

Chapter 1

SAFE DRINKING WATER: AN ONGOING CHALLENGE

*G.J. Medema, P. Payment, A. Dufour, W. Robertson,
M. Waite, P. Hunter, R. Kirby and Y. Andersson*

1.1 Introduction

1.1.1 Outbreaks of waterborne disease

The microbiological quality of drinking water is a concern to consumers, water suppliers, regulators and public health authorities alike. The potential of drinking water to transport microbial pathogens to great numbers of people, causing subsequent illness, is well documented in countries at all levels of economic development. The outbreak of cryptosporidiosis in 1993 in Milwaukee, Wisconsin, in the United States provides a good example. It was estimated that about 400 000 individuals suffered from gastrointestinal symptoms due, in a large proportion of cases, to *Cryptosporidium*, a protozoan parasite (MacKenzie *et al.*, 1994), although subsequent reports suggest that this may be a significant overestimation (Hunter and Syed, 2001). More recent outbreaks have involved *E. coli* O157:H7, the most serious of which occurred in Walkerton, Ontario Canada in the spring of 2000 and resulted in six deaths and over 2 300 cases (Bruce-Grey-Owen Sound Health Unit, 2000). The number of outbreaks that has been reported throughout the world demonstrates that transmission of pathogens by drinking water remains a significant cause of illness. However, estimates of illness based solely on detected outbreaks is likely to underestimate the problem. A significant proportion of waterborne illness is likely to go undetected by the communicable disease surveillance and reporting systems. The symptoms of gastrointestinal illness (nausea, diarrhoea, vomiting, abdominal pain) are usually mild and generally only last a few days to a week, and only a small percentage of those affected will see a doctor.

Among these, only a minor proportion will have their stools microscopically examined and the examination usually starts with bacterial food pathogens. The number of reported outbreaks differs substantially among countries (Stenström, 1994) even from comparable ones, like the Scandinavian countries (Norway, Sweden, Denmark, Finland). In many cases, this is likely to reflect the effectiveness of the reporting systems rather than a true difference in the number (or size) of outbreaks.

Most sporadic cases of waterborne intestinal illness will not be detected or, if detected, may not be recognised as water-related. In industrialised countries, drinking water that meets current water quality standards may still harbour low concentrations of pathogenic microorganisms. These will cause occasional illness throughout the community served. It is very difficult to relate these sporadic cases to drinking water, as they are buried within the endemic level of disease circulating in the population through other routes of transmission (person-to-person, food and animal contact). There are, however, data from epidemiological studies and seroprevalence studies that indicate that endemic transmission of disease through drinking water does occur (Payment *et al.*, 1991, 1997; Isaac-Renton *et al.*, 1996).

1.1.2 The disease burden is high

Several researchers have attempted to estimate the total burden of waterborne disease world-wide. Huttly (1990) reported a total number of 1.4 billion annual episodes of diarrhoea in children under five years of age, with an estimated 4.9 million children dying as a result (although these were due to all causes of diarrhoea and not just water-related cases). While Prüss *et al.*, (2002) estimated that water, sanitation and hygiene was responsible for 4.0% of all deaths and 5.7% of the total disease burden occurring worldwide (accounting for diarrhoeal diseases, schistosomiasis, trachoma, ascariasis, trichuriasis and hookworm disease). Clearly, in countries where a large part of the population does not have access to safe drinking water, a substantial number of these infections will be waterborne, indeed, Hunter (1997) estimated that waterborne disease might account for one-third of the intestinal infections world-wide.

Waterborne disease is not restricted to developing countries. Morris and Levine (1995) attempted to estimate the annual waterborne disease burden in the United States of America and indicated that 560 000 people may suffer from a moderate to severe waterborne infection and that 7.1 million suffer from a mild to moderate waterborne infection each year. All waterborne infections may lead to an estimated 1 200 deaths a year. Even if these rough figures are

overestimated, both the health and economic burden are considerable even for an industrialised society (Payment, 1997).

The diseases most frequently associated with water are enteric infections (such as infectious diarrhoea) that are also often associated with food (Mead *et al.*, 1999). In many cases, the disease is relatively mild and self-limiting. However, a proportion of the infected population will suffer more severe outcomes, especially when the health care system is lacking. Several waterborne pathogens, such as *Vibrio cholerae*, hepatitis E virus and *Escherichia coli* O157:H7, have high mortality rates (Hunter, 1997). In recent cholera outbreaks, for example, the fatality rate was generally 1-3%, but could be as high as 8-22%. Hepatitis E virus infections may lead to fulminant fatal hepatitis in 1-2% of cases, with pregnant women being especially at risk of more severe disease. Waterborne infections with *E. coli* O157:H7 are associated with haemorrhagic colitis and haemolytic uremic syndrome, both serious illnesses, with the latter occurring particularly in children. The fatality rate in waterborne outbreaks is 0.3-1.6% (Hunter, 1997; Bruce-Grey-Owen Sound Health Unit, 2000).

In the 1990s, evidence that microbial infections are associated with chronic disease started to accumulate. Several waterborne pathogens have been associated with serious sequelae (*i.e.* severe illness or chronic or recurrent disease that appears long after the initial exposure to contaminated water). Examples of sequelae that could potentially be associated with acute waterborne disease include:

- Diabetes, which has been linked to Coxsackie B4 virus (Roivainen *et al.*, 2000; Horwitz *et al.*, 1998).
- Myocarditis, which has been linked to echovirus (Ferreira Jr. *et al.*, 1995; Shanmugam *et al.*, 1986).
- Guillian-Barré syndrome associated with *Campylobacter* spp. (Prendergast and Moran, 2000).
- Gastric cancer, which has been linked to *Helicobacter* sp. (Uemura *et al.*, 2001).
- Reactive arthritis, which has been linked to *Klebsiella* sp. (Ebringer and Wilson, 2000).

With the exception of *Klebsiella*, the association of these microbes with acute waterborne disease has been well established. More remote connections between waterborne microbes and chronic disease has not been fully established, but is highly suspected (Hunter, 1997).

1.1.3 *New pathogens emerge*

Patterns of infection change over time and public health authorities can be faced with newly discovered or emerging pathogens that may be able to overcome the many barriers of the water treatment and distribution systems. Emerging pathogens are defined as microorganisms which are responsible for infectious diseases and which have appeared or increased in occurrence during the past two decades (CDR, 1998). The issue of emerging pathogens came to the fore in the 1990s when water suppliers were shocked by the discovery of previously, essentially, unknown microorganisms, which were responsible for a series of waterborne outbreaks of illness. This is likely to continue into the future as such emergence or re-emergence has been linked to intensive agriculture, increased growth and migration of human populations and climate change (US Department of Health and Human Services, 1998; WHO, 1998). Examples of enteric waterborne emerging pathogens include caliciviruses, *E. coli* O157:H7, *Helicobacter* sp., *Mycobacterium avium* complex (MAC) and the protozoa *Cryptosporidium* sp., *Cyclospora* sp. and *Toxoplasma* sp. This problem requires constant vigilance in terms of what may pose a ‘new threat’ and also constant development with regard to methodologies and techniques for the detection of such threats. As noted by LeChevallier *et al.* (1999a), “*knowledge is the first line of defense toward providing safe drinking water.*”

1.2 **A history of making water safer**

The recognition, in the 1800s that bacteria were agents of disease, along with the development of bacteriology as a science made it possible to use bacteria as tools to evaluate water quality and treatment. Essentially, non-pathogenic, easily detectable microorganisms were used to ‘indicate’ that contamination had taken place and, as such, there was a risk to public health. The need to be able to assess water quality, the fact that the majority of pathogens in drinking water are faecally derived and the ‘moving’ target presented by pathogens resulted in the idea to measure general levels of faecal contamination and the birth of the ‘indicator’ concept.

The presence of heterotrophic bacteria, measured by colony count following growth on a gelatine medium, has been used since the late 19th century to monitor general water quality as well as the function and efficiency of slow sand filtration. Koch (see Box 1.1) postulated that if the effluent of a slow sand filter contained less than 100 bacteria/ml, the water was suitable for drinking and presented no risk of cholera or typhoid. A number of findings paved the way for this development. The book *Microorganisms in Water*, for

example, published by the Franklands in 1894 contained several important findings, including:

- The number of bacteria in water is a measure of pollution, and the number of bacteria in seawater, groundwater and lakewater should be below 100/ml.
- Slow sand filtration reduces the number of bacteria from river water by more than 90% to below 100/ml.

The 100 bacteria/ml level became a standard in many European countries where it was accepted as an attainable objective, while the USA and Canada adopted a 500 bacteria/ml guideline. Although the level of heterotrophic bacteria in drinking water is not related to contamination by pathogens, it is still present in most national laws on drinking water quality as an indicator of the overall quality of the water (van der Kooij, 1993; Anon, 1999).

Box 1.1. Preventing disease transmission: the early years

As early as 400 BC, it was recognised that polluted water was associated with disease (Whitlock, 1954). The first demonstration that disease was transmitted by water took place over 2000 years later, when the first cholera pandemics, which originated in India, struck Europe and resulted in many victims. At the time, it was generally believed that the disease was spread through bad odours. Preventive measures were taken against such odours.

John Snow, a prominent epidemiologist, studied the cholera outbreaks in England. He found in several cases that sewage or night soil had contaminated the drinking water in wells from which cholera cases had drawn water. No cases of cholera were found in families whose water came from uncontaminated wells. In August and September 1854, a cholera epidemic raged in London, with 500 deaths within a range of 250 yards. By careful and systematic analysis, Snow observed that the only common factor was the consumption of water from the Broad Street pump. Two pieces of evidence were telling. Firstly, a man from Brighton came to visit his brother who was ill with cholera. The brother had already died and the man stayed at his house for only 20 minutes, but while he was there he consumed a brandy and water; the next day he died of cholera. Secondly, a lady who lived in another part of London, but preferred the water from Broad Street to her local water provided additional evidence. A carrier collected water at the Broad Street pump and brought it to her house. She and her niece drank the water and died of cholera within two days. The outbreak started on 31 August, it was thought that the well was contaminated by a local cesspit that received water from a household where a baby had developed cholera on 28 August. Snow postulated that transmission of cholera was due to some "morbid material" in cholera faeces that could contaminate drinking water and reproduce in the person who drank that water (Snow, 1855).

Box 1.1. Preventing disease transmission: the early years (continued)

Some 30 years later, Robert Koch developed solid media for the cultivation of some microorganisms and demonstrated that bacteria were the “morbid material” described by Snow. He isolated the bacterium that caused cholera from faeces of infected persons and from water and postulated that consumption of contaminated water could cause cholera (Koch, 1893). A similar mode of transmission was described for typhoid fever by William Budd (1873). In 1880, Eberth discovered that typhoid was produced by *Salmonella typhi* and four years later Gaffky isolated and cultured the organism (Edelman and Levine, 1986). Subsequent interest in the role of water in the transmission of disease initially focused on these two infections.

The demonstration of the link between faecal contamination of water and the transmission of typhoid and cholera focused attention at the end of the 19th century on water quality and the value of its purity. It soon became apparent from several studies that the use of unpolluted water sources or water treatment significantly reduced the incidence of disease and mortality, especially in terms of cholera and typhoid. A good example of which is provided by the Hamburg cholera epidemic of 1892. The city suffered an epidemic in which more than 17 000 people were affected and some 8 500 (13% of the total population) died. The city used water from the river Elbe for drinking and the only purification was sedimentation in three reservoirs. The neighbouring city of Altona used the same river water (with addition of Hamburg’s sewage) but had installed slow sand filtration. Only a small number of people from Altona contracted cholera, and most of them commuted to Hamburg (Koch, 1893). A year later, Mills and Reinke reported improvement in a community’s health after the contaminated source of drinking water had been replaced by a relatively uncontaminated one (White, 1999).

Other studies led to the concept of the faecal indicator. In 1885, Escherich described several microorganisms in the faeces of new-born and suckling babies. This included a motile, rod-shaped microorganism that could cause milk to clot, which he named *Bacterium coli commune* (commonly referred to as *Bacterium* or *Bacillus coli*). He observed that within a few weeks after birth, this bacterium became the dominant organism in the infant colon. Other workers showed that microorganisms consistent with Escherich’s description of *Bacterium coli* were invariably found in faeces. Schardinger proposed in 1892 that, since *Bacterium coli* was a characteristic component of the faecal flora, its presence in water could be taken as “*an indication of the presence of faecal pollution and therefore of the potential presence of enteric pathogens*”.

1.2.1 Refinement

The notion of examining microbial indicators of faecal pollution continued to be developed. Soon after the description of *Bacterium coli*, other Gram-negative, lactose-fermenting bacteria were isolated from stools and water (*Klebsiella* in 1882; *Aerobacter* [now *Enterobacter*] in 1890). Since 1901, these bacteria have been grouped under the name coliforms. The coliforms were defined as Gram-negative, non-spore-forming facultatively anaerobic bacilli

that ferment lactose with production of acid and gas within 48 hours at 35°C. The definition was based on detection methods that allowed for simple isolation and enumeration of coliforms. When this methodology was applied, it soon became apparent that many genera and species that meet the coliform definition are not, or are only rarely, related to faecal contamination (Geldreich *et al.*, 1962; Mack, 1977). Under certain conditions they are also able to multiply in the aquatic environment, thus reducing their value as an indicator of faecal contamination. Already in 1904, Eijkman adopted modifications in the detection methodology that included a higher incubation temperature, which improved the specificity of the indicator. Further modifications of his method have improved the methodology for detecting these thermotolerant coliforms (also called faecal coliforms, although this is not a proper description - see Chapter 2). Although significantly more specific for faecal contamination, this parameter also had similar shortcomings. It became apparent that other bacteria (mostly *Klebsiella*), which meet the criteria for thermotolerant coliforms, originate from non-faecal environments, such as paper mill or potato-industry wastewater and other high carbohydrate wastewater (Dufour and Cabelli, 1975).

It was eventually shown that among the thermotolerant coliforms *Escherichia coli* is the preferred microbial indicator of faecal pollution (Dufour, 1977), as it is the only member of the coliform group that is invariably found in faeces of warm-blooded animals and it outnumbers the other thermotolerant coliforms in both human and animal excreta. Other microorganisms have been suggested as microbial indicators of faecal pollution (see Chapter 2), such as enterococci (previously named faecal streptococci), coliphages and sulphite-reducing clostridial spores.

Although faecally derived coliforms, thermotolerant coliforms and/or *E. coli* have several drawbacks, they have historically been very useful and they are, undoubtedly, the most commonly used microbial parameters for testing drinking water quality. Their use has led to significant improvement in the safety of drinking water world-wide and they have been adopted in the World Health Organization (WHO) drinking water quality guidelines and all national drinking water quality standards. One of the main reasons for their success is the ease of the assay. In contrast with the approach to chemical contaminants of water, microbiologists soon realised the complexity that would be involved in trying to assay water for all enteric pathogens. As the common source of these pathogens was faecal pollution, microbiologists aimed for a universal microbial indicator of faecal contamination.

The ease and low cost of the assay means that it is possible to test water bodies frequently. Faecal contamination varies and it is likely that peak contamination will present the highest health risk. The importance of frequent testing has long been widely recognised:

“It is of the utmost importance for the control of the hygienic quality of the water supply that the bacteriological examination of both the water entering the distribution system and the water in the distribution system itself be carried out frequently and regularly” (WHO,1976);

and

“It is far more important to examine a (water) supply frequently by a simple test than occasionally by a more complicated test or series of tests” (Anon, 1969).

1.3 Defining the role of the indicator concept

The traditional role of indicator parameters in drinking water was as an index of faecal pollution and, therefore, likely health risk (see Box 1.2). The original microbial parameters were all bacteria that, to a greater or lesser degree, were derived from faecal contamination. Faecal-oral illness, however, is not only caused by enteric bacteria but may result from infection with pathogenic viruses or protozoa. The viruses and protozoa have different environmental behaviour and survival characteristics to bacteria, which means that faecal bacteria are not always an adequate indicator of their presence or absence. This is especially true for disinfected drinking water, as bacteria are very sensitive to disinfectants while viruses and parasites can be extremely resistant. Thus, the basic premise that the concentration of indicator organisms should be related to the extent of faecal contamination and by implication to the concentration of pathogens and the incidence of waterborne disease can not be maintained (Pipes, 1982). The roles of the indicator concept, however, are gradually expanding as is the number of possible indicator parameters. There is now a need better to define these specific roles such as in source assessment, validation of the drinking water treatment process, operational and routine monitoring as well as the traditional verification of the end product (see Chapter 1.4).

Box 1.2. Indicator concept and criteria

Microbial indicators of pollution have been in use for decades. They were originally developed as measures of faecal pollution of source waters and subsequently the same organisms were applied to measure efficiency of treatment and post-treatment contamination and deterioration. Mossel (1978) credited Ingram with recognising the different roles to which so-called indicators were being applied and proposing that the term 'indicator' should be used for the assessment of treatment process effectiveness, while 'index' should be used for the original role of indicators, that is as a measure of faecal pollution. The search for microbial faecal indicators was based on several criteria that were well accepted by the scientific community, but were based on the assumption that the same organism would serve as both index and indicator. The criteria were:

- The indicator should be absent in unpolluted water and present when the source of pathogenic microorganisms of concern is present.
- The indicator should not multiply in the environment.
- The indicator should be present in greater numbers than the pathogenic microorganisms.
- The indicator should respond to natural environmental conditions and water treatment processes in a manner similar to the pathogens of concern.
- The indicator should be easy to isolate, identify and enumerate.

Over time, the following criteria have been added to the original list:

- The test should be inexpensive thereby permitting numerous samples to be taken.
- The indicator should not be a pathogenic microorganism (to minimise the health risk to analysts).

The detection of pathogenic microorganisms is not normally associated with the indicator concept, as each pathogen essentially represents only itself and its absence is not an indication of the absence of other pathogens. The only current usage of a pathogen that meets the indicator concept is the detection of cryptosporidial oocysts as an indicator of treatment efficiency in the UK.

The list of microbial parameters has grown with time and these have been applied to a variety of environments, although in some instances their application strayed away from the original concept (i.e. relationship to faecal pollution), with indicators being used inappropriately.

Throughout this book, guidance is given on the best use of the various microbial and non-microbial parameters to fulfil the criteria for specific purposes. These purposes are outlined below, and in many cases may require the use of more than one microbial and/or non-microbial parameter.

- Index (or indicator) of faecal pollution in ambient waters not receiving any treatment (including water abstracted for drinking water purposes).
- Index (or indicator) of faecal pollution of groundwater.
- Indicator of treatment removal or disinfection efficiency.
- Indicator of recontamination of treated water within the distribution system.
- Models for pathogenic microorganisms.

Box 1.2. Indicator concept and criteria (continued)

In 1991, Waite reviewed the evolution of water bacteriology and proposed that the terms 'index' and 'indicator' should be adopted as originally suggested by Ingram. An index organism is, thus, any organism whose presence points to the possible occurrence of pathogenic similar organisms. Whereas an indicator organism is one where its presence represents a failure of Good Manufacturing Practice affecting the final product. The concept of index and indicator can be extended to cover non-microbial parameters.

Since an indicator is used as a surrogate to assess the efficacy of treatment it is preferable not to use the term in isolation, but in conjunction with what treatment is being considered (e.g. process indicator, disinfection indicator, turbidity as an indicator of filtration efficiency). Similarly, the term index may be qualified (e.g. an index of faecal pollution, conductivity in ground water as an index of deterioration).

1.3.1 Current practice

The basic idea behind the use of traditional faecal indicator parameters (*i.e.* when they are absent, pathogens are absent), while not universally valid, is still applied and useful today if the parameter is chosen correctly. The most common uses are for monitoring drinking water at the tap and as it leaves the treatment works. Despite the shortcomings that have been recognised for some time, in many jurisdictions this is still done by analysing for the absence of coliforms, with or without complementary testing for *E.coli* or thermotolerant coliforms. Once the water is distributed, a positive coliform test may indicate the presence of faecal contamination but could also be derived from a non-faecal origin. Thus, the test that is used as the primary warning of faecal contamination gives very little information on the presence or absence of a health risk. Confirmation of the faecal origin is embedded in most regulations and requires testing for thermotolerant coliforms or *E. coli*. WHO (1993) indicates that *E. coli* is the parameter of choice for monitoring drinking water quality (with thermotolerant coliforms as an alternative). Enterococci and sulphite-reducing clostridia are also used as additional parameters of faecal contamination or to monitor the integrity of the distribution or storage system. Less common is their use to classify the source water, with the level of treatment to produce safe drinking water being set accordingly (more details on the use of indicator parameters for specific purposes is given in Chapter 2).

The major problem, in terms of public health protection is that (for the most part) monitoring the safety of drinking water is reactive, in the sense that any event or breakdown in the system can occur many hours and sometimes days, before it is detected by monitoring for any of the microbial parameters. This is related to both the nature of the microbial testing, which currently requires at least a day to produce a result, and also to the monitoring strategy, which has traditionally focussed on water as it leaves the treatment works and on the distribution system.

1.3.2 *New challenges*

While the use of (thermotolerant) coliforms and enterococci as indices of faecal pollution has proved successful in preventing the spread of waterborne cholera and typhoid, in the 1960s a new challenge to public health was identified. It was increasingly recognised that enteric viruses, such as hepatitis A and other enteroviruses, could also be transmitted through drinking water (Anon, 1999). Viral contamination of water also originates from pollution with human excreta, but the nature of viruses is very different from that of bacteria. They are much smaller and therefore less likely to be removed during filtration or soil passage and their resistance to disinfection is typically greater. The occurrence of outbreaks of viral illnesses associated with drinking water meeting the coliform standards indicated that coliforms were an inadequate parameter to assess the virological quality of treated drinking water (Berg and Metcalf, 1978; Petrilli *et al.*, 1974; Melnick and Gerba, 1982). Water microbiologists sought suitable alternative microbial parameters and found several groups of viruses that infect bacteria, known as bacteriophages (phages), which have a similar size and also structure characteristics to human pathogenic viruses. These were suggested as being appropriate models for the potential presence of viruses and for their survival and behaviour in the environment, as well as their removal and inactivation by water treatment and disinfection processes (Grabow *et al.*, 1984; Havelaar *et al.* 1993).

More recently, a further challenge was identified with the outbreaks of intestinal illness due to the protozoa *Giardia* sp. and *Cryptosporidium* sp. As with viruses, outbreaks have occurred without any indication, from the coliform testing, that water quality was compromised (Barrell *et al.*, 2000). It was recognised that the failure of the coliform bacteria standard was due to the more robust nature of the protozoa to disinfection, resulting in inactivation of the indicator bacteria but not the viral and protozoan pathogens. Spores of the bacterium *Clostridium perfringens* and sulphite-reducing clostridia, which are also known to be robust and resistant to disinfection have been proposed as alternative microbial parameters for such protozoa. Other indicator parameters that have been suggested to assess treatment efficiency for the removal of pathogens are aerobic spores (Chapter 2, USEPA, 2000).

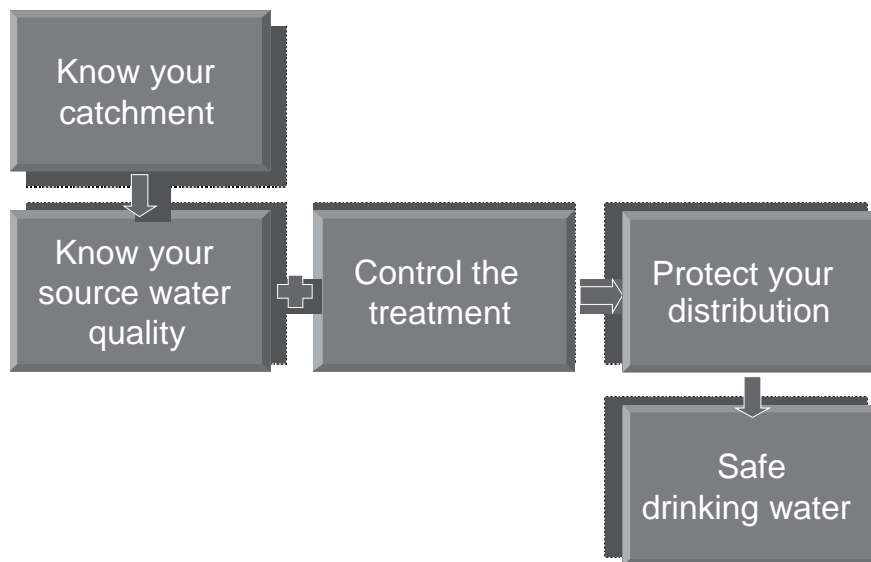
As mentioned earlier, a drawback to the current use of microbial parameters, in terms of public health protection, is the reliance on end-product monitoring. End-product monitoring cannot always safeguard health but acts to verify (or not) the effectiveness of the treatment barriers. This can provide important management information (see Chapter 7) and is a useful check, which will determine any performance deficiency and also allow an assessment of any

corrective procedures. Its main purpose, therefore, is to verify the efficiency of treatment and disinfection and detect post-treatment contamination.

While traditional microbial parameters have proved useful and still have an important role to play, monitoring of different aspects of the supply chain as well as possible health effects requires the use of different applications of traditional microbial parameters, different parameters and different approaches. There are two major initiatives that move to address this challenge:

- The development of water safety plans (see Box 1.3).
- The assessment of risk at all stages between catchment and consumer (Figure 1.1).

Figure 1.1. “Catchment to consumer” approach to risk management of the safety of drinking water



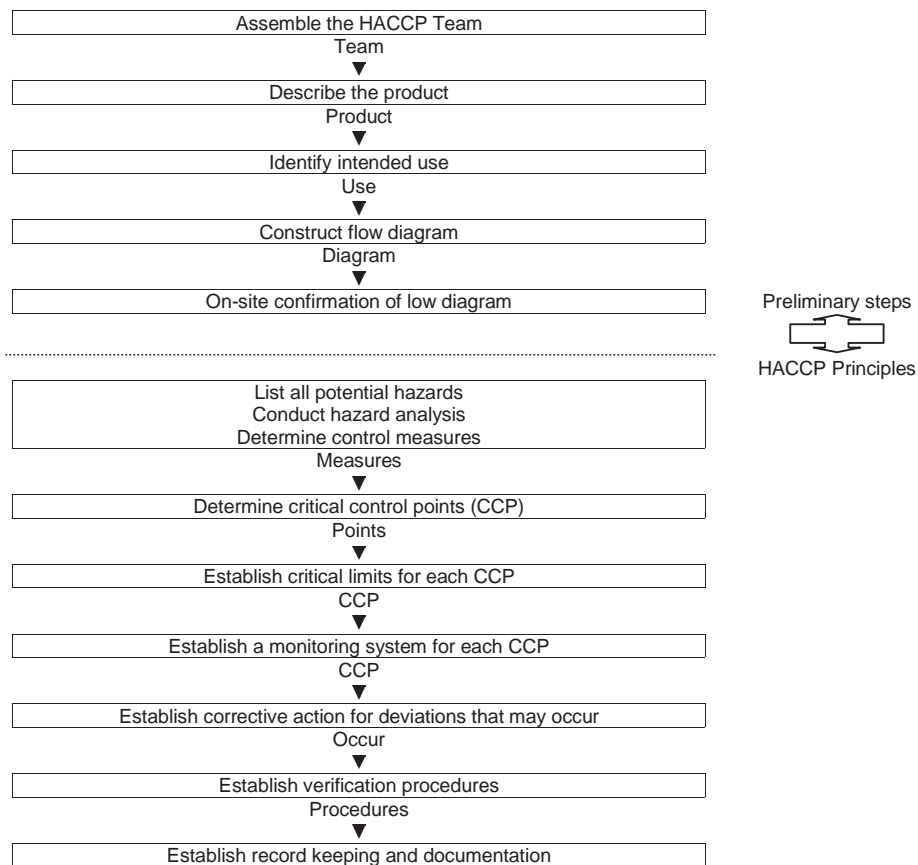
In addition, the development of rapid testing and molecular techniques for microbial parameters and pathogen detection (Chapter 1.5) may play a supporting role, particularly in risk assessment (Chapter 3) and outbreak investigation. For example, molecular techniques have, in a number of cases, allowed the identification of the source of contamination in waterborne outbreaks (Chapter 7).

1.4 Emergence of a new paradigm: “Due diligence”

The concept of due diligence, which means the prevention of foreseeable harm at reasonable cost, takes a significant step in changing the “reactive and sanctioning” paradigm under which suppliers (including water suppliers) operate. Demonstration of due diligence requires showing that all reasonable measures have been taken in advance to prevent the occurrence of negative health consequences. Thus, when a potentially adverse event is identified, a precautionary approach should be used. One such approach, which came out of the space programme in the 1960s, is HACCP (Hazard Analysis Critical Control Point), illustrated in Figure 1.2, which has been adapted for drinking water use and incorporated in ‘Water Safety Plans’ (Box 1.3 and Figure 1.3).

Figure 1.2. Steps in the development of a HACCP Plan

(Adapted from Deere *et al.*, 2001)



Box 1.3. Water safety plans for drinking water supply

The basis of ensuring water safety has five key components:

1. Water quality targets based on public health protection and disease prevention.
2. System assessment to determine whether the water supply chain (up to the point of consumption) as a whole can deliver water of a quality that meets the defined targets.
3. Monitoring the steps in the supply chain that are of particular importance in securing safe drinking water.
4. Management plans describing actions to be undertaken from normal conditions to extreme events.
5. Systematic independent surveillance that verifies that the above are operating properly.

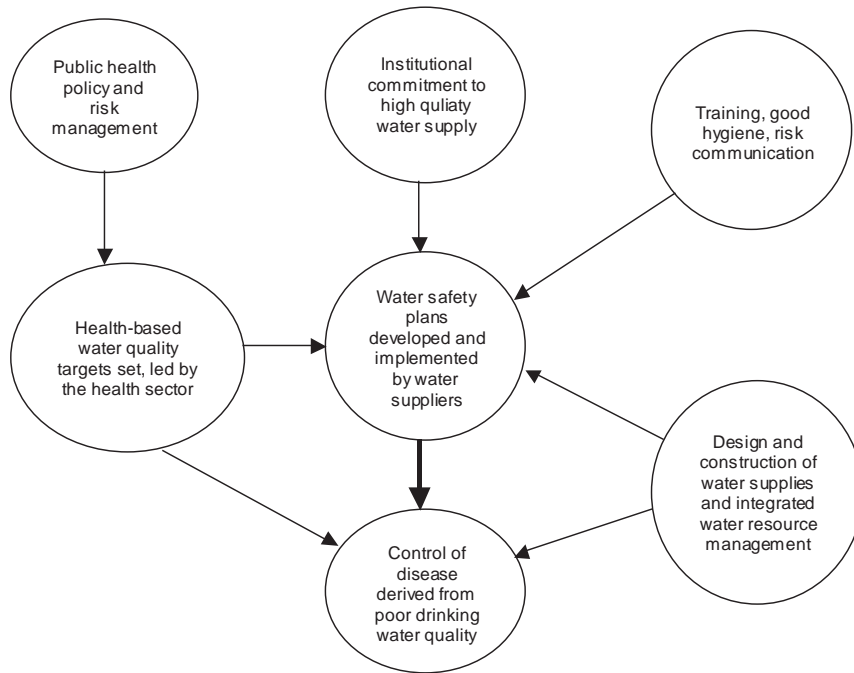
The management plans developed by water suppliers can be best termed a Water Safety Plan (WSP). The control of microbiological and chemical quality of drinking water requires the development of WSPs which, when implemented, provide the basis for process control to ensure pathogen and chemical loads are acceptable. Implicit within this process is that a tolerable disease burden has been identified at national and local levels and that water quality targets have been established for each type of technology used.

The delivery of 'safe' water, therefore, involves actions by a variety of stakeholders, as illustrated in Figure 1.3.

Water quality targets should be set to improve public health. Water suppliers have a basic responsibility to provide safe water and would be expected to undertake actions meeting points 2–4 above. The final component would normally be fulfilled by a regulatory body, which may in practice be the health, environment or local government sectors. All these processes are important in the delivery of good drinking water quality and are the subject of other texts linked to the WHO Guidelines for Drinking water Quality.

Figure 1.3. Protecting public health through ensuring drinking water quality

(Davison *et al.*, 2002)



In a drinking water application, HACCP is a source-to-tap system. Microbial safety is safeguarded through knowledge of the (variations in) quality of the source water, control of the treatment process and the integrity of the distribution or storage system. No single microbial (or non-microbial) parameter is adequate to determine if all steps in this system are working properly in all circumstances. Chapters 2, 4, 5 and 6 outline which parameters are most appropriate at each stage. The resulting data can be integrated into risk assessment models (quantitative or qualitative) or be used to complement epidemiological methods, as outlined in Chapter 3.

1.5 Direct pathogen testing

The discovery and characterisation of a significant number of gastrointestinal pathogens led to the development of a wide array of new methods to detect and identify them (Anon, 1999; Hurst *et al.*, 2001). In addition, methods for concentrating these pathogens from natural waters were developed in the 1970s and 1980s and applied to waters potentially contaminated by faecal pollution. Results indicate that viruses (Payment and Armon, 1989; Grabow *et al.*, 2001) and parasites (LeChevallier *et al.*, 1991) may still be present in otherwise apparently safe drinking water. This, together with the recognition that illness can be associated with drinking water that meets regulatory safety standards indexed by traditional bacterial parameters (Payment *et al.*, 1991, 1997), raised questions about the safety of drinking water (LeChevallier *et al.*, 1999 a,b). The United States of America extended the principle of “pathogen-free water” to a goal of zero level of pathogens, while recognising that, in reality, this implied a judgement on the level of treatment necessary to achieve minimal or tolerable risk. To determine these treatment goals, a survey of source water was conducted using standardised methods to obtain data on pathogen occurrence. The data revealed the difficulties associated with such testing even for gathering data on occurrence (Chapter 4; Rosen and Ellis, 2000; Allen *et al.*, 2000).

In the United Kingdom, testing for *Cryptosporidium* oocysts became mandatory at some sites (established on the basis of a risk assessment). The UK approach requires continuous sampling of a proportion of treated water as it leaves the treatment works, in view of the fact that spot sampling for pathogens would be liable to miss treatment deficiencies of short duration. Data from the current monitoring programme are used to verify compliance with a treatment standard of removal of oocysts to less than one oocyst in ten litres of treated water (HMSO, 1999).

In addition to what is required by regulations, a number of water suppliers have initiated some form of direct pathogen testing (Allen *et al.*, 2000). Pathogen testing can be a useful tool for sanitary surveys of catchment areas, for setting treatment goals, for laboratory or pilot-scale demonstration of the efficacy of existing or new water treatments and for investigation of outbreaks. In general, pathogen testing helps to sharpen practices for providing safe drinking water, and detection of (viable) pathogens in drinking water is a strong trigger for remedial action. Pathogen testing can be done in addition to standard monitoring, but it is not a simple monitoring tool. The methodological constraints require that a clear idea as to what is to be achieved by the exercise should be formulated prior to undertaking pathogen testing (Havelaar, 1993) and subsequent results should be interpreted with caution. Low levels of enteric

viruses and protozoan parasitic cysts have been found in drinking water in the absence of reported outbreaks (Gerba and Rose, 1990; Payment and Armon, 1989). This could be related to a lack of infection, mis-identification, acquired immunity or asymptomatic infections (Allen *et al.*, 2000; Issac-Renton *et al.*, 1994; Gerba and Rose, 1990; Payment *et al.*, 1991, 1997). Experience in Sydney (Australia) and Wyoming (USA), shed some light on the public, political, economic and legal implications of such findings even in the absence of any detectable health effects (Allen *et al.*, 2000). These cases show that unless it is known how to interpret the results and an emergency plan is in place to react to a positive result, the reaction may be inappropriate.

Methods for detecting pathogens in water are mostly still in the developmental stage (Chapter 8):

- Their sensitivity is still poor. The methods for detecting the pathogen are usually very sensitive, but because of the low level of pathogens in water, large volumes need to be analysed and as the detection methods can only effectively process small volumes a pre-concentration step must be undertaken.
- Only a few of the multitude of possible pathogens are currently detectable. Given that water may contain hundreds of different pathogens, and these may vary over time, the question of which pathogens to look for remains. Pathogen testing methods are relatively specific and will not detect all pathogens present. Molecular methods, coupled with high throughput parallel processing and bio-informatics, hold the promise of detecting a wider range of microorganisms, but are not yet practicable. One suggestion has been to search for the most significant pathogen as an indicator of treatment efficiency. The United Kingdom has adopted this approach and tests for *Cryptosporidium* oocysts in treated water from selected water treatment works.
- Analysis of water samples for pathogens requires a specialised laboratory, highly trained personnel and appropriate bio-safety containment. In industrialised countries few laboratories outside the clinical setting meet these requirements, and in many other countries such facilities do not exist. Pathogen testing may require growing and manipulating pathogens, thus the potential risk to analysts needs to be taken in consideration.
- Although some pathogens can be tested rapidly, most pathogen sampling and detection methods still have a time-to-(verified)-result of several days. Pathogen testing of treated water, therefore, does not escape the problems identified with end-product testing using traditional bacterial parameters, i.e. they signal that something is wrong after the problem has occurred.

These methodological limitations advocate the use of great care in the interpretation of results from pathogen testing. Any positive result may indicate water unsafe to drink and can be used to estimate the level of risk to consumers. Positive results should be used only in a well-managed, risk-based decision-making process. Negative results should always be viewed with some scepticism given the large number of possible pathogens and should not be used as an excuse for complacency.

Levels of pathogen testing may vary from simple occasional tests or planned experiments to routine monitoring of source water as well as treated water. However, if pathogen testing is included among the parameters, it is important that this is not done at the expense of essential basic monitoring. If pathogen testing is considered acceptable, where should it be done? Pathogen testing for an initial sanitary survey of source water is well accepted. After treatment at the waterworks, samples should be negative for infective pathogens, but what volume should be tested? How many samples should be taken to ensure that they are statistically representative? Familiarity with water treatment indicates that what is most to be feared is transient, short-lived failures of the system, which are difficult to detect. Given the high cost of pathogen testing – and this will not change in the near future – is the cost of routine pathogen testing justified? A good set of microbial and non-microbial parameters is probably more cost-effective.

Analysis of samples of distributed water presents a similar challenge. The objective is to detect recontamination of the water in the distribution system. How many samples should be taken, where should they be taken and for what pathogens? A good indicator of recontamination or disinfectant demand is probably more cost-effective as data can be obtained inexpensively for a large number of samples.

1.5.1 Dose-response relationships for pathogens

Determination of the effect of exposure to (different levels of) pathogenic microorganisms (*i.e.* dose-response functions) has allowed the design of a risk-based approach (Chapter 3), analogous to that taken against the risk of toxic chemicals in drinking water. Because the complete absence of pathogens in drinking water (zero risk) cannot currently be assured, this criterion has been replaced by the definition of an acceptable or tolerable risk level (Hunter and Fewtrell, 2001). Such a risk-based approach was developed by North American researchers in conjunction with the US Environmental Protection Agency (Haas, 1983; Rose and Gerba, 1991; Regli *et al.*, 1991). In this approach, a risk level of one infection per 10 000 persons per year is regarded as the acceptable

maximum for pathogens in drinking water. This was based on findings that during each reported waterborne outbreak of giardiasis, at least 0.5 % of the population (*i.e.* 50 or more per 10 000 people) were infected. Because public water supplies should provide greater protection from waterborne disease, water treatment should ensure less than one case of microbial illness per year per 10 000 people as a reasonable goal (USEPA, 1989). *Giardia* was chosen as the target organism because it is more resistant to disinfection than most other pathogens (Regli *et al.*, 1993). This approach has been adopted or is being considered by a number of countries. In the Netherlands, for example, guidelines have been issued for maximum acceptable pathogen concentrations in drinking water, based on the 10^{-4} infection risk level.

1.5.2 Molecular technologies

Currently (for the most part) microbial parameter detection involves sampling and filtration followed by cultivation of the chosen microorganism on selective media and then colony counting or, in some cases, the demonstration of growth (*e.g.* presence-absence tests) – a process that can take 24 to 72 hours and may not pick up a number of microorganisms. The last two decades of the 20th Century, however, saw the development of molecular biology and the promise for rapid testing (less than eight hours). This resulted in techniques, such as polymerase chain reaction (PCR), for the rapid, sensitive and specific detection of index/indicator microorganisms and pathogens. In the field of health-related water microbiology, this has allowed the development of detection methods for non-culturable viruses, such as Norwalk-like viruses. While the conventional *Cryptosporidium* detection methods do not discriminate between human-pathogenic and non-pathogenic oocysts, the specificity of PCR and subsequent verification methods (hybridisation, sequencing) have allowed much more specific detection of pathogenic species or genotypes within *Cryptosporidium* (see Chapter 7). Rapidity is another characteristic of PCR and related molecular technologies (see Chapter 8). Several researchers have developed PCR techniques for the rapid detection of *E. coli* and coliforms, which make detection possible within several hours (Bej *et al.*, 1991; Fricker and Fricker, 1994).

One of the challenges for molecular methods is to assess the viability/infectivity of the detected microorganisms, as currently they detect the presence of a nucleic acid sequence, which may have originated from a dead organism or even from DNA that has not been decomposed in the aquatic environment. Culture techniques provide this information as only viable microorganisms are detected. Several techniques or combinations of techniques are now available to overcome the viability/infectivity problem. Examples are

the use of inducible mRNA as target for RT-PCR (reverse transcriptase – polymerase chain reaction) or use of viability/infectivity methods to ‘pre-culture’ the target organism prior to PCR detection, such as the cell culture PCR method for *Cryptosporidium* and viruses (Chapter 8; Spinner and DiGiovanni, 2001), although pre-culture increases the overall assay time.

The taxonomy of microorganisms is now primarily based on genotyping, rather than on phenotypic characteristics. This genetic taxonomy allows the rapid characterisation and comparison of genotypes. This has been useful in investigations of outbreaks to determine the similarity of isolates from patients and suspected outbreak sources and in tracking the sources of contamination of a watershed or drinking water (Chapter 7; Kuhn *et al.*, 2000).

Advances in computer, chip, laser and optical technology have provided, and are providing, new opportunities for the detection and identification of microorganisms. New equipment has been used primarily in the research area, but several technologies are currently used in practice, such as flow cytometry and cell sorting, confocal laser scanning microscopy and laser scanning. Many methods are now being developed but, as yet, all are expensive as the equipment capital investment is high. One interesting development is the combination of computer chip technology and molecular microbiology, which should allow automated testing of multiple pathogens with a single DNA chip array (Chapter 8). The introduction of these technologies may allow rapid automated detection of (pathogenic or index/indicator) microorganisms in the near future.

The challenges that remain for these new methods are:

- Quantification. The quantitative aspects need to be improved as current molecular methods are, at best, only semi-quantitative.
- Infectivity. The viability and infectivity of the detected microorganisms is still uncertain.
- The “concentration issue”. Detection of (especially pathogenic) microorganisms in water requires assaying large volumes (0.1-100 litres or more), while the new technologies currently work with small volumes (0.00001-0.001 litres). This requires concentration methods that introduce recovery losses.
- The skills and novel infrastructure issue (both in personnel training and equipment). Further implementation of these technologies in practice ideally requires further simplification and also automation.

- Cost. The cost is still high and currently not amenable to frequent daily testing within the budget constraints of small water suppliers.

1.6 Information needs

The provision of safe drinking water is rarely the concern of a single body and in addition to the water supplier is likely to involve national, regional and/or local government, water authorities and public health authorities. In some cases, such as catchment control, there may be international involvement.

Traditionally, the main reason for monitoring water quality has been to verify whether the observed water quality is suitable for an intended use (in this case human consumption). Understanding the reasons for data collection (*i.e.* the purposes of the monitoring) helps to ensure that the data collected are appropriate for the management uses for which they are intended (Bartram and Helmer, 1996; Makela and Meybeck, 1996). The different information needs are described below.

1.6.1 Regulation

Many agencies, including the WHO (WHO, 1976, 1997) identify two complementary roles in drinking water monitoring: quality control by the supplier and independent surveillance by the regulatory body. There is an evident complementarity between them, but the merging of the two is inappropriate because of the conflict of interests that would emerge. Nevertheless, in recent years experience with increased data sharing, generation of data by third parties and audit-based approaches has begun to accumulate and contributes to minimising duplication of effort. Where regulation is based upon monitoring and analysis, it does not typically mimic the requirements of the water supplier but can be proactive and can single out problems, such as the areas of distribution most likely to cause problems or known to be problematic.

The regulatory agency is ultimately responsible for determining the required level of safety of drinking water. It does so by setting water quality standards and ensuring that water suppliers meet them. While requirements vary widely from country to country, compliance is generally based on standards (such as the European Community Directive 98/83/EC; Anon, 1998; WHO, 1993) that are intended to protect public health. Standards are typically specified in terms of sampling frequency and distribution (at fixed and/or variable points) in relation to the population supplied and/or high-risk points. Parameters typically include simple physical and chemical tests (disinfectant

residual, turbidity, etc.) and relatively frequent monitoring, with less frequent microbiological testing for indicators of faecal pollution (and sometimes other microbiological parameters) with specific follow-up requirements when standards are not met.

These standards clearly aim to limit the transmission of infectious disease through drinking water but they also influence the allocation of community resources to drinking water treatment. Drinking water is only one of the vectors of enteric infectious diseases. To be able to optimise the allocation of available resources to protection of drinking water, the regulator requires information on the contribution of drinking water to the overall disease burden of the population. At the next level of detail, policy makers require information about the most important threats to the safety of drinking water so that they can focus risk management options on the most relevant threats (Fewtrell and Bartram, 2001). For example, a system with a relatively high risk of post-treatment contamination should primarily focus on reducing this risk, rather than on the efficiency of source protection or on treatment.

1.6.2 Water supplier

Water suppliers require information on the microbiological quality of their source water. Information on the contamination level of source water is the basis for the design of an adequate treatment system. Information on sources of pollution in the catchment area of abstraction sites gives both an indication of the level of contamination that may be expected and potential risk events (such as heavy rainfall leading to agricultural run-off). A catchment survey will also yield information on opportunities for catchment control. This may not be the domain of water suppliers, but allows them to choose between installing treatment barriers or trying to implement source protection measures. In the design phase, a catchment survey will aid in the selection of the best site for abstraction (Chapter 4).

A water supplier also needs to know the efficiency of the treatment processes in eliminating microorganisms; initially, in the design phase, to be able to design an adequate treatment system and subsequently, in the production phase, to ensure its adequate operation. In the latter phase, detailed information on the elimination of microorganisms during the different operational phases may help to optimise the efficiency of treatment processes (Chapter 5; LeChevallier and Au, 2002).

To determine if a treatment is adequate and drinking water is safe, a water supplier also needs water quality targets (Box 1.3; Fewtrell and Bartram, 2001).

In the risk-based approach, water quality targets should be derived from a tolerable risk level. Water quality targets are usually set by national authorities, which should decide on the tolerable risk level and derive the water quality targets that result in compliance with this risk level. In the risk-based approach, targets could also be a maximum concentration of a pathogen but these are generally not intended to be a measured target.

For process operation, water suppliers rely on process parameters such as coagulant dose. To ensure that treatment is eliminating microorganisms adequately every hour of the day, they need information on the relationship between the operational parameters and the elimination of microorganisms (Chapter 5; LeChevallier and Au, 2002). Finally, the company/agency that distributes the water to the consumer needs information about the water-quality changes that occur during distribution so as to be able to detect and respond to any unacceptable deterioration of water quality (see Chapter 6 and Ainsworth, 2002).

1.6.3 Public health agencies

In most countries, public health agencies are no longer directly responsible for the management of water supply and distribution systems. Because of this, very few public health specialists will expect to see routine water quality data on a regular basis. On the other hand, most public health surveillance will be directed at detecting cases of infection in the community. Should outbreaks be detected that implicate the water supply, review of routine water quality monitoring data will be part of the subsequent outbreak investigation (see Chapter 7). Screening of the water supply for pathogens may also be undertaken in any investigation.

The most notable pathogen-monitoring scheme for a public water system, as mentioned previously, was introduced in the UK for *Cryptosporidium*. In England and Wales, it is now a criminal offence to supply water containing ≥ 100 *Cryptosporidium* oocysts/1 000 litres and supplies deemed to be at high risk have to be monitored continually (HMSO,1999). The standard chosen was based on operational rather than public health grounds and relating counts to public health risk has been difficult (Hunter, 2000).

With an overall responsibility towards public health, interest and indeed responsibility does not end at the quality of water leaving the water treatment works. Indeed, most public health interest may concern occurrences after the water supply enters the household and marginal or disadvantaged populations where no formal supply agency exists. Small community and particularly rural

water supplies are especially problematic and are of concern in countries at all levels of socio-economic development. While factors such as parameter selection may not be very different for such areas, overall approaches to monitoring are extremely problematic and innovative approaches are required for effective action (Bartram, 1998; Chapter 6).

Investigation of events of public health concern may be triggered by disease surveillance, data from water suppliers or others undertaking monitoring or through informal means. Since the effects are delayed, such investigation presents special problems and is an area where newer analytical methods, such as those outlined in Chapter 1.5.2, may make a particular contribution. This aspect is addressed further in Chapters 7 and 8.

1.7 The new approach: Total System Approach to Risk Management

The combination of these developments is leading towards a risk-based approach to ensure the safety of drinking water (see Box 1.3). Traditionally, drinking water was regarded as safe when monitoring of the treated water did not show the presence of coliforms in daily samples of drinking water. In quantitative risk assessment, the safety of drinking water is demonstrated by collecting quantitative information on quality of the source water, efficiency of treatment and integrity of the distribution system. This has the benefit of providing water suppliers and relevant agencies with insight into the level of consumer protection and providing information on the strengths and weaknesses of the systems installed to protect drinking water quality. End-product monitoring remains important for long term verification of the control system.

A long-established principle in drinking water risk management is not to rely on a single barrier against pathogenic microorganisms, but to use a multiple barrier approach. This implies not only multiple barriers in water treatment, but a more encompassing approach from source to the consumer's tap (see Figure 1.1). As suggested above, in order to design an effective risk management strategy, information is required on:

- Sources of contamination in the catchment area. This would also include the relative contribution of these sources to the overall contamination level. Knowledge of the nature, location and contribution made by individual sources of contamination means that it is possible to predict peak events and to determine effective catchment control measures.
- Microbiological quality of the source water and its variation. The quality of the source water, both under average conditions and during peak

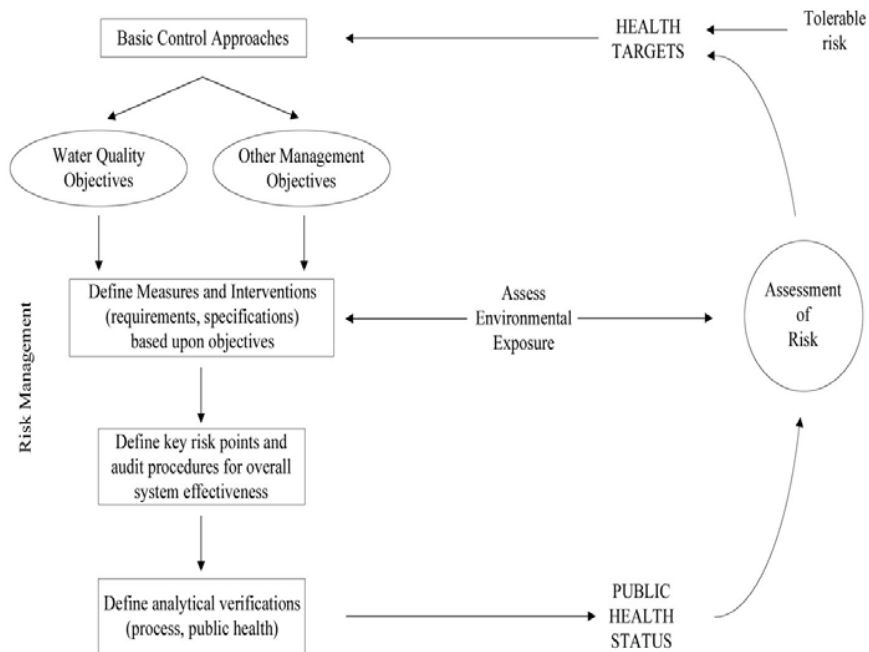
events, determines the required level of treatment. Information on the level of contamination at the abstraction point can be used to design appropriate treatment systems and to design operational procedures to deal with most eventualities.

- Efficiency of the water treatment process in eliminating microorganisms and its variation. Information is required on the effectiveness of different treatment processes (as unit process and in combination with other processes) in eliminating pathogens (Haas, 1999; USEPA, 1991).
- Sources and risk of post-treatment contamination.

Such considerations are essentially the risk management components of an even larger framework or approach, where consideration is also given to tolerable risk, water quality targets and public health status, as illustrated in Figure 1.4.

Figure 1.4. Decision-making framework

(Adapted from Bartram *et al.*, 2001)



Although some index/indicator parameters can serve multiple purposes, no single parameter can fill all the information needs. Later chapters give guidance on the application of parameters for specific information needs: catchment protection, source water quality assessment, assessment of treatment efficiency, monitoring the quality of drinking water leaving the treatment facility and in the distribution system. The emphasis is on their use for demonstrating the safety of drinking water and as basis for risk management decisions.

1.8 Summary

Drinking water that contains pathogenic microorganisms may cause illness and, as such, it is important to have some measure (or measures) that establishes whether it is safe to drink. For the most part there are too many different pathogens to monitor and as the majority of pathogens are derived from faecal material the idea of using non pathogenic bacteria as an index of faecal pollution was developed. Initially only a few such parameters were used, but now there are more techniques and methodologies available. It is possible to monitor a wide range of index/indicator parameters (microbial and non-microbial) and also pathogens and there is a move towards using a variety of different parameters throughout the water production process and, indeed, a catchment to consumer approach to water safety plans. New methods are constantly being developed, ranging from increased pathogen detection to more real-time microbial and non-microbial parameter monitoring. The development of new and improved methodologies, along with the need for vigilance with regard to emerging hazards, results in the need for frequent re-evaluation of the best approaches and indicator parameters.

REFERENCES

- Ainsworth, R.A. (2002) Water quality changes in piped distribution systems. World Health Organization.
- Allen, M.J., Clancy, J.L. and Rice, E.W. (2000) Pathogen monitoring – old baggage from the last millennium. *Journal of the American Water Works Association* **92**(9), 64-76.
- Anon (1969) Reports on Public Health and Medical Subjects No. 71. Her Majesty's Stationery Office, London.
- Anon (1998) European Council Directive 98/83/EC of 3 November 1998 on the quality of water intended for human consumption.
- Anon (1999) *Waterborne Pathogens*. AWWA Manual of Water Practices, M48. American Water Works Association, Denver, Colorado.
- Barrell, R.A.E., Hunter, P.R. and Nichols, G. (2000) Microbiological standards for water and their relationship to health risk. *Communicable Disease and Public Health* **3**(1), 8-13.
- Bartram, J. and Helmer, R. (1996) Introduction. In: *Water Quality Monitoring. A practical guide to the design and implementation of freshwater quality studies and monitoring programmes*. Bartram, J. and Balance, R. (Eds.) E & FN Spon, London. pp. 1-8.
- Bartram, J., Fewtrell, L. and Stenström, T-A. (2001) Harmonised assessment of risk and risk management for water-related infectious disease: an overview. In: *Water Quality: Guidelines, Standards and Health. Assessment of risk and risk management for water-related infectious disease*. Fewtrell, L. and Bartram, J. (Eds.) IWA Publishing, London. pp. 1-16
- Bartram, J. (1998) Effective monitoring of small drinking water supplies. In: *Providing Safe Drinking water in Small Systems*. Cotruvo, J., Craun, G.

- and Hearne, N. (Eds.) Lewis Publishers, Boca Raton, Florida. pp. 353-366.
- Bej, A.K., Dicesare, J.L., Haff, L. and Atlas, R.M. (1991) Detection of *Escherichia coli* and *Shigella* spp. in water using the polymerase chain reaction and gene probes for uid. *Applied and Environmental Microbiology* **57**, 1013-1017.
- Berg, G.T. and Metcalf, T. (1978) Indicators of viruses in waters. In: *Indicators of Viruses in Water and Food*. Ann Arbor Science.
- Bruce-Grey-Owen Sound Health Unit (2000) The investigative report on the Walkerton outbreak of waterborne gastroenteritis.
<http://www.publichealthbrucegrey.on.ca/private/Report/SPReport.htm>
- Budd, W. (1873) *Typhoid fever: its nature, mode of spreading and prevention*. Longmans, London. 193 pp.
- CDR (1998) Emerging pathogens and the drinking water supply. *CDR Weekly* **8**(33), 292.
- Davison, A., Howard, G., Stevens M., Callan, P., Kirby, R., Deere, D. and Bartram, J. (2002) *Water Safety Plans*. WHO/SHE/WSH/02/09 World Health Organization, Geneva, Switzerland.
- Deere, D., Stevens, M., Davison, A., Helm, G. and Dufour, A. (2001) Management Strategies. In: *Water Quality: Guidelines, Standards and Health. Assessment of risk and risk management for water-related infectious disease*. Fewtrell, L. and Bartram, J. (Eds.) IWA Publishing, London. pp. 257-288.
- Dufour, A.P. (1977) *Escherichia coli*: the fecal coliform. In: *Bacterial indicators/health hazards associated with water*. Hoadley, A.W. and Dutka, B.J. (Eds.) ASTM, Philadelphia. pp. 48-58.
- Dufour, A.P. and Cabelli, V.J. (1975) Membrane filter procedure for enumerating the component genera of the coliform group in seawater. *Applied Microbiology* **26**, 826-833.
- Ebringer, A. and Wilson, C. (2000) HLA molecules, bacteria and autoimmunity. *Journal of Medical Microbiology* **49**(4), 305-311.

- Edelman, R. and Levine, M.M. (1986) Summary of an international workshop on typhoid fever. *Reviews of Infectious Disease* **8**, 329-349.
- Eijkman, C. (1904) Die Gärungsprobe bei 46°C als Hilfsmittel bei der Trinkwasseruntersuchung. *Cbl. Bakteriol. Abth I. Orig.* **37**, 742-752.
- Escherich, T. (1885) Die Darmbakterien des Neugeborenen und Säuglings. *Fortschritte der Medizin* **3**, 515 and 547.
- Ferreira Jr., A.G., Ferreira, S.M., Gomes, M.L. and Linhares, A.C. (1995) Enteroviruses as a possible cause of myocarditis, pericarditis and dilated cardiomyopathy in Belem, Brazil. *Brazilian Journal of Medical and Biological Research* **28**(8), 869-874.
- Fewtrell, L. and Bartram, J. (2001) *Water Quality: Guidelines, Standards and Health. Assessment of risk and risk management for water-related infectious disease.* IWA Publishing, London, UK.
- Frankland, P. and Frankland, P. (1894) *Microorganisms in Water; Their Significance, Identification and Removal.* Longmans, Green & Co., London, UK.
- Fricker, E.J. and Fricker, C.R. (1994) Application of polymerase chain reaction to the identification of *Escherichia coli* and coliforms in water. *Letters in Applied Microbiology* **19**(1), 44-46.
- Geldreich, E.E., Huff, C.B., Bordner, R.H., Kabler, P.W. and Clark, H.F. (1962) The faecal coli-aerogenes flora of soils from various geographical areas. *Journal of Applied Bacteriology* **25**, 87-93.
- Gerba, C.P. and Rose, J.B. (1990) Viruses in source and drinking water. In: *Drinking Water Microbiology: Progress and Recent Developments.* McFeters, G.A. (Ed.). Springer-Verlag, New York, USA.
- Grabow, W.O.K., Coubrough, P., Nupen, E.M. and Bateman, B.W. (1984) Evaluation of coliphages as indicators of virological quality of sewage-polluted water. *Water SA* **10**(1), 7-14.
- Grabow, W.O.K., Taylor, M.B. and de Villiers, J.C. (2001) New methods for the detection of viruses: call for review of drinking water quality standards. *Water Science and Technology* **43**(12), 1-8.

- Haas, C.N. (1999) Disinfection. In: *Water Quality and Treatment: A Handbook of Community Water Supplies. Fifth Edition*. Letterman, R.D. (Ed.) McGraw-Hill, New York, USA. pp.14.1-14.6.
- Haas, C.N. (1983) Estimation of risk due to low doses of microorganisms: a comparison of alternative methodologies. *American Journal of Epidemiology* **118**, 573-582.
- Havelaar, A.H., van Olphen, M. and Drost, Y.C. (1993) F-specific RNA bacteriophages are adequate model organisms for enteric viruses in fresh water. *Applied and Environmental Microbiology* **59**, 2956-2962.
- Havelaar, A.H. (1993) The place of microbiological monitoring in the production of safe drinking water. In: *Safety of Water Disinfection. Balancing chemical and microbial risks*. Craun G.F. (Ed.) ILSI press, Washington, DC.
- HMSO (1999) The Water Supply (Water Quality) (Amendment) Regulations 1999, *Statutory Instrument 1999 No. 1524*. Her Majesty's Stationery Office, London.
- Horwitz, M.S., Bradley, L.M., Harbertson, J., Krahl, T., Lee, J. and Sarvetnick, N. (1998) Diabetes induced by Coxsackie virus: initiation by bystander damage and not molecular mimicry. *Nat. Med.* **4**(7), 781-785.
- Hunter, P.R. and Fewtrell, L. (2001) Acceptable risk. In: *Water Quality: Guidelines, Standards and Health. Assessment of risk and risk management for water-related infectious disease*. Fewtrell, L. and Bartram, J. (Eds.) IWA Publishing, London. pp.207-227.
- Hunter, P.R. and Syed, Q. (2001) Community surveys of self-reported diarrhoea can dramatically overestimate the size of outbreaks of waterborne cryptosporidiosis. *Water Science and Technology* **43**, 27-30.
- Hunter, P.R. (1997) *Waterborne Disease. Epidemiology and Ecology*, John Wiley and Sons, Chichester, United Kingdom.
- Hunter, P.R. (2000) Advice on the response to reports from public and environmental health to the detection of cryptosporidial oocysts in treated drinking water. *Communicable Disease and Public Health* **3**, 24-27.

- Hurst, C.J., Knudsen, G.R., McInerney, M.J., Stetzenbach, L.D. and Walter, M.V. (2001) *Manual of Environmental Microbiology*, 2nd Edition. American Society for Microbiology Press, Washington, DC.
- Huttly, S.R.A. (1990) The impact of inadequate sanitary conditions on health in developing countries. *World Health Statistics Quarterly* **43**,118-126.
- Isaac-Renton, J., Moorhead, W. and Ross, A. (1996) Longitudinal studies of *Giardia* contamination in two adjacent community drinking water supplies: cyst levels, parasite viability and health impact. *Applied and Environmental Microbiology* **62**, 47-54.
- Issac-Renton, J., Lewis, L., Ong, C. and Nulsen, M. (1994) A second community outbreak of waterborne giardiasis in Canada and serological investigation of patients. *Transactions of the Royal Society of Tropical Medicine and Hygiene* **88**, 395-399.
- Koch, R. (1893) Ueber den augenblicklichen stand der bakteriologischen Cholera diagnose. *Zeitschrift für Hygiene* **XIV**, 319.
- Kuhn, I., Iversen, A., Burman, L.G., Olsson-Liljequist, B., Franklin, A., Finn, M., Aerestrup, F., Seyfarth, A.M., Blanch, A.R., Taylor, H., Capllin, J., Moreno, M.A., Dominguez, L. and Mollby, R. (2000) Epidemiology and ecology of enterococci, with special reference to antibiotic resistant strains, in animals, humans and the environment. Example of an ongoing project within the European research programme. *Internal Journal of Antimicrobial Agents* **14**(4), 337-342.
- LeChevallier, M.W. and Au, K.K. (2002) Water treatment for microbial control: A review document. World Health Organization.
- LeChevallier, M.W., Abbaszadegan, M., Camper, A.K., Hurst, C.J., Izaguirre, G., Marshall, M.M., Naumovitz, D., Payment, P., Rice, E.W., Rose, J., Schaub, S., Slifko, T.R., Smith, D.B., Smith, H.V., Sterling, C.R. and Stewart, M. (1999a) Committee report: Emerging pathogens – bacteria. *Journal of the American Water Works Association* **91**(9),101-109.
- LeChevallier, M.W., Abbaszadegan, M., Camper, A.K., Hurst, C.J., Izaguirre, G., Marshall, M.M., Naumovitz, D., Payment, P., Rice, E.W., Rose, J., Schaub, S., Slifko, T.R., Smith, D.B., Smith, H.V., Sterling, C.R. and Stewart, M. (1999b) Committee report: Emerging

- pathogens - viruses, protozoa, and algal toxins. *Journal of the American Water Works Association* **91**(9),110-121.
- LeChevallier, M.W., Norton, W.D. and Lee, R.G. (1991) *Giardia* and *Cryptosporidium* spp. in filtered drinking water supplies. *Applied and Environmental Microbiology* **57**(9), 2617-2621.
- Mack, W.N. (1977) Total coliform bacteria. In: *Bacterial Indicators/Health Hazards Associated with Water*. Hoadley, A.W. and Dutka, B.J. (Eds.) ASTM, Philadelphia, pp. 59-64.
- MacKenzie, W.R., Hoxie, N.J., Proctor, M.E., Gradus, M.S., Blair, K.A., Peterson, D.E., Kazmierczak, J.J., Addiss, D.G., Fox, K.R., Rose, J.B. and Davis, J.P. (1994) A massive outbreak in Milwaukee of *Cryptosporidium* infection transmitted through the public water supply. *New England Journal of Medicine* **331**(3), 161-167.
- Makela, A. and Maybeck, M. (1996) Designing a monitoring programme. In: *Water Quality Monitoring*. Bartram, J. and Balance, R. (Eds.) E&FN Spon, London, pp. 35-59.
- McFeters, G.A. (1990) *Drinking Water Microbiology*. Springer-Verlag, New York.
- Mead, P.S., Slutsker, L., Dietz, V., McCraig, L.F., Bresee, J.S., Shapiro, C., Griffin, P.M. and Tauxe, R.V. (1999) Food-related illness and death in the United States. *Emerging Infectious Diseases* **5**(5), 607-625.
- Melnick, J.L. and Gerba, C.P. (1982) Viruses in surface and drinking waters. *Environmental International* **7**, 3-7.
- Morris, R.D. and Levine, R. (1995) Estimating the incidence of waterborne infectious disease related to drinking water in the United States. In: *Assessing and Managing Health Risks from Drinking Water Contamination: Approaches and Applications*. Reichard, E.G., Zappone, G.A. (Eds.) IAHS Press, Wallingford, Oxfordshire, United Kingdom, pp. 75-88.
- Mossel, D.A.A. (1978) Index and indicator organisms: a current assessment of their usefulness and significance. *Food Technology, Australia* **30**, 212-219.

- Payment, P. (1997) Epidemiology of endemic gastrointestinal and respiratory diseases – incidence, fraction attributable to tap water and costs to society. *Water Science and Technology* **35**, 7-10.
- Payment, P. and Armon, R. (1989) Virus removal by drinking water treatment processes. *CRC Critical Reviews in Environmental Control* **19**, 15-31.
- Payment, P., Richardson, L., Siemiatycki, J., Dewar, R., Edwards, M. and Franco, E. (1991) A randomized trial to evaluate the risk of gastrointestinal disease due to consumption of drinking water meeting currently accepted microbiological standards. *American Journal of Public Health* **81**, 703-708.
- Payment, P., Siemiatycki, J., Richardson, L., Renaud, G., Franco, E. and Prévost, M. (1997) A prospective epidemiological study of gastrointestinal health effects due to the consumption of drinking water. *International Journal of Environmental Health Research* **7**, 5-31.
- Petrilli, F.L., Crovari, P., DeFlora, S. and Vannucci, A. (1974) The virological monitoring of water. I. Drinking water. *Boll. Ist. Seiroter, Milan* **53**, 434-442.
- Pipes, W.O. (1982) Indicators and water quality. In: *Bacterial Indicators of Pollution*. Pipes W.O. (Ed.). CRC Press, Boca Raton. pp. 83-96.
- Prendergast, M.M. and Moran, A.P. (2000) Lipopolysaccharides in the development of the Guillain-Barré syndrome and Miller Fisher syndrome forms of acute inflammatory peripheral neuropathies. *Journal of Endotoxin Research* **6**(5), 341-359.
- Prüss, A., Kay, D., Fewtrell, L. and Bartram, J. (2002) Estimating the burden of disease due to water, sanitation and hygiene at global level. *Environmental Health Perspectives* IN PRESS.
- Regli, S., Berger, P. and Macler, B. (1993) Proposed decision tree for management of risks in drinking water: consideration for health and socioeconomic factors. In: *Safety of Water Disinfection: Balancing Chemical and Microbial Risks*. Craun G.F. (Ed.) ILSI Press, Washington, D.C. pp. 39-80.
- Regli, S., Rose, J.B., Haas, C.N. and Gerba, C.P. (1991) Modelling the risk from *Giardia* and viruses in drinking water. *Journal of the American Water Works Association* **83**(11), 76-84.

- Roivainen, M., Rasilainen, S., Ylipaasto, P., Nissinen, R., Ustinov, J., Bouwens, L., Eizirik, D.L., Hovi, T. and Otonkoski, T. (2000) Mechanisms of coxsackievirus-induced damage to human pancreatic beta-cells. *Journal of Clinical Endocrinology and Metabolism* **85**(1), 432-440.
- Rose, J.B. and Gerba, C. (1991) Use of risk assessment for development of microbial standards. *Water Science and Technology* **24**(2), 29-34.
- Rosen, J. and Ellis, B. (2000) The bottom line on the ICR Microbial data. Paper ST6-3 In: *Proceedings of AWWA Water Quality Technology Conference 2000*. Salt Lake City, Utah.
- Schardinger, F. (1892) Ueber das Vorkommen Gahrung Erregender Spaltpilze im drinkwasser und ihre Bedeutung for die Hygienische Beurthelung Desselben. *Wien. Klin. Wochschr.* **5**, 403-405.
- Shanmugam, J., Raveendranath, M. and Balakrishnan, K.G. (1986) Isolation of ECHO virus type-22 from a child with acute myopericarditis – a case report. *Indian Heart Journal* **38**(1), 79-80.
- Snow, J. (1855) *On the Mode of Communication of Cholera*. John Churchill, London.
- Spinner, M.L. and DiGiovanni, G.D. (2001) Detection and identification of mammalian reoviruses in surface water by combined cell culture and reverse transcription – PCR. *Applied and Environmental Microbiology* **67**(7), 3016-3020.
- Stenström, T.A. (1994) A review of waterborne outbreaks of gastroenteritis in Scandinavia. In: *Water and Public Health*. Golding, A.M.B., Noah, N. and Stanwell-Smith, R. (Eds.) Smith-Gordon & Co., London. pp. 137-143.
- Uemura, N., Okamoto, S., Yamamoto, S., Matsumura, N., Yamaguchi, S., Yamakido, M., Taniyama, K., Sasaki, N. and Schlemper, R.J. (2001) *Helicobacter pylori* infection and the development of gastric cancer. *New England Journal of Medicine* **345**(11), 784-789.
- US Department of Health and Human Services (1998) *Preventing emerging infectious diseases: A strategy for the 21st century*. US Department of Health and Human Services, Atlanta, Georgia.

- USEPA (1991) *Guidance Manual for Compliance with Filtration and Disinfection Requirements for Public Water Systems using Surface Water Sources*. US Environmental Protection Agency, Washington, DC.
- USEPA (1989) National Primary Drinking Water Regulations: filtration, disinfection, turbidity, *Giardia lamblia*, viruses, *Legionella*, and heterotrophic bacteria; Final Rule (40 CFR Parts 141 and 142). *Federal Register* **54**(124).
- USEPA (2000) National Primary Drinking Water Regulations: Long term I enhanced surface water treatment and filter backwash rule (40 CFR Parts 141 and 142). *Federal Register Proposed Rule* **65**(69).
- van der Kooij (1993) Importance and assessment of the biological stability of drinking water in the Netherlands. In: *Safety of Water Disinfection: Balancing Chemical and Microbial Risks*. Craun, G.F. (Ed.) ILSI Press, Washington, DC. pp. 165-179.
- Waite, W.M. (1991) Drinking water standards – a personal perspective. In: *Proceedings of the UK Symposium on Health-related Water Microbiology*. Morris R. et al. (Eds.). International Association for Water Pollution Research and Control, London. pp 52-65.
- White, G.C. (1999) *Handbook of Chlorination*. John Wiley & Sons Inc. New York.
- Whitlock, E.A. (1954) The waterborne diseases of microbiological origin. Paper presented at the Annual Conf. of the Nat. Assoc. of Bath Superintendents 1954, Anderson Ltd, Stepney Green, United Kingdom.
- WHO (1976) *Surveillance of Drinking water Quality*, World Health Organization, Geneva.
- WHO (1993) *Guidelines for Drinking water Quality. Volume ;, Recommendations. Second Edition*. World Health Organization, Geneva. Second Edition.
- WHO (1997) *Guidelines for Drinking water Quality, Volume 3: Surveillance and Control of Community Supplies. Second Edition*. World Health Organization, Geneva. Second Edition
- WHO (1998) Emerging and re-emerging infectious diseases. Fact sheet no. 97. (<http://www.who.int/inf-fs/en/fact097.html>).