Health Canada Santé Canada

Chlorine in Drinking Water

Document for public comment

Prepared by the Federal-Provincial-Territorial Committee on Drinking Water

Comment period ends
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Canada

Chlorine in Drinking Water
Public Comment Document

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Purpose of consultation

The Federal-Provincial-Territorial Committee on Drinking Water (CDW) has been assessing the available information on chlorine with the intent of developing a drinking water guideline. The purpose of this consultation is to solicit comments on the proposed Guideline Technical Document.

It should be noted that this Guideline Technical Document focuses on the health effects related to exposure to chlorine in drinking water supplies. It does not review the benefits or the processes of chlorination, nor does it assess the health risks related to exposure to by-products formed as a result of the chlorination process.

The CDW has requested that this document be made available to the public and open for comment. Comments are appreciated, with accompanying rationale, where required. Comments can be sent to the CDW Secretariat via email at water_eau@hc-sc.gc.ca. If this is not feasible, comments may be sent by mail to the CDW Secretariat, Water, Air and Climate Change Bureau, 3rd Floor, 269 Laurier Avenue West, A.L. 4903D, Ottawa, Ontario K1A 0K9.

It should be noted that this Guideline Technical Document on chlorine in drinking water will be revised following evaluation of comments received and that this document should be considered as a draft for comment only.

October 2007

Chlorine

Part I. Overview and Application

1.0 Proposed guideline

It is not considered necessary to establish a guideline for chlorine in drinking water, based on its low toxicity at concentrations found in drinking water as a result of treatment. Any measures taken to limit the concentration of chlorine or its by-products in drinking water supplies must not compromise the effectiveness of disinfection.

2.0 Executive summary

Most drinking water treatment plants in Canada use chlorine as a disinfectant. The use of chlorine in the treatment of drinking water has virtually eliminated waterborne diseases, because chlorine can kill or inactivate most microorganisms commonly found in water. Disinfection is essential to safeguard drinking water; the health risks from disinfection by-products are much less than the risks from consuming water that has not been disinfected. Free chlorine in Canadian drinking water distribution systems ranges from 0.04 to 0.8 mg/L.

This Guideline Technical Document focuses on the health effects related to exposure to chlorine in drinking water supplies. It does not review the benefits or the processes of chlorination, nor does it assess the health risks related to exposure to by-products formed as a result of the chlorination process.

Health Canada recently completed its review of the health effects associated with chlorine in drinking water, as well as taste and odour considerations. Based on this review, it is not considered necessary to establish a guideline for chlorine in drinking water. The Federal-Provincial-Territorial Committee on Drinking Water reviewed the proposed Guideline Technical Document for chlorine in drinking water and gave approval for it to undergo public consultation.

2.1 Health effects

Health Canada has classified chlorine as unlikely to be carcinogenic to humans. Studies in laboratory animals and humans indicate that chlorine exhibits low toxicity, regardless of the route of exposure (i.e., ingestion, inhalation, dermal). Studies in animals have not been able to identify a concentration of chlorine associated with adverse health effects, in part because of aversion to its taste and odour. No adverse health effects have been observed in humans from consuming water with high chlorine levels (up to 50 mg/L) over a short period of time.

2.2 Exposure

Human exposure to chlorine primarily results from the ingestion of free chlorine present in treated drinking water. However, because of chlorine's reactivity, only low levels of chlorine would reach the consumer's tap. Levels of chlorine in ambient air are estimated to be low. The use of chlorine is permitted in food processing, but the chlorine must be rinsed out or otherwise removed from contact with food. Chlorine and hypochlorite salts are not expected to accumulate or bioconcentrate in the food chain.

2.3 Treatment

An optimal operational range for chlorine in drinking water is between a detectable level and 5 mg/L. A minimum free chlorine residual of 0.2 mg/L at all points in the distribution system is considered desirable to prevent bacterial regrowth. As chlorine is added to drinking water as a disinfectant and to maintain a residual concentration in the distribution system, treatment of the water for chlorine removal is generally not required. However, for water treatment facilities that include a "superchlorination" treatment step, chlorine will need to be reduced to an appropriate level before distribution to the consumer. This is usually done by the addition of sodium bisulfite or sulfur dioxide to the water. In cases where an individual household obtains its drinking water from a chlorinated private well, a private residential drinking water treatment device may be an option for reducing chlorine concentrations in drinking water if the consumers find the taste objectionable. Because of the low toxicity of chlorine at concentrations found in drinking water, the applicable NSF/ANSI standard for the reduction of chlorine is based on aesthetic effects.

3.0 Application of the guideline

Note: Specific guidance related to the implementation of drinking water guidelines should be obtained from the appropriate drinking water authority in the affected jurisdiction.

Chlorine can be used in drinking water systems for both primary and secondary disinfection. Studies in laboratory animals and humans show that chlorine exhibits low toxicity. Health effects observed are generally attributed to the by-products of reactions between chlorine and the organic precursors naturally present in the raw water. As a result, it is important that the use of chlorine include strategies that reduce the formation of chlorinated disinfection by-products (CDBPs), without compromising the effectiveness of disinfection. To that end, it is recommended that chlorine used for primary disinfection be applied following the removal of organic precursors.

Chlorine used as a secondary disinfectant should be applied so as to maintain a sufficient residual concentration throughout the distribution system. Maintenance of an adequate free chlorine residual will minimize bacterial regrowth in the distribution system and provide a measurable level of chlorine; therefore a rapid drop in chlorine level suggesting unexpected changes in water quality could be immediately detected. A free chlorine residual of 0.2 mg/L is considered a desirable minimum level throughout the distribution system for control of bacterial regrowth. Requirements for chlorine residual concentrations are set by the regulatory authority

and may vary among jurisdictions. An optimal operational range for free chlorine residual is between a detectable level and 5 mg/L. Consumers may find the taste and odour of water containing higher concentrations of chlorine to be unacceptable.

Many Canadian drinking water supplies maintain a free chlorine residual in the 0.04–0.8 mg/L range in the distribution system. This level provides some protection against ingress of contaminated water and regrowth in the distribution system and is sufficient to be accurately measured by field instruments. Chlorine taste and odour at these concentrations are generally within the range of acceptability for most consumers.

3.1 Monitoring

For secondary disinfection using chlorine, it is recommended that a continuous analyser be used at the point of entry and throughout the distribution system or that grab samples be frequently tested. This ensures that the target chlorine level is being applied at all times and provides a comparison against residual levels observed throughout the distribution system. In addition, sample locations should be chosen to represent all areas of the distribution system, taking into account geographic location, age and materials of the water main, structural integrity of the distribution system, water storage, and retention times.

In general, water leaving a treatment plant should be tested daily for both chlorine residual and turbidity, and tested at least weekly for total coliforms and *E. coli* to confirm the microbiological safety of the supply. In the distribution system, the presence of adequate chlorine residuals should be confirmed when sampling for total coliforms and *E. coli*.

All field measurements of chlorine in the distribution system should be carried out on-site, since chlorine concentrations may decrease during transportation to the laboratory and storage. Several portable field instruments are available for this purpose, typically using a colorimetric method such as the N,N-diethyl-p-phenylenediamine (DPD) method for the determination. Sample locations should be free-flowing or flushed on-site to achieve a fresh sample that is reflective of the system water quality.

Part II. Science and Technical Considerations

4.0 Identity, properties, and uses

4.1 Occurrence and physical properties

Chlorine is a chemical element (symbol Cl) belonging to the halogen family, with an atomic weight of 35.457 (White, 1999). In nature, chlorine is found only as the combined chloride ion (Cl^-), with a valence of −1 (White, 1999). Chloride makes up much of the salt dissolved in the oceans (about 1.9% of the mass of seawater). Most chloride salts are soluble in water, so solid chloride is usually found in abundance only in dry climates or deep underground. Chloride ion is abundant in nature and necessary to most forms of life, including humans. Molecular or elemental chlorine (Cl_2) does not exist naturally, but can be produced industrially either by the electrolysis of sodium chloride dissolved in water or by the hydrogen chloride oxidation process (Connell, 1996; White, 1999). Once produced, it is collected, purified, compressed, and cooled; it is then stored and shipped as a pressurized liquified gas. Cl_2 solidifies at −101.5°C and boils at −34.0°C at standard atmospheric pressure (Connell, 1996; White, 1999). As chlorine gas, it is greenish yellow, 2.5 times as heavy as air (3.2 g/L at 0°C and 101.3 kPa), and extremely irritating to mucous membranes (White, 1999). Chlorine gas is considered to be slightly soluble in water: 14.6 g/L at 0°C (U.S. EPA, 1994c).

Hypochlorite salts (mostly sodium hypochlorite, NaOCl) are also commonly used in drinking water treatment. NaOCl, also known as chlorine bleach, bleach solution, or Javelle water, is commercially prepared by chlorinating aqueous sodium hydroxide solutions at reduced temperatures (IARC, 1991). For stability reasons, commercial solutions are prepared at concentrations of 5–15%. Calcium hypochlorite ($Ca(OCl)_2$), also known as chloride of lime, tropical bleach, bleaching powder, or granular bleach, is easy to handle and transport. Both $Ca(OCl)_2$ and NaOCl must be stored carefully to prevent deterioration; NaOCl must be protected from heat, light, pH, and the presence of heavy metal cations (Connell, 1996), whereas $Ca(OCl)_2$ must be protected from heat, organic materials, and humidity.

4.2 Chemistry in aqueous media

When added to water, chlorine gas (Cl_2) dissolves rapidly and establishes an equilibrium with hypochlorous acid (HOCl), according to chemical equation (1):

$$\mathbf{Cl_2} + H_2O \leftrightharpoons H^+ + Cl^- + \mathbf{HOCl} \qquad (1) \qquad pKa = 7.5$$

Addition of NaOCl and $Ca(OCl)_2$ to water achieves the same essential oxidizing agent, HOCl, according to chemical reactions (2) and (3) below (IARC, 1991; Connell, 1996; White, 1999), with the only difference being side reactions and end products:

$$NaOCl + H_2O \leftrightharpoons NaOH + \mathbf{HOCl} \qquad (2)$$

$Ca(OCl)_2 + 2H_2O \leftrightharpoons Ca(OH)_2 + 2\mathbf{HOCl}$ (3)

HOCl then dissociates to negative hypochlorite ion (OCl^-) according to chemical equation (4):

$\mathbf{HOCl} \leftrightharpoons H^+ + \mathbf{OCl^-}$ (4) pKa = 7.5

All of these chemical reactions, and thus disinfection effectiveness, are highly dependent upon the pH and temperature of the aqueous medium, which determine the extent of conversion between the three chlorine species Cl_2, HOCl, and OCl^-. HOCl is considered to be more effective at microbial inactivation and dominates at lower pH levels. For example, at a pH of 6.5 and temperatures of 0°C and 20°C, an aqueous solution of chlorine would contain about 95.5% and 92.4% HOCl respectively (4.5% and 7.6% of OCl^-); at a higher pH of 8.5, the equilibrium shifts to 17.5% and 10.8% of HOCl (82.5% and 91.2% of OCl^-). The typical pH range of drinking water is between 6.5 and 8.5 and it is recommended that chlorination of drinking water be conducted at pH levels below 8 for maximum disinfection efficiency (White, 1999; IPCS 2000; WHO, 2004).

4.3 Uses

The major uses of chlorine are in the manufacture of chlorinated organic chemicals (e.g., vinyl chloride monomer, carbon tetrachloride, perchloroethylene, 1,1,1-trichloroethane, and chlorobenzenes), non-chlorinated organic chemicals (e.g., propylene oxide and glycols), and chlorinated inorganic chemicals (e.g., sodium hypochlorite, hydrochloric acid, and hypochlorous acid). It is also widely used as a bleaching agent in the manufacture of pulp and paper; in bleaching textiles and fabrics; in the manufacture of pesticides, herbicides, adhesives, and pharmaceuticals; for drinking and swimming water purification; for the sanitation of industrial and sewage wastes; and in the degassing of aluminum metal (Curlin et al., 1991). In its liquid and solid forms, chlorine is a powerful oxidizing, bleaching, and disinfecting agent. In its gaseous and liquid forms, chlorine is commonly used to inactivate microbial pathogens found in drinking water supplies.

Only a small percentage of the chlorine produced worldwide is utilized in water treatment supplies; in the United States, 6% of all the chlorine produced domestically is used for the purposes of treating water (White, 1999). Chlorine is the most commonly used disinfectant in the world for treating drinking water (WHO, 1997) due to its effective and efficient germicidal properties, ease of application, measurement, and control, persistence, and low cost (IARC, 1991; Connell, 1996; White, 1999). The use of chlorine in the treatment of drinking water played a major role in reducing or even virtually eliminating waterborne diseases, such as typhoid fever, cholera, dysentery, and other gastroenteritic diseases, in developed countries (IARC, 1991; Bull, 2000). Chlorine can also be added to drinking water systems to prevent algal, fungal, and bacterial growth, to control slime growth in distribution systems, to maintain clean filter media at the treatment plant, to restore and preserve pipeline capacity, to restore well capacity, to disinfect water mains, and to control taste and odours (White, 1999; CCOHS, 2004a). Where

chlorine is used for drinking water treatment purposes, the type of chlorine selected depends on a number of factors, including cost, availability, equipment maintenance, and ease of application. For example, chlorine gas is approximately three times less expensive than hypochlorites (White, 1999), but may be more difficult to use than hypochlorite salts; calcium hypochlorite can contribute to scaling problems during water treatment but is less expensive than sodium hypochlorite. While sodium hypochlorite solution is difficult to transport, it is often preferred because it is more easily handled and gives the least maintenance problems with pumping and metering equipment (White, 1999). Calcium hypochlorite is most commonly used for the disinfection of rural and small community water supplies (WHO, 1997) and for swimming pool sanitation (CCOHS, 2004a; Wojtowicz, 2004).

Chlorine is used as a limited food additive such as a bleaching agent in flours and starches; its use in flours is regulated according to Good Manufacturing Practice Standards (Health Canada, 1993). Chlorine or hypochlorite salts are also commonly used during food processing to disinfect water supplies and control microbial agents. For example, chlorinated water is used in the washing and processing of red meat, poultry, and fish, as well as produce (CFIA, 2007, 2005, 2004). Chlorinated water is also used to sanitize food equipment and facilities during food processing (WHO, 1996; CFIA, 2004). Despite permitted use in food processing, chlorine must be rinsed out or otherwise removed from contact with food.

4.4 Application to drinking water treatment

This Guideline Technical Document focuses on the health effects related to exposure to chlorine in drinking water supplies. It does not review the benefits or the processes of chlorination, nor does it assess the health risks related to exposure to by-products formed as a result of the chlorination process.

4.4.1 Terminology

This section provides definitions for some relevant terms used in this document, as adapted from the American Water Works Association (AWWA, 1999; Symonds et al., 2000):

Chlorine residual: the concentration of chlorine species present in water after the oxidant demand has been satisfied.

Free chlorine: the amount of chlorine present in water as dissolved gas (Cl_2), hypochlorous acid (HOCl), and/or hypochlorite ion (OCl^-) that is not combined with ammonia or other compounds in water.

Combined chlorine: the sum of the species resulting from the reaction of free chlorine with ammonia (NH_3), including monochloramine (NH_2Cl), dichloramine ($NHCl_2$), and trichloramine (nitrogen trichloride, NCl_3).

Total chlorine: all chemical species containing chlorine in an oxidized state. Usually the sum of free and combined chlorine concentrations present in water.

Primary disinfection: the application of a disinfectant in the drinking water treatment plant, with a primary objective to achieve the necessary microbial inactivation.

Secondary disinfection: the subsequent application of a disinfectant, either at the exit of the treatment plant or in the distribution system, with a primary objective of ensuring that a disinfectant residual is present throughout the distribution system.

4.4.2 Chlorine in water treatment

Drinking water sources are often contaminated with a variety of pathogenic organisms, including enteric viruses, bacteria, and protozoa, that could be responsible for outbreaks of waterborne disease (White, 1999). The primary purpose of chlorinating drinking water is disinfection, through the destruction or inactivation of pathogenic organisms present (Connell, 1996). The U.S. Centers for Disease Control and Prevention (U.S. CDC, 1999) have identified the control of infectious diseases through clean water and improved sanitation as one of the 10 great public health achievements of the 20th century. The introduction of drinking water treatment early in the 20th century, including disinfection with chlorine, has yielded drastic reductions in the rates of illness and death from waterborne pathogens (Cutler and Miller, 2005).

Although not thoroughly understood, it is commonly believed that the effectiveness of a chemical disinfectant's biocidal properties may be attributed to its electrical charge. Since the outer coating of a microbial cell has a negative charge, it is believed that a disinfectant with a neutral or positive charge will penetrate the slime coating more rapidly. It will then attack the organism's enzyme groups and upset the natural life cycle processes, destroying or inactivating the organism, which will result in a microbiologically safe water (Connell, 1996; White, 1999). Chlorine can be used as both a primary and a secondary disinfectant.

Other chemical reactions occurring in the water will affect the amount of chlorine required for disinfection. Inorganic compounds, such as ammonia, iron, and manganese, will react rapidly with chlorine, whereas natural organic compounds, such as humic and fulvic acids and algal material, may react slowly. Therefore, chlorine is generally added in excess of the demand for disinfection throughout the distribution system (IPCS, 2000; Symonds et al., 2000).

4.4.3 Primary disinfection

Primary disinfection is the application of a disinfectant in the drinking water treatment plant, with a primary objective to achieve the necessary microbial inactivation. The efficacy of disinfection using chlorine can be predicted based on a knowledge of the residual chlorine concentration, temperature, pH, and contact time. This relationship is commonly referred to as the CT concept and is used by public drinking water suppliers as one tool for ensuring adequate inactivation of organisms during disinfection. CT is the product of the residual concentration of disinfectant (C), measured in mg/L at the outlet of the contact chamber, and the disinfectant contact time (T), measured in minutes. The CT values required to achieve the necessary inactivation will depend on the microorganism targeted. More information on CT values typically required to inactivate *Escherichia coli* (Health Canada, 2006a), enteric viruses (Health

Canada, 2004a), and certain protozoa (e.g. *Giardia*) (Health Canada, 2004b) can be found in the corresponding Guideline Technical Documents, available on-line at www.hc-sc.gc.ca/ewh-semt/pubs/water-eau/doc_sup-appui/escherichia_coli/index_e.html, www.hc-sc.gc.ca/ewh-semt/pubs/water-eau/doc_sup-appui/enteric-enterovirus/index_e.html, and www.hc-sc.gc.ca/ewh-semt/pubs/water-eau/doc_sup-appui/protozoa/index_e.html, respectively.

Other factors that may influence disinfection efficiency include contact chamber design, adequate mixing, and presence of sunlight (U.S. EPA, 1999; White, 1999).

4.4.4 Secondary disinfection

Secondary disinfection may be applied to the treated water as it leaves the treatment plant or at rechlorination points throughout the distribution system, to introduce and maintain a chlorine residual in the drinking water distribution system. Overall, a chlorine residual provides three main benefits:

1. It can limit the growth of biofilm within the distribution system and its associated taste and odour problems (LeChevallier, 1998; White, 1999).
2. It may provide some protection in the event of microbial contamination in the distribution system, depending on the magnitude of the event and the susceptibility of the contaminating microorganisms to chlorine.
3. Most importantly, a rapid drop in disinfectant residual may provide an immediate indication of treatment process malfunction or a break in the integrity of the distribution system (LeChevallier, 1998; Health Canada, 2002).

In general, water leaving a treatment plant should be tested daily for both chlorine residual and turbidity, and tested at least weekly for total coliforms, to monitor operational adequacy, and *E. coli* to confirm the microbiological safety of the supply (Health Canada, 2006a,b). In the distribution system, the presence of adequate chlorine residuals should be confirmed when sampling for total coliforms and *E. coli*.

Given the operational benefits of secondary disinfection, operators should strive to maintain a chlorine residual throughout the system to control regrowth and to provide an indication of system integrity. It is recognized that this may be difficult in low-flow areas such as dead-ends or extreme parts of the distribution system and that it may lead to the generation of unacceptable levels of chlorinated disinfection by-products (CDBPs) and could require the implementation of appropriate control strategies.

Canadian data obtained in 2005 from 3588 drinking water facilities located in nine provinces and territories indicate that sodium hypochlorite is the most common disinfectant used for secondary disinfection in 78% of the plants, whereas 19% used chlorine gas, 1.4% used calcium hypochlorite, and 1.6% applied alternative disinfectants. Typical levels of free chlorine in Canadian drinking water systems range from 0.4 to 2.0 mg/L at the treatment plant, from 0.4 to 0.8 mg/L at intermediate points within the distribution system, and from 0.04 to 0.8 mg/L at the far end of the distribution system. Requirements for chlorine residual concentrations are determined by the responsible authority and may vary amongst the provinces and territories. Most jurisdictions specify a minimum level of free chlorine residual that should be applied at the treatment plant and/or detectable within the distribution system. In most provinces and

territories, higher chlorine residuals can be permitted by the regulatory authority, as deemed necessary on a case-by-case basis. The U.S. EPA Surface Water Treatment Rule requires a minimum disinfectant residual of 0.2 mg/L for water entering the distribution system and that a detectable level be maintained throughout the distribution system (U.S. EPA, 2002). The World Health Organization (WHO) has suggested that, for areas with little risk of cholera or related outbreaks, a minimum free chlorine residual range of 0.2–0.5 mg/L be maintained at all points in the supply (WHO, 1997). In general, a free chlorine residual of 0.2 mg/L is considered a desirable minimum level for the control of bacterial regrowth in the distribution system (LeChevallier et al., 1996).

In some water systems, chlorine is combined with ammonia to form chloramines, which are used as a secondary disinfectant (Health Canada, 1996). More information on chloramine dosages, residuals, by-products, and operational issues can be found in the corresponding Guideline Technical Document (Health Canada, 2006c), available on-line at www.hc-sc.gc.ca/ewh-semt/pubs/water-eau/doc_sup-appui/chloramines/index_e.html.

4.4.5 Formation of chlorinated disinfection by-products

Some of the natural organic matter in the treated water has the potential to react with chlorine to form CDBPs at the plant and within the distribution system. The types and structures of CDBPs are complex and are a function of water quality and treatment conditions (IPCS, 2000). The most common CDBPs present in chlorinated waters include trihalomethanes (THMs) and halogenated acetic acids (HAAs). Because elevated levels of CDBPs may have adverse effects on health (WHO, 1995; U.S. EPA, 1999; Health Canada, 2000; IPCS, 2000), every effort should be made to maintain their concentrations as low as reasonably achievable, without compromising the effectiveness of disinfection. This can be done using strategies such as precursor control and removal or application of alternative/modified disinfection practices (IPCS, 2000), including optimization of the treatment process.

This document does not discuss the health effects of exposure to the various disinfection by-products. Guideline Technical Documents have been developed for specific CDBPs, including THMs (Health Canada, 2006c), available online at: http://www.hc-sc.gc.ca/ewh-semt/pubs/water-eau/doc_sup-appui/trihalomethanes/index_e.html, and chloramines (Health Canada, 1996), whereas others are being developed for HAAs (Health Canada, 2006d), chloral hydrate, and chlorite/chlorate (Health Canada, 2005). In addition, extensive reviews are available in the scientific literature (Cantor, 1997; IPCS, 2000; Villanueva et al., 2003).

4.4.6 Taste and odour considerations

While chlorination can help improve taste and odour through the reaction with organic materials and iron (Connell, 1996), it can also generate chlorinous flavours caused by the presence of the disinfectant itself or by the occurrence of other CDBPs formed by the reaction with other compounds in the water. For example, the reaction of chlorine with certain nitrogen compounds (e.g., amino acids, ammonium, urea) present in source water may lead to the formation of strong-smelling compounds such as aldehydes, nitriles, and some chloramines,

which can cause pronounced chlorinous tastes and odours, sometimes even at very low levels. Chlorophenols, resulting from the reaction of chlorine with phenolic compounds, can be formed at the plant or in the distribution system and can impart taste and odours to the water.

In an NTP (1992) rodent study, aversion to the taste of chlorine was demonstrated to be dose-related (U.S. EPA, 1994a). WHO (1997) noted that aversion to the taste of chlorine in drinking water could lead human populations to reject a source of water that is actually safe to drink. WHO (2004) also indicated an increased risk of unacceptability at free chlorine residuals between 0.6 and 1.0 mg/L. The Australia NHMRC (2004) reported an odour threshold of 0.6 mg/L for chlorine in drinking water, while a study by AwwaRF (2004) suggested thresholds as low as 0.05–0.1 mg/L. It is clear that there is a wide variability of taste and odour thresholds in the population, depending on individual sensitivities. However, a survey conducted in American and Canadian drinking water plants found that chlorine was still the dominant cause of odour complaints and a significant cause of taste complaints from consumers (Suffet et al., 1996).

A recent cross-country survey of 1750 people in the United States examined public perception of chlorinous flavours in tap water and found that the majority of consumers were satisfied with their municipal tap water. However, taste was often the cause of higher consumer dissatisfaction, and the most common off-flavour reported in drinking water was "chlorinous" (15.5% for taste, 14.8% for odour) (AwwaRF, 2004). In this study, average threshold sensitivity to the taste of free chlorine was 0.82 mg/L, with 46% unable to detect free chlorine at the highest residual concentrations (~1 mg/L) in their household tap water.

Suffet et al. (1996) indicated that it is possible that chlorine odour problems are produced by other compounds and are not a result of the disinfection process. Although consumers may report a taste or odour problem as "chlorinous," it should be noted that they may be confusing chlorinous taste and odour with tastes and odours from CDBPs (AwwaRF, 2004). Thus, in many cases, it is difficult to accurately identify consumers' taste and odour problems based solely on their description of the tastes and odours.

5.0 Exposure

Because chlorine is not stable under environmental conditions, exposure is not expected to be significant, and there are few data available.

5.1 Air

There are no data available on concentrations of chlorine in indoor air; however, gaseous chlorine is estimated to be at low levels in ambient air: 1–3.7 mg/m^3 (0.344–1.27 ppm) (U.S. EPA,1994b; WHO, 1996).

5.2 Drinking water

Human exposure to chlorine results primarily from the ingestion of free chlorine present in treated drinking water (U.S. EPA, 1994a; WHO, 1996). Vaporization of chlorine at low concentrations into the air from drinking water is not considered to be relevant (U.S. EPA, 1994b; IPCS, 2000). Similarly, the amount of chlorine released from dilute sodium hypochlorite solutions into the air under normal usage conditions is not considered significant (CCOHS, 2004c).

5.3 Food

Chlorine and hypochlorite salt solutions are commonly used during food processing to disinfect water supplies and control microbial agents. Fresh produce is permitted to be washed with chlorinated wash water containing free chlorine residual levels between 2 and 7 mg/L (or 100–150 mg total chlorine/L); excess amounts of wash water must be later removed from the produce (CFIA, 2005). Chlorinated water is also used in red meat, poultry, and fish processing. Water in contact with beef carcasses is permitted to contain a maximum total available chlorine level of 20 mg/L or a maximum level of 10 mg/L for total available chlorine as hypochlorous acid (CFIA, 2004). Beef must then be followed by a rinse with potable water or a similar appropriate measure to ensure that residues resulting from treatment are negligible (CFIA, 2004). Poultry carcasses and parts are also permitted to be dipped, sprayed, or washed with water containing 20–50 mg total available chlorine/L (CFIA, 2004) or up to 10 mg/L for total available chlorine as hypochlorous acid, provided that treatment is followed by a rinse with potable water. In fish processing, residual chlorine may not exceed 10 mg/L when the water will come into direct contact with fish; however, higher concentrations may be used for sanitation, provided that the water does not come into direct contact with fish (CFIA, 2007).

No data exist for chlorine residues in food. However, due to their water solubility and high reactivity, chlorine and hypochlorite salts are not expected to accumulate or bioconcentrate in the food chain (ATSDR, 2002a, 2002b; UNEP, 2003). Therefore, there is no reason to expect that residues above naturally occurring background levels would be found in foods (U.S. EPA, 1999).

5.4 Swimming pools and hot tubs

Chlorine and hypochlorite salts are also used for the disinfection of swimming pools and hot tubs. Those who swim or use a hot tub frequently could have greater dermal and possibly inhalation exposures to chlorine and CDBPs (U.S. EPA,1994a). However, for the purpose of this document, chlorine exposures from swimming pools and hot tubs will not be evaluated.

6.0 Analytical methods

The U.S. Environmental Protection Agency (EPA) has approved several methods, based on colorimetric (N,N-diethyl-p-phenylenediamine, or DPD), amperometric, iodometric, and syringaldazine methods, for the determination of free, total, and combined chlorine in drinking water.

The DPD colorimetric method for residual chlorine is the most widely used to determine free and total chlorine. The amperometric titration technique requires a higher degree of skill and care than the colorimetric method. The iodometric method is less sensitive than the amperometric method but is suitable for measuring total chlorine concentrations higher than 1 mg/L. The syringaldazine method is a colorimetric/spectrophotometric method specific for the analysis of free chlorine.

Table 1: U.S. EPA-approved analytical methods for chlorine

Methodology	Method	Chlorine residual measured / MDC	Comments
DPD colorimetric	SM 4500-Cl G[1]	Free, combined, total / 0.010 mg/L	Interferences: oxidized manganese; high organic content
	EPA 330.5[3]	Total / 0.2–4 mg/L	
DPD ferrous	SM 4500-Cl F[2]	Free, combined, total / 0.018 mg/L	Interferences: oxidized manganese and copper; combined chlorine of >0.5 mg/L can give high [Cl]
	EPA 330.4[3]	Total / NA	
Amperometric	SM 4500-Cl D[1]	Free, combined, total / NA	Interferences: chloramines can give high [Cl]; very low temperature requires long titration time; presence of copper and silver can cause electrode to malfunction; manganese, iron, and nitrite can be minimized by acidification
	ASTM D1253-03[4]	Total / NA	
	EPA 330.1[3]	Total / NA	
	EPA 330.2[3]	Total / NA	
Low-level amperometric	SM 4500-Cl E[1]	Total / 0.010 mg/L	Cannot differentiate between free and combined chlorine
Iodometric electrode	SM 4500-Cl I[1]	Total / >1 mg/L	Interferences: manganese and other oxidants
	EPA 330.3[3]	Total / >0.1 mg/L	
Syringaldazine (FACTS)	SM 4500-Cl H[1]	Free / 0.1 mg/L	Interferences: None reported

1. Standard Methods (APHA et al., 2005)
2. Standard Methods (APHA et al., 1998)
3. U.S. EPA methods are available for download at: http://www.nemi.gov/.
4. American Society for Testing and Materials (ASTM, 2006)

MDC, Minimum detectable concentration; NA, Not available

Other methods include Standard Method 4500-Cl B proposed by the American Public Health Association, where the minimum detectable concentration is approximately 0.04 mg/L. For this method, acid titration (pH 4) is preferred, because some forms of combined chlorine do not react at normal drinking water pH conditions (APHA et al., 2005). In addition, methods

approved by the International Organization for Standardization (ISO, 2006) for the determination of free chlorine and total chlorine include ISO 7393-1 (1985) (titrimetric), ISO 7393-2 (1985) (colorimetric), and ISO 7393-3 (1990) (iodometric titration).

Because chlorine is not stable in water, the chlorine content of samples will decrease with time. Therefore, chlorine should be determined immediately after sampling (APHA et al., 2005); for samples collected in the distribution system, it is preferable to conduct the analysis in the field using a field test kit (Harp, 2002). Field test kits are based on the DPD colorimetric method for measuring free or total chlorine in water. The use of automatic colorimeters eliminates the human error associated with colour matching. The visual comparators for free and total chlorine include colour cube (range 0.1–2.5 mg/L) and colour disc (0–3.5 mg/L). Pocket colorimeter kits may allow determination of free chlorine (0.02–2.0 mg/L) or total chlorine (0–4.5 mg/L), whereas spectrophotometers may allow chlorine analysis in the range 0.1–10 mg/L, depending on the model. A digital titrator based on the DPD-FAS (ferrous ammonium sulphate) method can also be used for field determination of chlorine in the concentration range 0.01–3.0 mg/L.

Special analysers are often used to control the feed rate of chlorine and chlorine residual on-line by a special analyser. The analysers use amperometric titration, colorimetric, or oxidation–reduction potential probe methods.

Free and combined forms of chlorine may be present simultaneously in chlorinated water. Chloramines are the combined forms resulting from the reaction of chlorine with the naturally occurring ammonia or ammonia added as part of the water treatment strategy. Total chlorine is the combination of free and combined chlorine.

7.0 Treatment technology

7.1 Municipal scale

The amounts of chlorine used have to be integrated in the overall optimization of the water treatment process. Removal of contaminants that increase the chlorine demand, including precursors of CDBPs, will reduce both the quantity of chlorine added to the water and the production of CDBPs.

Control of the chlorine dosage in drinking water requires effective control of the feed rate of chlorine. Computerized control systems have been developed to determine the amount of chlorine that needs to be applied for a given water by combining inputs from several measurements, including flow and residual level in treated water. This is usually known as compound loop control (MWH, 2005). Proper design, operation optimization, and equipment maintenance are the critical points identified for an efficient chlorination process, both at the treatment plant and in the drinking water distribution system.

Some water treatment facilities that include a “superchlorination” treatment step will subsequently require the dechlorination of the water to a desirable level before its distribution (White, 1999). This is usually done by the addition of chemicals, such as sulfur dioxide or sodium bisulfite, to the water.

Chemicals used for chlorination should be certified as meeting NSF International (NSF) / American National Standards Institute (ANSI) Standard 60: Drinking Water Treatment Chemicals — Health Effects, which is the recognized health effects standard for chemicals used to treat drinking water and includes certification criteria for chlorine, calcium hypochlorite, sodium hypochlorite, and dechlorination chemicals.

7.2 Residential scale

Generally, it is not necessary to use drinking water treatment devices with municipally treated water. The use of residential-scale treatment devices on municipally treated water is based primarily on individual choice. In cases where an individual household obtains its drinking water from a private well, there may be circumstances where chlorination of the well is warranted. In such cases, monitoring of the chlorine residual would also be recommended. Information about residential-scale water treatment devices for disinfection of drinking water can be found in the Health Canada Water Talk document, Water Treatment Devices for Disinfection of Drinking Water. The document is available on-line at http://www.hc-sc.gc.ca/ewh-semt/water-eau/drink-potab/disinfect-desinfection_e.html.

Private residential drinking water treatment devices may be an option for reducing chlorine concentrations in drinking water if the consumer finds the taste objectionable. Disinfection of a well with chlorine for emergency or regular maintenance requires higher chlorine concentrations than the dosage used for routine disinfection. However, treatment devices that reduce chlorine are not intended to be used to remove high concentrations of chlorine. Methodology for shock chlorination of a well is described in the Health Canada document, A Guide to Well Water Treatment and Maintenance, which is available on-line at www.hc-sc.gc.ca/ewh-semt/water-eau/drink-potab/well_water-eau_de_puits_e.html.

At the residential scale, the water treatment technology for the reduction of chlorine is adsorption onto activated carbon, and the applicable standard is NSF/ANSI Standard 42: Drinking Water Treatment Units — Aesthetic Effects. This standard establishes minimum requirements for materials, design and construction, and performance of drinking water treatment systems that are designed to reduce specific aesthetic-related (taste and odour) contaminants (NSF/ANSI, 2002). For a drinking water treatment device to be certified to NSF/ANSI Standard 42, the device should reduce the concentration of chlorine in water by a minimum of 50% from an influent concentration of 2 mg/L.

Treatment devices can lose their removal capacity through usage and time and need to be maintained or replaced. Consumers should verify the expected longevity of the components in their drinking water treatment device in the manufacturer's recommendations and service them when required.

Health Canada does not recommend specific brands of drinking water treatment devices, but it strongly recommends that consumers look for a mark or label indicating that the device or components have been certified by an accredited certification body as meeting the appropriate NSF/ANSI drinking water materials standards. These standards have been designed to safeguard drinking water by helping to ensure the material safety and performance of products that come into contact with drinking water. Certification organizations provide assurance that a product

conforms to applicable standards and must be accredited by the Standards Council of Canada (SCC). In Canada, the following organizations have been accredited by the SCC to certify drinking water devices and materials as meeting NSF/ANSI standards:

- Canadian Standards Association International (www.csa-international.org);
- NSF International (www.nsf.org);
- Water Quality Association (www.wqa.org);
- Underwriters Laboratories Inc. (www.ul.com);
- Quality Auditing Institute (www.qai.org); and
- International Association of Plumbing & Mechanical Officials (www.iapmo.org).

An up-to-date list of accredited certification organizations can be obtained from the SCC (www.scc.ca).

8.0 Kinetics and metabolism

Pharmacokinetic studies of Cl_2, HOCl, and OCl^- have been performed using radiolabelled chlorine compounds in rats and are summarized below. However, because chlorine molecules are so reactive in biological systems, the results may reflect the presence of metabolic reaction by-products and other chlorinated compounds more so than the specific toxicokinetics of free chlorine, due to the oxidative reactivity of the various compounds (U.S. EPA, 1994a, 1994c).

8.1 Absorption

In a number of studies, blood samples were collected from male rats administered radiolabelled HOCl by gavage (Abdel-Rahman et al., 1982a, 1983, 1984). These studies demonstrated that $HO^{36}Cl$ was quickly absorbed throughout the body at low doses. The rate constant for absorption of $HO^{36}Cl$ was determined to be 0.157 ± 0.001/hour, and the half-life of absorption was 4.42 ± 1.31 hours (Abdel-Rahman et al., 1982a). In another study of fasted and non-fasted Sprague-Dawley rats, oral absorption rate constants for $HO^{36}Cl$ were 0.322/hour and 0.316/hour for fasted and non-fasted rats, respectively, whereas absorption half-lives were 2.2 hours for both groups (Abdel-Rahman et al., 1983). It has been postulated that the range of differences in absorption of ^{36}Cl reflects the tendency for chlorine species to react with organic material in the blood or gastrointestinal tract of non-fasted animals to form diverse chlorine compounds (U.S. EPA, 1994c).

8.2 Metabolism

The oxidizing potential of chlorine was observed in the gastrointestinal tract of rodents. In a preliminary experiment with no quantitative analytical results given, six male Sprague-Dawley rats were given 56 mg of NaOCl solution (pH 7.9) by gavage, equivalent to 140 mg/kg body weight (bw) (Mink et al., 1983). After 1 hour, either trichloroacetic acid (TCA) or dichloroacetic acid (DCA) were detected in the stomachs of all six dosed rats and in certain plasma samples. Chloroform was detected in the stomachs of all rats, but only in the plasma of one rat. Dichloroacetonitrile (DCAN) was also found in the gut contents of two out of three non-fasted rats, but neither the fasted rats nor any plasma samples contained DCAN. Detection of

TCA (detection limit 1.3 µg/mL) and DCA (detection limit 0.3 µg/mL) in both fasted and non-fasted rats indicated that *in vivo* formation of these chlorinated acetic acids was not just dependent on the interactions of NaOCl with foreign organic material in the gut (Mink et al., 1983), although the mechanism of action for formation of these compounds has not been determined. It has been postulated that the bulk of the chlorinated by-products formed in the gastrointestinal tract remain as higher molecular weight products, which may have little toxicological significance (IPCS, 2000).

8.3 Distribution and excretion

Administered doses of $HO^{36}Cl$ were found to be quickly distributed throughout the body at low doses. After 72 hours, the highest concentrations of ^{36}Cl were found in plasma, at 0.77% of the initial dose, and decreasing doses were found in the stomach, testes, lung, kidney, duodenum, spleen, liver, bone marrow, carcass, and skin. The lowest concentration was found in the ileum, at 0.14% (Abdel-Rahman et al., 1982a). In another metabolism study in fasted and non-fasted Sprague-Dawley rats, after 96 hours, ^{36}Cl was distributed at highest levels in the plasma, at 1.92 µg/g, followed by whole blood, at 1.59 µg/g , with the lowest levels found in adipose tissue, at 0.09 µg/g (Abdel-Rahman et al., 1983). Plasma contained ^{36}Cl activity of 1.24% of the administered dose, and packed cells had an activity of 0.29%. The peak plasma level of ^{36}Cl reached 10.7 µg/mL at 4 hours in non-fasted rats, whereas the peak ^{36}Cl plasma level was obtained in 2 hours for the fasted rats at a concentration of 7.9 µg/mL (Abdel-Rahman et al., 1983).

A study of the subcellular distribution of ^{36}Cl compounds found that 75% of total ^{36}Cl activity was recovered in the cytosol, with 2.5% in the microsomal, 1.5% in the nuclear, and <0.1% in the mitochondrial fractions, respectively. It appears that a high percentage of total ^{36}Cl is loosely bound to the erythrocyte membrane or exchangeable with chloride in saline (Abdel-Rahman et al., 1983).

Blood samples following gavage dosage of $HO^{36}Cl$ to rats demonstrated that after 72 hours, the calculated rate constant for $HO^{36}Cl$ elimination from plasma was 0.009 ± 0.001/hour, and the half-life for elimination was 77.0 ± 8.8 hours (Abdel-Rahman et al., 1982a). In a later study, the elimination half-lives were determined to be 44.1 hours and 88.5 hours for fasted and non-fasted rats, respectively, whereas elimination rate constants were 0.016/hour and 0.008/hour for fasted and non-fasted rats, respectively (Abdel-Rahman et al., 1983).

Radiolabelled HOCl administered to fasted rats appeared to be converted to and eliminated completely in the form of chloride. The major route of excretion was via urine, as approximately three-quarters of the eliminated $HO^{36}Cl$ (21% of the initial dose) in the form of chloride ion was recovered in urine, whereas one-quarter (7% of the initial dose) was recovered in faeces (Abdel-Rahman et al., 1982a). In another study, within the first 24-hour period, 7.05% of the administered dose was excreted in urine and 7.45% was excreted via the intestinal route; by 96 hours, this proportion had increased to 36% in urine and 15% in faeces (Abdel-Rahman et al., 1983). ^{36}Cl compounds did not appear to be eliminated via expired air (Abdel-Rahman et al., 1982a).

9.0 Health effects

9.1 Effects in humans

Accidental ingestion of commercial sodium hypochlorite bleach (5.25% or 52 500 mg/L) is one of the most common poisoning events in young children. Intentional ingestion has also been reported frequently in adults. Poisonings have resulted in various degrees of toxicity, including mucosal irritation, nausea, vomiting, diarrhoea, corrosive injury to the oesophagus and gastrointestinal tract, acidosis, and even death (IPCS, 1997), although these effects appear to be due mainly to additional chemicals present or the extreme alkalinity of the product (Howell, 1991; IPCS, 1997). Even in the case of misuse, chlorine bleach has only slight toxicity and irritation potential, and recovery is often rapid and reversible (Racioppi et al., 1994; Babl et al., 1998).

Typical concentrations of free chlorine in drinking water are generally less than 1 mg/L, but humans have consumed hyperchlorinated water for short periods of time at levels as high as 50 mg/L with no apparent adverse effects (U.S. EPA, 1994c). An early anecdotal report noted that no adverse health effects were observed when 150 military personnel consumed water with chlorine levels of 50 mg/L during a period of water main disinfection (Muegge, 1956). Military personnel have also been reported to drink water containing up to 32 mg chlorine/L for several months with no ill effects (Australia NHMRC, 2004). Muegge (1956) also noted that army personnel drinking water containing chlorine at concentrations greater than 90 mg/L experienced momentary constriction of the throat and irritation of the mouth and throat (U.S. EPA, 1994c). The toxicity of chlorine at levels normally found in drinking water appears to be relatively low (Cotruvo and Regelski, 1989; WHO, 1995), and humans appear to tolerate highly chlorinated water well (Muegge, 1956).

In a clinical study, physical and biochemical parameters were measured in 10 healthy male volunteers after they drank increasing concentrations of chlorine in water, ranging from 0.1 to 24.0 mg/L, for 18 days. No treatment-related health effects or toxicity were observed (Lubbers et al., 1982). A subsequent study in 60 men demonstrated minor but statistically significant changes in selected blood and biochemical parameters; however, owing to the short duration of the study and rising dose tolerance, the changes were not necessarily of clinical importance (Lubbers and Bianchine, 1984).

In a study by Wones et al. (1993), test groups of men and women consumed 1.5 L of chlorinated (20 mg/L) water (pH 8.0) daily for 4 weeks. Small decreases in triiodothyronine (T3) and thyroxine (T4) in the chlorinated water group were of borderline significance in men only, but were judged not meaningful by the authors, as levels of thyroid-stimulating hormone did not change. Overall, the authors concluded that there was no significant impact on lipid or thyroid metabolism in healthy adults ingesting 20 mg chlorine/L in drinking water (Wones et al, 1993).

High concentrations of chlorine, such as undiluted NaOCl bleach, have been reported to be severely dermally irritating in humans (Nixon et al., 1975), although this has been strongly linked to the high alkalinity of the solution (Hostynek et al., 1990) rather than the presence of chlorine itself. Habets et al. (1986) reported a few cases of dermatitis in the form of dry, red,

itchy, cracked skin following exposure to household bleaching agents containing sodium hypochlorite. Although reports of allergic contact hypersensitivity to hypochlorite exist (e.g., Eun et al., 1984; Salphale and Shenoi, 2003), these sensitive individuals are often predisposed to allergies (Hostynek et al., 1989; Racioppi et al., 1994; Salphale and Shenoi, 2003; CCOHS, 2004b, 2004c). Despite the frequent use of sodium hypochlorite in bleaching agents, dermal sensitization to hypochlorite salts is expected to be quite rare or even negligible (Stotts, 1980; Habets et al., 1986; Racioppi et al., 1994).

The general population is not expected to spontaneously develop dermal allergies to the low levels of chlorine observed in drinking water. No published reports were found regarding dermal or ocular irritation following bathing or showering with chlorinated tap water. Furthermore, no adverse dermal effects from exposure to chlorine in swimming pools have been reported; for swimming pools, minimum free chlorine concentrations between 1 and 3 mg/L (PMRA, 1999; Queensland Health, 2004; U.S. CDC, 2005; WHO, 2006) and total chlorine concentrations up to 10 mg/L (City of Sydney, 1996) are recommended. In addition, no information is available on any potential systemic toxicity that can be caused by exposure to chlorine via the dermal route (UNEP, 2003).

Adverse health effects from inhalation of high concentrations of chlorine gas, including bronchospasm (wheezing, throat irritation, and hypoxia), have been well documented (IPCS, 1982). After mild exposures, clinical symptoms usually resolve within 6 hours (Deschamps et al., 1994; Sexton and Pronchik, 1998; UNEP, 2003). As with dermal hypersensitivity, adverse reactions following inhalation or ingestion of small amounts of chlorine tend to occur in those with chlorine allergies or asthma (Penny, 1983; Potts, 1996). There is no human or laboratory animal information available regarding health effects from short-term or long-term inhalation exposure to mists from sodium hypochlorite solutions (CCOHS, 2004c). However, under normal conditions of use, the inhalation of mists or vapours from bleach solutions or other dilute chlorine solutions is not expected to be significant or to result in any health effects in the general population (Racioppi et al., 1994; CCOHS, 2004c).

Epidemiological studies have noted an association between the use of chlorine as a drinking water disinfectant and long-term health effects, including increased risks for cancer and other health effects (AwwaRF, 1991; IPCS, 2000; Arbuckle et al., 2002). However, these studies have examined only the broad exposure to chlorinated water, and generally links have been made between health effects and exposure to CDBPs rather than exposure to free chlorine residuals. Since chlorine is intentionally added to drinking water and is highly reactive, its effects have been difficult to separate from those of its by-products. There have not been any epidemiological studies that have specifically examined free chlorine concentrations in water and long-term health effects in the human population (CCOHS, 2004c).

9.2 Effects on laboratory animals and *in vitro* test systems

9.2.1 Acute toxicity

The oral LD_{50} for chlorine, in the form of calcium hypochlorite, is 850 mg/kg bw in rats (NIOSH, 1984; WHO, 2003) and 880 mg/kg bw in mice (U.S. EPA, 1994a).

Fasted Sprague-Dawley rats were administered 4 mL of sodium hypochlorite at either 200 mg/L or 1000 mg/L by gavage (Scully et al., 1984). Analysis of stomach fluids formed within 10 minutes showed that derivatives of chloramines were produced in the stomach when chlorinated water was administered in conjunction with amine compounds. In addition, very low levels of chlorinated nitrogen compounds were found in blood plasma as soon as 30 minutes later. In a subsequent study, Scully et al. (1986) treated rat stomach fluid *in vitro* with 100, 200, 400, 600, 800, 1000, or 1200 mg Cl_2/L. At concentrations between 200 and 1000 mg chlorine/L, organic N-chloramines were identified. However, the authors cautioned that the high dosage levels administered could be overwhelming existing mechanisms that would reduce or deactivate chlorine or chloramines at lower concentrations in the stomach *in vivo*.

Male Sprague-Dawley rats were administered 3-mL single doses of hypochlorous acid at 0, 10, 20, or 40 mg/L (0, 0.19, 0.38, or 0.75 mg/kg bw) by gavage, and blood samples were obtained at 15, 30, 60, and 120 minutes following administration. Blood glutathione (GSH) measurements decreased 30 minutes after administration in the 10 and 40 mg/L dose groups; at 60 minutes, maximum decreases in blood GSH occurred in all treatment groups, but blood GSH was within the control range after 2 hours. Similarly, blood haemolysis (osmotic fragility) increased significantly in all groups within 15 minutes, but returned to normal within an hour (Abdel-Rahman et al., 1984).

9.2.2 Short-term exposure

Reversible effects on the liver were observed in adult male Sprague-Dawley rats given 5 mL of sodium hypochlorite solution containing the equivalent of 1% (10 000 mg/L) free chlorine (Chang et al., 1981). An increase of long-chain polyunsaturated fatty acids was observed in the liver; after 10 days, there were no obvious differences between liver triacylglycerol levels in treated groups and those of controls.

Groups of six female C57BL/6 mice were administered sodium hypochlorite in drinking water at levels of 7.5, 15, or 30 mg/L for 2 weeks (French et al., 1998). No consistent differences were observed in mesenteric lymph node, lymphocyte proliferation, spleen or thymus weight, antibody titres, or number of antibody-forming cells between treated mice and control mice. Some inconsistent increases in baseline spleen lymphocyte proliferation and T cell mitogen response were observed, but these did not appear to be dose-related. The study was repeated with an extended duration of 6 weeks, and the experimental outcome was identical. The authors concluded that neither Cl^- nor chlorinated by-products formed in the gut adversely affect immune function and that the immune system does not appear to be a sensitive target for Cl^- toxicity.

In another immunotoxicity study, male Sprague-Dawley rats were exposed to sodium hypochlorite in drinking water at concentrations of 5, 15, or 30 mg/L from weaning to 12 weeks of age. Statistically significant changes in spleen weight and delayed-type hypersensitivity reactions were observed in rats at the highest dose. Rats in the mid- and high-dose groups demonstrated delayed macrophage oxidative metabolism, as well as elevated prostaglandin E production. It was hypothesized that direct cytotoxicity to immune system cells (via oxidative stress-type mechanisms) might be responsible for immune effects. The authors concluded that some macrophage function could be affected by subchronic exposure to chlorine-based disinfectants, but only at relatively high doses, and that chlorine-based disinfectants were generally not particularly strong immunodepressants (Exon et al., 1987).

Chlorinated drinking water was administered to 210 male CR1:CD1 mice for 120 days at concentrations up to 30 mg/L. After 48 hours, mice previously immunized with sheep erythrocytes and receiving the 30 mg/L water treatment showed a slight, but not statistically significant, increase in foot pad thickness (a test for delayed-type hypersensitivity) compared with other immunized treatment groups. No other differences in immune response were observed, and the authors concluded that hyperchlorinated drinking water did not appear to significantly affect the *in vivo* immune function of mice (Hermann et al., 1982).

No hormonal effects were observed in Sprague-Dawley rats intubated with sodium hypochlorite for 1 week (Vogt et al., 1982), and monkeys treated with chlorinated drinking water did not demonstrate changes in serum T4 levels (Condie and Bercz, 1985).

Revis et al. (1986) exposed white rabbits for 3 months to chlorine in drinking water at a concentration of 15 mg/L (pH 6.5 or 8.5) administered in conjunction with a diet of 300 µg iodide/kg (sufficient) and 950 µg iodide/kg (high) in a reduced-calcium diet (previous studies had shown dietary calcium to enhance the effect of drinking water disinfectants on plasma cholesterol, T3, and T4). No statistically significant changes in free and bound plasma iodide *in vivo* were observed, and chlorine did not appear to significantly alter plasma iodide levels at low iodide treatment levels (Revis et al., 1986). A subsequent study in rabbits given 0, 0.5, 2, 6, or 15 mg chlorine/L in drinking water for 9 months also found no statistically significant effects on plasma cholesterol or T4 levels. When effects were observed, they did not appear to be dose-dependent. The authors suggested that there is no firm evidence of a causative link between chlorinated drinking water and elevated plasma cholesterol or T4 levels (Holdsworth et al., 1990).

In a subchronic toxicity test, F344 rats ingested chlorine at concentrations of 500, 1000, 2000, or 4000 mg/L in drinking water for 92 days. A trend of decreased water consumption with increasing chlorine dose was observed in both males and females, and body weight in females at the highest dose was decreased compared with controls. A few organ weights were decreased at the highest dose, including thymus and lung in both males and females, liver and spleen in males, and heart, brain, and salivary glands in females. The authors noted that no remarkable pathological changes were observed (Furukawa et al., 1980).

New Zealand white rabbits were exposed to 0, 0.5, 2, 6, or 15 mg chlorine/L in drinking water for 9 months, along with both a normal and a calcium-deficient diet. Liver cholesterol levels were increased in the two highest dose groups, and liver triglycerides were significantly increased in the highest dose group. Under microscopic examination, there was an increased appearance of lipid droplets in the hepatocytes of treated rabbits. The presence of calcium did not significantly alter the effect of chlorine on lipid metabolism. The authors suggested that chlorine affects the excretion of cholesterol from the liver, although the mechanism is not known (Revis et al., 1990).

Drinking water with chlorine concentrations of 0, 25, 100, 175, and 250 mg/L at pH 9.4 was administered to Crl:CDBR Sprague-Dawley rats (10 per sex per group) for 90 days (Daniel et al., 1990). Statistically significant decreased water consumption was observed in a dose-dependent manner, likely due to taste aversion. Although some changes were observed in organ weight and haematological and clinical parameters, these appeared to be sporadic. No treatment-related effects were seen upon gross and microscopic observation. The authors established a no-observed-adverse-effect level (NOAEL) of 250 mg/L (24.9 mg/kg bw per day for females, 16.7 mg/kg bw per day for males), the highest dose tested, in this study.

Chlorine was administered via drinking water to B6C3F1 mice for 90 days, at concentrations of 12.5, 25, 50, 100, and 200 mg/L (Daniel et al., 1991). One female in the highest dose group died towards the end of the study, necropsy revealing mild congestion of the lung and bronchus, which could not be identified as treatment-related. Decreased water consumption was observed in both sexes, with statistical significance in females at the two highest doses. Some changes were observed in organ weights and serum enzymes, but the authors attributed observed changes to decreased water and nutrient consumption and altered electrolyte balance, since there were no overt clinical signs of toxicity and no detectable treatment-related histopathologies. No gross or microscopic lesions were observed. It was concluded that drinking water disinfectants such as chlorine induce a mild, non-specific toxicity via indirect mechanisms — for example, through nutritional deficiencies — rather than by direct toxicological effects on specific organs or tissues. A NOAEL of 50 mg/L (10–12 mg/kg bw per day) was established, based on reduction in heart weight in females and other decreased organ weights in males at 100 mg/L (Daniel et al., 1991).

In a preliminary subchronic toxicity study (Hasegawa et al., 1986), 120 F344 rats (10 per sex per group) were exposed to sodium hypochlorite concentrations of 0, 250, 500, 1000, 2000, and 4000 mg/L in distilled water for 13 weeks. Body weight gain was decreased in all groups, but the decrease was statistically significant only in the two highest dose groups in males and the highest dose group in females. Some of the high-dose rats were emaciated. No decrease in water consumption or aversion to the taste of water was reported by the authors. Absolute lung, liver, and spleen weights in males and salivary gland, lungs, heart, and brain weights of females were significantly lower than controls. No histological changes were evident; biochemical examination showed signs of slight damage to the liver in the two highest dose groups of both sexes. A long-term follow-up experiment did not indicate any notable differences in previous

results obtained. The authors concluded that despite the lack of clear gross or histopathological changes in organs or survival rates, a sodium hypochlorite dose greater than 1000 mg/L was suggestive of subchronic toxicity to the rat, based on decreases in body weight.

9.2.3 Dermal effects

Five female ICR mice (CD-1 strain) had hair removed and abdomens sprayed with 0.8 mL of 0.525% (5250 mg/L) sodium hypochlorite bleach eight times per day for two consecutive days. After treatment, skin had a dry appearance, with scattered brown crusty patches. Tissue changes were moderate, with a few areas showing more severe changes. The authors postulated that the high pH may have been responsible for some of the observed effects (Hess et al., 1991).

Robinson et al. (1986) treated Sencar mice with a single solution of 1000 mg/L hypochlorous acid (pH 6.5) and sodium hypochlorite (pH 8.5) by whole-body exposure and examined them on days 1, 2, 3, 4, 5, 8, 10, and 12 following treatment. The maximum response was observed on the 8th day after hypochlorous acid treatment but waned as time passed, although it was still above baseline on the last day of examination. Treatment with sodium hypochlorite (OCl^- ion) also caused increases in skin thickness, but the maximum increase of 18.2 µm occurred on day 10. Results demonstrate that brief exposures to high concentrations of hypochlorous acid and sodium hypochlorite result in dermal hyperplastic responses in mice, although the increase in thickness of the epidermal layer for OCl^- was only 40% of that of hypochlorous acid, thus indicating that hypochlorous acid is a more potent hyperplasiogenic compound.

Robinson et al. (1986) also treated female Sencar mice dermally with solutions of 1, 10, 100, 300, or 1000 mg hypochlorous acid/L (pH 6.5) by whole-body exposure for a 10-minute period for 4 days. In addition, OCl^- was tested at 1000 mg/L (pH 8.5). Skin thickness was assessed the day after the last treatment. Morphological changes observed were thickened epidermis and elongated basal cells. The maximum epidermal thickness of 38.7 µm was observed following 1000 mg hypochlorous acid/L treatment, compared with 13.8 µm in controls. Increased doses were linked to increased thicknesses, although the two low-dose treatment groups of 1 and 10 mg/L were similar to controls. However, for 1000 mg OCl^-/L treatment, epidermal thickness increased to only 25 µm, even though corresponding cell counts from other treatments indicated that the thickness should be greater. Hypochlorous acid did appear to be hyperplasiogenic; however, repeated high-dose applications did not appear to be particularly effective at maintaining the maximum response, which may indicate either an adaptive response through ineffective penetration of the hyperplastic skin or toxicity to the surface layer of cells, causing more rapid loss with subsequent treatments.

Dermal application of 10% (100 000 mg/L) sodium hypochlorite in combination with 4-nitroquinoline 1-oxide to ddN female mice was performed. Application of hypochlorite itself, in 60 applications over 300 days, did not induce skin tumours. However, skin tumours were induced in 9/32 (4 malignant, 5 benign) mice following 45 applications of sodium hypochlorite

given after submanifestational doses of 4-nitroquinoline 1-oxide, and one lymphatic leukaemia was observed. The authors noted, however, that it seemed unlikely that sodium hypochlorite constitutes a practical carcinogenic hazard (Hayatsu et al., 1971).

Kurokawa et al. (1984) examined both promoter and complete carcinogenic properties of sodium hypochlorite in female Sencar mice. In the promotion studies, the dorsal skin of 20 mice was shaved prior to single topical application of acetone (control) or 20 nmol dimethylbenzanthracene (initiator) in 0.2 mL acetone. One week later, 0.2 mL of a solution of 1% (10 000 mg/L) hypochlorite solution dissolved in acetone was applied; the application was repeated at the rate of twice per week for 1 year. The number and diameter of all skin tumours were recorded weekly, and body weight was recorded monthly. The incidence of skin tumours was not statistically different from controls. In the complete carcinogenicity skin tests, sodium hypochlorite was topically applied alone (dissolved in acetone) for 51 weeks (Kurokawa et al., 1984). None of the mice developed tumours within the 1-year period, and no epidermal hyperplasia was seen. Overall, sodium hypochlorite did not appear to be either a promoter or a complete carcinogen.

9.2.4 Long-term exposure and carcinogenicity

Male Sprague-Dawley rats were administered hypochlorous acid at 0, 1, 10, or 100 mg/L daily in drinking water for 1 year, with blood GSH and osmotic fragility measurements taken at 2, 3, 4, 6, 10, and 12 months (Abdel-Rahman et al., 1984). After 10 and 12 months of treatment, GSH decreased in higher-dose groups compared with controls, which was thought to be due to possible impacts on the oxidation–reduction cycle by the GSH-dependent system. Interim examination at 3 months revealed statistically significant decreases in red blood cell count and haematocrit percentage in the high-dose group, but this was not observed at a later time period. Decreases in osmostic fragility values were observed at 6 months of treatment, but may have been due to lower-than-normal haemolysis occurring in the controls. No significant chloroform concentrations were found in blood during the 1 year of treatment; therefore, it does not appear that THMs are formed as a result of reactions between chlorinated water and organic material in the body. The study results revealed that changes in haematological parameters were inconsistent and did not indicate a dose–response pattern (IRIS, 1994).

Groups of 50 male rats were administered concentrations of 0.05% and 0.1% (500 and 1000 mg/L) sodium hypochlorite in drinking water, and groups of 50 female rats were administered 0.1% and 0.2% (1000 and 2000 mg/L) sodium hypochlorite in drinking water, for 2 years (Hasegawa et al., 1986; Kurokawa et al., 1986). Animals were observed daily; body weight was measured weekly during the first 6 weeks and then every 4 weeks until the end of the experiment. Rats of both sexes showed a dose-related reduction in body weight gain. No decreases in water consumption were noted. Relative liver weights were similar to controls, but absolute liver weights were significantly lower in treated groups. Statistically significant decreases in male brain and heart weights at the high dose were observed; in females, decreased salivary gland weights at both dose levels and in kidneys at the highest dose were observed. Sporadic changes in blood parameters occurred but did not appear to be dose-related. No treatment-related increase in non-neoplastic lesions was observed; in fact, non-neoplastic lesions

were decreased. Tumours were found to occur in a number of organs, but no statistically significant difference between treated groups and controls was observed for any type of tumour. Most of the tumours found were reported to be those that occur as common spontaneous tumours in F344 rats. The authors concluded that sodium hypochlorite was not a carcinogen in F344 rats and that all dose levels had no carcinogenic effect. However, body weight reduction was suggestive of chronic toxicity, with females at the highest dose weighing up to 20% less than controls after 2 years of treatment (Hasegawa et al., 1986).

Groups of 50 male and 50 female mice were administered sodium hypochlorite at 0, 500, or 1000 mg/L in drinking water for 2 years (Kurokawa et al., 1986). Survival rates did not differ between the treated and control groups. Dose-related reductions in body weight gain occurred and were particularly evident in the high-dose group. Tumours were observed in a number of organs, including hyperplastic nodules and hepatocellular carcinomas in livers of all males. However, similar to rats, none of these were statistically significant or appeared to be dose-related. The authors concluded that sodium hypochlorite was not a carcinogen in mice.

Chlorinated water at concentrations of 0, 70, 140, and 275 mg/L was given to F344/N rats for 2 years (NTP, 1992). Doses were equivalent to 0, 4.2, 7.3, and 13.6 mg/kg bw per day for male rats and 0, 4.2, 7.8, and 14.4 mg/kg bw per day for female rats (IRIS, 1994). Groups of 10 rats were sacrificed at weeks 14, 66, and 104 of the study. There was evidence of a dose-related decrease in water consumption in both sexes, but no differences in survival rates were observed. Mean body weights were between 5 and 8% less than in controls throughout the study (U.S. EPA, 1994c), but there were no biologically significant differences in organ weights or organ to body weight ratios. No haematological changes or clinical findings attributable to treatment were observed. The incidence of mononuclear cell (MNC) leukaemia in mid-dose, but not high-dose, females was significantly greater than in controls, according to the life table test ($p = 0.014$). However, the incidence of MNC leukaemia in the control group (16%) was less than that in historical controls (25%), suggesting that the marginal increase observed may have been spurious and not due to treatment. Furthermore, there was no clear dose–response relationship or reduced latency evident in those receiving lower doses of chlorinated water. Other neoplastic lesions were observed in the kidney, pancreas, oral cavity, and spleen, but these were not considered to be related to the consumption of chlorinated water. No gross or microscopic lesions were attributable to chlorinated water consumption. Overall, the authors reported that evidence was weak in support of an association between MNC leukaemia in female rats and consumption of chlorinated water.

Chlorinated water was also given to B6C3F1 mice for 2 years at concentrations of 0, 70, 140, and 275 mg/L (NTP, 1992). Doses were equivalent to 0, 8, 15, and 24 mg/kg bw per day for male mice and 0, 1, 13, and 22 mg/kg bw per day for female mice (WHO, 2003). Groups of 10 mice were sacrificed at weeks 15, 66, and 104 of the study. Survival rates among treated mice were not different from controls, but water consumption was decreased in both sexes. Mean body weights were within 10% of controls throughout the study; however, at the interim evaluation of 66 weeks, body weights of high-dose male mice were statistically significantly lower than controls. There were no biologically significant differences in organ weights or organ to body weight ratios between treated and control groups; lower brain and liver weights were

thought to be due to the lower body weights observed. No alterations in haematology or clinical chemistry were observed, and no gross or microscopic lesions were attributable to chlorinated water.

Groups of 50 male and 50 female Sprague-Dawley rats were exposed to sodium hypochlorite concentrations of 0, 100, 500, and 750 mg/L in drinking water (Soffritti et al., 1997). Animals were allowed to live out the entire lifespan, with the death of the last animal at 151 weeks. Mean daily water consumption was decreased in treated animals in a dose-related fashion. Body weights were slightly decreased in those treated with the highest dose of chlorine; this was more evident in males. Statistical significance was not given in this study. No non-neoplastic changes were observed by gross inspection or histological examination. Tumour development was not dose-related, and there was a greater than expected increase in tumours at the lowest dose. Increased incidences of lymphomas and leukaemias were observed in female rats; however, these were not dose-related, and the incidence of leukaemias was unusually low in the control groups. Males at the highest and lowest doses had increase incidences of relatively rare stomach tumours (squamous cell carcinoma of the forestomach and leiomyosarcomas); in the lowest-dose females, three rare lung adenomas were observed (Soffritti et al., 1997). Although tumour incidence was not dose-related, the authors suggested further research examining the oncogenic risks related to the chlorination of drinking water.

9.2.5 Mutagenicity/genotoxicity

In an *in vitro* Ames assay, sodium hypochlorite was found to be marginally positive for reverse mutation. The number of mutants obtained was variable and was not dose-related. In a repeated test, sodium hypochlorite was mutagenic for *S. typhimurium* strain TA 1530, but not for strain TA 1538 (Wlodkowski and Rosenkranz, 1975). Alternately, in a bioassay examining inhibition of an *E. coli* strain deficient in polymerase I, it was observed that sodium hypochlorite consistently inhibited pol A_1^- strain (Rosenkranz, 1973; Rosenkranz et al., 1976) but did not induce mutation in the Ames assay (Rosenkranz et al., 1976).

A metabolic activation system with rat liver microsome fraction plus cofactors (S9 mix) was applied to *in vitro* chromosomal aberration tests. Sodium hypochlorite tested positive (10–19.9%) for chromosomal aberration tests on activation with S9 mix (Matsuoka et al., 1979). Both calcium hypochlorite and sodium hypochlorite tested positive in another Ames test, as well as in chromosomal aberration tests in Chinese hamster cells (Ishidate et al., 1984). Calcium hypochlorite demonstrated a high frequency of cells with exchange-type aberrations (per unit dose, mg/mL), which generally show carcinogenic potential in animals.

A micronucleus test in six ddY mice was negative for both sodium hypochlorite and calcium hypochlorite (Hayashi et al., 1988). In another micronucleus test, there were no biologically significant differences observed, and no significant differences in chromosomal aberrations were observed for chlorine at pH 6.5 or 8.5 (Meier et al., 1985).

The SOS Chromotest, an *in vitro* assay showing primary damage on *E. coli*, was negative for sodium hypochlorite. The Ames fluctuation test, an *in vitro* test detecting point mutations on *S. typhimurium*, was also negative for sodium hypochlorite (Le Curieux et al., 1993). The authors concluded that no detectable mutation was induced by sodium hypochlorite following

these two tests. A newt micronucleus test, for *in vivo* clastogenic effects on peripheral blood erythrocytes of amphibian newt larvae, was positive for sodium hypochlorite (Gauthier et al., 1989). However, the authors did not rule out the possibility that chlorinated compounds might be partially responsible for the observed effects (Gauthier et al., 1989).

A test for unscheduled DNA synthesis was performed, using cultured mammalian cells to assess *in vitro* genotoxicity of 12.6% sodium hypochlorite solution in Syrian hamster embryo cells. Sodium hypochlorite did not appear to induce unscheduled DNA synthesis (Hamaguchi and Tsutsui, 2000).

Mutagenicity tests are often preliminary screening tests, and positive results do not always correlate with long-term *in vivo* toxicity (Ishidate et al., 1984). Generally, the above tests indicate that neither chlorine nor sodium hypochlorite is considered to be genotoxic.

9.2.6 Reproductive and developmental toxicity

Several studies have demonstrated that chlorine has few effects on reproductive or developmental health in rodents. Drinking water containing free chlorine concentrations of 100 mg/L was given daily over the entire lifetime of 236 BD II rats, in seven consecutive generations. Chlorine was reported to be well tolerated, and there were no adverse effects on fertility, lifespan, growth pattern, haematology, or histology of the liver, spleen, kidney, or other organs. The incidence of malignant tumours was identical in experimental and control groups of rats (Druckey, 1968).

Two strains of mice, C3H/HeJ and C57BL/6J, were used to test the reproductive effects of acid chlorine-treated water for 6 months. Treated water contained 10–13 mg residual chlorine/L acidified to a pH of 2.5, whereas tap water (chlorine residual unspecified) at a pH of 9.6 was used as a control. One hundred and sixty-eight pairings of C3H/HeJ mice (1 male and 1 female) occurred, along with 168 matings of C57BL/6J mice (45 pairings of 1 male and 1 female, and 123 trio matings of 1 male with 2 females). In the C3H/HeJ strain, the total number of mice born, total number weaned, and both the number born per dam and the number weaned per dam were statistically significantly greater in the treated water group than in the control group. In the C57BL/6J strain, the percentage weaned in the control group was slightly greater than that in the treatment group if only paired matings were considered; however, if the type of mating was disregarded, reproductive performance in the treated group exceeded that in the control group in every parameter. The number of C3H/HeJ mice weaned was 5.7% greater in the treated group than in the control group; the number of C57BL/6J mice weaned was 17.5% greater in the treated group than in the control group. The author noted, therefore, that chlorine or hydrochloric acid treatment of water was not detrimental to reproductive outcomes in mice (Les, 1968).

In a study by Chernoff et al. (1979), approximately 500 CD-1 mice were allowed to consume distilled (control) or municipal (treated) water for a minimum of 2 weeks during acclimatization and then were bred at various intervals over a period of 8 months. Levels of chlorine in the treated water were not specified. Analysis of fetal parameters indicated no significant treatment-related effects in number of implants, mortality, weight, or degree of ossification. However, an increased incidence of fetal supernumerary ribs in the tap water group

was observed ($p < 0.005$). The authors noted that considerable variations in the parameters studied in both control and tap water groups indicated that fluctuations were random and not related to water quality. Visceral analysis showed no significant differences in either the type or occurrence of teratological effects. No water-related differences in fetal mortality or occurrence of malformations were observed (Chernoff et al., 1979).

Female Sprague-Dawley rats, six per group, were administered 0, 1, 10, or 100 mg hypochlorous acid/L in drinking water for 2.5 months prior to insemination (day 0). Treatment continued during gestation until day 20, when rats were sacrificed and live and dead fetuses as well as resorptions were noted. Individual fetal weights were recorded, and gross examinations for malformations were made. Fetuses were also examined for skeletal anomalies and soft-tissue defects. All fetuses were viable and normal in external appearance, with no gross abnormalities. Two early resorption sites were discovered in one female in the high-dose group, but this was not significantly different from control. No significant effects on fetus weights were observed. Although the high-dose group had higher percentages of both skeletal and soft-tissue defects, there were no statistically significant differences. The number of anomalies in control and low/mid-dose groups was similar, with the low-dose group actually showing lower percentages of total defects compared with the control. The authors concluded that hypochlorous acid is not embryotoxic or teratogenic, and chlorine in drinking water at the concentrations noted is relatively harmless to the rat when fed to pregnant dams (Abdel-Rahman et al., 1982b).

In a test for sperm-head abnormalities in B6C3F1 mice, a significant dose-related increase was observed for the 100 and 200 mg/L doses (equivalent to 1.6 and 4.0 mg/kg bw per day) of OCl^- (pH 8.5) at a lag period of 3 weeks following dosing (Meier et al, 1985). At the highest dose of 400 mg/L (8.0 mg/kg bw per day), the incidence of sperm-head abnormalities was significantly increased compared with controls, but it was not further increased above that found with the 200 mg/L dose. However, hypochlorous acid (pH 6.5) did not produce any sperm-head abnormalities at any dose, nor were there consistent positive results for OCl^- at lag periods of 1 and 5 weeks. Effects on reproductive outcome and genetic damage were not assessed in this study (CCOHS, 2004c).

Long-Evans rats were administered hypochlorous acid by gavage at 0, 1, 2, or 5 mg/kg bw for 56 days prior to breeding and throughout a 10-day breeding period for males; and for 14 days prior to breeding and throughout breeding, gestation, and lactation (until pups were weaned 21 days following parturition) for females. Following breeding, males were given a gross necropsy of the whole reproductive tract and were evaluated for sperm morphology. Dams were observed for fertility, length of gestation, body weight gain, maternal behaviour, and reproductive tract evaluation. Litters were evaluated for viability, litter size, day of eye opening, body weight gain, and gross external abnormalities. Results indicated no clinical signs of toxicity, haematological changes, or body weight depression, even at the highest dose up to 76 days of exposure. The observed fertility rate and vaginal patency (sexual maturation) were normal in females; in litters, day of eye opening, litter survival, litter size, and pup weight were also normal. In male rats, sperm motility, progressive movement, and abnormal sperm

morphology were comparable across all groups. No histopathological lesions were observed in any of the rats. It was noted that chlorine demonstrated no teratogenic effects (Carlton et al., 1986).

9.2.7 Mode of action

The pharmacokinetics and mode of action of free chlorine on the human body are not fully understood. Researchers have postulated that any observed effects on biological systems result not from free chlorine itself, but rather from chlorinated organics formed in the body due to its reactivity (Mink et al., 1983; Meier et al., 1985; Exon et al., 1987; U.S. EPA, 1994a, 1994c). These chlorinated compounds appear to form in the gastrointestinal tract (Mink et al., 1983); however, the mechanism of formation has not been determined. Although CDBPs appear to be formed in the gastrointestinal tract, it has been postulated that the bulk of them remain as higher molecular weight products, which may have little toxicological significance (IPCS, 2000). Based on the available studies, the toxicity of free chlorine in drinking water is low, and effects observed, if any, appear to be transient and reversible.

10.0 Classification and assessment

The International Agency for Research on Cancer (IARC, 1991) has classified hypochlorite salts in Group 3, not classifiable as to carcinogenicity to humans, due to inadequate evidence in experimental animals and no data in humans. Similarly, the U.S. EPA (1994a) has classified chlorine in Group D, not classifiable as to human carcinogenicity. Based on its lack of toxicity in rodents, Health Canada classifies free chlorine in the form of hypochlorite ion or hypochlorous acid in Class IV D, unlikely to be carcinogenic to humans (data from epidemiological studies are inadequate to assess carcinogenicity; there is no evidence of carcinogenicity in well-designed and properly conducted carcinogenicity bioassays in two species of animals) (Health Canada, 1994).

The NTP (1992) study concluded that there was equivocal (marginal) evidence of carcinogenic activity in female rats based on an increase in the incidence of MNC leukaemia and that there was no evidence of carcinogenic activity of chlorinated water in male rats, male mice, or female mice. Although an increased incidence of MNC leukaemia in female rats was observed, there was no evidence of a dose–response relationship and no evidence of a temporal relationship between increasing dose and incidence of tumours. In addition, it was noted that MNC leukaemia has a high spontaneous rate of occurrence in female F344 rats and that the levels reported in the NTP study were within the historical control range of incidence for the sex and strain of rat (U.S. EPA, 1994a). The U.S. EPA (1994a) stated that the incidence of MNC leukaemia in female rats cannot be solely attributed to exposures to chlorine in drinking water, but rather may reflect the high background rate of MNC leukaemia in the test species. Other long-term studies have not found any adverse effects resulting from ingestion of chlorine in drinking water (Hasegawa et al., 1986; Kurokawa et al., 1986) and thus support the findings of the NTP study (IRIS, 1994).

A decrease in animal body weight, at times accompanied by a decrease in certain organ weights, has been observed in a number of studies. Authors have suggested that this response is linked to decreased water consumption by the rodents (Hasegawa et al., 1986; Kurokawa et al., 1986; Daniel et al., 1991; NTP, 1992; Soffritti et al., 1997), most likely due to aversion to the taste of high levels of chlorine in drinking water (Daniel et al., 1991; IRIS, 1994). Daniel et al. (1991) suggested that inadequate water consumption results in altered electrolyte balance and nutritional deficiencies, which in turn can affect body and organ weights. Hasegawa et al. (1986) demonstrated that once treatment with chlorinated drinking water was terminated, rodents showed rapid and remarkable body weight gains, thus suggesting that any effects on body weight are indeed transient and reversible.

In humans, the general population is not expected to experience dermal effects or to spontaneously develop dermal allergies to chlorine in drinking water, particularly at the low levels observed. There have been no published reports of dermal or ocular irritation in humans following bathing or showering with chlorinated tap water. In addition, no information is available on any potential systemic toxicity that can be caused by exposure to chlorine via the dermal route (UNEP, 2003).

10.1 Aesthetic considerations

While chlorination can help improve taste and odour through the reaction with organic materials and iron (Connell, 1996), it can also generate chlorinous flavours caused by the presence of the disinfectant itself or by the occurrence of other chlorinated by-products formed by the reaction with other compounds in the water. A survey conducted in American and Canadian drinking water plants found that chlorine was the dominant cause of odour complaints and a significant cause of taste complaints from consumers (Suffet et al., 1996).

WHO (1997) noted that aversion to the taste of chlorine in drinking water could lead human populations to reject a source of water that is actually safe to drink. The reported threshold ranges for unacceptability vary from as low as 0.05–0.1 mg/L (AwwaRF, 2004) to between 0.6 and 1.0 mg/L (WHO, 2004).

11.0 Rationale

The use of chlorine in the treatment of drinking water has virtually eliminated waterborne diseases, because chlorine can kill or inactivate most microorganisms commonly found in water. The majority of drinking water treatment plants in Canada use some form of chlorine to disinfect drinking water. Health risks from chlorine, or from any of its disinfection by-products, are much less than the risks from consuming water that has not been disinfected.

Health-based values have been established in other countries or by international organizations. WHO and Australia have established standard or guideline values of 5 mg chlorine/L in drinking water, while the U.S. EPA has set a maximum residual disinfectant level of 4 mg/L for chlorine. These health-based values have all been derived from the same NTP (1992) study, from a NOAEL at the highest dose tested in rodents, but with some differences: WHO and Australia assume that drinking water is the sole source of exposure to chlorine for the

general population, whereas the U.S. EPA assumes that drinking water contributes 80% of chlorine exposure. Based on the lack of toxicity observed in animal studies, these standards or guidelines are considered to be extremely conservative.

Based on the low toxicity of chlorine at concentrations found in drinking water as a result of treatment (generally less than 5 mg/L) and on the toxicological information available, the Federal-Provincial-Territorial Committee on Drinking Water has deemed that there is no need to establish a guideline for chlorine in drinking water. Where chlorine is used as a drinking water disinfectant, it is recommended that an appropriate concentration be maintained to ensure that adequate disinfection has occurred, that a residual can be maintained in the distribution system, and that the levels of free chlorine are sufficiently low at the tap so as not to be unpalatable to consumers.

12.0 References

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Appendix A: List of acronyms

ANSI	American National Standards Institute
ASTM	American Society for Testing and Materials
AWWA	American Water Works Association
bw	body weight
CDBPs	chlorinated disinfection by-products
DCA	dichloroacetic acid
DCAN	dichloroacetonitrile
DNA	deoxyribonucleic acid
DPD	N,N-diethyl-p-phenylenediamine
FAS	ferrous ammonium sulphate
GSH	glutathione
HAAs	haloacetic acids
HOCl	hypochlorous acid
IARC	International Agency for Research on Cancer
ISO	International Organization for Standardization
LD_{50}	median lethal dose
MDC	minimum detectable concentration
MNC	mononuclear cell
NOAEL	no-observed-adverse-effect level
NSF	NSF International
NTP	National Toxicology Program (United States)
ppm	parts per million
SCC	Standards Council of Canada
SM	Standard Methods
T3	triiodothyronine
T4	thyroxine
TCA	trichloroacetic acid
THMs	trihalomethanes
U.S. CDC	United States Centers for Disease Control and Prevention
U.S. EPA	United States Environmental Protection Agency
WHO	World Health Organization