United States
Environmental Protection
Agency

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HEALTH RISKS TO FETUSES, INFANTS, AND CHILDREN (FINAL STAGE 1 D/DBP RULE)

October 29, 1998

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ABBREVIATIONS

BDCM	bromodichloromethane
BMD	benchmark dose
DBCM	dibromochloromethane
DBP	disinfectant byproduct
DCA	dichloroacetic acid
D/DBP	disinfectant and disinfectant byproduct
DWEL	drinking water equivalent level
LOAEL	lowest-observed-adverse-effect level
MCLG	maximum contaminant level goal
MCL	maximum contaminant level
MF	modifying factor
MRDLG	maximum residual disinfectant level goal
NOAEL	no-observed-adverse-effect level
OR	odds ratio
RfD	reference dose
TCA	trichloroacetic acid
THM	trihalomethane
TTHM	total THM
UF	uncertainty factor

PLAIN LANGUAGE SUMMARY OF EVALUATION OF CHILDREN'S RISK

Executive Order 13045: Protection of Children from Environmental Health Risks and Safety Risks

1. Does E.O. 13045 apply to this rule?

This final rule is not subject to E.O. 13045, entitled "Protection of Children from Environmental Health Risks and Safety Risks" (62 FR 19885, April 23, 1997), because the environmental health or safety risks addressed by this action do not have a disproportionate effect on children.

2. Children's Health Protection

In accordance with section 5(501), the Agency has evaluated the environmental health or safety effects of disinfectants and disinfectant byproducts (DBPs) on children. We conclude that the Maximum Contaminant Level Goals (MCLGs) are protective of children. For some chemicals this is because the MCLG was based on a study of reproductive effects or effects on developing organisms. For the other chemicals it was determined that the toxic effects were not more likely to occur in children or to affect children at lower doses than would affect adults. In making these determinations we reviewed all available data and asked the following questions for each disinfectant or DBP:

1. Is there information which shows that the disinfectant or DBP causes effects in the developing fetus or impairs ability to conceive and bear children? If it causes this type of problem will it occur at a lower dose than that which will cause other types of effects?

2. If the disinfectant or DBP causes cancer, are children more likely to be affected by it than are adults?

3. If the disinfectant or DBP causes some noncancer toxic effect, are children more likely to be affected by it than are adults?

The answers to these questions can be found in this document.

The disinfection of public drinking water supplies to prevent waterborne disease is the most successful public health program in U.S. history. However, numerous chemical byproducts (DBPs) result from the reaction of chlorine and other disinfectants with naturally occurring organic and inorganic material in source water, and these may have potential health risks. Thus, maximizing health protection for sensitive subpopulations requires balancing risks to achieve the recognized benefits of controlling for waterborne pathogens while minimizing risk of potential DBP toxicity. Human experience shows that waterborne disease from pathogens in drinking water is a major concern for children and other subgroups (elderly, immune compromised, pregnant women) because of their greater vulnerabilities (Gerba et al., 1996). Based on animal studies, there is also a concern for potential risks posed by DBPs to children and pregnant women (EPA, 1994a, 1998a).

1. INTRODUCTION

1.1. RISK TO CHILDREN

In 1995, EPA's Administrator established an Agencywide policy that calls for consistent and explicit consideration of the risk to infants and children in all risk assessments and characterizations, as well as in environmental and public health standards (Memorandum from the Office of the Administrator, October 20, 1995). The Safe Drinking Water Act amendments of 1996 also stipulated that in establishing maximum contaminant levels (MCLs) the Agency should consider "the effects of the contaminant on the general population and on groups within the general population such as infants, children, pregnant women, the elderly, individuals with a history of serious illness or other subpopulations that are identified as likely to be at greater risk of adverse health effects due to exposure to contaminants in drinking water than the general population." On April 21, 1997, the President signed an Executive Order (13045) that federal health and safety standards must include an evaluation of the potential risks to children in planned regulations. EPA's Office of Water is following the above policy and order and has historically considered risks to sensitive populations (including fetuses, infants, and children) in establishing drinking water assessments, advisories or other guidance, and standards (Ware, 1989; EPA, 1991).

The Office of Water is charged with ensuring that the U.S. population has clean water and safe drinking water. This mandate covers chemical, physical, and biological water pollutants. The disinfection of public drinking water supplies to prevent waterborne disease is the most successful public health program in U.S. history. The diarrheal diseases most commonly associated with waterborne infectious agents have a more severe outcome for children and debilitated adults. Thus, the savings in lives from disinfected water has largely been in those populations. However, chemical disinfectant byproducts (DBPs) result from the reaction of chlorine and other disinfectants with naturally occurring organic materials in source water, and these have potential health risks. Thus, to provide for maximum human health protection it is necessary to balance risks from water pathogens and DBPs.

A growing body of scientific knowledge demonstrates that children may suffer disproportionately from some environmental health risks. These risks may arise because children's neurological, immunological, and digestive systems are still developing. In addition children may incur greater exposure, because they eat more food, drink more fluids, and breathe more air in proportion to their body weights than do adults. Waterborne disease is a major concern for infants, children, and other sensitive subgroups (elderly, immune-compromised, pregnant women) because of their greater vulnerabilities (Gerba et al., 1996). Based on animal

studies, there is a concern for risks posed by DBPs to children and the developing fetus (EPA, 1994a, 1998a).

1.2. RISK ASSESSMENT METHODS

Risk assessment is a process by which judgments are made about an agent's potential to cause harm to humans. Risk assessment of chemicals follows the process developed by the National Academy of Sciences/National Research Council (NAS/NRC). The process is based on analysis of scientific data to determine the likelihood, nature, and magnitude of harm to public health associated with exposure to environmental agents (NRC, 1983, 1994). The NAS paradigm defines four steps:

- Hazard Assessment—Does the chemical produce adverse health effects?
- Dose-Response Assessment—How does the frequency of adverse effects change with dose?
- Exposure Assessment—How much chemical are humans exposed to in various environments?
- Risk Characterization—Summarizes and integrates the scientific findings of the hazard, dose-response, and exposure assessments to determine the potential human risk.

To evaluate the toxicity of disinfectants and disinfectant byproducts (D/DBPs) for fetuses, infants, and children, the following three types of toxicity studies were evaluated:

1. Developmental and reproductive toxicity including both prenatal and postnatal exposures and effects
2. Systemic toxicity
3. Carcinogenicity

Developmental toxicity is defined as the occurrence of adverse effects in the developing organism that may result from exposure before conception, during prenatal development, or postnatally to the time of sexual maturation. Adverse effects may be detected at any point in the life span of the organism (i.e., in the developing organism, neonate, adolescent, or even in the elderly as a late-age-onset disorder). There are a number of developmental abnormalities of concern, including these: spontaneous abortions, stillbirths, malformations, premature mortality, reduced birth weight, mental retardation, and sensory loss, as well as other adverse functional

and physical effects. Developmental abnormalities are extremely common and present an enormous burden for society.

Risk assessment methodologies have been developed to estimate the magnitude of potential harm to humans from carcinogenic and noncarcinogenic effects. Most chemicals that do not produce carcinogenic effects are believed to have a low dose below which no adverse, noncarcinogenic effects occur with incidence above background. On the other hand, carcinogenic chemicals may cause some effect at low doses or not, depending on the way in which they affect the carcinogenic process.

For contaminants with carcinogenic potential, chemical levels are measured against a cancer potency slope factor or a unit risk value together with an assumption of lifetime exposure from ingestion of water. The cancer unit risk has often been derived from a linearized multistage model with a 95% upper confidence limit providing a low-dose estimate. The interpretation of this number when derived from animal studies is that the true risk to humans is not likely to exceed the upper-limit estimate at low doses and, in fact, may be lower. Excess cancer risk estimates have also been calculated using other models such as the one-hit, Weibull, logit, and probit.

In 1996, EPA proposed revisions to the 1986 EPA Guidelines for Carcinogenic Assessment (EPA, 1996). Rather than relying exclusively on tumor findings, the new Guidelines include an expanded weight-of-evidence approach that emphasizes understanding mode of action, conditions of expression of carcinogenicity (e.g., route and magnitude of exposure), and consideration of all other relevant data. Dose-response assessment is now done in two steps: appropriate models are fitted to data in the empirical range of observation in animal studies to determine a point of departure for the second step, which is extrapolation below the observable range. The 1996 revisions to the Guidelines include several default procedures (linear, nonlinear, or both), rather than relying on the linear multistage (LMS) model as the only default for extrapolation of dose-response relationships. The EPA is currently applying the principles of the new Guidelines, consistent with the 1986 Guidelines. The proposed 1996 Guidelines are not inconsistent with the 1986 Guidelines, and findings under the 1986 Guidelines are considered to be based on sound science for decisionmaking.

In the quantification of noncarcinogenic effects, a reference dose (RfD) is most often calculated. The RfD is an estimate (with uncertainty spanning an order of magnitude) of a daily exposure to the human population (including sensitive subgroups) that is likely to be without an appreciable risk of deleterious health effects during a lifetime. The RfD is derived from a no-observed-adverse-effect level (NOAEL), or lowest-observed-adverse-effect level (LOAEL), identified from a chronic or subchronic study, divided by an uncertainty factor (UF) or factor

times a modifying factor (MF). The RfD is calculated as follows:

$$RfD = \frac{(NOAEL \text{ or } LOAEL)}{[Uncertainty\ Factor(s) \times Modifying\ Factor]} = \text{—} \ mg/kg/day.$$

When the data support it, a benchmark dose (BMD) may be calculated by applying an appropriate mathematical curve-fitting procedure. The BMD can then be used as a NOAEL estimate. The NOAEL, LOAEL, or BMD is divided by a UF times an MF to calculate the RfD. Selection of the UF to be employed in the calculation of the RfD/RfC is based on professional judgment, which considers the entire database of toxicologic effects for the chemical. To ensure that UFs are selected and applied in a consistent manner, the EPA (1994a) employs a modification to the guidelines proposed by the NAS, as shown in the box on the next page (NAS 1977, 1980).

1.3. MAXIMUM CONTAMINANT LEVEL GOAL AND MAXIMUM RESIDUAL DISINFECTANT LEVEL GOAL

Maximum contaminant level goals (MCLGs) are nonenforceable health goals. They are set at concentration levels at which no known or anticipated adverse effects on the health of persons occur, and which includes an adequate margin of safety. Establishment of an MCLG for each specific contaminant is based on the available evidence of carcinogenicity or noncancer adverse health effects from drinking water exposure using EPA's guidelines for risk assessment (see the proposed rule at 59 FR 38677 for a detailed discussion of the process for establishing MCLGs). They can be based on an RfD for general systemic effects, on a quantitative estimate for developmental or reproductive effects, or on a consideration of a cancer quantitative assessment. For carcinogenicity, when a linear low-dose extrapolation is done, the MCLG is set at zero. The maximum residual disinfectant level goal (MRDLG) concept was introduced in the proposed rule for disinfectants to reflect the fact that these substances have beneficial disinfection properties. As with MCLGs, MRDLGs are established at the level at which no known or anticipated adverse effects on the health of persons occur and which allows an adequate margin of safety. MRDLGs are nonenforceable health goals based only on health effects and exposure information and do not reflect the benefit of the addition of the chemical for control for waterborne microbial contaminants.

Uncertainty Factors Used in RfD/RfC Calculations

Standard Uncertainty Factors (UFs)

Use a 10-fold factor when extrapolating from valid experimental results from studies using prolonged exposure to average healthy humans. This factor is intended to account for the variation in sensitivity among the members of the human population.

Use an additional 10-fold factor when extrapolating from valid results of long-term studies on experimental animals when results of studies of human exposure are not available or are inadequate. This factor is intended to account for the uncertainty in extrapolating animal data to risks for humans. A 3-fold uncertainty factor is used for extrapolating from inhalation studies on experimental animals to humans for the derivation of an inhalation RfC. This difference is because dosimetric adjustments reduce the uncertainty associated with extrapolation between experimental animals and humans.

Use an additional 10-fold factor when extrapolating from less than chronic results on experimental animals when there are no useful long-term human data. This factor is intended to account for the uncertainty in extrapolating from less than chronic NOAELs to chronic NOAELs.

Use an additional 10-fold factor when deriving an RfD from a LOAEL instead of a NOAEL. This factor is intended to account for the uncertainty in extrapolating from LOAELs to NOAELs.

Modifying Factor (MF)

Use professional judgment to determine another uncertainty factor (MF) that is greater than zero and less than or equal to 10. The magnitude of the MF depends upon the professional assessment of scientific uncertainties of the study and data base not explicitly treated above, e.g., the completeness of the overall data base and the number of species tested. The default value for the MF is 1.

The MCLG or MRDLG for drinking water is calculated from the RfD for a 70 kg adult consuming 2 L of water per day and also taking into consideration the relative contribution from drinking water. The Agency views the use of 2 L per day adult drinking water consumption to derive the MCLG from an RfD appropriate, because it represents the 84th percentile of adult drinking water consumption. Recent analyses show that the 90th percentile for tap water consumption for persons in the United States aged 11–19 is 1.38 L/day. A conservative estimate is that children may be exposed about 3.5-fold more than adults relative to their water intake–body weight ratio (Draft Water Quality Criteria Methodology Revisions; Human Health, August 1998). The Agency believes that the use of 2 L to calculate the MCLG provides sufficient protection to fetuses and children.

The final Stage 1 D/DBP Rule contains MCLGs for the following disinfectant byproducts: four trihalomethanes (THMs) (chloroform, bromodichloromethane, dibromochloromethane, and bromoform), two haloacetic acids (dichloroacetic acid and trichloroacetic acid), bromate, and chlorite, as shown in Table 1. The final Rule contains MRDLGs for the following disinfectants: chlorine, chloramines, and chlorine dioxide, as depicted in Table 2.

1.4. DETERMINING RISK TO CHILDREN

In developing this regulation, risks to sensitive subpopulations including fetuses and children were taken into account in the assessments of D/DBPs. To determine whether fetuses and children are more sensitive than adults, the following issues were considered:

1. Do D/DBPs cause reproductive and developmental effects at doses below those causing systemic toxicity or cancer?
2. Are fetuses and children more susceptible than adults to the systemic toxicity of D/DBPs?
3. Are fetuses and children more susceptible than adults to cancer from D/DBPs?

For questions 2 and 3 above, mechanistic data were considered when available, that is, for the THMs.

Table 1. Disinfectant Byproducts and their MCLGs
Cited in the Final Stage 1 D/DBP Rule

Disinfectant Byproducts	MCLG (mg/L)
Chloroform	0
Bromodichloromethane (BDCM)	0
Dibromochloromethane (DBCM)	0.06
Bromoform	0
Dichloroacetic acid (DCA)	0
Trichloroacetic acid (TCA)	0.3
Chlorite	0.8
Bromate	0

Table 2. Disinfectants and their MRDLGs
Cited in the Final Stage 1 D/DBP Rule

Disinfectants	MRDLG (mg/L)
Chlorine	4 (as Cl_2)
Chloramine	4 (as Cl_2)
Chlorine dioxide	0.8 (as ClO_2)

1.5. SUMMARY AND CONCLUSIONS

This document evaluates the available data for each of the D/DBPs used for deriving the MCLG or MRDLG, to determine if the derived MCLGs and/or MRDLGs are protective for the fetuses and children. Table 3 summarizes the comparison of toxicity endpoints for the various D/DBPs. As can be seen in the table, chloroform, BDCM, bromoform, DCA, and bromate are considered likely to be carcinogenic for humans. MCLGs of zero were selected after consideration of the potential carcinogenicity of these chemicals. This MCLG of zero would protect both children and adults. The MCLG/MRDLGs for DBCM and chloramine were based on systemic toxicity. The NOAEL/LOAEL used to derive the numbers are lower than the NOEL/LOAELs for developmental effects; therefore, the MCLG/MRDLG would be protective of infants and children. In the case of chlorine, the MCLG is based on systemic toxicity, because the NOAEL of 5 mg/kg/day based on developmental effects is the highest dose tested and

Table 3. Comparison of Toxicity Endpoints

Disinfectant	Systemic Toxicity		Developmental Toxicity		Carcino-genicity	MCLG[b] mg/L
	NOAEL[a] mg/kg/day	LOAEL[a] mg/kg/day	NOAEL mg/kg/day	LOAEL mg/kg/day		
Chloroform	—	12.90	35–50	—	probable	0
Bromodichloro-methane (BDCM)	—	25.00	50.00	—	probable	0 (Ca)
Dibromochloro-methane (DBCM)	21.00	—	200.00	—	possible	0.06 (sys tox & Ca)
Bromoform	25.00	—	50.00	—	probable	0 (Ca)
Dichloroacetic acid (DCA)	—	—	14.00	—	probable	0 (Ca)
Trichloroacetic acid (TCA)	36.5	—	—	330.00	—	0.3 (devel tox)
Bromate	—	—	15.00	—	probable	0 (Ca)
Chlorite	—	—	3.00	0	—	0.8 (devel tox)
Chlorine dioxide	—	—	3.00	—	—	0.8 (devel tox)
Chlorine	14.00	—	5.00[c]	—	—	4.00 (sys tox)
Chloramine	9.5	—	10.00	—	—	3.00 (sys tox)

[a]NOAEL = No observed adverse effect level; LOAEL = Lowest observed adverse effect level.
[b]MCLG = Maximum contaminant level goal; (Ca) = basis for MCLG is carcinogenic effects; (sys tox) = basis for MCLG is systemic toxic effects; (devel tox) = basis for MCLG is developmental effects.
[c]Highest dose tested.

therefore not a true NOAEL. For chlorine dioxide, chlorite, and TCA, the MCLG/MRDLGs are calculated on data from developmental studies; hence the numbers derived would be protective for both children and adults. It can be concluded that the MCLG/MRDLGs of all the D/DBPs in the Stage 1 D/DBP Rule are protective of fetuses, infants, and children.

2. ESTIMATES OF RISK TO CHILDREN FOR STAGE 1 DISINFECTANTS/DISINFECTANT BYPRODUCTS

Descriptions of the data available for evaluating risks to children and conclusions drawn for each MCLG or MRDLG contained in this regulation are given in the following sections.

2.1. CHLORINATED DRINKING WATERS

This section considers studies on exposure to chlorinated drinking water rather than to individual D/DBPs. Several epidemiology studies have reported an association between exposure to THMs and developmental/reproductive effects (Bove et al., 1995; Nuckols et al., 1995; Savitz et al., 1995; Waller et al., 1998). Two studies reported a relationship specifically between developmental/reproductive toxicity and BDCM. An epidemiological study (Kramer et al., 1992) reported an increased risk of intrauterine growth retardation with exposure to drinking water with BDCM concentrations greater than or equal to 10 µg/L compared with drinking water with undetectable BDCM concentrations (odds ration [OR] = 1.7, 95% confidence interval [CI] = 0.9–2.9). Three new reproductive epidemiology studies were published recently.

Klotz and Pyrch (1998) examined the potential association between neural tube defects and certain drinking water contaminants, including some DBPs. In this case-control study, births with neural tube defects reported to New Jersey's Birth Defects Registry in 1993 and 1994 were matched against control births chosen randomly from across the State. Birth certificates were examined for all subjects, as were drinking water data corresponding to the mother's residence in early pregnancy. The authors reported elevated ORs, generally between 1.5 and 2.1, for the association of neural tube defects with total THMs (TTHMs). The only statistically significant results were seen when the analysis was isolated to those subjects with the highest THM exposures (greater than 40 ppb) and was limited to those subjects with neural tube defects in which there were no other malformations (OR = 2.1, 95% CI = 1.1–4.0).

Two other studies investigated early-term miscarriage risk factors. The first of these studies examined the potential association between early-term miscarriage and exposure to THMs (Waller et al., 1998). The second study examined the potential association between early-term miscarriage and tap water consumption (Swan et al., 1998). Both studies used the same group of 5,144 pregnant women living in three areas of California. They were recruited from the Santa Clara area, the Fontana area in southern California, or the Walnut Creek area. The women were all members of the Kaiser Permanente Medical Care Program and were offered a chance to participate in the study when they called to arrange their first prenatal visit. In the Waller et al. study, additional water quality information from the women's drinking water utilities were

obtained so that THM levels could be determined. The Swan et al. study provided no quantitative measurements of THMs (or DBPs), and thus provided no additional information on the risk from chlorination byproducts.

In the Waller et al. (1998) study, utilities that served the women were identified. Utilities provided THM measurements taken during the time period when participants were pregnant. The TTHM level in a participant's home tap water was estimated by averaging water distribution system TTHM measurements taken during a participant's first 3 months of pregnancy. This "first trimester TTHM level" was combined with self-reported tap water consumption to estimate a TTHM exposure level. Exposure levels of the individual THMs (e.g., chloroform, bromoform) were estimated in the same manner. Actual THM levels in the home tap water were not measured.

Women with high TTHM exposure in home tap water (drinking five or more glasses per day of cold home tap water containing at least 75 µg/L of TTHM) had an early-term miscarriage rate of 15.7%, compared with a rate of 9.5% among women with low TTHM exposure (drinking fewer than five glasses per day of cold home tap water or drinking any amount of tap water containing less than 75 µg/L of TTHM). An adjusted OR for early-term miscarriage of 1.8 (95% confidence interval = 1.1–3.0) was determined.

Only high BDCM exposure was associated with early-term miscarriage. This was defined as drinking five or more glasses per day of cold home tap water containing >18 µg/L BDCM. An adjusted OR for early-term miscarriage of 3.0 (95% CI = 1.4–6.6) was determined.

2.2. TRIHALOMETHANES

2.2.1. Chloroform

Chloroform is one of the best studied DBPs and has a very extensive toxicological database. Chloroform and its metabolites have been shown to cause liver and kidney toxicity and tumors as its primary adverse effects.

2.2.1.1. *Developmental/Reproductive Effects*

Three developmental toxicity studies (two in rats and one in rabbits) by the oral route of administration (Thompson et al., 1974; Ruddick et al., 1983), and three developmental toxicity studies (two in rats and one in mice) by the inhalation route of administration (Schwetz et al., 1974; Murray et al., 1979) were investigated.

Thompson et al. (1974) studied the effects of chloroform on embryonic and fetal development of Sprague-Dawley rats. In a range-finding study, pregnant Sprague-Dawley rats were administered gavage doses of 79, 126, 300, 316, or 516 mg/kg/day chloroform in corn oil on days 6–15 of gestation. The dams developed alopecia, rough hair, and eczema. Food consumption and body weight gain were significantly decreased at 126 mg/kg/day and higher, and 316 mg/kg/day resulted in severe maternal toxicity and death, as well as fetotoxicity. In the main study, groups of 25 pregnant rats (181–224 g) were gavaged with chloroform in corn oil at total daily doses of 0, 20, 50, or 126 mg/kg/day by oral intubation on days 6–15 of gestation, administered in two doses/day. Dams receiving 50 or 126 mg/kg/day displayed signs of maternal toxicity (decreased weight gain, mild fatty changes in the liver). There was no evidence of maternal toxicity at 20 mg/kg/day, although microscopic examinations were conducted on only 2 dams/group. Fetuses were removed by caesarean section 1 or 2 days prior to expected parturition and examined for external, skeletal, and/or soft tissue abnormalities. There were no fetal malformations. The incidence of fetuses with bilateral extra lumbar ribs was significantly ($p<0.05$) increased at the high dose, but the increase in affected litters was not statistically significant. Fetal weight was also reduced at the high dose ($p<0.05$). This study identified a maternal NOAEL of 20 mg/kg/day and a LOAEL of 50 mg/kg/day in rats. For developmental effects, the NOAEL was 50 mg/kg/day, with a LOAEL of 126 mg/kg/day.

In the same study, Thompson et al. (1974) administered chloroform (in corn oil) to Dutch-Belted rabbits. In a preliminary range-finding study, doses of 0, 25, 63, 100, 159, 251, or 398 mg/kg/day were administered to pregnant rabbits on days 6–18 of gestation. High levels of maternal death (60–100%) were observed at doses of 100 mg/kg/day and above. Adverse effects at 63 mg/kg/day included anorexia, weight loss, diarrhea, abortion, and one maternal death. No overt signs of toxicity other than mild diarrhea and intermittent anorexia were observed in dams dosed with 25 mg/kg/day. In the main study, groups of 15 pregnant dams (1.7–2.2 kg) were dosed by oral intubation with chloroform at 0, 20, 35, or 50 mg/kg/day on days 6–18 of gestation. Decreased maternal weight gain was observed in dams given 50 mg/kg/day. Four high-dose dams died from hepatotoxicity, but no evidence of hepatotoxicity was observed in surviving rabbits. Four high-dose dams aborted, but this was not considered to be a treatment-related effect because three control animals aborted. Histopathology examinations revealed no evidence of maternal toxicity at 35 mg/kg/day. Small reductions in body weights (7.5% and 12%, respectively) were observed in fetuses from dams administered 20 or 50 mg/kg/day ($p<0.05$), whereas only a 5.5% decrease in fetal weight was observed at 35 mg/kg/day. At least some of the decrease in fetal weight at the high dose may be attributable to the larger litter size at the high dose (7.4, vs. 6.4 in the controls), although the mean litter size at

the mid dose was only 4.5. An increased incidence of fetuses with incompletely ossified skull bones (usually parietals) was observed at 20 and 35 mg/kg/day ($p<0.05$); a smaller increase at the high dose was not statistically significant. This study is limited by the high incidence of abortions and mortality in the control group. Due to the absence of a clear dose response, no definitive NOAEL or LOAEL for developmental effects can be identified for this study, although the NOAEL appears to be in the range of 35–50 mg/kg/day.

Ruddick et al. (1983) investigated the potential developmental toxicity of chloroform in groups of 15 mated Sprague-Dawley rats. Pregnant dams (8–14 animals per dose group) were given 0, 100, 200, or 400 mg/kg chloroform in corn oil on days 6–15 of gestation. Maternal weight gain was depressed by at least 20% at all dose levels. In addition, all dose levels of chloroform produced maternal liver enlargement, decreased hemoglobin, and decreased hematocrit. Levels of serum inorganic phosphorus and cholesterol were elevated in the dams at the highest exposure level. Fetal weight was decreased by about 19% at the highest dose level. There were no fetal malformations, but sternebra aberrations were observed with a dose-dependent incidence at 200 mg/kg/day and 400 mg/kg/day. Interparietal deviations also occurred at the high dose. There was a clear increase in the incidence of these variations indicating a potential developmental effect.

In a study via the inhalation route, Schwetz et al. (1974) exposed pregnant female Sprague-Dawley rats to chloroform at target concentrations of 0, 30, 100, or 300 ppm (actual concentrations of 0, 30, 95, or 291 ppm; 0, 146, 464, or 1420 mg/m^3) for 7 hours/day from gestation days 6 through 15. The numbers of pregnant rats exposed in each group were 68, 22, 23, and 3, respectively. Although the study authors attributed the low percentage of pregnancy at the high concentration (15%) to chloroform exposure, this result can not be treatment-related, in light of the timing of exposure. The small number of pregnant animals at 300 ppm reduced the study sensitivity. Dams were sacrificed on gestation day 21, and fetuses delivered by caesarian section. Numbers of live, dead, and resorbed fetuses were determined. Fetuses were weighed, measured, and sexed. Half were examined for skeletal anomalies, and the other half were examined for organ anomalies. High-exposure dams lost weight during exposure and ate minimal amounts of food (approximately 1 g/rat/day); concentration-related decreases in body weight gain and food consumption were observed at lower exposure levels. Relative liver weights were significantly increased in dams exposed to 100 and 300 ppm at study termination, with a significant decrease in absolute liver weight at 300 ppm. At 300 ppm, 61% of the implantations were resorbed, a statistically significant increase. This high resorption rate was not observed in the "starved" control group, suggesting that weight loss cannot account for the observed effect, although the starved control group was provided more food (3.7 g/rat/day) than

was consumed by the 300 ppm group. Fetal body weights were significantly decreased (40%) at 300 ppm. and fetal crown-rump lengths were slightly, but significantly, decreased (2%) at 30 ppm and significantly decreased (15%) at 300 ppm. The frequencies of litters with acaudia or imperforate anus were significantly increased at 100 ppm. Malformations were not observed at 300 ppm, but there were only three litters at this concentration. The frequency of litters with delayed ossification was elevated in all exposure groups. In addition, there were statistically significant increases in wavy ribs at 30 ppm, and in missing ribs and subcutaneous edema at 100 ppm. The authors concluded that chloroform exposures of 100 and 300 ppm were highly embryotoxic and fetotoxic, with embryolethality a significant effect at 300 ppm.

Murray et al. (1979) found that 100 ppm chloroform was teratogenic in CF-1 mice exposed on gestation days 8–15, but fetotoxic in mice exposed on gestation days 1–7 or 6–15. Groups of 34–40 pregnant females (as determined by vaginal plug) were exposed to 0 or 100 ppm (0 or 490 mg/m^3) for 7 hours/day on gestation days 1–7, 6–15, or 8–15, and sacrificed on gestation day 18. The ability of the mice to maintain pregnancy was significantly decreased in the groups exposed on gestation days 1–7 or 6–15, and there was a slight, but not statistically significant, decrease in pregnancies in the group exposed on gestation days 8–15. Statistically significant decreases in fetal weight and fetal length were observed in the groups exposed on gestation days 1–7 and 8–15, but not on days 6–15. Cleft palate was observed at a statistically significant increased incidence in litters of mice exposed on gestation days 8–15, but not in the other groups. Cleft palate was seen predominantly in fetuses with retarded growth. No other malformations were significantly increased in any group, although increased incidences of two skeletal variations were observed. Delayed ossification of skull bones was significantly increased in all exposed groups, and delayed ossification of sternebrae was significantly increased in the groups exposed on gestation days 1–7 and 8–15, but not 6–15. The study authors suggested that the lack of malformations in the group exposed on gestation days 6–15 may have resulted from the lethality to the early embryo obscuring other effects. Maternal toxicity was evident as increased liver weight and increased serum glutamate-pyruvate transaminase activity.

Based on the findings of animal studies discussed above, developmental effects have been found after chloroform exposure (e.g., pup weight reduction, skeletal variations). These prenatal effects, however, were typically associated with doses causing maternal toxicity, and occurred at doses above those causing hepatotoxicity. The oral NOAEL for developmental toxicity is in the range of 35–50 mg/kg/day, and the oral LOAEL for hepatotoxicity is 12.9 mg/kg/day for chloroform. Therefore, the RfD and MCLG based on a LOAEL of 12.9 mg/kg/day for liver effects would be sufficiently protective.

A multigeneration reproductive assay was conducted with chloroform in CD-1 mice (NTP, 1988). This assay evaluated reproductive effects in two successive generations, as well as systemic effects in the second generation (i.e., F1 animals). In the first phase, mice were administered chloroform by gavage in corn oil at 6.6, 16, or 41 mg/kg/day, 7 days/week for 18 weeks. In the second phase, the last litter of the control and of the high-dose groups were retained. After weaning, the mice were administered the same chloroform dose as their parents, and dosing continued through mating and parturition, when the study was terminated. No adverse effects on fertility or reproduction of the F1 generation were observed, although increased liver weight and liver lesions (degeneration of centrilobular hepatocytes, accompanied by occasional single cell necrosis) were observed in all females exposed to the single dose tested. The degeneration was characterized as minimal in 2/20, mild in 9/20, and moderate in 9/20 animals. Thus, a dose of 41 mg/kg/day caused mild to moderate liver histopathology in F1 females. No NOAEL can be identified for this effect, because the low- and mid-dose groups were not evaluated histopathologically. However, no adverse effects on fertility or reproduction were found.

2.2.1.2. *Systemic Effects*

Numerous animal studies in several species (rats, mice, and dogs) have shown that liver and kidney toxicity are primarily target sites for the systemic effects of chloroform. Nasal toxicity is also found in the rat following inhalation exposure. Organ toxicity (and liver and kidney tumor response) following chloroform treatment vary with the exposure route, vehicle of administration, and strain of rat or mouse. The sensitivity to the organ toxicity induced by chloroform is associated with oxidative metabolism. These results were summarized in two EPA documents (EPA, 1994b, 1998b), and will not be discussed in this paper. Organ toxicity that results from chloroform is considered to be part of the continuum that leads to tumor development. The organ toxicity is thus discussed below in the context of the mode of carcinogenic action for chloroform.

2.2.1.3. *Carcinogenicity*

Chloroform has been found to cause liver and kidney tumors in rodents (discussed in EPA, 1994a, 1998a). To explore the issue of whether fetuses or children are at increased cancer risk compared with adults, the mode of carcinogenic action of chloroform was examined. Several issues (e.g., differences in rate of cell proliferation and metabolism in children versus adults) were explored concerning susceptibility of fetuses or children in the context of what is known about the carcinogenic mode of action of chloroform. A substantial body of data

indicates that chloroform is not a DNA-reactive mutagen. Thus, mutagenicity is not the key influence of chloroform on the carcinogenic process. Chloroform induces liver and kidney tumors at doses that cause cell injury or organ toxicity. Numerous studies have shown that organ toxicity and regenerative proliferation are associated with tumorigenicity of chloroform, and thus are key steps in its carcinogenic mode of action (EPA, 1998b; ILSI, 1997).

Organ toxicity from chloroform is dependent on oxidative metabolism primarily by cytochrome P450 CYP2E1 (as discussed in EPA, 1998b). The oxidative metabolism of chloroform generates highly tissue reactive metabolites, phosgene and HCl, which produce cytotoxicity (cell death) and regenerative hyperplasia (EPA, 1998b; ILSI, 1997). This process may lead to tumor development if sustained. Given that oxidative metabolism is key to the carcinogenic potential of chloroform, studies on CYP2E1 in fetal and adult tissues were evaluated (EPA, 1998b). The status of CYP2E1 in human fetuses remains unclear, with conflicting studies. In those studies showing expression of CYP2E1, levels lower than those in adults were found (Vieira et al., 1996; Boutelet-Bochan et al., 1997; Carpenter et al., 1996; Hakkola et al., 1998). Regardless of fetal CYP2E1 expression, the enzyme is rapidly induced upon birth. Animal studies of CYP2E1 provide evidence of rapid induction of this gene soon after birth (Song et al., 1986; Umeno et al., 1988; Schenkman et al., 1989; Ueno and Gonzalez, 1990).

The study by Schenkman et al. (1989) indicated that CYP2E1 protein is present in low levels in neonates, rises to a peak level at age 2 weeks, and subsequently decreases to adult levels by puberty. Analysis of protein levels quantified from western blots showed a maximum at 2 weeks with decreasing levels at 4 and 12 weeks. The protein level at 12 weeks was approximately 50% of the level at 2 weeks. The authors did not provide a statistical analysis of this result, but it appears from the error bars that the 2-week and 12-week levels (but not 4-week levels) were significantly different.

Song et al. (1986) conducted a similar analysis and reported a rapid transcriptional induction of CYP2E1 (P450) within 1 week following birth that remained elevated throughout 12 weeks. The authors did not quantitate the western blots. However, in this same study, enzyme activity gradually increased over time, reaching a maximum at adulthood.

Ueno and Gonzalez (1990) showed that extracts from 3-day-old and 12-week-old rat liver, but not those from fetal or newborn rat liver, were able to generate significant CYP2E1 transcription in vitro. The ability of the extract to result in transcription of CYP2E1 was slightly greater at 12 weeks.

Taken together, the animal studies do not provide conclusive evidence of an early period of increased enzymatic activity. If, however, the twofold increase in CYP2E1 induction in animals were verified, its importance in terms of chloroform toxicity would depend on the dose. Under low-dose conditions (e.g., much lower than the Km) it is possible that an increase in the level of enzyme would not have any effect on active metabolite formation, because the amount of chloroform, and not CYP2E1, would control the rate of the enzyme activity. On the other hand, under saturating doses of chloroform, all the available enzyme would be active; thus a twofold increase in CYP2E1 could result in greater activation of the compound. Although the animal data remain unclear regarding the potential for a neonatal period of increased CYP2E1 activity above that in the adult, the data in humans show a gradual increase of CYP2E1 activity throughout childhood with a maximum level at adulthood, as demonstrated by Vieira et al. (1996). Therefore, although children may have capacity to metabolize chloroform, data on CYP2E1 activity provide no evidence to suggest that children have an increased susceptibility to chloroform toxicity compared with adults (EPA, 1998b). Furthermore, the animal data from Schenkman et al. (1989) fall within the UF for intraspecies variability.

The next issue to examine is whether the developing fetus may be more susceptible to the toxicity of chloroform because of its greater rate of cell proliferation. There are very few data for pre- and postnatal exposures to chloroform and resultant organ toxicity. Liver toxicity was found in a multigeneration reproductive assay in CD-1 mice (NTP, 1988). The period of exposure includes prenatal, postnatal, and adult stages. The liver toxicity from this multigeneration reproductive study was compared with that of a comparable 90-day study in adult B6C3F1 mice for liver toxicity (Bull et al., 1986). The similarity of effects at comparable doses from these two studies suggests that there is not an increased susceptibility to chloroform that results from pre- or postnatal exposures (EPA, 1998b). It should be noted that there are limitations in this comparison; different strains of mice were used, and only LOAELs were identified in these two studies (EPA, 1998b).

2.2.1.4. *Children's Risk in Relation to the MCLG*

Because a substantial database indicates that tumor development for chloroform is secondary to organ toxicity and regenerative proliferation, a nonlinear dose-response approach is viewed as more appropriate. Use of a low dose-linear approach is considered overly

conservative for extrapolating cancer risk associated with chloroform exposure (EPA, 1998a,b; ILSI, 1997). In the nonlinear approach, the MCLG is based on liver toxicity as the most sensitive effect for chloroform (as it is the lowest possible MCLG for any organ) and as a precursor response to a key step to its carcinogenicity. This approach is considered equally protective of both adults and children because the database on chloroform does not indicate that children are more susceptible than adults to liver toxicity. The mode of action by which chloroform produces organ toxicity and carcinogenicity is, thus, the same for children and adults. An oral study in dogs (Heywood et al., 1979) was used to derive the RfD of 0.01 mg/kg/day (EPA, 1994b). This RfD is based on a LOAEL (12.9 mg/kg/day) for hepatotoxicity and application of a UF of 1,000 (100 was used to account for inter- and intraspecies differences and a factor of 10 for use of a LOAEL) to calculate the RfD (0.01 mg/kg/day).

The MCLG is calculated to be 0.07 mg/L by assuming an adult tap water consumption of 2 L per day for a 70 kg adult, and by applying a relative source contribution of 20%.

The MCLG of 0.07 mg/L is considered protective of both adults and children, given that developmental effects occurred at doses above those causing hepatotoxicity. Also, the mode of action data indicates that children are not uniquely sensitive to the organ toxicity caused by high doses of chloroform.

Under the linear approach, the MCLG would be zero because under that approach it is assumed that there is no dose that is without some risk. An MCLG of zero would be a zero risk for either children or adults.

2.2.2. Brominated Trihalomethanes

There is sufficient evidence for carcinogenicity via ingestion of bromoform and BDCM to consider them probable human carcinogens. The evidence is limited for DBCM. Based on available data, mechanism of action involving mutagenicity was postulated for the brominated THMs, implying linear low-dose extrapolation as a reasonable approach. The proposed mechanism of carcinogenicity for these compounds was examined to determine if this would provide any reason for concern that children or fetuses may be more susceptible to development of cancer following exposure. If carcinogenicity is the result of mutations by either the parent compound or a metabolite, children or the developing fetus could be more susceptible to the carcinogenicity of brominated THMs due to a higher rate of cell proliferation in the target organs. However, an increased risk of this type would be true for all genotoxic carcinogens and not

specific to brominated THMs. There are no data currently available for brominated THMs to permit quantification of a possible increase in risk to the developing fetus or children.

Brominated THMs are extensively metabolized via oxidative and reductive pathways in humans and animals, primarily in the liver, but also in the kidney (EPA, 1994b). Oxidative metabolism (which requires the presence of oxygen) results in the production of a dihalocarbonyl (CX2O) intermediate. Under conditions of low oxygen (reductive metabolism), the metabolic reaction products appear to be free radical species such as the dihalomethyl radical (CHX2.). Both dihalocarbonyls and dihalomethyl radicals are reactive species and may cause direct/indirect damage to cellular components including DNA. For BDCM, the genotoxicity is associated with the glutathione conjugation pathway (Pegram et al., 1997).

Studies have investigated CYP isozyme involvement in THM metabolism. Thorton-Manning et al. (1994) found that BDCM dosing decreased the activity of CYP1A and CYP2B, but not CYP2E1. The cytochrome P450 CYP2E1 subfamily of enzymes undergo changes during development. Activity is absent during gestation, with onset at birth in rats. Limited studies compare the enzyme activity at different stages of life for CYP2E1 (Song et al., 1986; Umeno et al., 1988; Schenkman et al., 1989; Ueno and Gonzalez, 1990; Ronis et al., 1996; Vieira et al., 1996). There are even fewer data for other P450 isozyme families and glutathione transferase in this aspect.

Several epidemiology studies have reported an association between exposure to THMs and developmental/reproductive effects (Bove et al., 1995; Savitz et al., 1995; Nuckols et al., 1995; Waller et al., 1998). Two studies have reported a relationship specifically between developmental/reproductive toxicity and BDCM. An epidemiological study (Kramer et al., 1992) reported an increased risk of intrauterine growth retardation with exposure to drinking water with BDCM concentrations greater than or equal to 10 µg/L compared with drinking water with undetectable BDCM concentrations (OR = 1.7, 95% CI = 0.9–2.9). Waller et al. (1998) reported an increased risk of spontaneous abortion with consumption of greater than or equal to five glasses of cold water with a BDCM concentration greater than or equal to 18 µg/L (OR = 2.0, 95% CI = 1.2–3.5). Because the subjects were exposed to other contaminants and disinfection byproducts in the drinking water, correlation of the effects directly to individual brominated THM exposure is difficult.

2.2.2.1. *Bromodichloromethane*

Developmental/Reproductive Effects

Developmental and reproductive toxicity data are available for BDCM and were considered in the derivation of the MCLG. Two oral studies evaluated the developmental toxicity of BDCM (Ruddick et al., 1983; Narotsky et al., 1997). Ruddick et al. investigated developmental toxicity in pregnant Sprague-Dawley rats (9–14/group) administered BDCM by gavage in corn oil at dose levels of 0, 50, 100, or 200 mg/kg/day from gestation days 6–15. Maternal toxicity at the high dose was indicated by a significantly decreased body weight gain. A NOAEL for developmental effects (sternebra aberration) in this study was 50 mg/kg/day. To determine the effect of vehicle on BDCM toxicity, Narotsky et al. administered BDCM by gavage in either corn oil or an aqueous vehicle with Emulphor® to pregnant F344 rats (12–14/group) at dose levels of 0, 25, 50, or 75 mg/kg/day during gestation days 6–15 . Decreased maternal weight gain and full-litter resorption were observed at 50 and 75 mg/kg/day. The incidence of full-litter resorption was significantly higher in the corn oil vehicle (83%) compared with the aqueous vehicle (21%) at the high dose. Accordingly, the NOAEL for developmental and maternal effects would be 25 mg/kg/day.

Systemic Effects

BDCM causes decreased weight gain and various adverse effects in the nervous and immune systems, thyroid, kidney, and liver. The NTP (1987) chronic study was selected by EPA as the most appropriate study for derivation of the RfD. The lowest LOAEL value of 25 mg/kg/day for lesions (in liver, thyroid, and kidney) observed in treated mice was selected to derive a RfD value of 0.02 mg/kg/day or 0.7 mg/L for a 70 kg adult drinking 2 L of water/day. A UF of 1,000 was employed for use of a LOAEL, extrapolation from an animal study to humans, and to account for variation in sensitivity among members of the human population.

Carcinogenicity

There is sufficient animal evidence to consider BDCM a possible human carcinogen by ingestion. Tumors were observed in the large intestine and kidneys of male and female rats, kidneys of male mice, and livers of female mice when these rodents were treated with BDCM in corn oil in a 2-year bioassay (NTP, 1987). Consideration of mode of action involving irreversible changes (mutations) led to use of a linear low-dose extrapolation. The Agency calculated a cancer oral slope factor based on renal tumors in treated male mice (6.2×10^{-2} per mg/kg/day). The concentration for 10^{-5} lifetime risk is 6 µg/L.

Children's Risk in Relation to the MCLG

The Agency has proposed an MCLG of zero for BDCM based on its probable carcinogenicity. The consideration of mode of action involving irreversible changes (mutations) led to the subsequent use of a linear low-dose extrapolation. The Agency believes that the proposed MCLG of zero is protective of both child and adult health.

2.2.2.2. *Dibromochloromethane*

Developmental/Reproductive Effects

Ruddick et al. (1983) investigated developmental toxicity in pregnant Sprague-Dawley rats (10–12/group) administered DBCM by gavage in corn oil at dose levels of 0, 50, 100, or 200 mg/kg/day from gestation days 6 to 15. Although maternal toxicity was indicated by a significant decrease in body weight gain at the highest dose, no treatment-related developmental toxicity was observed for DBCM. Therefore, the NOAEL in this study for developmental effects would be 200 mg/kg/day, which is approximately 10-fold higher than the NOAEL used to derive the RfD based on liver toxicity. Borzelleca and Carchman (1982) conducted a two-generation reproductive study in ICR Swiss mice. Nine-week-old mice (10 males and 30 females per dose group) were continuously maintained on drinking water containing 0, 0.1, 1.0, or 4.0 mg/mL DBCM (0, 17, 171, or 685 mg/kg/day). Based on maternal toxicity (weight loss, liver pathology) and possible fetotoxicity (decreased pup weight and viability in some generations), this study identified a NOAEL of 17 mg/kg/day and a LOAEL of 171 mg/kg/day for DBCM. This NOAEL is similar to the NOAEL used to derive the RfD based on liver toxicity (a duration-adjusted NOAEL of 21 mg/kg/day with a duration-adjusted LOAEL of 43 mg/kg/day).

NTP (1996) conducted a short-term reproductive toxicity study on Sprague-Dawley male and female rats. These rats were treated with DBCM in drinking water at concentrations of 0, 50, 150, or 450 ppm during a study period of 35 days (from gestation day 6 through parturition). Based on measured water consumption, the authors estimated dose levels for the treated males to be 4.2, 12.4, and 28.2 mg/kg/day, and for treated females 6.3, 17.4, and 46.0 mg/kg/day. The developmental toxicity of the offspring from these treated rats are compared with those from the control group. After a through examination, no significant reproductive/developmental toxicity was observed at any dose level; the NOAEL for reproductive/developmental effects identified in this study is 28.2 mg/kg/day.

Systemic Effects

DBCM causes decreased weight gain and various adverse effects in the nervous and immune systems, kidneys, and liver. The NTP subchronic study in rats was selected by EPA as the most appropriate basis for derivation of the RfD and DWEL (NTP, 1985). The lowest NOAEL value of 30 mg/kg/day in rats (for absence of clinical and histological changes) was selected to derive the RfD value of 0.02 mg/kg/day (see also Children's Risk section).

Carcinogenicity

Evidence is limited for DBCM carcinogenicity via ingestion. Tumors occurred in the livers of female and male mice when these rodents were treated with BDCM in corn oil for 2 years (NTP, 1985). Consideration of mode of action involving irreversible changes (mutations) led to use of a linear low-dose extrapolation. The Agency used a cancer oral slope factor of 8.4×10^{-2} per mg/kg/day based on liver tumors in treated female mice. The concentration for 10^{-5} lifetime risk is 4 µg/L.

Children's Risk in Relation to the MCLG

The proposed MCLG of 0.06 mg/L for DBCM is based on noncarcinogenic endpoints (the RfD) with an additional safety factor to account for possible carcinogenicity. An RfD of 0.02 mg/kg/day was derived from a duration-adjusted NOAEL of 21 mg/kg/day for liver toxicity in exposed rats from a subchronic study (NTP, 1985) and divided by a UF of 1,000. The UF accounts for use of a less-than-lifetime study, interspecies extrapolation, and variability among members of the human population. An additional safety factor of 10 for possible carcinogenicity is used to calculate the MCLG along with an assumed drinking water contribution of 80% of total exposure to DBCM: MCLG = (30 mg/kg/d × 5/7 × 70 kg × 0.8)/(1,000 × 2 L/d × 10) = 0.06 mg/L. Developmental and reproductive toxicity data are available for DBCM and were considered in the derivation of the RfD value for BDCM. The Agency believes that the MCLG of 0.06 mg/L is protective of children's health because no developmental or reproductive effects have been found to occur below the level of the critical effect (liver toxicity) used to derive the current RfD.

2.2.2.3. *Bromoform*

Developmental/Reproductive Effects

Ruddick et al. (1983) investigated developmental toxicity in pregnant Sprague-Dawley rats (14–15/group) administered bromoform by gavage in corn oil at dose levels of 0, 50, 100, or 200 mg/kg/day from gestation days 6 to 15. No maternal toxicity was observed at any dose

level: some fetal skeletal anomalies were observed. Incidences of both fetuses and litters with interparietal deviations were increased at the mid- and high-dose groups compared with the controls. Furthermore, incidences of both fetuses and litters with sternebra aberrations increased in a dose-related fashion. A NOAEL for developmental effects would be 50 mg/kg/day. The prenatal and postnatal effects of bromoform on fertility and reproduction was investigated by NTP (1989a) in Swiss CD-1 mice using a continuous reproductive breeding protocol. Mice were administered bromoform by gavage in corn oil for 105 days at dose levels of 0, 50, 100, or 200 mg/kg/day. No effect on any fertility or reproductive parameter (numbers of litters per pair, litter size, proportion of live pups, sex ratio of live pups, and pup body weight) was observed in the F_0 generation. The effect of bromoform administration was also evaluated in the control and high dose group of the F_1 generation. No effect was observed in the standard reproductive endpoints (mating index, fertility index, litter size, proportion of live pups, sex ratio, or pup body weight). Furthermore, no effect was observed on any sperm parameters evaluated (density, motility, or morphology). Therefore, the NOAEL for reproductive effects would be 200 mg/kg/day.

Systemic Effects

Bromoform causes decreased weight gain and various adverse effects in the nervous and immune systems, kidney, and liver. The NTP (1989b) subchronic study was selected by EPA as the most appropriate basis for derivation of the RfD and DWEL. The lowest NOAEL value of 25 mg/kg/day for absence of clinical and histological effect observed in treated rats was selected to derive the RfD value of 0.02 mg/kg/day. A UF of 1,000 was based on NAS/OW guidelines for use of a NOAEL from a less-than-lifetime study, extrapolation from an animal study to humans, and for variation in sensitivity among members of the human population.

Carcinogenicity

Evidence is sufficient to consider bromoform a possible human carcinogen via ingestion. Tumors occurred in the large intestine in female and male rats when these rodents were treated with bromoform in corn oil (NTP, 1989b). Consideration of mode of action involving irreversible changes (mutations) led to use of a linear low-dose extrapolation. The Agency used a cancer oral slope factor of 7.9×10^{-3} per mg/kg/day based on liver tumors in treated female mice. The concentration for 10^{-5} lifetime risk is 40 µg/L.

Children's Risk in Relation to the MCLG

The Agency has proposed an MCLG of zero for bromoform based on its probable carcinogenicity. The consideration of mode of action involving irreversible changes (mutations) led to the use of a linear low-dose extrapolation. The Agency believes that the proposed MCLG of zero is protective of both child and adult health.

2.2.3. Haloacetic Acids

An MCL of 0.06 mg/L for a combined total of five haloacetic acids (mono-, di-, and trichloroacetic acids and mono- and dibromoacetic acids) has been determined by the EPA. The MCLG for DCA has been set at zero based on its potential human carcinogenicity. The MCLG for TCA has been set at 0.3 mg/L, based on developmental and possible carcinogenic effects (EPA, 1994a).

2.2.3.1. *Dichloroacetic Acid*

The MCLG for DCA has been set at zero based on evidence of potential carcinogenicity in humans and, consideration of mode of action involving irreversible changes (mutations) (EPA, 1994a). The available animal studies also suggest a potential for developmental toxicity; however, the doses used in these animal studies were very high.

Reproductive/Developmental Effects

In two studies reported by Smith et al. (1992), pregnant Long-Evans female rats (approximately 20/dose) received DCA by gavage on gestation days 6–15 doses of 900, 1,400, 1,900, or 2,400 mg/kg/day (first study) or 14, 140, or 400 mg/kg/day (second study). Dams were sacrificed on gestation day 20, and both maternal toxicity and fetal toxicity were assessed. Dose-related increases in mortality occurred in dams dosed at 1,400 mg/kg/day and above, body weight gain was significantly reduced at 140 mg/kg/day and above, significant implantation loss occurred at 900 mg/kg/day and above, and the number of live fetuses per litter was reduced at 900 mg/kg/day and above. Fetal weight and crown length were significantly lower at levels of 400 mg/kg/day and above. Dose-related increases were also reported for external, soft tissue, cardiovascular, urogenital, and orbital malformations in the developing fetuses at doses of 140 and above. No malformations were observed in 507 fetuses (39 litters) in the control group. The authors identified a developmental NOAEL of 14 mg/kg/day and LOAEL of 140 mg/kg/day. Because maternal toxicity was observed at all doses, a LOAEL of 14 mg/kg/day was identified

for maternal toxicity. The extent to which the developmental effects were attributable to maternal toxicity is unknown.

Epstein et al. (1992) reported findings from a similar series of experiments in pregnant Long-Evans rats exposed to DCA by gavage. There were three separate, sequential phases of this study; in each phase, the dams were exposed for a specific 1- to 3-day period during gestation and were sacrificed on gestation day 20. Both maternal and fetal toxicity were assessed,
including histological examination of the fetuses. In all three phases of the study, no treatment-related maternal toxicity was observed (based on body weight and organ weight data).

In the first phase of the study, dams were exposed to 1,900 mg/kg/day during gestation days 6–8, 9–11, or 12–15, in order to observe the effects of DCA during specific periods of organogenesis. A decrease in average fetal weight was reported in the dose group exposed on days 6 to 8, but no malformations were reported in this dose group. In the groups dosed on days 9–11 and 12–15, the mean percentage of cardiac malformations per litter was significantly ($p \leq 0.001$) increased.

In the second phase of the study, pregnant dams were administered a single dose of 2,400 mg/kg on gestation days 10, 11, 12, or 13. Fetal weights in each exposure group were similar to control values. Significant ($p \leq 0.05$) increases in cardiac malformations were reported in groups exposed on day 10 or day 12. In the third phase of the study, a single dose of 3,500 mg/kg was administered to dams on gestation days 9, 10, 11, 12, or 13. This higher dose level resulted in a slightly higher incidence of cardiac defects (2.9–3.6%), and the increase was significant ($p \leq 0.05$) on day 12. These experiments suggest that increasing the dose has a minimal effect on incidence of cardiac defects.

Saillenfait et al. (1995) studied the potential developmental toxicity of DCA in a rat whole embryo culture system. Groups of 10 explanted embryos from Sprague-Dawley rats were cultured for 46 hours in 0, 1.0, 2.5, 3.5, 5.0, 7.5, or 10 mM DCA. A significant, dose-dependent decrease in crown-rump length was seen at 3.5 mM and above. A similar study with CD-1 mouse whole embryo culture exposed to DCA for 24 hours found significant increases in neural tube defects at treatment concentrations of 5.9 mM and above (Hunter et al., 1996). These in vitro studies cannot be used for determination of a LOAEL or NOAEL; effects observed in these experiments cannot be translated to in vivo effects due to pharmacokinetic and pharmacodynamic uncertainties. However, these results do support the in vivo observations.

Systemic Effects

In a 90-day study in beagle dogs administered DCA by capsule, Katz et al. (1981) observed effects on body weight gain, hematology, and clinical chemistry parameters at doses of 50, 75, and 100 mg/kg/day; neurotoxic and ocular effects were also observed. In a study by Cicmanec et al. (1991), in which dogs were administered 12.5–72 mg/kg/day DCA by capsule for 90 days, a LOAEL of 12.5 mg/kg/day was identified based on degeneration of germinal epithelium and syncytial giant cell formation in testes, and vacuolization of white myelinated tracts of cerebrum and cerebellum. The RfD was calculated from this LOAEL of 12.5 mg/kg/day using the UF of 3,000 for use of a study with a less-than-lifetime duration, for extrapolation from an animal study to humans, and to account for variation in sensitivity among members of the human population.

Carcinogenicity

Several lifetime drinking water studies indicate that DCA is carcinogenic in both rats and mice (Bull et al., 1990; Daniel et al., 1991; DeAngelo et al., 1991), these studies indicate that DCA induces liver tumors. Based on these results, the Agency has classified DCA in Group B2: probable human carcinogen. DCA has been found to be mutagenic and clastogenic, but responses generally occur at high dose levels. It appears that a mutagenic mechanism for DCA carcinogenicity may not be important because of the low exposure levels likely to be found in drinking water. Evidence is still accumulating that suggests a mode of carcinogenic action through modification of cell signaling systems, with down-regulation of control mechanisms in normal cells providing a growth advantage to initiated cells (EPA, 1998c). EPA considers that a contribution of cytotoxicity and compensatory cell proliferation at high doses cannot be ruled out either. It appears that the shape of tumor dose-response curves for DCA is nonlinear. Currently, there is an insufficient database for understanding the mode of action or the appropriate basis for low-dose extrapolation. At this point, EPA has chosen to use an MCLG of zero for DCA, as was proposed in 1994 (EPA, 1994a). NTP is conducting a 2-year rodent bioassay that will include full histopathology, and additional mode of action studies are being done by various investigators, including those at the EPA National Health Effects Research Laboratory.

Children's Risk in Relation to the MCLG

The Agency has proposed an MCLG of zero for DCA based on the considerations described above. The Agency believes that the proposed MCLG of zero is protective of both

children and adults. Developmental effects were found at higher doses and thus are of secondary concern compared with the carcinogenic effects of DCA.

2.2.3.2. *Trichloroacetic Acid*

The MCLG for TCA has been set at 0.3 mg/L based on developmental and possible carcinogenic effects (EPA, 1994a).

Developmental/Reproductive Effects

In a study by Smith et al. (1989), pregnant Long-Evans rats (20 animals/dose) received TCA at doses of 0, 330, 800, 1,200, or 1,800 mg/kg/day in drinking water during gestation days 6–15. Maternal spleen and kidney weights were increased significantly in all dose groups in a dose-dependent manner ($p=0.0001$); liver weights of dams were not affected by TCA treatment. Postimplantation loss increased at dose levels of 330 mg/kg/day and higher. Fetal body weight and crown–rump length were significantly ($p<0.05$) lower than controls for all dose groups. Soft-tissue malformations in the cardiovascular system were increased for all treatment groups in a dose-dependent manner. Levocardia (primarily a defect between the ascending aorta and right ventricle) occurred in 0%, 32%, 71%, 71%, and 88% of the litters in the 0-, 330-, 800-, 1,200-, and 1,800 mg/kg/day groups, respectively. The lowest dose, 330 mg/kg/day, was considered the LOAEL for this study, based on the dose-dependent maternal effects (increased kidney and spleen weights) and developmental effects (decreased fetal weight and crown–rump length and increased incidences of levocardia in litters).

Saillenfait et al. (1995) and Hunter et al. (1996) studied developmental toxicity of TCA in rat whole embryo or mouse whole embryo culture systems. Groups of 10 explanted embryos from Sprague-Dawley rats were cultured for 24 hrs and groups of 10 from CD-1 mice for 46 hrs in 0–6 mM DCA. A significant dose-related decrease in yolk sac diameters was seen at 1.0 mM and above, and a significant dose-related decrease was seen in crown–rump length, head length, somite (embryonic segments) number, protein content, and DNA content at 2.5 mM and above in rats and at 1.0 mM and above in mice. These in vitro studies cannot be used for determination of a LOAEL or NOAEL; effects observed in these experiments cannot be translated to in vivo effects because of pharmacokinetic and pharmacodynamic uncertainties. However, these results do support the in vivo observations.

Systemic Effects

In a 90-day study by Mather et al. (1990), Sprague-Dawley rats (10 males/group) received TCA in drinking water at dose levels of 0, 50, 500, or 5,000 ppm (0, 4.1, 36.5, or 355

mg kg day). No effects were seen on body weight or liver and kidney weights at all doses. At 355 mg/kg day, spleen weight was reduced and the relative kidney and liver weights were increased. Also, at the high dose, hepatic peroxisomal beta-oxidation activity was increased, but no effects were seen on hepatic microsomal enzyme activity. The NOAEL from this study is 36.5 mg/kg/day, and the LOAEL is 355 mg/kg/day.

Bull et al. (1990) treated groups of mice with TCA in their drinking water at 0 mg/L or 1 g/L for 52 weeks, at 2 g/L for 37 weeks with a 15-week recovery period, or at 2g/L for 52 weeks. Doses calculated were approximately 164 or 329 mg/kg/day for 52 weeks or 309 mg/kg/day for 37 weeks. Small increases in liver size, some accumulation of lipofuscin, and focal necrosis were seen in all groups. The LOAEL for hepatic lesions from this study is 164 mg/kg/day.

Carcinogenicity

A number of studies have been done to test the carcinogenic potential of TCA, with conflicting results. A study by DeAngelo et al. (1993) treated groups of 50 male Fischer 344 rats with 0, 0.05, 0.5, and 5 g/L TCA in drinking water for 104 weeks (0, 2.83, 26, and 283.6 mg/kg/day). Body weight and body weight gain were significantly reduced in the high-dose group. Decrease of 10% in absolute liver weight was reported in the high-dose group compared with controls. Histopathology revealed an increase in cytoplasmic necrosis in the high-dose group but no evidence of hyperplastic nodules in the livers. There was no evidence of carcinogenicity in any treatment group compared with controls.

An earlier study by DeAngelo and colleagues (1991) in $B6C3F_1$ mice showed hyperplastic nodules and hepatocellular tumors, both adenomas and carcinomas, mostly in male mice. The authors noted that the female mice appear to be less sensitive than the male mice to the carcinogenic potential of TCA. Bull et al. (1990) found that exposure to TCA via drinking water resulted in induction of liver tumors in male $B6C3F_1$ mice; female mice, however, did not show these effects.

The Agency has classified TCA in category C—a possible human carcinogen based on limited evidence of carcinogenicity from experimental studies. As discussed in the 1997 DBP NODA (EPA, 1997), there have also been several recent studies examining the mode of carcinogenic action for TCA. These new studies suggest that TCA does not operate through mutagenic mechanisms. At this time, EPA has determined that the data are not sufficient to support a choice of mode of action or appropriate basis for a low-dose extrapolation.

Children's Risk in Relation to the MCLG

The Agency has proposed an MCLG of 0.3 mg/L for TCA based on a developmental study in animals. Because this MCLG is based on a NOAEL derived from a reproductive/development study in animals, the Agency believes that the MCLG of 0.3 mg/L is protective of children and adults. This number was derived with a consideration of the increased potential risk to the developing fetus, and is considered to be protective of this subpopulation.

2.2.4. Bromate

Bromate is formed in water following disinfection through ozonation of water containing bromide ion. In laboratory studies, the rate and extent of bromate formation depends on the ozone concentration used in disinfection, pH, and contact time.

2.2.4.1. *Reproductive/Developmental Effects*

Potassium bromate has been studied for prenatal and postnatal reproductive effects in multigeneration reproductive studies on rats and mice (EPA, 1993). In a study by Kurokawa et al. (1990) (cited in EPA, 1993, Final Draft Drinking Water Criteria Document), mice and rats were fed flour treated with 15 ppm potassium bromate (15 mg/kg/day in diet) over five and eight generations, respectively. No effects were observed on weight gain, reproductive performance, or survival in mice or rats. It appeared that 15 mg/kg/day in diet was the NOAEL. A literature search from 1993 to 1998 did not reveal any new developmental toxicity data for bromate.

2.2.4.2. *Systemic Toxicity*

The available data are considered insufficient to ascertain noncancer toxic effects of bromate. Only one toxicity study not dealing with carcinogeincity was located in the literature. The study failed to provide dose response data and did not identify a NOAEL (EPA, 1993).

2.2.4.3. *Carcinogenicity*

In the 1994 proposal, EPA concluded that bromate was a probable human carcinogen (Group B2) under the 1986 EPA Guidelines for Carcinogen Risk Assessment weight of evidence classification approach, and hence MCLG was set at zero. Cancer is the critical effect. The new rodent cancer study by DeAngelo et al. (1998) contributes to the weight of evidence for the potential human carcinogenicity of bromate and confirms the study by Kurokawa et al. (1986a,b). Under the principles of 1996 EPA Proposed Guidelines for Carcinogen Risk Assessment weight of evidence approach, bromate is considered to be a likely human carcinogen. This weight of evidence conclusion is based on sufficient experimental findings that include the

following: tumors at multiple sites in rats, tumor responses in both sexes, and evidence of mutagenicity including point mutations and chromosomal aberrations in vitro (EPA, 1998d).

Consideration of mode of action involving irreversible changes (mutations) led to use of a linear low-dose extrapolation. Cancer risk estimates were derived from the DeAngelo et al. (1998) study by applying the one-stage Weibull model for the low-dose linear extrapolation. Bromate was administered to male F344 rats or B6C3F1 Mice in drinking water at concentrations of 0, 0.02, 0.1, 0.2, and 0.4 g/L or 0, 0.08, 0.4, and 0.8 g/L, respectively, for 108 weeks. The upper bound cancer potency for bromate ion is estimated to be 0.7 per mg/kg/day-1. Assuming a daily water consumption of 2 L for a 70 kg adult, lifetime risks of 10^{-4}, 10^{-5}, and 10^{-6} are associated with bromate concentrations in water of 5, 0.5, and 0.05 µg/L, respectively. This estimate of cancer risk from the DeAngelo et al. study is similar to the risk estimate derived from the Kurokawa et al. (1986a) study presented in the 1994 proposed rule.

2.2.4.4. *Children's Risk in Relation to the MCLG*

The Agency has proposed an MCLG of zero for bromate based on its probable carcinogenicity. Consideration of mode of action involving irreversible changes (mutations) led to use of a linear low-dose extrapolation. The Agency believes that the proposed MCLG of zero is protective of both children and adults.

2.2.5. Chlorite/Chlorine Dioxide

Chlorite and chlorine dioxide are evaluated together in this assessment for children's risk because the studies conducted with chlorite, the predominant degradation product of chlorine dioxide, are likely relevant to characterizing the toxicity of chlorine dioxide. In addition, studies conducted with chlorine dioxide may be relevant to characterizing the toxicity of chlorite. Chlorine dioxide is fairly unstable and rapidly dissociates, predominantly into chlorite and chloride, and to lesser extent, chlorate. There is a ready interconversion among these species in water (before administration to animals) and in the gut (after ingestion) (EPA, 1994c). Therefore, what exists in water or stomach is a mixture of these chemical species (i.e., chlorine dioxide, chlorite, and chlorate) and possibly their reaction products with the gastrointestinal contents. As a result, the toxicity data for one compound are considered applicable for assessing toxicity for the other.

2.2.5.1. *Reproductive/Developmental Effects*

EPA (1994c) and the Report of Health Risk Assessment/Characterization of the Drinking Water Disinfection Byproducts Chlorine Dioxide and Chlorite (EPA, 1998e) describe at length the reproductive and developmental effects observed in chlorine dioxide and chlorite studies; the reader is directed to these documents for detailed information. Several developmental toxicity studies are available on chlorite and chlorine dioxide in addition to the recent two-generation reproductive rat study designed to evaluate the effects of chlorite on reproduction and pre- and postnatal development when administered orally via drinking water for two successive generations (CMA, 1997). The NOAEL from the two-generation reproductive study was determined to be 35 ppm (3 mg/kg/day) based on neurodevelopmental effects. The data considered to support this NOAEL are summarized in EPA (1998e) and included the CMA study as well as previous reports on developmental toxicity. For both chlorite and chlorine dioxide, a NOAEL dose of 3 mg/kg/day from the CMA two-generation study in rats was considered for the derivation of an RfD. For chlorite, using a NOAEL of 3 mg/kg/day and applying a UF of 100 to account for inter- and intraspecies variation in response to toxicity, the MCLG is calculated to be 0.8mg/L. Using a NOAEL of 3 mg/kg/day from the CMA study and applying a UF of 100 for inter- and intraspecies variation response to toxicity, the revised MRDLG for chlorine dioxide is calculated to be 0.8 mg/L.

2.2.5.2. *Systemic Toxicity*

EPA (1994c) and the Report of Health Risk Assessment/Characterization of the Drinking Water Disinfection Byproducts Chlorine Dioxide and Chlorite (EPA, 1998e) discuss at length the subchronic and chronic toxic effects observed in chlorine dioxide and chlorite studies utilizing various exposure durations; the reader is directed to these documents for detailed information about these studies. In general, the systemic toxicity studies presented in these documents are of limited quality.

2.2.5.3. *Carcinogenicity*

There have been no long-term oral bioassays for carcinogenicity of chlorine dioxide, and long-term studies in rats and mice do not provide sufficient evidence to support conclusions as to the carcinogenic potential of chlorite. In accordance with the 1986 cancer guidelines, chlorine dioxide and chlorite were categorized in Group D, "Not classifiable as to human carcinogenicity" (EPA, 1986). In accordance with the 1996 proposed cancer guidelines, the carcinogenicity of these chemicals is considered, "Cannot be determined" (EPA, 1996).

2.2.5.4. *Children's Risk in Relation to the MCLG*

The MCLG and MRDLG calculated for chlorite and chlorine dioxide are considered to be protective of susceptible groups, including children, given that the RfD is based on a NOAEL derived from developmental testing, which includes a two-generation reproductive study. In the case of chlorite and chlorine dioxide a factor of 10 was used to account for variability between the average human response and the response of more sensitive individuals including deficiency associated with glucose-6-phosphate dehydrogenase.

2.2.6. Chlorine

Chlorine forms elemental chlorine (Cl_2), chloride ion (Cl^-), and hypochlorous acid (HOCl) in pure water. As pH increases, hypochlorous acid dissociates to hypochlorite ion (OCl^-). Several factors, including chlorine concentration, pH, temperature, exposure to light, and presence of catalysts or organic material affect the stability of free chlorine in aqueous solution. Because hypochlorite solutions are more stable than hypochlorous acid, calcium hypochlorite and sodium hypochlorite are often used as chlorine sources for disinfection of drinking water (EPA, 1994d). Chlorine and hypochlorites are very reactive and thus can react with the constituents of saliva and possibly food and gastric fluid to yield a variety of reaction byproducts. Thus, the health effects associated with administration of high levels of chlorine and/or the hypochlorites in various animal studies may be due to these reaction byproducts and not the disinfectant itself (EPA, 1994d).

Scully and White (1991) noted that reactions of aqueous chlorine with sulfur-containing amino acids appear to be so fast in saliva that all free available chlorine is dissipated before water is swallowed (EPA, 1994d). Therefore, the possibility of oral exposure to chlorine by fetuses, infants and children is very limited.

2.2.6.1. *Developmental/Reproductive Effects*

Animal studies have demonstrated no evidence of developmental effects associated with chlorine (EPA, 1994d). In a study by Carlton et al. (1986), developmental landmarks such as the mean day of eye opening and the average day of observed vaginal patency were compared across groups with no statistical differences detected. In this study, chlorine was administered by gavage in deionized water at doses of 1.0, 2.0, and 5.0 mg chlorine per kg/day to male and female Long-Evans rats for 66–76 days. No statistical differences were observed between the control and dosed groups in litter survival, litter size, and pup weight. The NOAEL in this study is 5 mg/kg/day; however, higher doses were not tested (IRIS, 1994).

In a multigenerational study, rats were given drinking water chlorinated to a

concentration of 100 mg free chlorine/L (14 mg/kg/day) (Druckrey, 1968). The term "free chlorine" (free available chlorine, free residual chlorine) refers to the concentrations of elemental chlorine, hypochlorous acid, and hypochlorite ion that collectively occur in water. Animals were mated repeatedly and continued to drink the test water throughout gestation and lactation. Microphthalmia of one or both eyes was noted in 17 treated progeny but it was stated that this condition has been known to occur spontaneously in BDII rats. No adverse reproductive or developmental effects were observed.

Meier et al. (1985) demonstrated that oral administration of a sodium hypochlorite solution resulted in dose-related increases in the number of sperm-head abnormalities in male B6C3F1 mice. Ten animals/group were given 1 ml of a free residual chlorine solution daily for 5 days. Test solutions were prepared by bubbling Cl_2 into a 1 M solution of NaOH and adjusted to a pH of either 8.5 (predominant species OCl^-) or 6.5 (predominant species HOCl). The solutions were diluted with distilled water to 200 mg/L, 100 mg/L, and 40 mg/L chlorine equivalents (8.0, 4.0, or 1.6 mg/kg bw/day, respectively). The mice were then sacrificed at 1, 3, or 5 weeks after the last dose was administered. In mice given OCl^-, significant increases in sperm-head abnormalities were observed only at the 3-week interval at doses of 1.6 and 4.0 mg/kg bw/day. These results were reproduced in retrials of the experiment. No dose of HOCl was associated with increases in sperm-head abnormalities (EPA, 1994d).

Six Sprague-Dawley rats (Abdel-Rahman et al., 1982) were administered 0, 1, 10, or 100 mg HOCl/L in drinking water for 2.5 months prior to mating. Animals were maintained on the treated water after pregnancy was confirmed (day 0) and killed on day 20. Maternal weight at time of death was not reported. Incidence of fetal anomalies associated with exposure to hypochlorous acid solutions was not found to be statistically significant. Mean fetal weights from the 10 and 100 mg/L groups were less than the control, but this decrease was not statistically significant. Neither was there a significant difference in numbers of resorptions between control and treated groups. Examination of general trends in the study indicated an increase (not significant) in skeletal anomalies in animals treated with 10 mg HOCl/L. Soft tissue anomalies for the 100 mg HOCl/L treatment group were increased significantly compared

with the control. The findings of these experiments were limited by the small number of study animals; in addition, some calculations of anomaly percentages were reported incorrectly.

2.2.6.2. *Systemic Toxicity*

In the National Primary Drinking Water Regulations; Disinfectants and Disinfection Byproducts; Proposed Rule, issued on July 29, 1994, the MRDLG and the MRDL of 4.0 mg/L for chlorine was proposed. The study selected for determining a RfD is the 2-year rodent bioassay that was conducted by the National Toxicology Program (NTP, 1990). In this study, male and female F344 rats and B6C3F1 mice were given chlorine in distilled drinking water at levels of 0, 70, 140, and 275 mg/L (0, 4, 8, and 14 mg/kg/day) for 2 years. No effects on body weight or survival were observed for any of the treated groups of animals; it should be noted that dosing begins when rats and mice are as young as 7 weeks old.

Using a NOAEL of 14 mg/kg/day identified from female rats in the NTP (1990) study, an MRDLG of 4 mg/L based on lack of systemic toxicity was derived. A UF of 100 was applied to account for inter- and intraspecies extrapolation in accordance with EPA guidelines when a NOAEL from a chronic animal study is the basis for the RfD (EPA, 1994d). Given the UF that is factored into the estimation of the MRDLG and MRDL for chlorine, the value of 4.0 mg/L is considered protective of sensitive subpopulations.

2.2.6.3. *Carcinogenicity*

No apparent carcinogenic potential was demonstrated following oral exposure to chlorine in distilled drinking water as hypochlorite, at levels up to 275 mg/L over a 2-year period (NTP, 1990). The EPA has categorized chlorine in Group D, not classifiable as to human carcinogenicity (EPA, 1994a).

2.2.6.4. ***Children's Risk Relative to the MRDLG***

The Agency believes that the proposed MRDLG of 4 mg/L is protective of children's health.

2.2.7. Chloramines

Inorganic chloramines are alternative disinfectants that are rapidly formed when free chlorine is added to water containing ammonia. Monochloramine is the principal chloramine formed in chlorinated natural and wastewaters at neutral pH and is much more persistent in the environment (EPA, 1994e).

2.2.7.1. *Developmental Effects*

In a developmental study (Abdel-Rahman et al., 1982), the authors investigated the effects of monochloramine administered in drinking water to female Sprague-Dawley rats. Rats were administered 0, 1, 10, or 100 mg/L monochloramine daily in drinking water for 2.5 months before and throughout gestation. On the 20th day of gestation, animals were sacrificed for soft tissues and skeletal examination of the progeny. Monochloramine did not produce any significant changes in rat fetuses at any dose level; there was a slight nonsignificant increase in fetal weight in all chloramine-treated groups compared with controls. The NOAEL identified in this study is 100 mg/L (~10 mg/kg/day), which is slightly higher or almost the same as the NOAEL identified in the systemic toxicity study (see below).

2.2.7.2. *Systemic Toxicity*

EPA selected the lifetime study in rats (NTP, 1990) as the basis for calculating the MRDLG for chloramines. F344/N rats were administered 0, 50, 100, and 200 ppm (2.8, 5.3, and 9.5 mg/kg/day) chloramine in drinking water. Although at the highest dose tested (9.5 mg/kg/day) there were statistically significant changes in body and several organ weights, the biological significance of these changes is unclear. The test animals consumed a reduced amount of water, which was perhaps due to unpalatability, and NTP does not consider these changes in body weight biologically significant. The NOAEL identified is this study is 9.5 mg/kg/day. An MRDLG of 3 mg/L for chloramine (4 mg/L measured as total chlorine) was derived, based on the lack of toxic effects, for a 70 kg adult consuming 2 L/day of water and assuming a relative source contribution (RSC) from drinking water of 80%. A UF of 100 was applied to the NOAEL of 9.5 mg/kg/day (10 for intra- and 10 for interspecies variation).

2.2.7.3. *Carcinogenicity*

The EPA has categorized monochloramine in Group D, not classifiable, because of inadequate human and animal evidence (EPA, 1994a).

2.2.7.4. *Children's Risk in Relation to the MRDLG*

The Agency believes that the MRDLG of 3 mg/L is protective of children's health because no developmental effects were found to occur below the systemic NOAEL of 9.5 mg/kg/day used to derive the current RfD.

3. SUMMARY AND CONCLUSIONS

In developing the Final Stage 1 D/DBP Rule, risks to sensitive subpopulations including fetuses and children were taken into account in the assessments of D/DBPs. To determine whether fetuses and children are more sensitive than adults, the following issues were considered:

- Do D/DBPs cause developmental and reproductive effects at doses below those causing systemic toxicity or cancer?
- Are fetuses and children more susceptible than adults to cancer from D/DBPs?
- Are fetuses and children more susceptible than adults to the systemic toxicity of D/DBPs?

This document evaluates the available data for each of the D/DBPs used for deriving the MCLG or MRDLG, to determine if the derived MCLGs and/or MRDLGs are protective for the fetuses and children. Table 3 summarized the comparison of toxicity endpoints for the various D/DBPs. As can be seen in the table, chloroform, BDCM, bromoform, DCA, and bromate are considered to be probable human carcinogens. MCLGs of zero were selected after consideration of the potential carcinogenicity of the chemicals. This MCLG of zero would protect both children and adults. The MCLG/MRDLGs for DBCM, and chloramine were based on systemic toxicity. The NOAEL/LOAEL used to derive the numbers are lower than the NOEL/LOAELs for developmental effects; therefore, the MCLG/MRDLG would be protective of infants and children. In the case of chlorine, the MCLG is based on systemic toxicity because the NOAEL of 5 mg/kg/day based on developmental effects is the highest dose tested and is therefore not a true NOAEL. For chlorine dioxide, chlorite, and TCA, the MCLG/MRDLGs are calculated on data from developmental studies; hence the numbers derived would be protective for both children and adults. It can be concluded that the MCLG/MRDLGs of all the D/DBPs in the Stage 1 D/DBP Rule are protective of fetuses, infants, and children.

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