referred to as DPD. Methods in which *o*-tolidine is employed were formerly recommended, but this substance is a recognized carcinogen, and the method is inaccurate and should not be used. Analysis using starch–potassium iodide is not specific for free chlorine, but measures directly the total of free and combined chlorine; the method is not recommended except in countries where it is impossible to obtain or prepare DPD.

Procedures for the determination of free chlorine residual are described in Annex 9.

4.3.2 pH

It is important to measure pH at the same time as chlorine residual since the efficacy of disinfection with chlorine is highly pH-dependent: where the pH exceeds 8.0, disinfection is less effective. To check that the pH is in the optimal

range for disinfection with chlorine (less than 8.0), simple tests may be conducted in the field using comparators such as that used for chlorine residual. With some chlorine comparators, it is possible to measure pH and chlorine residual simultaneously. Alternatively, portable pH electrodes and meters are available. If these are used in the laboratory, they must be calibrated against fresh pH standards at least daily; for field use, they should be calibrated immediately before each test. Results may be inaccurate if the water has a low buffering capacity.

Procedures for measuring pH using a comparator are described in Annex 10.

4.3.3 Turbidity

Turbidity is important because it affects both the acceptability of water to consumers, and the selection and efficiency of treatment processes, particularly the efficiency of disinfection with chlorine since it exerts a chlorine demand and protects microorganisms and may also stimulate the growth of bacteria.

In all processes in which disinfection is used, the turbidity must always be low—preferably below 1 NTU or JTU (these units are interchangeable in practice). It is recommended that, for water to be disinfected, the turbidity should be consistently less than 5 NTU or JTU and ideally have a median value of less than 1 NTU.

Turbidity may change during sample transit and storage, and should therefore be measured on site at the time of sampling. This can be done by means of electronic meters (which are essential for the measurement of turbidities below 5 NTU). For the monitoring of small-community water supplies, however, high sensitivity is not essential, and visual methods that employ extinction and are capable of measuring turbidities of 5 NTU and above are adequate. These rely on robust, low-cost equipment that does not require batteries and is readily transportable in the field, and are therefore generally preferred.

Procedures for measuring turbidity in the field using a simple "turbidity tube" are described in Annex 10.

4.4 Aesthetic parameters

Aesthetic parameters are those detectable by the senses, namely turbidity, colour, taste, and odour. They are important in monitoring community water supplies because they may cause the water supply to be rejected and alternative (possibly poorer-quality) sources to be adopted, and they are simple and inexpensive to monitor qualitatively in the field.

4.4.1 Colour

Colour in drinking-water may be due to the presence of coloured organic matter, e.g. humic substances, metals such as iron and manganese, or highly coloured industrial wastes. Drinking-water should be colourless. For the purposes of surveillance of community water supplies, it is useful simply to note the presence or absence of observable colour at the time of sampling. Changes in the colour of water and the appearance of new colours serve as indicators that further investigation is needed.

4.4.2 Taste and odour

Odours in water are caused mainly by the presence of organic substances. Some odours are indicative of increased biological activity, others may result from industrial pollution. Sanitary inspections should always include the investigation of possible or existing sources of odour, and attempts should always be made to correct an odour problem. Taste problems (which are sometimes grouped with odour problems) usually account for the largest single category of consumer complaints.

Generally, the taste buds in the oral cavity detect the inorganic compounds of metals such as magnesium, calcium, sodium, copper, iron, and zinc. As water should be free of objectionable taste and odour, it should not be offensive to the majority of the consumers. If the sampling officer has reason to suspect the presence of harmful contaminants in the supply, it is advisable to avoid direct tasting and swallowing of the water. Under these circumstances, a sample should be taken for investigation to a central laboratory.

4.5 Other analyses of relevance to health

Although the great majority of quality problems with community drinking-water are related to faecal contamination, a significant number of serious problems may occur as a result of chemical contamination from a variety of natural and man-made sources. In order to establish whether such problems exist, chemical analyses must be undertaken. However, it would be extremely costly to undertake the determination of a wide range of parameters on a regular basis, particularly in the case of supplies that serve small numbers of people. Fortunately, such parameters tend be less variable in source waters than faecal contamination, so that alternative strategies can be employed.

The range of health-related parameters may include:

- fluoride (where it is known to occur naturally)
- nitrate (where intensification of farming has led to elevated levels in groundwater)
- lead (in areas where it has been used in plumbing)
- chromium (e.g. in areas where it is mined)
- arsenic (in areas where it is known to occur naturally)
- pesticides (where local practices and use indicate that high levels are likely).

If these or any other chemicals of health significance are thought to be present, they should be monitored and the results examined in the light of the WHO guideline values and any relevant national standards (see Volumes 1 and 2).

Some health-related parameters may be measured in community supplies by means of portable test kits based on conventional titrations, comparators, or photometers. If this is done, the reagents must be of high quality and carefully standardized. Other parameters require conventional laboratory analysis by spectrophotometry, atomic absorption spectroscopy, or chromatography, using standard methods.

4.6 Analytical quality assurance and quality control

Standard methods for drinking-water analysis should be tested under local conditions for accuracy and precision, agreed at national level, and applied universally by both water-supply and regulatory agencies. However, the use of standard methods does not in itself ensure that reliable and accurate results will be obtained.

In the context of analytical work, the terms quality assurance and quality control are often treated as synonymous. In fact, they are different concepts.

Analytical quality control is the generation of data for the purpose of assessing and monitoring how good an analytical method is and how well it is operating. This is normally described in terms of within-day and day-to-day precision.

Analytical quality assurance, by contrast, comprises all the steps taken by a laboratory to assure those who receive the data that the laboratory is producing valid results. Quality assurance thus encompasses analytical quality control but also includes many other aspects such as proving that the individuals who carried out an analysis were competent to do so, and ensuring that the laboratory has established and documented analytical methods, equipment calibration procedures, management lines of responsibility, systems for data retrieval, samplehandling procedures and so on.

A checklist for effective analytical quality assurance is given in Table 4.6.

Quality assurance as applied to conventional laboratories is relatively straightforward. It is also important in field testing in view of the more exacting conditions under which it takes place and the unspecialized nature of the responsible staff. Paradoxically, therefore, quality assurance has the greatest importance in circumstances where it is most difficult to undertake. The following are among the possible approaches to overcoming the problem:

- Supervision. An effective network for on-site testing cannot function without adequate supervision, which should cover all field activities, including waterquality testing. This helps to maintain adequate standards of analysis.
- Blank sample analysis. It is unlikely that staff will be willing to submit reports from the field which question their own ability. Furthermore, it is often impractical to prepare, distribute, and collect the results of known quality-

Table 4.6 Checklist for effective analytical quality assurance

Do laboratory personnel have:

- clearly defined responsibilities?
- qualifications?
- experience?
- training?

Is space:

- adequate for the types and number of analyses being undertaken?

Is equipment:

- adequate?
- regularly serviced and maintained?
- calibrated and used only by authorized personnel?

Are materials:

- bought from a reliable supplier, who carries out quality control?

Are there proper facilities:

 for the receipt and storage of samples, and systems for coding and identifying them?

Are data:

- archived?
- retrievable?

Are methods:

- validated?
- documented?
- monitored (i.e. the results subjected to analytical quality control)?

Is safety assured by:

- adequate working and waste-disposal procedures?
- training of staff?
- proper maintenance of equipment?
- proper supervision of staff?

control samples, which would anyway receive especially careful treatment in the field. An alternative strategy is therefore to encourage staff to process sterile distilled water in place of the sample from time to time. If contamination does occur, analysts should then recognize the inadequacies in their own technique and question their own work accordingly. Similarly, samples known to be contaminated may be processed to provide a crude positive control.

• Equipment review. A commitment to decentralized testing with field test kits and other portable equipment normally results in a larger quantity of equipment being in use. Regular review of the equipment (e.g. temperature checking of incubators) is essential. To ensure standardization, this should be undertaken by supervisory staff from a control laboratory. The applicability of methods under field conditions should be assessed by a central laboratory.

4.7 Safety

The safety of staff undertaking analytical procedures, both in the field and in the laboratory, is of the greatest importance. All staff should be trained in safety procedures relevant to their work. In the laboratory, individual staff members should be authorized to undertake procedures involving risk of any type only after appropriate training; unauthorized staff should not be allowed to undertake analyses.

All laboratories should formulate and implement a safety policy that should cover cleaning, disinfection, and the containment of hazardous substances. Safety equipment such as fire extinguishers, safety glasses, and first-aid kits should be suitably located, and readily available; they should be routinely checked and all staff should be trained in their use.