

**Estimation of the Health Risks and Safety
from Exposures to Chlorine and Chloroform
for Swimmers in Pools**

A Study Performed for
US Environmental Protection Agency
Office of Pesticide Programs
Washington, DC

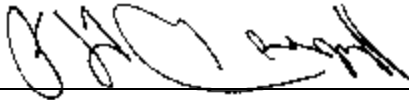
On Behalf of
National Association of Gas Chlorinators
Chlorine Chemistry Council
California International Chemical Company, Inc.

by
The Sapphire Group, Inc.
Bethesda, Maryland

November 2000

Forward

This study was conducted under the auspices of *The Sapphire Group, Inc.*, an international public-health oriented, risk management consultancy. The firm recognizes the primary authorship of Robert G. Tardiff, Richard P. Hubner, Carol G. Graves, Christopher R. Kirman, and Stacey A. Galik, and it gratefully acknowledges the assistance of Leslie K. Dreves. Quality assurance is credited to Carol G. Graves. Information about this report may be obtained by contacting the undersigned at 301-657-8008 or rgt@thesapphiregroup.com.



Robert G. Tardiff, Ph.D., ATS
President and CEO

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Estimation of the Health Risks and Safety from Exposures to Chlorine and Chloroform for Swimmers in Pools

Executive Summary

The Sapphire Group, Inc. conducted this study to determine scientifically (1) whether the concentration of 10 ppm chlorine is reasonably expected to pose no harm (*i.e.*, “safe”) for re-entry of swimmers into pools that have been superchlorinated with gaseous and other forms of chlorine as part of the accepted course of disinfection and (2) whether exposure to chloroform, a byproduct of chlorination, also poses no undue health risk to swimmers during the accepted course of disinfection. The results from the critical examination and analysis of the data on chlorine and on chloroform indicate that 10 ppm chlorine in pool water is a safe re-entry level for swimmers.

USEPA, as part of its process to re-register chlorine as a disinfectant under the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) has proposed a re-entry standard of 4 ppm free available chlorine (FAC) (USEPA, 1999).

Chlorination of swimming pools is an essential public health measure to protect against exposures to dangerous pathogens such as *E. coli* (0157:H7), *Giardia*, Hepatitis A, and *Cryptosporidium*. According to the Centers for Disease Control and Prevention (CDC), approximately ten outbreaks of waterborne gastroenteritis per year in 1997 and 1998 were caused by the parasite *Cryptosporidium*, and 90% were “associated with recreational use of treated water in venues such as swimming pools...” (CDC, 2000a). The Washington State Environmental Health Association has compiled a list of the types of illnesses related to improperly disinfected pools (WSPHA, 1997).

To be fully effective, chlorine (and any other) disinfectant must be present continually in sufficient concentrations to act as a barrier against the survival of newly introduced pathogens. Achieving this goal requires continuous maintenance of an optimum FAC residual of 2-5 ppm with a maximum of 10 ppm (NSPI, 1999). Periodic superchlorination¹ to about 10 ppm such

¹“Superchlorination” is defined as the periodic addition of chlorine at ten times the amount of “combined” chlorine, also called chloramines (Williams, 1999). For example, if the combined chlorine in pool water was 1 ppm, then 10 ppm (10 × 1) chlorine would be added. In residential swimming pools, disinfection often relies solely on the periodic (*e.g.*, weekly or biweekly) application of gaseous and other forms of chlorine which is applied in quantities sufficient to achieve the benefits of superchlorination and to maintain an adequate measure of health protection for swimmers. between applications.

as that recommended by the National Spa and Pool Institute (NSPI, 1995, 1999) and the National Swimming Pool Foundation (NSPF, 1990) is needed to ensure that the minimum chlorine level is always present. Accordingly, this study has evaluated the safety of a 10 ppm upper limit² of chlorine for re-entry in pools.

The application of chlorine to swimming pools is generally of two types: (1) direct addition of chlorine (including gas, liquid bleach, tablet and/or granular chlorine) to residential pools by a homeowner or pool company, and (2) a combination of periodic superchlorination and continuous feed of chlorine through a fixed chlorinator as is usually found at commercial pools. This study focused on a chlorine level for re-entry by swimmers regardless of how the chlorine was applied.

To be effective against disease-causing organisms that can be present in pools, sufficient chlorine must be added to destroy not only the pathogens but also to control or remove algae and any other organic, oxidizable material in the water. This process, referred to as superchlorination (including “breakpoint” chlorination), facilitates the maintenance of efficacious concentrations of free available chlorine³ between superchlorination events and minimizes the formation of chloramines whose presence is undesirable.

Thus, setting 4 ppm FAC as a maximum can make it difficult for pool operators to maintain sufficient residual chlorine in some parts of a pool when temperatures and organic loads from bathers and other sources are high. Furthermore, a maximum of 4 ppm FAC also discourages “shock” treatment and superchlorination which are needed to achieve adequate public health protection. Indeed, the public health community has observed that some managers of public pools, concerned that the level of FAC might exceed 4 ppm the day following superchlorination, will refrain from the superchlorination practice, thereby appreciably increasing the risk that disinfection will be inadequate to prevent waterborne illness among swimmers. The unquestioned public health benefits of adequate chlorination for disinfection of drinking water reported by the International Life Sciences Institute (Craun, 1993) offer a valuable parallel for the proper use of chlorine to protect those who swim in chlorinated pools, whether residential or public.

Since USEPA first recommended its re-entry guideline, additional scientific information has become available to warrant a present re-examination of that re-entry value. USEPA has also indicated that whereas the original guideline had been based on considerations of the hazards

²No concentration higher than 10 ppm was addressed.

³“Free available chlorine” (FAC, also free chlorine) is defined as the combination of hypochlorous acid and hypochlorite ion (NSPI, 1995).

posed solely by chlorine, the Agency is considering expanding its evaluation to include chloroform, a byproduct of chlorine combining with natural organic material.

This study was undertaken to examine all relevant information to determine whether the re-entry guideline could be justifiably raised from the proposed 4 ppm to 10 ppm chlorine. The study first examined the data on chlorine and then on chloroform.

Chlorine

The results indicate that dermal and ingestion exposures to chlorine at 10 ppm in swimming pool water pose no undue risk of adverse health outcomes and that inhalation by swimmers of chlorine gas in pool water is unlikely to occur at the concentrations used for disinfection. For incidental ingestion of chlorinated water, the margin of exposure is amply protective of public health. Although, the margin of exposure for skin contact cannot be estimated because of limited toxicity information, sufficient indirect evidence (including the 4 ppm MRDLG for chlorine that is considered safe with an adequate margin of safety by USEPA for bathing and/or showering) suggests that skin exposures up to 10 ppm chlorine will cause no harm. Furthermore, the volume of water ingested during swimming (*i.e.*, 0.05 L) is considerably smaller than that on which USEPA's 4 ppm MRDLG is based (*i.e.*, 2L), and, therefore, ingestion of chlorinated water during swimming is unlikely to pose any health risk.

Since chlorine gas once in water at the concentrations used in pools is not apt to volatilize; inhalation exposure is considered to be essentially absent and thus represents no risk of adverse health consequences.

Consequently, a re-entry of 10 ppm chlorine is not only safe for swimmers of all ages but also a major safeguard against exposures to harmful pathogens. Therefore, appropriate justification exists on chlorine to revise the re-entry guideline of 4 ppm to 10 ppm of free available chlorine. Such a change would foster the continued public health benefit of controlling human pathogens in swimming pool water.

Chloroform

The results for chloroform are presented separately for users of outdoor and indoor pools, because the majority of pools that are chlorinated with the gaseous form are outdoors and because the statistical distributions of the data for each group appear to be different.

The maximum dose obtained by swimmers at outdoor pools (which comprise the majority of residential pools that employ gaseous chlorine as the mode of application) produced a margin of exposure from 5200 to 30400 for skin contact; 100 for inhalation; and 600 to 3500 for

ingestion. The margin of exposure (MoE) is based on adjusted human NOAELs that incorporate uncertainty factors that address differences in susceptibility between species and among humans. These MoE values are conservative inasmuch as they incorporate uncertainty factors in adjusting the observed values to human situations, and they rely on maximum measures of exposure. Had the mid-point of the concentration ranges been used to estimate MoE, the MoE would have been even greater, suggesting that the majority of the population that is exposed below the maximum measured would obtain an additional measure of safety at 10 ppm chlorine, while being protected against pathogens through the use of regular superchlorination. Had the USEPA approach for estimating the RfD and RfC been applied, the MoE for each value would be approximately 100 times greater than our values.

Similar comparisons were made for the chloroform data from indoor pools, which comprise mostly commercial pools. The results were comparable for skin contact (8000-46600) and ingestion (400-2430); however, the MoE for inhalation exposures were somewhat lower (15-75) than for outdoor pools. This result may well be related to minimal air flow indoors to dissipate the air above the pool water, thereby increasing the concentration of chloroform above the water and in the breathing zone of swimmers. Had the USEPA approach for estimating the RfD and RfC been applied, the MoE for each value would be approximately 100 times greater than our values.

The evaluation of risks and safety of chloroform is somewhat more complex than that for chlorine for several reasons. Unlike chlorine, chloroform is a systemic toxicant; therefore, the doses by various routes for exposure are summed to reflect full exposure. The systemic doses of chloroform were estimated by individual routes of exposure and then summed to produce cumulative systemic doses, the highest doses were compared to the most conservative NOAEL for chloroform (chronic ingestion). The resulting MoE was 14, which provides an additional margin of safety to that incorporated in the adjusted human NOAEL, a value that USEPA had recently recommended as the basis for an MCLG of 300 ppb for chloroform in drinking water. Had the USEPA approach for estimating the RfD and RfC been applied, the MoE for each value would be approximately 100 times greater than our value (*i.e.*, 1400).

It is noteworthy that the concentrations of chloroform measured in and around swimming pools were obtained at facilities that are believed to have followed the traditional chlorination procedures including regular superchlorination. The amount of chloroform produced during the superchlorination events with gaseous chlorine should be no greater than those reported, because at that level of chlorination, the oxidation of byproducts (of which chloroform is one) would be far greater than at 3 to 5 ppm FAC found in pools much of the time. Thus these results should apply correspondingly to pools in which superchlorination (whether through periodic gas chlorination or some other process) is the means of proper disinfection and health protection.

Consequently, the levels of chloroform to which swimmers are exposed in a pool having a concentration of 10 ppm chlorine, regardless of the detailed means of chlorination, provide a reasonable certainty of no harm to the health of swimmers. These findings strongly support the reliance on 10 ppm chlorine as guidance for re-entry of swimmers into chlorine-treated pools. Adopting 10 ppm chlorine as the re-entry value will ensure continued public health protection without contributing an undue health risk to swimmers.

Estimation of the Health Risks and Safety from Exposures to Chlorine and Chloroform for Swimmers in Residential and Commercial Pools

1 Introduction

The Sapphire Group, Inc. conducted this study to determine the scientifically supported (1) concentrations of chlorine for safe re-entry of users of any pool that has been disinfected with chlorine, and (2) levels of exposure to chloroform that pose no undue health risk to swimmers in pools disinfected with chlorine.

Chlorination of swimming pools is an essential public health measure to protect against exposures to dangerous pathogens such as *E. coli* (0157:H7), *Giardia*, Hepatitis A, and *Cryptosporidium*. According to the Centers for Disease Control and Prevention (CDC), approximately ten outbreaks of waterborne gastroenteritis per year in 1997 and 1998 were caused by the parasite *Cryptosporidium*, and 90% were “associated with recreational use of treated water in venues such as swimming pools...” (CDC, 2000a). In these cases, fecal accidents that overwhelmed disinfectant capacity of the pools are strongly suspected. Outbreaks of dermatitis of bacterial origin have also been associated with deficiencies in chlorination of water in pools and spas (CDC, 2000a). The Washington State Environmental Health Association has compiled a list of the types of illnesses related to improperly disinfected pools (WSPHA, 1997).

To be fully effective, chlorine (and any other) disinfectant⁴ must be present continually in sufficient concentrations to act as a barrier against the survival of newly introduced pathogens. Achieving this goal requires continuous maintenance of an optimum free available chlorine (FAC) residual of 2-5 ppm with a maximum of 10 ppm (NSPI, 1999). Periodic superchlorination⁵ to about 10 ppm such as that recommended by the National Spa and Pool Institute (NSPI, 1995, 1999) and the National Swimming Pool Foundation (NSPF, 1990) is

⁴All chlorine, whether introduced as a gas or as a dry or liquid compound, when added to water, forms hypochlorous acid (HOCl) and hypochlorite ion (OCl⁻) (NSPF, 1990), which together represent free available chlorine. HOCl is the form of chlorine that kills pathogens.

⁵“Superchlorination” is defined as the periodic addition of chlorine at ten times the amount of “combined” chlorine, also called chloramines (Williams, 1999). For example, if the combined chlorine in pool water were 1 ppm, then 10 ppm (10 × 1) chlorine would be added. In residential swimming pools, disinfection often relies solely on the periodic (e.g., weekly or biweekly) application of gaseous or other forms of chlorine which is applied in quantities sufficient to achieve the benefits of superchlorination and to maintain an adequate measure of health protection for swimmers between applications.

needed to ensure that the minimum chlorine level is always present. While *E. coli* and Hepatitis A may be destroyed relatively quickly by chlorine at 1 ppm, nearly seven days of contact time would be required at that level of chlorine to kill *Cryptosporidium*. With superchlorination to 10 ppm, however, that disinfection time is reduced to about 16 hours (CDC, 2000b), enabling safe re-entry of swimmers into chlorinated pools.

The application of chlorine to swimming pools is generally of two types: (1) direct addition of chlorine (including gas, liquid bleach, tablet and/or granular chlorine) to residential pools by a homeowner or pool company, and (2) a combination of periodic superchlorination and continuous feed of chlorine through a fixed chlorinator as is usually found at commercial pools. This study focused on a chlorine level for re-entry by swimmers regardless of how the chlorine was applied.

Maintaining water quality is an important part of the disinfection process. It requires control of several parameters under the supervision of a trained professional: (1) stabilization with cyanuric acid, (2) pH control, (3) total alkalinity, (4) calcium hardness, and (5) total dissolved solids. Maintaining recommended FAC levels will prevent most bacterial outbreaks. Compromising any of the essential conditions can reduce chlorine's effectiveness as a disinfectant, a condition that could endanger the health of swimmers.

Without superchlorination, maintaining this minimum throughout the pool and throughout the day without exceeding the maximum of 4 ppm recently proposed by USEPA is unlikely at public and residential pools with high temperatures and high bather loads. A 4 ppm maximum provides operators with little safety margin, and it also discourages shock treatment that is needed regularly to oxidize chloramines and maintain FAC at public pools. Indeed, NSPI (1995, 1999) recommends "regular superchlorination" under conditions of hot weather and heavy use. Regrettably, when faced with such a limitation, public pools often fail to shock treat because chlorine levels will not return to 4 ppm before the pool is scheduled to open. Indeed, the public health community has observed that some managers of public pools, concerned that the level of FAC might exceed 4 ppm the day following superchlorination, will refrain from the superchlorination practice, thereby appreciably increasing the risk that disinfection will be inadequate to prevent waterborne illness among swimmers. Thus, in the United States as in many other parts of the world, these disinfection practices, including superchlorination up to 10 ppm, allow for the safe recreational use of water at home and at public pools.

USEPA, as part of its process to re-register chlorine as a disinfectant under the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) has asked whether re-entry, when levels of chlorine are as high as 10 ppm following superchlorination of residential pools, is safe or poses an unacceptable risk to those who swim in pools that rely on this method for disinfection.

Historically, USEPA had no upper limit on chlorine levels in residential pools. Recently, USEPA has proposed a guideline of 4 ppm, of free available chlorine as a level considered safe for swimmers to re-enter a chlorine-treated pool (USEPA, 1999). Since USEPA first recommended its re-entry guideline, additional scientific information has become available to warrant a present re-examination of that re-entry value. USEPA has also indicated that whereas the original guideline had been based on considerations of the hazards posed solely by chlorine, the Agency is considering expanding its evaluation to include chloroform, a byproduct of chlorine combining with natural organic material.

In a spirit of cooperation, several industry groups with interest in this matter have sponsored this independent evaluation of safety and risk to assist USEPA in its regulatory decision making. While the issue with respect to the appropriate re-entry level for free available chlorine has arisen in connection with the re-registration of gaseous chlorine for the disinfection of residential swimming pools, the question may be just as suitable in the commercial pool context.

The study reported herein thus addresses the safety and risks of chlorine and chloroform to users of residential and commercial swimming pools containing FAC. This study was undertaken to examine all relevant information so as to determine whether the maximum re-entry guideline could be justifiably raised from 4 ppm to 10 ppm. The study first examined the data on chlorine and then on chloroform. The methodological approach of this study is that of risk assessment as originally defined by the National Research Council (NRC, 1983) and as subscribed to by USEPA for over two decades. This methodology systematically analyzes data of three types (human exposure⁶, hazards⁷, and toxic potency⁸), and assimilates the findings in a risk characterization step that includes a weight-of-evidence analysis.

The method used herein to estimate safety is a modification of what USEPA calls its reference dose (RfD) and reference concentration (RfC) methodology (USEPA, 1993). In our study, the experimentally derived no-observed-adverse-effect level (NOAEL) was converted to an “adjusted human NOAEL” through the application of uncertainty factors identical to those applied by USEPA. The Agency usually refers to these values as either RfD for ingested

⁶“Exposure assessment” determines who gets exposed to how much of specific substances and under what circumstances.

⁷“Hazard identification” is the step that defines the toxic properties that are known or anticipated to be produced in humans.

⁸“Dose-response assessment” or “toxic potency assessment” estimates the amounts needed to produce toxic effects under specific circumstances.

substances and RfC for inhaled substances. The “adjusted human NOAEL” and the RfD or RfC have the same scientific meaning in risk analysis, namely that exposures at and below each value have a reasonable certainty of no harm.

Judging the safety of specified exposures can be accomplished by estimating the margin of exposure (MoE). We chose to use the “adjusted human NOAEL” as the value against which to determine how much additional safety was present in addition to that which is defined by the composite uncertainty factors. USEPA, by contrast, prefers to describe the MoE by starting from the experimental NOAEL, which is not a direct index of safety for humans. The advantage of our procedure over that of the Agency in this case is that it more clearly defines safe levels of exposure to humans. In addition, it then provides, at a subsequent step, a quantitative basis for estimating the additional measures of safety and provides a level of confidence whether or not exposures are more or less likely to produce any undue risk to the health of swimmers.

The practical difference between the two approaches is illustrated by the following. For a substance for which a rodent NOAEL has measured as 10,000 mg/kg-day, for which human exposure was estimated to be 5 mg/kg-day, and for which the applicable uncertainty factors were a product of 100 (10×10), we would describe the MoE as 20 ($10,000 \div 100 \div 5$) times above the level unlikely to harm humans. Using the USEPA approach, the MoE would be 2,000 ($10,000 \div 5$) times greater than the rodent NOAEL; and if the Agency specified 100 as an adequate uncertainty factor to protect humans against harm, then the justifiable conclusion would be that the exposure level had a reasonable certainty of no harm by a factor of 20.

This report first characterizes exposures of swimmers to chlorine and chloroform (Chapter 2), since exposure has a major impact on delineating the nature and magnitude of safety and risk. Next, the relevant hazard properties and toxic potency of chlorine and chloroform are described, and their relevance to swimmers is identified (Chapter 3). The adjusted human NOAELs (referred to by some as “virtually safe doses”) of each compound are characterized using appropriate metrics including a concentration of chlorine considered safe for re-entry of swimmers in such treated pools. Finally, the toxic potency of each compound is compared with estimates of actual exposure to obtain the margins of exposure (Chapter 4).

2 Exposure Assessment

Exposure of humans to the compounds of interest must be characterized in order to estimate levels of chlorine and chloroform that are judged to be safe for re-entry into pools treated with gaseous and other forms of chlorine. Evaluating exposure requires identifying, for the substances of interest, demographics of the exposed population, the magnitude and frequency of contact with the substances, the routes of exposure, and the durations of contact.

In this case, the compounds of interest are, by definition, chlorine and chloroform. The routes of exposure considered herein are skin contact while in the pool, inhalation of compounds in the breathing zone of pool users, and incidental ingestion of water during swimming. For substances absorbed into the systemic circulation (specifically, chloroform), the rates of absorption by tissues serving as portals of entry into the body can be used to integrate doses systemically. Other characteristics that make up the exposure scenarios are defined below.

Data on the presence of chlorine and chloroform are available on commercial indoor and outdoor pools that use chlorine for disinfection. These data provide an indication of the range of concentrations of chloroform that can be expected in pool water and in the air in the breathing zone of swimmers. They serve as a means of approximating median concentration levels for long durations as well as peak levels for short times. The data from commercial indoor pools can serve as an approximate upper-bound (*e.g.*, 90th percentile), because commercial indoor pool conditions favor higher concentrations of chloroform in the air than are usually present at outdoor pools. Since only outdoor residential pools use gaseous chlorination and since the fraction of outdoor pools is much greater than indoor pools, relying in part on concentrations from indoor pools overstates the size of the population exposed to the concentrations at the higher end of the range of exposure. The data from commercial pools provide a basis for making reasonable, conservative approximations of what concentrations of chlorine and chloroform are likely to be present with the application of chlorine to residential pools, virtually all of which are outdoors.

An overview of exposure to chloroform via ingestion of drinking water and via skin contact has been prepared by ILSI (Olin, 1999).

2.1 Populations Exposed

The population of swimmers includes people of all ages (including the elderly) and of both genders. Swimmers in pools are expected to be, for the most part, involved in recreational water activities, some even in therapeutic swimming. The individuals exposed are children and adults who swim. The exposure to chlorine and chloroform at residential pools of competitive

swimmers for a portion of their lives is expected to be limited. Competitive swimmers may use residential pools for some training; however, they would not use residential pools for competitive events, because in part residential pools are relatively small compared to pools used for competitive swimming. Also, competitive swimmers are usually on teams that train together at commercial or institutional pools. These individuals might use residential pools for occasional practice to supplement the team training elsewhere, but, in general, this use would likely be minimal.

Various unpublished surveys⁹ provide insight into the extent to which people swim:

- In the U.S. in 1993, nearly half of working women exercise at least once a week, and 30% of white collar working women exercise three times per week. Of the many types of exercise undertaken by these individuals, swimming ranks fourth and accounts for half of those who exercise; therefore, about 25% of working women swim approximately once a week.
- In the United States in 1991, approximately 10 million individuals exercised by both swimming and running. In 1988, approximately 71 million individuals participated in swimming.
- Among “baby-boomers” in the U.S. prior to 1984, 46 percent swam.
- In the United States in 1984, swimmers swam an average of 40 times per year and 41 percent of Americans swam.
- In California in 1994 and in 1991, most residential outdoor pools were open for six months of the year, and approximately 30 percent were open year-round (Occidental Chemical Corporation, 1991, 1994).

2.2 Swim Conditions

The range of exposure scenarios is very broad: from those who do not go near the water to those who swim nearly every day for extended periods of their lives. No one knows who or what scenario is “average.” The backdrop, however, is important in making reasonable estimates of contact frequencies and durations. The temperate climate and culture in the southern States promote swimming activities. Many homes have private swimming pools, many families belong to swim clubs (some open year round), and many children learn to swim early in life and later join swim teams (swimming for one hour for three to five days per week). Some have suggested that anyone who enjoyed swimming as a child is likely to continue to do so for health and/or recreational reasons as adults. From personal observations, we are aware of adults who swim an hour or so daily for physical fitness.

⁹Personal communication. Chlorine Chemistry Council, Arlington, Virginia, 1995.

For this analysis, the authors have defined the “average” exposure for a swimmer as swimming in a chlorinated pool one hour per day, three days per week, 39 weeks per year (for outdoor pools) or 50 weeks per year (indoor pools), for 35 years. Since use of outdoor pools is certainly temperature and weather dependent, the frequency of use was reduced from 50 to 39 weeks per year. The selection of 39 weeks of use was predicated on information provided on California pools from Occidental Chemical Data Scope for the years 1991 and 1994 (two surveys; Occidental Chemical Corporation, 1991, 1994). According to these data, approximately half of the pools surveyed in California were open six months of the year; and about one-third were open the full year, with an average of approximately eight months of the year. In our judgment, a mid-point in duration (*i.e.*, nine months) between a half year and a full year seems a reasonable approximation, based on limitations of the data, recognizing that a worst case assumption is that of the yearly duration of swimming. For this analysis, we have selected 39 weeks (117 events per year) as the basis for yearly duration of swimming in outdoor chlorinated pools, and 50 weeks (150 events per year) as an upper-bound.

We selected 35 years (*i.e.*, 12,775 days per 70-year lifetime) as a reasonable mid-point in a 70-year lifetime, because it appears counter-intuitive to assume daily swimming from cradle to grave. Interests change during people’s lives: students go to college (most are not on a swim team), children and adults go on vacation; women have babies; people lose interest; people are hospitalized, etc. This value is compatible with using 70 years of use as an upper-bound. The default duration of exposure identified in USEPA draft SWIMODEL is 32 years (12 as a child and 20 as an adult) (Dang, 1996).

Another condition used herein is an inhalation rate for swimmers of 1 m³/hour, which represents the weighted (9:1) average of recreational swimmers and competitive swimmers, based on breathing rates found in USEPA’s Exposure Factors Handbook for light and heavy activity, respectively (USEPA, 1995).

2.3 Concentrations in the Breathing Zones and Water of Swimmers

Estimates of the degree to which individuals are exposed to chlorine and chloroform from chlorinated pools were obtained by considering several types of data, including the populations exposed, swim conditions that influenced breathing rates, and the concentrations of chloroform in the breathing zone of those who use pools. This step describes the information defining the concentrations of the compounds in water used for swimming and in the air in the breathing zone of swimmers.

Major limitations of the exposure assessment include (1) the relatively few data characterizing the concentrations of chloroform above the water of swimming pools; (2) the relatively large

variation in measured chloroform above water in swimming pools; and (3) the measurement of chlorine in pool water.

2.3.1 Chlorine

2.3.1.1 Concentrations in water: By design, superchlorination with gaseous or other forms of chlorine is performed regularly to achieve an initial concentration of at least 10 ppm chlorine in pools that have proper pH balance and stabilization, and thereby maximize public health protection. If the initial concentration of chlorine exceeds 10 ppm, within a few hours the chlorine concentration has generally been reduced to about 10 ppm¹⁰. The decay rate thereafter can be in the range of 0.5-1 ppm per day⁵; however, the rate is highly dependent upon several factors, particularly organic loading, temperature, and sunlight (outdoor pools) that consume available chlorine. Proper pool maintenance should achieve chlorine levels between two and five ppm between superchlorination events (NSPI, 1999).

The measured concentrations of chlorine reported in commercial indoor pool water of swimmers are presented in **Table 1**. Two data sets described the concentrations of chlorine in the water immediately prior to or after individuals swam in the pool water. The types of measurement, range, midpoint, average, and citation are presented. The results indicated concentrations of 0.5 and 0.35 ppm of chlorine.

No published data were found on chlorine levels in outdoor pools. To assure protection of health, chlorine levels are to be kept between two and five ppm (NSPI, 1999).

2.3.1.2 Concentrations in air: Gaseous chlorine and hypochlorite are highly water soluble, and are added to pool water by techniques that promote retention in water rather than release to the atmosphere. Chlorine in a balanced pool should volatilize little, if any, as unreacted chlorine, although some aerosolization of water containing chlorine occurs when the water is highly agitated as in diving and splashing. No reports were found indicating that gaseous chlorine was measured in the air above the treated pool water.

At times, an odor of “chlorine” can be detected in the air at some pools. Some individuals have erroneously concluded that such odor is due to chlorine gas; however, such odor results from chloramines that are present only when the pool water is not balanced properly.

When pool water is being re-balanced, swimming is not recommended, and no exposure to chlorine is likely to occur at that time. Since chloramines represent an aberrant and undesired situation in swimming pools and since pools are treated to prevent the formation of

¹⁰Personal communication, National Association of Gas Chlorinators, September 2000.

chloramines, these compounds are not included in this evaluation of safety and risk to human health.

2.3.2 Chloroform

2.3.2.1 Concentrations in water: Concentrations of chloroform reported in commercial pool water of swimmers are presented in **Tables 1** and **2**. Several sets of data (some citations contain more than one data set) describe the concentrations of chloroform in the water immediately prior to or after individuals swam in the pool water. The types of measurement, range, midpoint, average, and citation for twelve of the data sets are presented in these tables. Of these data sets, ten describe chloroform levels in indoor pools, and two in commercial outdoor pools.

The preferred statistic of each data set is the mean, as a partial definition of the measure of “average” specified by USEPA. If no mean was available or can be calculated, the median is calculated and used. If neither is available or capable of being calculated, the midpoint of each range is an appropriate value for an evaluation such as this one, since we are seeking to approximate the average. Data between pool types (indoor vs. outdoor) or between individual reports (even if by the same authors) were not averaged.

The range of concentrations of chloroform in water at commercial indoor pools was reported to be from 3 ppb to about 140 ppb. At commercial outdoor pools, the concentrations ranged from 4 ppb to approximately 127 ppb. One data set (Levesque *et al.*, 1994) was judged to be invalid for this analysis, because the investigators spiked the pool water with chloroform to test a hypothesis about the toxicokinetics of chloroform among swimmers. Regardless, these data suggest a 50-fold range of concentrations (3-140 µg/L), reflecting environmental conditions. Although not reported explicitly, chlorine was believed to have been added continuously to these public pools at 2-5 ppm, and routine superchlorination was believed to have been practiced.

2.3.2.2 Concentrations in air: Several sets of data (some citations contain more than one data set) were obtained describing the concentrations of chloroform in the air immediately above or adjacent to the pool water. The types of measurement, range, midpoint, average, and citation for six of the data sets are presented in **Tables 3** and **4**. Of these data sets, twelve address indoor pools, and five address outdoor pools. In addition, Matthiessen and Jentsch (1999) reported on the presence of trihalomethanes (THM) in the air above indoor swimming pools, but without differentiating the individual compounds of that group; therefore, these results were not included in **Table 3**.

The preferred statistic of each data set is the mean, as a partial definition of the measure of “average” specified by USEPA. If a mean was unavailable or could not be calculated, the median was calculated and used. If neither was available nor capable of being calculated, the midpoint of each range was an appropriate value for an evaluation such as this one, since we are seeking to approximate the average. Note that several papers report data points as averages of several measurements, usually for one location or time of sampling; in this instance, these individual data averages found within one report were further averaged to obtain estimates of exposure. Data between pool types (indoor vs. outdoor) or between individual reports (even if by the same authors) were not averaged.

The range of concentrations of chloroform in water at commercial indoor pools was reported to be from 0.5 ppb to about 640 ppb. At commercial outdoor pools, the concentrations ranged from 0.1 ppb to 140 ppb. No U.S. standard exists for chloroform in swimming pools against which to compare these reported concentrations. However, these values can be compared to USEPA’s short-term (10-day) health advisory of 4 mg/L (ppm) of chloroform in drinking water (USEPA, 2000); this health advisory was developed to be protective for children (10-kg body weight) as well as adults. All reported concentrations in pool water are well below this health advisory.

2.4 Estimation of Human Doses

The metrics to express human doses included the following: (1) mg/kg-day for absorbed doses (ingestion, inhalation, skin contact), and (2) mg/m³ for skin contact with chlorine.

The average daily doses from skin contact with water containing chloroform were calculated using the matrix and equation¹¹ presented in **Table 5**.

The average daily doses from inhalation of air were calculated using the following equation¹²:

$$\text{dose } (\mu\text{g/person-day}) = \text{concentration } (\mu\text{g}/\text{m}^3) \times 1 \text{ m}^3 \text{ inhaled/hr} \times 1 \text{ hr/day} \times 3 \text{ days/7 days} \times 50 \text{ weeks/52 weeks} \times 35 \text{ yrs/ 70yrs}$$

The average daily doses from ingestion of water were calculated using the following equation:

¹¹Note: The length of the equation to derive dermal doses precluded its inclusion in the text; rather, this equation is located in Table 5.

¹²Duration of swimming = 39 weeks per year for outdoor pool use and 50 weeks per year for indoor pool use; no other differences between outdoor and indoor pools.

$$\text{dose } (\mu\text{g/person-day}) = \text{concentration } (\mu\text{g/L}) \times 0.05 \text{ L ingested/hr} \times 1 \text{ hr/day} \times 3 \text{ days/7 days} \times 50 \text{ weeks/52 weeks} \times 35 \text{ yrs/70 yrs}$$

The ingestion rate of 50 ml/hr was based on 95th percentile exposures as estimated by USEPA (1995).

2.4.1 Chlorine

Chlorine in water at such low concentrations as those encountered in chlorinated pools may interact with the tissues with which it comes directly into contact; however, it is not absorbed into the systemic circulation as chlorine but rather as chloride which has no toxicological significance. Therefore, the doses are estimated at each portal of entry (lungs, gastrointestinal tract, and skin), and these respective doses are not summed.

2.4.1.1 Skin Contact: The dose of chlorine to the skin is presented as the concentration that comes into contact with it, with 10 ppm as the re-entry level shortly after superchlorination. One hundred percent of the body surface area is estimated to come into contact with the chlorine in pool water. When establishing the MRDLG for chlorine, USEPA has determined that exposure to 4 ppm chlorine is unlikely to cause harm, with an adequate (magnitude undetermined) margin of safety, to those who bathe and/or shower with such water.

2.4.1.2 Inhalation: Because the chlorine gas is unlikely to be present above the surface of the water in a properly maintained pool, this exposure is judged to be essentially zero and thus of no toxicological significance.

2.4.1.3 Incidental Ingestion: Free chlorine does not survive long enough in saliva to be available for absorption and to produce any direct systemic effects of toxicological significance. According to USEPA's Science Advisory Board (USEPA, 1990), very high doses of chlorine (>100 ppm) might be sufficient to produce some internal exposure to free chlorine ion, but no data exist to suggest that the resulting concentrations would be sufficient to produce significant toxicological effects.

2.4.1.4 Combined Systemic Doses: The doses from each portal of contact are not summed because ample understanding exists to indicate that chlorine is not absorbed systemically. This assumption is based on laboratory kinetic data that suggest that at the point of contact between free chlorine and tissues, the free chlorine is converted to chloride ion (USEPA, 1994a). This small amount of chloride ion then becomes part of the larger volume of chloride ion in the body, and is of no toxicological consequence.

2.4.2 Chloroform

The systemic doses of chloroform were estimated first by each route of contact (skin contact, inhalation, ingestion), and then the doses from each route were summed to obtain estimates of systemically absorbed doses.

2.4.2.1 Skin Contact: The dose of chloroform in pool water from contact with the skin is a function of the surface area of the skin, the duration of contact, and the rate of absorption through the skin. The integration of this information is presented in a model described in **Table 5**. The following data (two studies of humans and two in rats) provide the underpinnings for the rates of absorption, a key parameter.

A shower study was conducted, where humans breathed purified air while being exposed to chloroform via skin contact (Weisel and Jo, 1996). The concentration of chloroform in water was measured during the experiment: With whole body skin contact, the amount of chloroform expired per μg chloroform in one liter of water ranged from 0.33 (10 minute shower) to 0.56 (60 minute shower) μg . The amount of chloroform expired via inhalation and dermal contact were similar, leading the authors to conclude that the internal dose via dermal contact was 30 μg and 80 μg for 10 and 30 minute contact times, respectively.

In another study, humans exposed dermally to chloroform via bathwater showed a significant decrease in the amount of chloroform exhaled as exposure temperature decreased from 40°C to 35°C to 30°C. The skin permeability coefficient was calculated to be 0.06 cm/hr at 40°C for both males and females (Corley *et al.*, 1999).

Some laboratory animal data are available for comparison. Chloroform was rapidly cleared from the blood of rats exposed dermally for 1, 3, or 8 min of 99% chloroform (with 1% ethanol) solution; and the terminal elimination rate constant was estimated to be 0.009/min (Islam *et al.*, 1999). Of the total uptake (29 mg) from a dilute stirred solution of chloroform (0.44 $\mu\text{g}/\text{L}$) at 36°C, 95% was absorbed after a 30 minute exposure to hairless rats in a bath. (Islam *et al.*, 1995).

The results of applying the equation on **Table 5** and described in **Table 6** indicate that the systemic dose of chloroform by skin contact ranged from 0.03 to 0.23 $\mu\text{g}/\text{person-day}$ when the concentration of chloroform in pool water ranged from 14 to 140 $\mu\text{g}/\text{L}$.

2.4.2.2 Inhalation: The dose of chloroform in pool water from inhalation is a function of the breathing rate of a swimmer, the duration of swimming, and the rate of absorption through the lungs. The following data (one study of humans) provide the underpinnings for the rate of absorption, a key parameter.

A shower study was conducted where humans wore waterproof clothes while being exposed by inhalation (Weisel and Jo, 1996). The concentration of chloroform was measured during the experiment. The amount of chloroform expired per μg chloroform in one liter of water ranged from 0.33 (10 minute shower) to 0.56 (60 minute shower) μg after inhalation exposure. The inhalation and dermal expiration doses were similar, leading the authors to conclude that the internal dose via inhalation contact was 30 μg and 80 μg for a 10 and 60 minute contact times, respectively.

Using the equation above and the relevant parameters, results described in **Table 6** indicate that the systemic dose of chloroform by inhalation ranges from about 0.08 to about 43 $\mu\text{g}/\text{person-day}$ when the concentration of chloroform in pool water ranged from about 0.5 to slightly over 200 $\mu\text{g}/\text{L}$. The levels measured at outdoor pools (midpoint = 110) were much lower than those at indoor pools (midpoint = 23).

2.4.2.3 Incidental Ingestion: The dose of chloroform in pool water from incidental ingestion is a function of the volume taken into the mouth, the fraction swallowed, the frequency of occurrence per unit of time, the duration of swimming, and the rate of absorption through the buccal membranes and the gastro-intestinal tract. The following data (one study of humans) provide the underpinnings for the rate of absorption, a key parameter.

In a study by Fry *et al.* (1972), a single oral dose of either 0.5 or 1.0 g chloroform was administered to 12 male and female volunteers (six per test group). The 0.5 g test group received their dose via a gelatin capsule while the 1 g group received their dose via an olive oil solution. For eight hours post-exposure, the test subjects wore a sealed gas-mask and expired gases were measured at 10 minute intervals. Urine and blood samples were also collected with the latter sampling continuing 28 hour post-exposure. The authors reported a linear relationship between rate of pulmonary excretion and concentration in blood. After peak blood levels of up to 5 μg chloroform/ml, the proportion of the dose excreted in eight hours via the lungs ranged from 17.8 to 66.6 percent. Further, chloroform could be detected at some concentration (but below measurable limits) up to 24 hours post exposure.

Using the equation above and the relevant parameters, results described in **Table 6** indicate that the systemic dose of chloroform by incidental ingestion ranges from about 0.15 to about 1.4 $\mu\text{g}/\text{person-day}$ (= 0.002-0.02 $\mu\text{g}/\text{kg-day}$) when the concentration of chloroform in pool water ranged from about 14 to 140 $\mu\text{g}/\text{L}$.

2.4.2.4 Combined Systemic Doses: Because chloroform is a systemic toxicant, the doses obtained simultaneously from multiple routes should be summed to accurately characterize the magnitude of the internal doses obtained by swimmers. Given the wide range of measured values for indoor and outdoor pools using continuous application of chlorine, the maximum

cumulative dose presented in **Table 12** was 42.5 µg/person per day, which is comprised of 0.23 µg from skin contact, 40.8 from inhalation, and 1.44 µg from incidental ingestion.

Since most residential pools, particularly those treated by gas chlorination, are outdoors, the more likely doses to the majority of swimmers will be at the lower end of this range. These data will serve as a basis for risk characterization in **Chapter 4**.

Table 1. Concentrations of Chloroform and Free Chlorine in the Water of *Indoor Pools*

<u>Facility</u>	<u>Monitoring Location</u>	<u>Chloroform Concentration (?g/L)</u>		<u>Free Chlorine Concentration (mg/L)</u>		<u>Citation</u>
		<u>Range</u>	<u>Average</u>	<u>Range</u>	<u>Average</u>	
12 public	in water (N=127 people)	17-47	30.9	NA NA		Aggazzotti <i>et al.</i> , 1995
1 public	in water (N=10 people)	NA	24.0	NA NA		Aiking <i>et al.</i> , 1994
1 public	in water (N=11/7 sessions)	158.6-567.5	364.8	NA NA		Levesque <i>et al.</i> , 1994
1 public	in water (N=5/4 sessions)	25-43	33.7	NA NA		Aggazzotti <i>et al.</i> , 1998
1 public	in water (N=28 people)	3.04-27.8	14.14	NA NA		Cammann & Hubner, 1995
8 public	in water (N=11/pool)	NA	NA	NA 0.51		Lahl <i>et al.</i> , 1981
2 public	in water (N=8/3 sessions)	18.9-93.6	37.23	NA NA		Aggazzotti <i>et al.</i> , 1993
1 public	in water (N=?)	16.8-46.98	32.6	NA NA		Faust, 1993
1 public	in water (N=5)	32-150	85	NA NA		Weisel & Shepard, 1994
4 public	in water (N=?)	3-580	140.25	NA NA		Armstrong and Golden, 1986
4 public	in water (N=22)	NA	NA	NA 0.35		Drobnic <i>et al.</i> , 1996

N= number of detectors or people wearing detectors

Table 2. Concentration of Chloroform and Free Chlorine in the Water of *Outdoor Pools*

<u>Facility</u>	<u>Monitoring Location</u>	<u>Chloroform Concentration (µg/L)</u>		<u>Free Chlorine Concentration (mg/L)</u>		<u>Citation</u>
		<u>Range</u>	<u>Average</u>	<u>Range</u>	<u>Average</u>	
1 public	in water (N=8 people)	NA	18.4	NA	NA	Aiking <i>et al.</i> , 1994
5 public	in water (N=?)	4-402	127.6	NA	NA	Armstrong and Golden, 1986

N= number of detectors or people wearing detectors

Table 3. Concentrations of Chloroform in the Air of *Indoor Pools*

<u>Facility</u>	<u>Monitoring Location</u>	<u>Concentration ($\mu\text{g}/\text{m}^3$)</u>		<u>Citation</u>
		<u>Range</u>	<u>Average</u>	
12 public	above water (N=127 people)	66-650	179	Aggazzotti <i>et al.</i> , 1995
1 public	above water (N=28 people)	8-191	79	Cammann & Hubner, 1995
4 public "baths"	10-20 cm above water	80-102	92	Ullrich, 1982
8 public	above water (N=11/pool)	33-241	117	Lahl <i>et al.</i> , 1981
2 public	above water (N=8/pool x 3 sessions)	48-277	136	Aggazzotti <i>et al.</i> , 1993
2 public	2 cm above water (N=3/pool)	21-331	198	Armstrong & Golden, 1986
3 public	above water (N=18/pool x 3 sessions)	65-642	211	Aggazzotti <i>et al.</i> , 1990
1 public	above water (N=?)	32-90	[62=MP]	Faust, 1993
2 public	above water (N=?)	28-34	31	Jo, 1994
4 public	above water (N=?)	0.5-331	108.75	Armstrong and Golden, 1986
* * * * *				
2 public	lifeguards personal air (N=3)	46-95	70	Wallace, 1997
1 public	6 cm above floor; 3 m from edge (N=5)	23-120	87	Weisel & Shepard, 1994
2 public	2 m above floor; 2 m from edge (N=3/pool)	0.5-260	128	Armstrong & Golden, 1986

MP = midpoint of range (used in the absence of an average or median); N= number of detectors or people wearing detectors

Table 4. Concentration of Chloroform in the Air of *Outdoor Pools*

<u>Facilities Monitored</u>	<u>Monitoring Location</u>	<u>Concentration ($\mu\text{g}/\text{m}^3$)</u>		<u>Citation</u>
		<u>Range</u>	<u>Average</u>	
4 public	2 cm above water (N=5/pool)	4-140	62	Armstrong & Golden, 1986
1 public	0.5 m above water (N=?)	0.4-1.2	0.8 [MP]]	Chou, 1991
5 public	above water (N=?)	4-140	48	Armstrong and Golden, 1986
* * * * *				
1 public	lifeguards personal air (N=3)	2.0-5.2	3.1	Wallace, 1997
4 public	2 m above edge 2 m from edge (N=4/pool)	0.1-1	0.5 [MP]	Armstrong & Golden, 1986

MP = midpoint of range (used in the absence of an average or median); N = number of detectors or people wearing detectors

Table 5. Dermal Exposure Model for Chloroform in Water

This Model was used to estimate the Absorbed Dose of Chloroform per cm² of Exposed Skin Resulting from Single Water Contact. Minimal Assumptions Needed for Use of this Model:

1. Skin is undamaged and 2. Chloroform is Dissolved in a Water Vehicle

For Example:

Compound:	Chloroform	Skin Surface Area (cm²):	18150	Molec. Wt.:	119.39
Event Freq. (events/day):	1	Stratum Corneum Thickness (cm):	0.00	Exposure Freq. (days/yr):	150
Concentration (mg/ml):	0.00	Exposure Duration (Days/year):	150/365	Carcinogenic Potential (Yes or No?):	No
LogKow:	1.97	Body Weight (kg):	70.00	Averaging Time (days):	150
Water Contact Time (Tevent; hr):	1				

Using the following equation:

$$\text{dose } (\mu\text{g/person-day}) = \text{concentration } (\mu\text{g/L}) \times \text{skin surface area } (\text{cm}^2) \times \text{dermal absorption } (\text{mg/cm}^2\text{-event}) \times 1 \text{ event/day} \times 3\text{day/7day} \times 50\text{weeks/52weeks} \times 35\text{yrs/70yrs}$$

Then the Data Calculation would be:

<i>Compound</i>	<i>Molecular Weight</i>	<i>Octanol-Water Partition Coef. (Kow)</i>	<i>logKow</i>	<i>Skin Permeability Coef. (Kp)</i>	<i>Dimensionless Partitioning Constant (B)</i>	<i>Stratum Corneum Thickness (cm)</i>
Chloroform	119.39	9.33E+01	1.97	8.92E-03	9.33E-03	1.00E-03
<i>Stratum Corneum Diffusivity (cm²/hr)</i>	<i>Lag Time (hour)</i>	<i>Time to Reach Steady-State (hour) - T*</i>	<i>Constants Used for Determination of T* (Only Used When B>1.17)</i>	<i>Dermal Absorption per Event (μg/cm²-event)</i>	<i>Dermally Absorbed Dose (μg/kg-day)</i>	
3.56E-07	4.68E-01	1.12E+00	3.06E-01	3.43E-01	1.69E-05	1.20E-05

Table 6. Average Daily Doses of Chloroform Obtained in Indoor or Outdoor Pools

Inhalation		
<i>Swimmers Indoor - Chloroform</i>		<i>Data Source</i>
<i>Concentration</i> ($\mu\text{g}/\text{m}^3$)	<i>Dose</i> ($\mu\text{g}/\text{person-day}$)	
79.00	16.28	Cammann and Hubner, 1995
92.00	18.96	Ullrich, 1982
117.00	21.11	Lahl <i>et al.</i> , 1981
136.00	28.02	Aggazzotti <i>et al.</i> , 1993
198.00	40.80	Armstrong and Golden, 1986
211.00	43.48	Aggazzotti <i>et al.</i> , 1990
62.00	12.77	Faust, 1993
31.00	6.39	Jo, 1994
108.75	22.41	Armstrong and Golden, 1986
70.00	14.42	Wallace, 1997
87.00	17.93	Weisel and Shepard, 1994
128.00	26.37	Armstrong and Golden, 1986
178.65	36.81	Aggazzotti <i>et al.</i> , 1995
<i>Swimmers Outdoor - Chloroform</i>		<i>Data Source</i>
<i>Concentration</i> ($\mu\text{g}/\text{m}^3$)	<i>Dose</i> ($\mu\text{g}/\text{person-day}$)	
62.00	9.96	Armstrong and Golden, 1986
0.80	0.13	Chou, 1991
48.00	7.71	Armstrong and Golden, 1986
3.10	0.50	Wallace, 1997
0.50	0.08	Armstrong and Golden, 1986

Ingestion

<i>Swimmers Indoor - Chloroform</i>		
<i>Concentration</i> ($\mu\text{g/L}$)	<i>Dose</i> ($\mu\text{g/person-day}$)	<i>Data Source</i>
24.00	0.25	Aiking <i>et al.</i> , 1994
33.70	0.35	Aggazzotti, <i>et al.</i> , 1998
14.14	0.15	Cammann and Hubner, 1995
37.23	0.38	Aggazzotti <i>et al.</i> , 1993
32.60	0.34	Faust, 1993
85.00	0.88	Weisel and Shepard, 1994
140.25	1.44	Armstrong and Golden, 1986
30.9	6.32	Aggazzotti, <i>et al.</i> , 1995

<i>Swimmers Outdoor - Chloroform</i>		
<i>Concentration</i> ($\mu\text{g/L}$)	<i>Dose</i> ($\mu\text{g/person-day}$)	<i>Data Source</i>
18.40	0.15	Aiking <i>et al.</i> , 1994
127.60	1.03	Armstrong and Golden, 1986

Dermal Contact

<i>Swimmers Indoor - Chloroform</i>		
<i>Concentration</i> ($\mu\text{g/L}$)	<i>Dose</i> ($\mu\text{g/person-day}$)	<i>Data Source</i>
24.00	0.04	Aiking <i>et al.</i> , 1994
33.70	0.06	Aggazzotti, <i>et al.</i> , 1998
14.14	0.03	Cammann and Hubner, 1995
37.23	0.07	Aggazzotti <i>et al.</i> , 1993
32.60	0.06	Faust, 1993
85.00	0.15	Weisel and Shepard, 1994
140.25	0.03	Armstrong and Golden, 1986
30.9	0.05	Aggazzotti <i>et al.</i> , 1995

<i>Swimmers Outdoor - Chloroform</i>		
<i>Concentration</i> ($\mu\text{g/L}$)	<i>Dose</i> ($\mu\text{g/person-day}$)	<i>Data Source</i>
18.40	0.03	Aiking <i>et al.</i> , 1994
127.60	0.23	Armstrong and Golden, 1986

3 Hazard Identification and Toxic Potency Assessment

The toxic properties of chlorine and chloroform are summarized below, with an emphasis on those properties that are known and likely to be manifest in humans. Also, where known, the toxic potency of each compound for each of three routes of exposure and for three durations of exposure is presented. The toxic potency has two aspects: (1) it describes how much of a substance is needed to elicit an adverse response; and (2) it also describes doses at and below which no adverse response is observed and expected. The observed value is often referred to as the “no-observed-adverse-effect level (NOAEL), which can then be used to estimate the adjusted human NOAEL by taking into account several scientific considerations.

The hazard information is described first by route of exposure (skin contact, inhalation, and ingestion—from most to least likely form of contact), and then by duration of exposure generally encountered in laboratory toxicity studies (*viz.*, less-than-lifetime also called “subchronic;” lifetime, referred to as “chronic;” and a few, brief exposures, called “acute”). The order of presentation by durations of exposure was chosen deliberately to reflect the most to the least likely scenarios for most swimmers. Most swimmers will likely fall within the less-than-lifetime or subchronic category. For instance, residential and commercial pools are likely to be used by individuals intermittently (not every day) for several years, perhaps even a few decades. The ends of the distributions of the use of pools include situations where a few individuals swim only once or twice over their lifetime in properly chlorinated pools or those few who swim in chlorinated pools very frequently throughout their lifetime.

For each category of study, when multiple studies relevant to this risk assessment were identified, the one that possessed a combination of the lowest NOAEL and the strongest scientific characteristics of design, execution, and relevance to humans was selected. Only that most relevant study in each category and with the lowest NOAEL is summarized in this report. The major toxic properties reported in these studies were listed.

To address the toxic potency of chlorine and chloroform by the exposure routes of interest, the relevant NOAEL was identified, and through the application of appropriate uncertainty factors, was converted, when needed, to adjust NOAELs for humans. The adjusted NOAELs are levels at and below which the risk of health injury is highly unlikely; therefore, these values are sometimes referred to as “safe” levels. This information is used in the risk characterization section (Chapter 4). NOAELs for chlorine and chloroform by route of exposure are presented in **Tables 7 and 8**.

Table 7. Actual and Adjusted NOAELs for humans exposed to chlorine

Route	Duration	Observed NOAEL	Uncertainty Factor(s)	Adjusted NOAEL	Basis (Reference)
Dermal	Subchronic	> 10 mg/L (H ₂ O)	not calculated	> 10 mg/L (H ₂ O)	Human anecdotal information; inferred from ACGIH, 1996a
	Chronic	> 10 mg/L (H ₂ O)	not calculated	> 10 mg/L (H ₂ O)	Human anecdotal information; inferred from ACGIH, 1996a
	Acute	> 10 mg/L (H ₂ O)	not calculated	> 10 mg/L (H ₂ O)	Human anecdotal information; inferred from ACGIH, 1996a
Inhalation	Subchronic	2433 µg/m ³ (air)	1+ ¹³	2433 µg/m ³ (air)	Human irritation (Anglen 1981); supported by ACGIH, 1996a
	Chronic	2433 µg/m ³ (air)	1+	2433 µg/m ³ (air)	Human irritation (Anglen 1981); supported by ACGIH, 1996a
	Acute	4866 µg/m ³ (air)	1+	4866 µg/m ³ (air)	Human irritation (Anglen 1981); supported by ACGIH, 1996a
Ingestion	Subchronic	14 mkd	100	140 µgkd	Animal chronic toxicity (NTP, 1992); supported by several subchronic studies (Daniel <i>et al.</i> , 1991) and USEPA, 1998a
	Chronic	14 mkd	100	140 µgkd	Animal chronic toxicity (NTP, 1992); supported by USEPA, 1998a

¹³ACGIH recommended an 8-hour TLV of 0.5 ppm and a 15-minute STEL of 1 ppm based on the human NOAELs reported by Anglen (1981).

	Acute	900 mkd	100	9000 μ gkd	Animal acute toxicity (Cunningham, 1980)
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mkd = mg chemical per kilogram of body weight per day

μ gkd = μ g chemical per kilogram of body weight per day

Table 8. Observed/Estimated and Adjusted NOAELs for humans exposed to chloroform

Route	Duration	Observed NOAEL	Uncertainty Factor(s)	Adjusted NOAEL	Source Reference
Dermal	Subchronic	160 mkd (est.)	300	3600 $\mu\text{g}/\text{person-day}$	estimated from USEPA, 1998b
	Chronic	160 mkd (est.)	1000	1200 $\mu\text{g}/\text{person-day}$	estimated from USEPA, 1998b
	Acute	70 mkd (est.)	100	7000 $\mu\text{g}/\text{person-day}$	estimated from USEPA, 1994b
Inhalation	Subchronic	50 mg/m^3	100	1000 $\mu\text{g}/\text{person-day}$	animal study; Larson <i>et al.</i> , 1996
	Chronic	80 mg/m^3 (est.)	1000	600 $\mu\text{g}/\text{person-day}$	estimated from USEPA, 1998b
	Acute	15 mg/m^3	100	3000 $\mu\text{g}/\text{person-day}$	animal study; Mery <i>et al.</i> , 1994
Ingestion	Subchronic	80 mkd	300	1800 $\mu\text{g}/\text{person-day}$	Animal studies (Jorgenson <i>et al.</i> , 1985); supported by USEPA, 1998b
	Chronic	80 mkd	1000	600 $\mu\text{g}/\text{person-day}$	Animal studies (Jorgenson <i>et al.</i> , 1985); USEPA, 1998b
	Acute	35 mkd	100	3500 $\mu\text{g}/\text{person-day}$	Animal studies (Larson <i>et al.</i> , 1993); One- and 10-day health advisories (USEPA, 1994b)

3.1 Chlorine

The toxicity literature for chlorine has been comprehensively summarized elsewhere (ACGIH, 1996a; USEPA, 1994a; WHO, 1996; WHO, 2000). According to the latest review by WHO (2000) related to drinking water, “evidence from these [discussed elsewhere in its report] animal and human studies suggest that chlorine, hypochlorite solutions, chloramine, and chlorine dioxide themselves probably do not contribute to the development of cancer or any toxic effects.” The following briefly describes the chlorine data determined to be particularly germane to the estimation of the health safety and risk from exposures to chlorine for swimmers in residential and commercial pools.

3.1.1 Skin Contact

No human or laboratory animal studies were located regarding dermal exposures to chlorine. However, limited human experiences suggest that chlorine may be capable of causing “itching” at concentrations much greater than 10 ppm (a case report of a skin patch test of an individual who was exposed to 400 to 600 ppm of sodium hypochlorite and who was shown to have skin irritation as a result; a report of an occupational exposure to sodium hypochlorite (concentration unspecified) causing dermatitis; and a secondary reference stating that 5%-10% available chlorine is allegedly classified in Europe as an “irritant” whereas <5% does not require classification as to irritancy). This information has not been confirmed. Because some microbes present in pool water are known to be stimuli for itching, it is unclear what, if any, role chlorine might have in this phenomenon.

An indirect measure of a human NOAEL for chlorine in contact with skin is the ACGIH 8-hour TLV of 0.5 ppm of air (2433 $\mu\text{g}/\text{m}^3$) for inhalation. This TLV is meant to guard against irritation of respiratory mucous membranes that are far more (perhaps greater than a factor of two) sensitive to the irritant effects of chlorine than is skin. The skin is perhaps two-100 times less responsive to the irritant effects of chlorine gas than are mucous membranes. Thus, 10 ppm of chlorine in water represents a plausible and conservative lower bound on a human NOAEL, even for annoying and non-health-threatening responses.

3.1.2 Inhalation

3.1.2.1 Subchronic: No human or laboratory animal studies were located regarding subchronic inhalation exposures to chlorine. However, repeated exposures to high (undefined) concentrations in various occupational settings have been noted to result in itching or burning of the eyes, nose, and throat, cough, runny nose, nausea, headache, reduced pulmonary function, and general discomfort at undefined, and presumably high, concentrations over varying exposure durations (ACGIH, 1996a).

The human study of Anglen (1981) provides some limited insight on safe levels of exposure to chlorine in air. Anglen (1981) exposed males and females, smokers and nonsmokers, to 0, 0.5, 1, or 2 ppm (0, 2433, 4866, and 9734 $\mu\text{g}/\text{m}^3$) of chlorine for four- and eight-hour intervals. The 29 subjects were interviewed at 15, 30, 60, 90, 120, 180, and 240-minute intervals. The test subjects reported a wide range of sensations including smell, taste, itching and/or burning of the eyes, production of tears, urge to cough, runny nose, nausea, headache, dizziness, shortness of breath, and drowsiness. In addition, pulmonary tests including vital capacity and forced expiratory volume were conducted prior to exposure, two hours into the four-hour exposure period, and at the end of the exposure. Study results indicated that the male subjects experienced greater discomfort to the chlorine exposure than did females. The highest doses (1 and 2 ppm for eight hours) elicited mild (subjective irritation) and serious (decrements in pulmonary function) responses, respectively, in the test subjects, while the lowest concentration produced no response. At four hours of exposure, only mild subjective responses were noted.

The observed NOAEL for subchronic exposure was found to be 2433 $\mu\text{g}/\text{m}^3$. Because the information was obtained in humans exposed repeatedly in a workplace setting and because this level has proven to be safe for individuals exposed for long periods of time, the actual uncertainty factor is likely to be larger than one. Thus, the adjusted human NOAEL was also selected as 2433 $\mu\text{g}/\text{m}^3$.

3.1.2.2 Chronic: Data addressing chronic inhalation exposure to chlorine are limited. An analysis by Henderson and Haggard (1943) suggested 0.35 ppm (1703.5 $\mu\text{g}/\text{m}^3$) be a maximal concentration for prolonged inhalation exposures. The authors reached this conclusion after reviewing older literature that suggested that concentrations at or above 5 ppm resulted in respiratory complaints, dental decay, and the inflammation of mucous membranes of the nose. This conclusion has not been confirmed.

A two-year inhalation study of exposure of female and male B6C3F1 mice and F344 rats to chlorine gas has been reported (Wolf, *et al.*, 1995). Female and male B6C3F1 mice and F344 rats were exposed to chlorine gas for up to two years to determine chronic toxicity and carcinogenicity. Groups of approximately 70 each of female and male mice and rats were exposed to 0, 0.4, 1.0, or 2.5 ppm chlorine gas for six hr/day, five days/week (mice and male rats), or three alternate days/week (female rats) for two years, with an interim necropsy of rats at 12 months (10 rats/sex/concentration group). Histological examination was performed on all organs from high-concentration and control animals and selected target organs from mid- and low-concentration groups. Exposure-dependent lesions were confined to the nasal passages in all sex and species groups. Chlorine-induced lesions, which were most severe in the anterior nasal cavity, included respiratory and olfactory epithelial degeneration, septal fenestration, mucosal inflammation, respiratory epithelial hyperplasia, squamous metaplasia

and goblet cell hypertrophy and hyperplasia, and secretory metaplasia of the transitional epithelium of the lateral meatus. Intracellular accumulation of eosinophilic proteinaceous material was also a prominent response involving the respiratory, transitional, and olfactory epithelia, and in some cases the squamous epithelium of the nasal vestibule. Many of these nasal lesions exhibited an increase in incidence and/or severity that was related to chlorine exposure concentration and were statistically significantly increased at all chlorine concentrations studied. Male mice and female rats appeared more sensitive to chlorine than female mice and male rats, respectively. The incidence of neoplasia was not increased by exposure, indicating that inhaled chlorine in rats and mice is an upper respiratory tract toxicant but not a carcinogen.

The data of Anglen (1981) provide the basis for a comparable NOAEL of 2433 $\mu\text{g}/\text{m}^3$, which is also ACGIH's 8-hour TLV. This value was selected as the adjusted human NOAEL.

3.1.2.3 Acute: Data regarding acute inhalation exposures to chlorine consist primarily of observations from workplace exposures and studies conducted on human volunteers. Thirteen human inhalation studies were located which investigated acute inhalation exposure to chlorine. The study of Anglen (1981) provides support for a safe level of acute exposure at 4866 $\mu\text{g}/\text{m}^3$, which is equivalent to ACGIH's 15-minute STEL. This value was selected as the adjusted NOAEL for humans.

3.1.3 Ingestion

3.1.3.1 Subchronic: No human studies have been reported. Data regarding subchronic oral exposures to chlorine consist primarily of 90-day drinking water studies in laboratory animals. Nine studies were identified which investigated subchronic oral exposure to chlorine. The one reliable study that reported the lowest NOAEL value is summarized below.

In a study by Daniel *et al.* (1991), chlorine was added to drinking water that was then administered to adult male and female B6C3F1 mice. Ten animals/sex/dose received doses of 0, 12.5, 25, 50, 100, or 200 mg/l (corresponding to 0, 2.7, 5.1, 10.3, 19.8, or 34.4 mg/kg-day for males and 0, 2.8, 5.8, 11.7, 21.2, or 39.2 mg/kg-day for females) for 90 days. Mortality, food and water consumption, hematology, clinical chemistry, body weight, histopathology, and organ weights were monitored. The authors reported concentration-dependant decreases water consumption in males and females, with a statistically significant decrease in females at the two highest dose levels. In addition, a concurrent decrease in body weight gain was also observed. The authors concluded that these effects, along with inconsistent reductions in organ weights and serum enzymes, were consistent with decreased water consumption rather than chlorine-related toxicity, particularly in light of the fact that no

histopathological changes or clinical toxicity were noted. Therefore, a dose of 50 mg/L (ppm; 10.3 mg/kg-day) was identified as the NOAEL.

The basis for judging safe levels of subchronic exposure to ingested chlorine is the lifetime exposure study of NTP (1992), because of its greater robustness and longer duration showing a slightly higher NOAEL (14 mkd). Therefore, this value was selected for our evaluation as the basis of the risk assessment. To this NOAEL, an uncertainty factor of 100 is applied to obtain a NOAEL adjusted for humans of 140 µg/kg-day.

3.1.3.2 Chronic: No human studies have been reported. Data regarding chronic oral exposures to chlorine consist primarily of two-year drinking water studies. Six studies were located which investigated chronic oral exposure to chlorine. The most reliable study that reported the lowest NOAEL value is summarized below.

NTP (1992) conducted a two-year bioassay on F344 rats and B6C3F1 mice to evaluate the chronic toxicity and carcinogenicity of chlorinated drinking water. The results of only the mouse¹⁴ portion of this study are reported herein. In this study, 70/sex/dose mice were administered chlorinated drinking water at 0, 70, 140, or 275 ppm (0, 7.4, 14.0, or 24 mg/kg-day for males and 0, 7.6, 14.2, or 24.2 mg/kg-day for females) available chlorine, as sodium hypochlorite, for 104 weeks. Interim sacrifices were performed at 14 and 66 weeks on 10 animals/sex/dose. Body weights, organs weights, histopathology and hematology and food and water consumption were evaluated throughout the study.

Results indicated that body weights were 5-8% lower in high-dose males and 5-7% lower in females. No treatment-related significant differences were noted in organ weights or organ-to-body weight ratios. No alterations were reported in hematologic or gross or microscopic histopathology. Although a high rate of mortality had occurred, the survival rate was similar for all groups of controls and treated animals. Significant decreases in the weight of whole body, salivary gland, and kidneys were observed in females. In the males, a significant decrease in brain and heart weights was observed. No significant increase was found in the incidence of gross or histopathologic nonneoplastic lesions of any type in treated animals. No changes in serum chemistry, food consumption, or water consumption were observed. The biological significance of the reduced organ and body weights remains unclear although the authors concluded that these effects may be suggestive of chronic toxicity. Based on these results, a NOAEL of 275 ppm (24 mg/kg-day) was concluded.

Supporting evidence for the safe ingestion of chlorine in water comes from (1) the study of Druckrey (1968) who exposed seven generations of rats to 100 mg/L (10 mg/kg-day) of

¹⁴The part of the study performed with rats found no NOAEL.

chlorine in their water with no signs of systemic or reproductive toxicity and (2) from the study of Hasegawa *et al.* (1986) who demonstrated that at a concentration of chlorine as high 0.2% in drinking water consumed for 102 weeks produced no compound-related tumors in F344 rats.

The adjusted human NOAEL is based on the NOAEL (14 mg/kg-day) identified in the NTP (1992) study to which an uncertainty factor of 100 was applied to account for inter-species and inter-human variability is susceptibility. The resulting adjusted human NOAEL is 140 µg/kg-day (9,800 µg/person-day for a 70-kg individual).

3.1.3.3 Acute: No human studies have been reported. Data regarding acute oral exposures to chlorine are limited primarily to determinations of lethality (*i.e.*, LD₅₀) in laboratory animals. However, six studies were found that investigated toxicologic endpoints other than lethality. Of these six studies, only one involved a mode of administration considered appropriate for this assessment; that study is summarized below.

Cunningham (1980) conducted a study on weanling Wistar rats and albino guinea pigs using cow's milk treated with sodium hypochlorite. In this study, cow's milk was mixed on a daily basis with nominal concentrations of 0, 40, 200, or 1000 mg/l (0, 36.1, 180.5, or 902.4 mg/kg-day). Five male rats per treatment group were given the milk *ad libitum* for nine days. Body weights were recorded for five of the nine test days. Rats also received a commercial rat diet, but no water during the exposure period. Weight gain was found to increase at the two lowest dose levels, although not at statistically significant levels. Organ weights (liver, kidneys, heart, and brain) as a percentage of body weight were not affected. Based on these results, a NOAEL of 1000 mg/L was identified.

The adjusted human NOAEL is based on the NOAEL (900 mg/kg-day) identified in the Cunningham (1980) study to which an uncertainty factor of 100 was applied to account for inter-species and intra-species variability is susceptibility. The resulting adjusted human NOAEL is 9000 µg/kg-day (630,000 µg/person-day for a 70-kg individual).

3.2 Chloroform

The toxicity literature for chloroform has been comprehensively summarized elsewhere (ACGIH, 1996b; USEPA, 1994b, 1998b; WHO, 1994, 1996, 2000). Because of the robust database available regarding chloroform, the following briefly describes the data determined to be most germane to the estimation of the health risks and safety from exposures to chloroform for swimmers in residential pools. The cornerstone for estimating safe levels of exposure to chloroform in drinking water has been amply described by USEPA (1998b), and

the following evaluation relies on the same data for chronic and subchronic exposures from all routes to chloroform when swimming.

3.2.1 Skin Contact

No human or laboratory animal studies were found regarding subchronic or chronic dermal exposures to chloroform. Since some studies have reported that chloroform can be absorbed through the skin to a limited degree, the ingestion data (summarized below and including an understanding of the mode of toxic action) were used to estimate NOAELs and adjusted NOAELs for humans. Based on the weight-of-evidence, the target organs by skin contact should be the same as for ingestion, namely the liver and kidneys. However, the potency would be tempered substantially by the rate and degree of absorption.

3.2.1.1 Subchronic The NOAEL for the subchronic duration of exposure by skin contact was estimated from the chronic ingestion study of Jorgenson *et al.* (1985). The original NOAEL was multiplied by a factor of two to account conservatively for the degree and rate of skin penetration of chloroform. An uncertainty factor of 300 includes consideration of inter-species (rat to human) and inter-human differences in susceptibility as well as differences in duration of exposure in the study (lifetime) and in among swimmers (less than lifetime). The result is a human adjusted NOAEL of 3600 µg/person-day.

3.2.1.2 Chronic The NOAEL for the chronic duration of exposure by skin contact was estimated from the chronic ingestion study of Jorgenson *et al.* (1985). An uncertainty factor of 1000 includes consideration of inter-species (rat to human) and intra-species (among humans) differences in susceptibility as well as differences in duration of exposure in the study (lifetime) and in among swimmers (less than lifetime). The result is a human adjusted NOAEL of 1200 µg/person-day.

3.2.1.3 Acute One acute dermal study has been reported. In that study, no deaths resulted from dermal exposure of rabbits exposed to doses of up to 3980 mg/kg chloroform for 24 hours (Torkelson *et al.*, 1976).

The NOAEL for the acute duration of exposure by skin contact was estimated from the acute ingestion study of Larson *et al.* (1993), one that USEPA relied upon for the one- and ten-day health advisories for chloroform in drinking water. The original NOAEL was multiplied by a factor of two to account conservatively for the degree and rate of skin penetration of chloroform. An uncertainty factor of 100 includes consideration of inter-species (rat to human) and inter-human differences in susceptibility. The result is a human adjusted NOAEL of 7000 µg/person-day.

3.2.2 Inhalation

Most of the data describing inhalation exposure to chloroform, and any resultant toxicity, in humans were obtained from clinical studies pertaining to chloroform-related health effects in patients under anesthesia or occupational exposures. In most instances, these reports noted concurrent exposures to medications, in the case of anesthesia, or other airborne chemicals, in occupational settings, which confound study results. Most of these reports failed to report exposure concentrations or durations. However, chloroform-related effects in humans are supported by laboratory animal data such that a qualitative expression of health hazard can be ascribed to chloroform.

3.2.2.1 Subchronic: Data regarding subchronic inhalation exposures in humans to chloroform involve case studies and clinical reports from occupational and hospital sources, respectively (ACGIH, 1996b; ATSDR, 1998). Several laboratory animal studies describing exposure duration and concentration as well as pathological lesions have been reported. These studies were often performed at concentrations unlikely to be experienced by humans. Some studies conducted at high concentrations found no NOAEL. However, two studies were located that reported a NOAEL. A reliable study that reported the lowest NOAEL value is summarized below.

Larson *et al.* (1996) investigated the ability of intermediate exposure to chloroform vapors to produce toxicity and regenerative cell proliferation in the nasal passage of male and female B6C3F1 mice. Groups of eight animals of each sex were exposed to 0, 0.3, 2, 10, 30, or 90 ppm (0, 1.46, 9.7, 48.7, 146, or 438 $\mu\text{g}/\text{m}^3$) chloroform via inhalation for six hours a day, seven days a week for three, six, or 13 weeks; additional groups of eight animals of each sex were exposed for six hours a day, five days a week for 13 weeks. Alterations in nasal tissues were noted at the 30 and 90 ppm exposure level, and the exposure level of 10 ppm (approximately 50 mg/m^3) was considered a NOAEL.

The NOAEL for the subchronic duration of exposure by inhalation was estimated from the subchronic ingestion study of Larson *et al.* (1996). An uncertainty factor of 100 includes consideration of inter-species (rat to human) and inter human differences in susceptibility. The result is a human adjusted NOAEL of 1000 $\mu\text{g}/\text{person-day}$.

3.2.2.2 Chronic: Evaluation of the chronic toxicity of chloroform includes consideration of systemic toxicity and of tumor formation. Data regarding chronic inhalation exposures in humans to chloroform are limited to case studies and clinical reports from occupational and hospital sources, respectively. (ACGIH, 1996b; ATSDR, 1998). Some data from laboratory animals are available (ATSDR, 1998). One occupational study, that described both exposure duration and concentration as well as a NOAEL, is summarized below.

In an occupational study, gastrointestinal disturbances (nausea, dry mouth, and fullness of the stomach) were reported in female workers exposed to 22-71 ppm chloroform for 10-24 months and 77-237 ppm chloroform for 3-10 years (Challen *et al.*, 1958). The authors derived a NOAEL for chloroform of 71 ppm (345.6 µg/m³).

No data have indicated that inhaled chloroform may produce tumors. Nevertheless, since ingested chloroform has produced tumors systemically in laboratory rats, the possibility that it might do so has been raised. Therefore, the toxicological data on chloroform have been examined to address that possibility, and some specific aspects of its potency as it relates to all routes of administration that yield systemic doses is also addressed.

The following analysis demonstrates that (1) chloroform has been shown to be a carcinogen only in animals and only by ingestion, although limited and unverified information suggests that chloroform inhaled at very high doses elicited kidney tumors in one gender of a uniquely susceptible strain of mice; (2) chloroform elicits tumors in rodents, regardless of route, only at doses that first cause injury to vital tissues, and without damaging genetic material (*viz.*, DNA); correspondingly, at doses below those that cause cellular and tissue injury, chloroform would not elicit tumors; (3) the carcinogenic potency of chloroform by inhalation may be inferred from scientifically defensible studies in which chloroform has been ingested at very high doses; (4) the carcinogenic potency of chloroform by inhalation should be estimated by the use of a non-linear method, such as that which relies on NOAELs and uncertainty factors (UF), and not by use of linear low-dose extrapolation models.

The weight-of-evidence has led to the following conclusions:

1. Chloroform has been shown to be a carcinogen only in animals and only by ingestion, although limited and unverified information suggests that chloroform inhaled at very high concentrations elicited kidney tumors in one gender of a uniquely susceptible strain of mice.

The basis for this conclusion is as follows. High doses of chloroform induced liver tumors in male and female B6C3F1 mice when administered by gavage in a corn oil vehicle (NCI, 1976), kidney tumors in male Osborne-Mendel rats when given by gavage or in drinking water (NCI, 1976; Jorgenson *et al.*, 1985), and kidney tumors in male BDF1 mice, a strain particularly susceptible to the nephrotoxic properties of chloroform administered by inhalation (Yamamoto *et al.*, 1994; Matsushima, 1994). However, the male BDF1 mice had to have exposed concentrations increased slowly over a period of weeks in order to adapt to a concentration high enough to produce cancer. Exposing these animals directly to atmospheres of 30 or 90 ppm resulted in significant deaths from acute kidney failure (Yamamoto *et al.*, 1994; Matsushima, 1994; Larson *et al.*, 1996).

2. Chloroform elicits tumors in rodents, regardless of route, only at doses that first cause injury to vital tissues, and without damaging genetic material (DNA).

This conclusion is based on the following. The current overall weight-of-evidence indicates that chloroform is a rodent carcinogen only at high-dose exposures and that its carcinogenic effects are secondary to cytotoxicity-induced regenerative cell proliferation occurring in response to cell injury, subsequent cell death, and the need for cell replacement. Three lines of evidence are strongly consistent with this mode of action for chloroform in the liver and kidney:

First, genotoxicity studies demonstrate that chloroform is likely to be a carcinogen by acting through a nongenotoxic mode of action (Butterworth and Eldridge, 1992). Bacterial assays for mutation induction are negative (*e.g.*, Kirkland *et al.*, 1981; Uehleke *et al.*, 1977; Van Abbe *et al.*, 1982; Gocke *et al.*, 1981). Chloroform does not generally induce mutations in sister chromatid exchanges in human lymphocytes (Kirkland *et al.*, 1981), and no evidence demonstrated that chloroform is a DNA alkylating agent (Reitz *et al.*, 1980, 1982; Pereira *et al.*, 1982). More recently, tests have been conducted that investigate unscheduled DNA synthesis (UDS)—that is a measure of DNA repair and thus an indirect, more sensitive measure of covalent binding of chemicals or their metabolites to DNA. Earlier studies have shown that chloroform does not induce UDS in the *in vivo* hepatocyte DNA repair assay in male F-344 rats at doses of 40 or 400 mg/kg (Mirsalis *et al.*, 1982), or in human hepatocytes exposed *in vitro* to concentrations of 0.01 to 1.0 Mm chloroform (Butterworth *et al.*, 1989). Current studies by Larson *et al.* (1994a) have shown similar findings for *in vitro* assays of primary hepatocyte cultures from female B6C3F₁ mice incubated with 0.01 to 10 Mm chloroform. Larson *et al.* (1994b) also conducted an *in vivo* DNA repair assay in which animals were treated by gavage with 238 and 477 mg/kg-day chloroform in corn oil (*i.e.*, the doses that induced tumors in NCI's 1976 mouse chronic bioassay). Primary hepatocyte cultures were prepared two and 12 hours later. No DNA repair activity was seen at either dose or at either time point. Since these assays are sensitive to covalent binding with DNA, the negative results strongly support the conclusion that chloroform does not damage genetic material. An expert committee recently conducted a critical review, and concluded that neither chloroform nor its metabolites appear to interact directly with DNA or possess genotoxic activity (WHO, 1994).

Second, toxicokinetic studies have indicated that chloroform metabolism is essential for toxicity, that tissues most actively metabolizing chloroform, (*i.e.*, the liver and kidney) are also the primary target organs, and that species- and gender-related differences in metabolism are associated with similar differences in toxicity (USEPA, 1994b).

Thus, the cytotoxicity of chloroform is influenced by factors that affect the detoxification capacity of the target organ. Since the oxidative metabolism of chloroform is enzyme-mediated and thus saturable, high doses of chloroform result in a high rate of production of toxic metabolites that can overwhelm cellular defense mechanisms, and induce cell injury and cell killing (*e.g.*, Reitz *et al.*, 1980). This toxicokinetic behavior coherently explains the rodent bioassay findings in which daily, single, high-dose bolus administration of chloroform (by gavage) induced liver tumors in mice (NCI, 1976), whereas similar daily doses ingested in small amounts throughout a 24-hour day did not (Jorgenson *et al.*, 1985). Reitz *et al.* (1990) used toxicokinetic data to develop a pharmacokinetic model that was based on macromolecular binding, and predicted the cytotoxic effects of chloroform in long-term rodent bioassays. The model predicted that tumor incidence was correlated with the instantaneous rate of chloroform metabolism and with cytotoxicity, but not with the total amount of chloroform metabolized in the liver. Thus, the predicted peak rate of chloroform metabolism following bolus gavage dosing was much higher than that for continuous exposure in drinking water, and could account for the disparate results seen in cancer bioassays when the mode of compound administration was drinking water versus gavage in a corn oil vehicle (Jorgenson *et al.*, 1985; NCI, 1976). Because inhalation produces a continuous systemic dose as does consumption of drinking water, the effects observed in the ingestion studies should be directly relevant to estimating the toxicokinetic behavior of chloroform by inhalation. However, in less-than-lifetime inhalation studies at acutely toxic doses, the pattern of pathology in the kidneys, which included severe necrosis, cell killing, and regenerative cell proliferation, was more comparable to that in the bolus-dosing (gavage) studies (Larson *et al.*, 1996). These findings provide support for a non-DNA-reactive-cytotoxic mode of action (Templin *et al.*, 1995, 1996).

Third, mode of action studies have examined changes in cell proliferation in rodent liver and kidney as a function of chloroform dose and found a consistent pattern demonstrating that cellular injury and proliferation mediated by high, toxic doses are most likely the obligatory steps to tumorigenesis.

Increased cell proliferation is utilized as an early indicator of cytotoxicity and cytolethality, and is measured using a labeling index (LI; defined as percent nuclei in the S-phase of mitosis as indicated by 5-bromo-2³deoxyuridine immunohistochemical staining) (Larson *et al.*, 1993). The results show that organ- and species-specific patterns of cytotoxicity and resulting cell proliferation parallel those of tumorigenicity at similar doses administered via gavage in corn oil (Larson *et al.*, 1993).

This pattern was observed for the induction of liver tumors. A more detailed dose-response study with female mice, using gavage administered in a corn oil vehicle, also showed dose-dependent changes in cellular proliferation and centrilobular necrosis of the liver. Tumors

were induced at these doses in the NCI (1976) chronic animal bioassay. In contrast, mice administered doses in drinking water similar to those of the Jorgenson *et al.* (1985) chronic bioassay showed no histological changes or cell proliferation in the liver at any dose, even though the cumulative daily amount of chloroform ingested was similar to the amount given in the gavage study. In summary, the results of metabolic, toxicokinetic, and mechanistic investigations are strongly consistent with a cytotoxicity/cytolethality mode of action for the high-dose carcinogenicity of chloroform in the liver.

For the induction of kidney tumors, significantly increased cell proliferation accompanied by proximal tubular necrosis was observed in male F344 rats treated with single high doses of chloroform administered by gavage in corn oil (180 and 477 mg/kg). Lipsky *et al.* (1993) exposed F344 rats for five days per week for four weeks at doses of 90 or 180 mg/kg-day, administered via gavage in corn oil or water. Animals were sacrificed, and the kidneys were examined for histopathologic changes and for increased DNA synthesis. Dose-dependent and segment-specific epithelial cell necrosis was observed in the kidney of rats exposed via corn oil. Necrosis was localized primarily in the second segment of the proximal tubules. DNA synthesis was significantly increased in the corn oil vehicle group. In contrast, rats exposed via water showed neither significant histopathologic changes nor any changes in DNA synthesis—a situation expected to be the same when chloroform is inhaled. These data extend findings from studies on the liver to the kidney, (*i.e.*, that a correlation exists between cell toxicity and increased cell proliferation in the renal proximal tubules, which are the site of tumor induction and that the cytotoxicity of chloroform is potentiated by the corn oil vehicle when administered to rodents via gavage.

Inhalation studies of four days to 13 weeks were conducted with B6C3F1 male and female mice at doses of 0.3, 2, 10, 30 and 90 ppm to develop indices of cytotoxicity, cytolethality, and regenerative cell proliferation in the liver and the kidney (Larson *et al.*, 1996). Animals were exposed to a range of airborne concentrations similar to those employed in the inhalation bioassay (Yamamoto *et al.*, 1994; Matsushima, 1994). Exposure duration was six hours/day for either five or seven days/week. A stop-exposure study, where animals were exposed seven days/week for six weeks, kept for an additional seven weeks, and sacrificed at week 13, was also conducted to assess the reversibility of inhalation effects. In addition to standard pathology and histopathology, increased cell proliferation, as indicated by changes in the labeling index (LI), were also measured in the liver, kidney and nasal passages.

In female mice, liver lesions were observed that were both dose- and duration-dependent, and histopathologically similar to those observed in female mice administered chloroform via gavage (Larson *et al.*, 1996). Increases in hepatic LI occurred as early as four days in the 90 ppm group and persisted for 13 weeks. Increased hepatic LI was observed at 30 ppm during week three and week six, but not at week 13. No increases in cell proliferation were observed

at doses of 10 ppm or lower. Therefore, Larson *et al.* (1996) considered continuous exposure to 10 ppm (air) chloroform to be a NOAEL. This NOAEL is conservative, since sustained cell proliferation was not observed at week 13 in the 30 ppm group. A less conservative LOAEL for hepatic effects would be 90 ppm, with a corresponding NOAEL of 30 ppm. No increases in LI were observed in the female mouse kidney. Transient nasal passage toxicity and associated increased cell proliferation were observed at 10, 30, and 90 ppm after four days of exposure.

In male mice, liver lesions were very similar to those occurring in male and female mice exposed to chloroform by gavage, and were both dose- and time-dependent. Significant increases in hepatic LI occurred only at 90 ppm. Kidney lesions were the same as those described in male mice exposed to chloroform by gavage. The increase in frequency and severity of nephropathy increased with increasing dose and exposure duration. The results of one-week studies (Larson *et al.*, 1994c), in which mice and rats were administered doses that were higher than those in the three- to 13-week investigations (Larson *et al.*, 1996) support these findings.

Increased renal cell proliferation was confined to the proximal convoluted tubules at 30 and 90 ppm, beginning at three weeks and continuing to week 13. Nasal lesions were similar in duration and severity to those observed in female mice.

These studies collectively indicate that mice exposed to 90 ppm chloroform consistently demonstrated increased hepatic LI and histopathological changes that were reversible and represented regenerative growth in response to the continual cell death produced by repeated chloroform exposures. Chloroform also produced histologic changes, necrosis and regenerative cell proliferation in the kidneys of male but not female mice. Thus, the potential exists for chloroform to induce kidney tumors in male mice by a nongenotoxic-cytotoxic mode of action.

From these overall findings, one can infer with a reasonable degree of scientific certainty that the continuous inhalation of high doses of chloroform would produce pathological outcomes similar to those observed as result of high-dose ingestion. For systemic toxicants such as chloroform, *in vivo* genotoxicity is not a route-dependent phenomenon; therefore, the absence of genotoxicity by chloroform via ingestion raises no legitimate concern that it might be genotoxic via inhalation. Therefore, inhaled chloroform would likely function as a non-genotoxic carcinogen with a non-linear dose-response relationship.

Indeed, USEPA (1998a) has acknowledged a newly achieved consensus that chloroform exhibits a non-linear dose-response relationship with regard to chronic toxicity including cancer, that the most appropriate means of extrapolating to low doses is one that relies on the

application of uncertainty factors and not the linear low-dose models, and that chloroform is unlikely to pose a cancer risk at a dose of approximately 90 µg/kg-day (600 µg/person-day).

The inhalation carcinogenic potency of chloroform can be estimated from studies in which chloroform has been inhaled at very high doses. The strength of scientific evidence indicates that chloroform is tumorigenic via a nongenotoxic/cytotoxic mode of action at high doses only and that low-dose effects are nonlinear. Furthermore, the LI is a reliable indicator of cytotoxicity and regenerative cell proliferation. Therefore, an animal NOAEL of 10 ppm (50 mg/m³) in air (Larson *et al.*, 1996) may be used conservatively to estimate a NOAEL for chloroform inhalation exposure.

The carcinogenic potency of chloroform by inhalation can be inferred from scientifically defensible studies in which chloroform has been ingested at very high doses. The threshold for chloroform's cell toxicity that might precipitate a tumorigenic response is expected to be dependent largely on target absorbed dose, particularly target tissue concentration. The absorbed or internal dose can be normalized by expressing the dose in units of "mg of chemical per kg of body weight per day" with appropriate adjustment for degree of absorption via the respective routes. Once normalized, such doses can also be legitimately compared using the metameter¹⁵ of "mg of chemical per person per day" by adjusting for the number of kilograms that make up the mass of one or a group of persons. Therefore, one could select the most relevant ingestion study to estimate a NOAEL by another route such as inhalation.

However, recent studies by Wang *et al.* (1994, 1995), in which dose- and route-dependent changes in the metabolism and toxicity of chloroform were investigated, indicate that greater toxicity, at equivalent doses, is produced following ingestion, as compared with either inhalation or intraperitoneal administration. The authors attribute these findings to first-pass metabolism in the liver, unique to ingestion. Thus, the use of ingestion studies to estimate toxic potency by inhalation will likely overestimate the carcinogenic potency of chloroform by inhalation.

Chloroform elicits tumors only at doses that first cause injury to vital tissues and correspondingly at doses below those that cause cellular and tissue injury, chloroform would not elicit tumors. In the only inhalation bioassay of chloroform (a study performed in Japan and of which only an abstract is available), compound-related tumors were observed only at the highest dose tested (126 mg/kg-day); the highest dose at which no tumors were observed was 42 mg/kg-day (Yamamoto *et al.*, 1994; Matsushima, 1994).

¹⁵"Metameter" is a dose metric or a unit of measure for a dose; for instance, "mg/kg-day" is metameter.

By comparison, chloroform administered in drinking water produced kidney tumors at 160 mg/kg-day in male rats but none at 80 mg/kg-day; in mice no tumors were observed with doses as high as 263 mg/kg-day (Jorgenson *et al.*, 1985). In corresponding studies of chloroform's mode(s) of action, it was demonstrated that the doses causing tumors in the lifetime studies also caused tissue damage in target organs, whereas at doses at which no tumors were observed, no compound-related tissue damage was reported.

As a result, the carcinogenic potency of chloroform by inhalation should be estimated by the use of a non-linear method, such as that which relies on NOAELs and uncertainty factors (UF), and not by use of linear low-dose extrapolation. The toxicokinetic, mechanistic, and genotoxic data are compelling in their support of a threshold mode of action for the carcinogenicity of chloroform. The weight-of-evidence indicates that (1) chloroform induces tumors through a nongenotoxic/cytotoxic mechanism, and (2) tumor formation occurs only at doses that produce cytotoxicity and cell death, and is secondary to cytolethality-induced, regenerative cell proliferation. Thus, no increased risk of tumor formation would be anticipated at doses that do not induce cell death, and the region of a threshold dose may be defined. This approach is scientifically justified with chloroform because it is most likely a nongenotoxic, threshold carcinogen with a mode of action that is reasonably well understood (USEPA, 1998b). As a result, considerable confidence exists that 600 µg/person-day or approximately 90 µg/kg-day would be protective against all forms of chronic toxicity including cancer in humans.

The NOAEL for the chronic duration of exposure by inhalation was estimated from the chronic ingestion study of Jorgenson *et al.* (1985). An uncertainty factor of 1000 includes consideration of inter-species (rat to human) and intra-species (among humans) differences in susceptibility as well as differences in duration of exposure in the study (lifetime) and in among swimmers (less than lifetime). The result is a human adjusted NOAEL of 600 µg/person-day. This value is consistent with USEPA's evaluation of a safe level of exposure to chloroform for a lifetime of ingestion via drinking water.

3.2.2.3 Acute: Data regarding acute inhalation exposures to chloroform are limited primarily to determinations of lethality (*i.e.*, LD₅₀) in laboratory animals; however, twelve studies were located investigating toxicologic endpoints other than lethality. A reliable study that reported the lowest NOAEL value is summarized below.

The study of Mery *et al.* (1994) demonstrated that acute inhalation exposure to chloroform induced site-specific as well as biochemical changes in the nasal region of female B6C3F1 mice and male Fischer 344 rats. Mice were exposed to 1.2, 3, 10, 29.5, 101, and 288 ppm (5.8, 14.6, 48.7, 143.6, 491.6, or 1401.7 µg/m³) chloroform, and rats were exposed to 1.5, 3.1, 10.4, 29.3, 100, and 271 ppm (7.3, 15.1, 50.6, 142.6, 486.7, or 1319 µg/m³) for six hours a day for seven days to determine the nasal cavity site-specific lesions and the occurrence of

cell induction/proliferation associated with these varying concentrations of chloroform. Results in males indicated that bone formation within the nasal region was prominently seen at 10.4 ppm and above, and it followed a concentration-response curve. In addition, the Bowman's glands were markedly reduced in size in a dose-dependent manner. Lastly, exposure to chloroform at 10.4 ppm (50.6 $\mu\text{g}/\text{m}^3$) and higher resulted in a dramatic increase in the number of S-phase nuclei, with the proliferative response confined to activated periosteal cells, including both osteogenic (round) and preosteogenic (spindle) cells. The only detectable treatment-related histologic change observed in female mice was a slight indication of new bone growth in the proximal part of the first endoturbinate in one mouse exposed to 288 ppm chloroform. In addition, the S-phase response was observed at chloroform concentrations of 10.4 ppm and higher. Based on these results, a concentration of 3.1 ppm (15.1 mg/m^3) was identified by the authors as the NOAEL.

The NOAEL for the acute duration of exposure by inhalation was estimated from the acute ingestion study of Mery *et al.* (1994). An uncertainty factor of 100 includes consideration of inter-species (rat to human) and intra-species (among humans) differences in susceptibility. The result is a human adjusted NOAEL of 3000 $\mu\text{g}/\text{person-day}$.

3.2.3 Ingestion

Numerous epidemiology studies have investigated whether chloroform and other chlorination byproducts in drinking water are capable of causing disease, particularly cancer and disruption of the reproductive process including damage to the developing fetus (ATSDR, 1998; USEPA, 1998b). However, these studies often address the full suite of disinfection by-products and are, therefore, of limited utility for this report. In addition, of the epidemiologic studies that isolated chloroform exposure, limitations in study design, failure to report exposure duration or concentration, and/or other biases (*i.e.*, socio-economic factors, smoking) have provided scant evidence of a clear association between chloroform exposure via ingestion and the aforementioned adverse health effects. However, the laboratory animal database for oral exposure to chloroform is quite robust, such that a qualitative expression of health risk can be ascribed to chloroform.

3.2.3.1 Subchronic: Studies of subchronic oral exposure to chloroform by humans consist mainly of epidemiologic studies that examine impacts on reproductive functions (ATSDR, 1998; USEPA, 1998b). Data from laboratory animal studies, in particular 90-day drinking water studies, have been reported, and are useful in assessing chloroform-related toxicity via oral exposure of swimmers. These studies were often performed at concentrations much higher than those that humans encounter while swimming. Seven laboratory animal studies have been reported. The most reliable one that reported the lowest NOAEL value is summarized below.

Chu *et al.* (1982) exposed male weanling Sprague-Dawley rats to chloroform via drinking-water for 28 days at doses of 0, 0.1, 1.3, 1.3 and 11 mg/L (corresponding to 0, 0.7, 7.4 and 63 mg/kg-day, respectively). The only treatment-related effect observed was a decrease in the neutrophils in the 11 mg group. Therefore, a dose of 1.3 mg/L (7.4 mg/kg-day) was considered by the authors to be the NOAEL.

The NOAEL for the subchronic duration of exposure by ingestion was estimated from the chronic ingestion study of Jorgenson *et al.* (1985). An uncertainty factor of 300 includes consideration of inter-species (rat to human) and intra-species (among humans) differences in susceptibility as well as differences in duration of exposure in the study (lifetime) and in among swimmers (less than lifetime). The result is a human adjusted NOAEL of 1800 µg/person-day.

3.2.3.2 Chronic: Chronic oral exposure to chloroform in humans has been investigated epidemiologically, many exploring a causal link with several forms of cancer (ATSDR, 1998; USEPA, 1998b). These studies, as well as the results of laboratory animal studies that consist primarily of two-year drinking water studies, describe the chronic toxicity of chloroform as consisting of both non-cancer pathology and cancer.

Modest associations were noted in some epidemiologic studies between chlorinated water and bladder and colo-rectal cancer (McGeehin, 1993; Vena *et al.*, 1993; King and Marrett, 1996; Doyle, 1997; Freedman, 1997; Cantor, 1998; and Hildeheim, 1998). The association for bladder has been reported by several investigators; however, methodological limitations preclude any conclusions as to causation. In an effort to address this uncertainty, the Agency evaluated several epidemiology studies which examined the association between chlorinated water and cancer.

In an attempt to increase the power of several modest associations, a meta-analysis was conducted (Morris *et al.*, 1992). However, the inherent uncertainties in studies were soon recognized, and the utility of the approach was generally unsuitable to illuminate the question of causation. As a result, when USEPA promulgated the final Stage 1 Disinfection Byproducts Rule, the Agency concluded:

“The cancer epidemiology studies are insufficient (at this time) to establish a causal relationship between exposure to chlorinated drinking water and cancer; and are thus considered limited for use in quantitative risk assessment.”

“This judgement of causality was based on evaluating the existing cancer epidemiological database for the following criteria: strength of association, consistency of the findings, specificity of the association, as well as other

information concerning the temporal sequence and presence of a dose response relationship and biological plausibility. USEPA applied the criteria stated above to assess the possible causality of cancer using the best available cancer epidemiology studies (Cantor *et al.*, 1987; McGeehin, 1993; King and Marrett, 1996; Cantor, 1998; Freedman, 1997; Hildeheim, 1998; Doyle, 1997). These studies found a weak association for bladder cancer, although the findings were not consistent within and among studies. The specificity of association, temporal association, and dose response relationship remain unknown. In addition the biological mode of action has not been determined. Using the criteria for causality, the present epidemiologic data does not support a causal relationship between exposure to chlorinated drinking water and the development of cancer at this time.” (USEPA, 1998b).

With regard to reproductive and developmental toxicity, epidemiologic and laboratory animal studies have reported some associations. To date, the findings are considered inconclusive, and the subject is remanded to further investigation.

Three particular reproductive and developmental epidemiologic studies (Klotz and Pynch, 1998; Swan *et al.*, 1998; and Waller *et al.*, 1998) relate to chloroform (USEPA, 1998b). Of these, only Waller *et al.* was considered appropriate due to a lack of quantifiable DBP data in the other two studies. This study, which reviewed trihalomethane exposures in drinking water and the incidence of spontaneous abortions, indicated that an association could exist between high trihalomethane exposures and early term miscarriages. USEPA’s recent conclusion is particularly revealing in this respect:

“The reproductive developmental epidemiology data provide important information that contributes to the weight-of-evidence evaluation on the potential risks from exposures to chlorinated drinking water. However, the reproductive epidemiology studies are insufficient to establish a causal relationship between exposure to chlorinated drinking water and reproductive and developmental effects” (USEPA, 1998b).

As an independent effort, the International Life Sciences Institute (ILSI, 1997) also reviewed these data to ascertain the strength of the association between chloroform exposure and cancer. ILSI concluded that there was sufficient evidence that chloroform is carcinogenic in mice and rats. However, evidence for carcinogenicity in humans is inadequate stating:

“...the epidemiological studies of bladder and colo-rectal cancer have generally shown an increased risk associated with the consumption of chlorinated surface water, although a causal association has not been conclusively established.”

Based on the most recent draft of the Agency's Cancer Assessment Guidelines, the weight-of-evidence for the carcinogenesis and non-cancer toxicity of chloroform provided the Agency with the scientific justification to recommend an MCLG of 300 ppb for chloroform in drinking water. While chloroform is regarded qualitatively as a carcinogen, sufficient evidence of its mode of action exists, in the judgement of an assortment of scientific authorities, that "a reasonable scientific basis for support" exists to rely on a nonlinear dose-response model as most appropriate for determining the risk of chloroform at low doses. A NOAEL of 80 mg/kg-day was derived based on the tumor kidney response data in Osborne-Mendel rats from Jorgenson *et al.* (1985).

The NOAEL for the chronic duration of exposure by ingestion was derived from the chronic ingestion study of Jorgenson *et al.* (1985). An uncertainty factor of 1000 includes consideration of inter-species (rat to human) and inter-human differences in susceptibility. The result is a human adjusted NOAEL of 600 µg/person-day. This value is identical to that estimated by USEPA for lifetime exposure to chloroform via drinking water (USEPA, 1998b).

3.3.3.3 Acute: The data regarding acute oral exposures to chloroform are limited primarily to determinations of lethality (*i.e.*, LD₅₀ in laboratory animals). Three studies were located which investigated toxicologic endpoints other than lethality. Of these three, only one reliable study involved a mode of administration which would be appropriate for this assessment. That study is summarized below.

Differences in chloroform toxicity have been noted in female mice when chloroform was administered in different vehicles and by different dosing regimes (Larson *et al.*, 1994b). Mice were treated orally with 3, 10, 34, 90, 238, or 477 mg/kg-day of chloroform in corn oil, or with 16, 26, 53, 81, or 105 mg/kg-day in the drinking water, for four days. The results of the drinking water portion of this study are presented herein. Results indicated that serum ALT or SDH were not different from controls at any dose. At the highest test doses *i.e.*, 53, 81, and 105 mg/kg-day), two showed significant histologic changes in the liver when compared to controls. Therefore a dose of 26 mg/kg-day was considered to be a NOAEL by the authors.

The NOAEL for the acute duration of exposure by ingestion was estimated from the acute ingestion study of Larson *et al.* (1993). An uncertainty factor of 100 includes consideration of inter-species (rat to human) and inter-human differences in susceptibility. The result is a human adjusted NOAEL of 3500 µg/person-day.

4 Risk Characterization: Safe Re-Entry Levels of Chlorine for Swimmers

This evaluation was undertaken to determine whether any undue risk to health may be present when individuals are exposed to chlorine and chloroform when they re-enter swimming pools that are periodically superchlorinated up to 10 ppm of chlorine to control for human pathogens. This information is to guide in establishing a re-entry level of chlorine that balances the public health values of disinfection with safety of the disinfectant and one of its byproducts (chloroform).

Determining whether swimming in water that periodically contains chlorine at 10 ppm requires thoughtfully combining information about the toxicity of chlorine and chloroform (a byproduct of chlorine interacting with organic matter) and the degree of human exposure by all relevant routes of exposure.

The following are the results of combining the exposure information (Chapter 2) with the hazard information (Chapter 3). The results are presented as a matrix for each compound that takes into account routes and durations of exposure—factors that are related directly to the estimations of safety and risk for humans. The measure used to define safety is the margin of exposure or MoE between actual exposure under actual conditions of disinfection practice in the U.S. and the relevant adjusted human NOAELs¹⁶.

4.1 Chlorine

The safety and risk to those who swim in pools superchlorinated periodically with gaseous and other forms of chlorine is characterized by comparing the anticipated levels of exposure to aqueous chlorine considered unlikely to pose a danger to health. The results are presented in **Table 9**.

The results indicate that, for skin contact, exposure occurs up to 10 ppm from time to time; however, the true NOAEL is somewhat greater than that concentration. However, it is not possible to determine the MoE in this case, since the toxicological findings are so limited. Nevertheless, over the course of its use, few, even anecdotal, reports of adverse effects on the skin have been documented, suggesting that chlorine at relatively high concentrations does not cause skin injury. When establishing a 4-ppm MRDLG for chlorine, USEPA has determined that such a concentration is reasonably unlikely to cause harm, with an adequate margin of safety to the skin of those who bathe and/or shower with such treated water.

¹⁶In contrast to the approach used herein to estimate the MoE, USEPA divides the experimentally derived NOAEL by the dose.

As noted previously, chlorine gas, once in water at the concentrations used in pools, is not apt to volatilize; therefore, inhalation exposure is considered to be essentially absent.

Some water containing chlorine is ingested while swimming. However, ample margins of exposure and safety exist to assure that this route of exposure poses no risk to human health. The volume of water ingested is considerably smaller than that on which USEPA's 4 ppm MRDLG is based that the exposure is unlikely to pose any health risk.

Consequently, a re-entry of 10 ppm chlorine is not only safe for swimmers of all ages but also a major safeguard against exposures to harmful pathogens.

4.2 Chloroform

The decision on a re-entry guideline for chlorine in periodically treated pools is likely to consider the possible health risks, if any, that chloroform, a byproduct of chlorination, may pose. The safety and risk to those who swim in pools treated periodically with gaseous or other forms of chlorine up to 10 ppm is characterized by comparing the anticipated doses to chloroform considered unlikely to pose a danger to health. The results are presented in **Tables 10-12**.

The results for chloroform are presented separately for users of outdoor and indoor pools, because the majority of pools that are chlorinated with the gaseous form are outdoors and because the statistical distributions of the data for each group appear to be different. The maximum dose obtained by swimmers at outdoor pools (which comprise the majority of residential pools that employ gaseous chlorine as the mode of application) produced a margin of exposure from 5200 to 30400 for skin contact; 100 for inhalation; and 600 to 3500 for ingestion (**Table 10**). The margin of exposure (MoE) is based on adjusted human NOAELs that incorporate uncertainty factors that address differences in susceptibility between species and among humans. If the mid-point of the concentration ranges had been used to estimate MoE, the MoE would have been even greater, suggesting that the majority of the population that is exposed below the maximum measured would obtain an additional measure of safety at 10 ppm chlorine, while being protected against pathogens through the use of regular superchlorination. Had the USEPA approach for estimating the RfD and RfC been applied, the MoE for each value would be approximately 100 times greater than our values.

Similar comparisons were made for the chloroform data from indoor pools, which comprise mostly commercial pools. The results were comparable for skin contact (8000-46600) and ingestion (400-2430); however, the MoE for inhalation exposures were somewhat lower (15-75) than for outdoor pools. This result may well be related to minimal air flow indoors to dissipate the air above the pool water, thereby increasing the concentration of chloroform

above the water and in the breathing zone of swimmers. Had the USEPA approach for estimating the RfD and RfC been applied, the MoE for each value would be approximately 100 times greater than our values.

The evaluation of risks and safety of chloroform is somewhat more complex than that for chlorine for several reasons. Unlike chlorine, chloroform is a systemic toxicant; therefore, the doses by various routes of exposure are summed to reflect full exposure. The systemic doses of chloroform were estimated by individual routes of exposure and then summed to produce cumulative systemic doses, the highest doses were compared to the most conservative NOAEL for chloroform (chronic ingestion). The resulting MoE was 14, which provides an additional margin of safety to that incorporated in the adjusted human NOAEL, a value that USEPA had recently recommended as the basis for an MCLG of 300 ppb for chloroform in drinking water. Had the USEPA approach for estimating the RfD and RfC been applied, the MoE for each value would be approximately 100 times greater than our value (*i.e.*, 1400).

It is noteworthy that the concentrations of chloroform measured in and around swimming pools were obtained at facilities that are believed to have followed the traditional chlorination procedures including regular superchlorination. The amount of chloroform produced during the superchlorination events with gaseous chlorine should be no greater than those reported, because at that level of chlorination, the oxidation of byproducts (of which chloroform is one) would be greater than at 2 to 5 ppm FAC found in pools much of the time. This conclusion is substantiated by the work of Clark and Sivaganesan (1998) whose studies showed the amount of total trihalomethane (of which chloroform is one) formed at constant total organic carbon (TOC), pH, and temperature increased relatively little; changes in temperature had a much greater impact on formation of TTHM than did the amount of chlorine. Thus, these results should apply correspondingly to pools in which periodic gas chlorination is the means of proper disinfection and health protection.

Consequently, the levels of chloroform to which swimmers are exposed in a chlorinated pool, regardless of the detailed means of chlorination, pose no health risk to the swimmers. These findings strongly support the reliance on 10 ppm chlorine as guidance for re-entry of swimmers into chlorine-treated pools. Adopting 10 ppm chlorine as the re-entry value will ensure continued public health protection without contributing an undue health risk to swimmers.

Table 9. Comparison of upper-bound exposure to chlorine by pool users with adjusted human NOAELs

Scenario	Dermal			Inhalation			Ingestion		
	Exposure (mg/L H ₂ O)	Adjusted Human NOAEL ¹ (mg/L H ₂ O)	MoE ²	Exposure (µg/m ³)	Adjusted Human NOAEL ¹ (µg/m ³)	MoE ²	Exposure (µg/k-d)	Adjusted Human NOAEL ¹ (µg/kg-d)	MoE ²
Sub-Chronic	1-10	>10	?	0 (est.)	2433	?	0.02	140	7000
Chronic	1-10	>10	?	0 (est.)	2433	?	0.02	140	7000
Acute	1-10	>10	?	0 (est.)	4866	?	0.02	9000	450,000

¹Uncertainty factors are incorporated in these values.

² MoE = margin of exposure which is obtained by dividing the adjusted human NOAEL by the corresponding exposure estimate.

Table 10. Comparison of upper-bound exposure to chloroform by outdoor pool users with adjusted human NOAELs

Scenario	Dermal			Inhalation			Ingestion		
	Exposure (µg/p-d)	Adjusted Human NOAEL ¹ (µg/p-d)	MoE ²	Exposure (µg/p-d)	Adjusted Human NOAEL ¹ (µg/p-d)	MoE ²	Exposure (µg/p-d)	Adjusted Human NOAEL ¹ (µg/p-d)	MoE ²
Sub-Chronic	0.23	3600	15650	10	1000	100	1.03	1800	1800
Chronic	0.23	1200	5200	10	600	100	1.03	600	600
Acute	0.23	7000	30400	10	3000	100	1.03	3500	3500

¹Uncertainty factors are incorporated in these values.

² MoE = margin of exposure which is obtained by dividing the adjusted human NOAEL by the corresponding exposure estimate.

µg/p-d = micrograms of chloroform per person per day.

Table 11. Comparison of upper-bound exposure to chloroform by indoor pool users with adjusted human NOAELs

Scenario	Dermal			Inhalation			Ingestion		
	Exposure (µg/p-d)	Adjusted Human NOAEL ¹ (µg/p-d)	MoE ²	Exposure (µg/p-d)	Adjusted Human NOAEL ¹ (µg/p-d)	MoE ²	Exposure (µg/p-d)	Adjusted Human NOAEL ¹ (µg/p-d)	MoE ²
Sub-Chronic	0.15	3600	24000	40.8	1000	24	1.44	1800	1250
Chronic	0.15	1200	8000	40.8	600	15	1.44	600	400
Acute	0.15	7000	46600	40.8	3000	75	1.44	3500	2430

¹Uncertainty factors are incorporated in these values.

² MoE = margin of exposure which is obtained by dividing the adjusted human NOAEL by the corresponding exposure estimate.

µg/p-d = micrograms of chloroform per person per day.

Table 12. Comparison of upper-bound cumulative exposure to chloroform by pool users with the most conservative adjusted human NOAEL

Exposure (µg/p-d)				Adjusted Human NOAEL ³ (µg/p-d)	MoE ⁴
Dermal ¹	Inhalation ²	Ingestion ²	TOTAL		
0.23	40.8	1.44	42.5	600	14

¹Based on maximum concentration reported at outdoor pools.

²Based on maximum concentration reported at indoor pools.

³Adjusted human NOAEL for chronic ingestion of chloroform; includes uncertainty factor of 100.

⁴ MoE = margin of exposure which is obtained by dividing the adjusted human NOAEL by the corresponding exposure estimate. Using USEPA's RfD and RfC method, the MoE would be 1400.

µg/p-d = micrograms of chloroform per person per day.

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