FINAL
9-30-2009

Provisional Peer-Reviewed Toxicity Values for

Dibromochloromethane
(CASRN 124-48-1)

Superfund Health Risk Technical Support Center
National Center for Environmental Assessment
Office of Research and Development
U.S. Environmental Protection Agency
Cincinnati, OH 45268

COMMONLY USED ABBREVIATIONS

BMD	Benchmark Dose
IRIS	Integrated Risk Information System
IUR	inhalation unit risk
LOAEL	lowest-observed-adverse-effect level
$LOAEL_{ADJ}$	LOAEL adjusted to continuous exposure duration
$LOAEL_{HEC}$	LOAEL adjusted for dosimetric differences across species to a human
NOAEL	no-observed-adverse-effect level
$NOAEL_{ADJ}$	NOAEL adjusted to continuous exposure duration
$NOAEL_{HEC}$	NOAEL adjusted for dosimetric differences across species to a human
NOEL	no-observed-effect level
OSF	oral slope factor
p-IUR	provisional inhalation unit risk
p-OSF	provisional oral slope factor
p-RfC	provisional inhalation reference concentration
p-RfD	provisional oral reference dose
RfC	inhalation reference concentration
RfD	oral reference dose
UF	uncertainty factor
UF_A	animal to human uncertainty factor
UF_C	composite uncertainty factor
UF_D	incomplete to complete database uncertainty factor
UF_H	interhuman uncertainty factor
UF_L	LOAEL to NOAEL uncertainty factor
UF_S	subchronic to chronic uncertainty factor

PROVISIONAL PEER-REVIEWED TOXICTY VALUES FOR DIBROMOCHLOROMETHANE (CASRN 124-48-1)

Background

On December 5, 2003, the U.S. Environmental Protection Agency's (U.S. EPA) Office of Superfund Remediation and Technology Innovation (OSRTI) revised its hierarchy of human health toxicity values for Superfund risk assessments, establishing the following three tiers as the new hierarchy:

1. U.S. EPA's Integrated Risk Information System (IRIS).
2. Provisional Peer-Reviewed Toxicity Values (PPRTVs) used in U.S. EPA's Superfund Program.
3. Other (peer-reviewed) toxicity values, including:
   - Minimal Risk Levels produced by the Agency for Toxic Substances and Disease Registry (ATSDR),
   - California Environmental Protection Agency (CalEPA) values, and
   - EPA Health Effects Assessment Summary Table (HEAST) values.

A PPRTV is defined as a toxicity value derived for use in the Superfund Program when such a value is not available in U.S. EPA's IRIS. PPRTVs are developed according to a Standard Operating Procedure (SOP) and are derived after a review of the relevant scientific literature using the same methods, sources of data, and Agency guidance for value derivation generally used by the U.S. EPA IRIS Program. All provisional toxicity values receive internal review by two U.S. EPA scientists and external peer review by three independently selected scientific experts. PPRTVs differ from IRIS values in that PPRTVs do not receive the multiprogram consensus review provided for IRIS values. This is because IRIS values are generally intended to be used in all U.S. EPA programs, while PPRTVs are developed specifically for the Superfund Program.

Because new information becomes available and scientific methods improve over time, PPRTVs are reviewed on a 5-year basis and updated into the active database. Once an IRIS value for a specific chemical becomes available for Agency review, the analogous PPRTV for that same chemical is retired. It should also be noted that some PPRTV documents conclude that a PPRTV cannot be derived based on inadequate data.

Disclaimers

Users of this document should first check to see if any IRIS values exist for the chemical of concern before proceeding to use a PPRTV. If no IRIS value is available, staff in the regional Superfund and Resource Conservation and Recovery Act (RCRA) program offices are advised to carefully review the information provided in this document to ensure that the PPRTVs used are appropriate for the types of exposures and circumstances at the Superfund site or RCRA facility in question. PPRTVs are periodically updated; therefore, users should ensure that the values contained in the PPRTV are current at the time of use.

It is important to remember that a provisional value alone tells very little about the adverse effects of a chemical or the quality of evidence on which the value is based. Therefore, users are strongly encouraged to read the entire PPRTV document and understand the strengths

and limitations of the derived provisional values. PPRTVs are developed by the U.S. EPA Office of Research and Development's National Center for Environmental Assessment, Superfund Health Risk Technical Support Center for OSRTI. Other U.S. EPA programs or external parties who may choose of their own initiative to use these PPRTVs are advised that Superfund resources will not generally be used to respond to challenges of PPRTVs used in a context outside of the Superfund Program.

Questions Regarding PPRTVs

Questions regarding the contents of the PPRTVs and their appropriate use (e.g., on chemicals not covered, or whether chemicals have pending IRIS toxicity values) may be directed to the U.S. EPA Office of Research and Development's National Center for Environmental Assessment, Superfund Health Risk Technical Support Center (513-569-7300), or OSRTI.

INTRODUCTION

Dibromochloromethane is a trihalomethane found in drinking water that has been extensively studied in laboratory animals but not humans. IRIS provides a chronic oral RfD of 2×10^{-2} mg/kg-day (U.S. EPA, 1991). The critical study used by IRIS (NTP, 1985) is based on a subchronic rat gavage bioassay with a NOAEL of 30 mg/kg-day converted to a 21.4 mg/kg-day human equivalent concentration (HEC) and a LOAEL of 60 mg/kg-day converted to 42.9 mg/g-day for rat hepatic lesions with a composite uncertainty factor (UF) of 1000. The composite UF is composed of 10 for subchronic assay, 10 for extrapolation from animal data, and 10 for sensitive human populations. The value is of medium confidence (study and database). IRIS also examined a chronic NTP study (NTP, 1985) that gave LOAELs of 40 mg/kg-day for rats and 50 mg/kg-day for mice. IRIS concluded that the subchronic NOAEL was a better basis for the RfD than the chronic LOAEL because of greater confidence in the subchronic NOAEL since the chronic LOAEL was associated with several effects. Due to a lack of inhalation toxicity information, an inhalation RfC is not derived (U.S. EPA, 2009).

HEAST provides a subchronic oral RfD of 2×10^{-1} mg/kg-day (HEAST, 1997) based on a NOAEL of 21.4 mg/kg-day for liver lesions based on the same study used for IRIS (NTP, 1985). ATSDR (2005) provides acute and chronic oral MRLs of 1×10^{-1} and 9×10^{-2} mg/kg-day based on the same NTP study (NTP, 1985). Health advisories include an MCLG of 0.06 mg/L and an MCL of 0.08 mg/L (Final Rule for Disinfectants and Disinfection by-products). IRlS provides a carcinogenicity assessment (U.S. EPA, 1992) of Group C (possible human carcinogen) based on inadequate human data and limited evidence of carcinogenicity in animals; namely, positive carcinogenic evidence in B6C3F1 mice (males and females), positive mutagenicity data, and structural similarity to other trihalomethanes (which are known animal carcinogens). IRIS provides an oral slope factor (OSF) of 8.4×10^{-2} per mg/kg-day based on a gavage female mouse (B6C3F1) study (NTP, 1985). Drinking water Unit Risks are provided as 2.4×10^{-6} per µg/L. A linearized multistage procedure, extra risk model was utilized in the determination. IARC indicated that no epidemiological data relevant to the carcinogenicity of chlorodibromomethane were available and that limited evidence exists in experimental animals for the carcinogenicity of chlorodibromomethane. IARC's overall evaluation is that chlorodibromomethane is not classifiable as to its carcinogenicity to humans (Group 3).

Metabolism studies indicate that the oxidation of dibromochloromethane to bromide and carbon monoxide is mediated primarily by cytochrome P450 2E1, 2B1 and 2B2. Excretion of unmetabolized parent compound and carbon monoxide was observed in expired air of both rats and mice.

Several animal bioassays have been reported for dibromochloromethane using oral gavage, drinking water administration, or dietary exposure as the routes of exposure. No studies of the inhalation route of exposure have been reported in the literature. The primary target tissues in these oral studies are the liver and kidney. NTP (1985) conducted 13 week and 2 year oral gavage studies that found noncancer effects such as hepatic fatty metamorphosis, hepatocellular centrilobular necrosis, and toxic nephropathy. Rats failed to demonstrate dibromochloromethane carcinogenicity in this study. Male mice exhibited hepatocellular carcinomas, while female mice displayed hepatocellular adenomas and carcinomas, and thyroid follicular cell hyperplasia. A 90-day drinking water study of dibromochloromethane in rats resulted in hepatotoxicity and nephrotoxicity. Significant increases in the incidence, multiplicity and size of preneoplastic colon lesions known as aberrant crypt foci (ACF) were found in two subchronic drinking water studies of dibromochloromethane. Shorter-duration studies support the findings of liver and kidney toxicity in rats and mice. Only one two-generation reproduction study has been reported for dibromochloromethane. This study reports hepatotoxicity and reduced fertility.

Computer searches were initially conducted through March 2007 but have since been updated to include information published through May 2009. The databases include health effects and toxicity information available from the U.S. EPA (IRIS), ATSDR, and other relevant federal, state, or international governmental or quasi-governmental agencies: ACGIH, NIOSH, OSHA, NTP, IARC and WHO. Additional sources include CURRENT CONTENTS, TOXLINE, MEDLINE, CANCERLIT, RTECS, HSDB, TSCATS, CCRIS, GENETOX, EMIC, EMICBACK, DART, CalEPA and ETICBACK. Unpublished potentially relevant information from industry or academic laboratories was also sought.

REVIEW OF PERTINENT LITERATURE

Human Studies

There are no epidemiologic data directly relevant to dibromochloromethane in humans (IARC, 1999; 1991).

Animal Studies

NTP (1985) conducted gavage studies with a corn oil vehicle (vehicle corn oil) of dibromochloromethane in F344/N rats and B6C3F1 mice. The exposure durations evaluated in this study included a single exposure, 14-day exposure, 13-week subchronic exposure, and 2-year chronic exposure. The single and 14-day exposure durations were range finding studies that are not relevant to the derivation of subchronic and chronic noncancer toxicity values and cancer slope factors. U.S. EPA (1997) derived an oral RfD from the subchronic exposure study of rats (NTP, 1985). Rats and mice (10/sex/dose) were administered 0, 15, 30, 60, 125 or 250 mg/kg dibromochloromethane by gavage (5 days/week) for 13 weeks. In rats, the 250-mg/kg group (the high-dose group) exhibited high mortality in both sexes (9/10) prior to

study termination. Body weight (bw) was depressed by 47% for males and 25% for females. Fatty metamorphosis of the liver, characterized by intracytoplasmic clear vacuoles in hepatocytes, was observed in all of the high-dose males and females (Table 1). Control and lower dose male rats also had high incidences of this effect; however, a Fisher Exact test concluded that only liver lesions in the dose groups 60 mg/kg or higher were statistically significantly elevated (Table 1). The LOAEL was 60 mg/kg and the NOAEL was 30 mg/kg. This NOAEL was converted to a daily dose of 21.4 mg/kg-day (5/7 days of dosing), which was used by U.S. EPA to derive the chronic oral RfD. Fatty metamorphosis of the liver was observed in 1/10 female control rats, 0/10 in the 125-mg/kg dose group, and 9/9 in the 250 mg/kg-day dose group; the other female dose groups were not evaluated for this endpoint. Hepatocellular centrilobular necrosis was observed in high-dose male rats (8/10) and high-dose female rats (7/9) but not in the other lower dose groups examined. Toxic nephropathy was observed in high-dose male rats (8/10) and high-dose female rats (9/9) but not in the other lower-dose groups examined. Acute inflammation and squamous metaplasia of the salivary gland were observed only in the high-dose groups of both sexes.

Table 1. Incidences of Noncancer Effects in Male and Female Rats in the 13-Week Study

	Control	15 mg/kg	30 mg/kg	60 mg/kg	125 mg/kg	250 mg/kg
Male rats						
Liver						
Fatty metamorphosis	4/10	7/10	8/10	10/10	10/10	10/10
Centrilobular necrosis	0/10	1/10	0/10	0/10	0/10	8/10
Kidney						
Toxic nephropathy	0/10	NE	NE	NE	0/10	8/10
Salivary gland						
Inflammation	0/10	NE	NE	NE	0/10	5/10
Squamous metaplasia	0/10	NE	NE	NE	0/10	9/10
Female rats						
Liver						
Fatty metamorphosis	1/10	NE	NE	NE	0/10	9/9
Bile duct hyperplasia	1/10	NE	NE	NE	0/10	6/9
Centrilobular necrosis	0/10	NE	NE	NE	0/10	7/9
Kidney						
Toxic nephropathy	0/10	NE	NE	NE	0/10	9/9
Salivary gland						
Inflammation	0/10	NE	NE	NE	0/10	5/8
Squamous metaplasia	0/10	NE	NE	NE	0/10	6/8

NE, not evaluated.

In mice, fatty metamorphosis (necrosis and vacuolar change) was observed only in the 250-mg/kg dose group of male mice (5/10). Toxic nephropathy, characterized by tubular degeneration or mineralization of the kidney, was exhibited by 5/10 male mice in the high-dose group. The 125-mg/kg dose group did not exhibit these endpoints. The lower dose groups (15, 30 and 60 mg/kg) were not evaluated for liver or kidney effects. Female mice did not exhibit these lesions.

In the 2-year gavage studies (NTP, 1985), F344/N rats were administered 0, 40 mg/kg (low dose) or 80 mg/kg (high dose) dibromochloromethane (50/sex/dose) for 5 days/week. At study termination, mean body weights of males were decreased by 7.7% in the high-dose group. Mean bw of females were not different between treated and control groups. No statistically significant differences in survival were observed between groups; survival of males was 34/50 in controls, 38/50 in the low-dose group, and 43/50 in the high-dose group. Survival of females was 39/50 in controls, 37/50 in the low-dose group, and 41/50 in the high-dose group. Incidences of neoplastic lesions were not statistically significantly elevated at any site in either male or female rats. Statistically significant negative trends were observed for the incidences of thyroid gland follicular cell carcinomas in male rats, follicular cell adenomas or carcinomas (combined) in male rats, thyroid gland C-cell carcinomas in female rats, C-cell adenomas or carcinomas (combined) in female rats, mononuclear cell leukemia in male rats, mammary gland fibroadenomas in female rats, endometrial stromal polyps in female rats, and testis interstitial cell tumors in male rats. Negative trends in the thyroid gland, the hematopoietic system, and testis were not considered to be chemical-related since incidences in the dosed groups were not different from historical rates at this laboratory.

Noncancer effects in dibromochloromethane-treated rats include fatty metamorphosis in the liver, ground-glass cytoplasmic changes of the liver, and nephrosis (Table 2). In male rats, fatty metamorphosis incidences were 27/50 in controls, 47/50 in the low-dose group, and 49/50 in the high-dose group. Female rats had fatty metamorphosis incidences of 12/50 in controls, 23/50 in the low-dose group, and 50/50 in the high-dose group. The incidences of ground-glass cytoplasmic changes in male rats were 8/50 in controls, 22/50 in the low-dose group, and 34/50 in the high-dose group; female rats had incidences of 0/50 in controls, 1/50 in the low-dose group, and 12/50 in the high-dose group. Kidney nephrosis in male rats was comparable in control, low-dose and high-dose groups (42/50, 44/50 and 41/50, respectively). Female rats exhibited a dose-related increase in the incidence of nephrosis: 7/50 in controls, 11/50 in the low-dose group, and 14/50 in the high-dose group.

Table 2. Incidences of Noncancer Effects in Male and Female Rats in the Chronic Exposure Study

	Male rats			Female rats		
	Controls	**40 mg/kg (low dose)**	**80 mg/kg (high dose)**	**Controls**	**40 mg/kg (low dose)**	**80 mg/kg (high dose)**
Liver fatty metamorphosis	27/50	47/50	49/50	12/50	23/50	50/50
Liver group-glass cytoplasmic changes	8/50	22/50	34/50	0/50	1/50	12/50
Kidney nephrosis	42/50	44/50	41/50	7/50	11/50	14/50

In the chronic study, B6C3F1 mice were administered by gavage 0, 50 mg/kg (low dose) or 100 mg/kg (high dose) dibromochloromethane (50/sex/dose) for 2 years (5 days/week). Mean body weights of the high-dose mice of both sexes were lower than controls for most of the study. Mean bw of low-dose mice were lower than controls beginning in week 60. Survival of the low-dose and high-dose mice was significantly lower than the controls. An accidental overdose caused the death of 35 low-dose male mice during weeks 58–59, rendered the analysis inadequate for analysis. Female low-dose mice also received this overdose, however, no adverse effects were observed. Nine high-dose males died at week 82 due to unknown causes. Survival rates for the female mice at study termination were 32/50 in controls, 27/50 in the low-dose group, and 36/50 in the high-dose group.

Table 3 presents the effects observed in male mice, including those accidentally killed. The incidence of hepatocellular carcinomas was significantly increased in the high-dose male mice compared to the controls. Liver effects were observed in the low-dose male mice that died at weeks 58–59 including centrilobular necrosis and fatty metamorphosis. The incidence of malignant lymphomas in male mice was significantly lower in the high-dose mice compared to the control: 9/50 in control and 0/50 in high-dose male mice.

Table 3. Chronic Effects of Male Mice Administered Dibromochloromethane				
		50 mg/kg (low dose)		
	Control	Animals killed weeks 58–59	Animals surviving to terminal kill	100 mg/kg (high dose)
Liver				
Necrosis	2/50	28/35	1/15	9/50
Fatty metamorphosis	13/50	32/35	0/15	20/50
Hepatocytomegaly	0/50	0/35	0/15	12/50
Adenoma	14/50	3/35	2/15	10/50
Carcinoma	10/50	2/35	7/15	19/50
Adenoma/carcinoma	23/50	5/35	9/15	27/50
Kidney				
Nephrosis	0/50	35/35	10/15	37/50
Tubular calcification	0/50	12/35	1/15	0/50

Female mice exhibited significant positive trends for hepatocellular adenomas, hepatocellular adenomas, and carcinomas combined (Table 4). Incidences of follicular cell hyperplasia of the thyroid also occurred with a dose-response relationship. U.S. EPA (1992) derived an oral cancer slope factor (OSF) from the incidences of hepatocellular adenoma or carcinoma (combined) in female mice.

Table 4. Chronic Effects in Female Mice Administered Dibromochloromethane			
	Control	50 mg/kg (low dose)	100 mg/kg (high dose)
Liver			
Adenoma	2/50	4/49	11/50
Carcinoma	4/50	6/49	8/50
Adenoma/carcinoma	6/50	10/49	19/50
Thyroid			
Follicular cell hyperplasia	1/49	13/46	31/50

NTP (1985) concluded that "Under the conditions of these gavage studies, there was no evidence of carcinogenicity in male or female F344/N rats receiving dibromochloromethane at doses of 40 or 80 mg/kg five times per week for 104 weeks. Fatty metamorphosis and ground-glass cytoplasmic changes of the liver in male and female F344/N rats were related to administration of dibromochloromethane. There was equivocal evidence of carcinogenicity for male B6C3F1 mice; dibromochloromethane caused an increased incidence of hepatocellular carcinomas, whereas the combined incidence of hepatocellular adenomas or carcinomas was

only marginally increased. Some evidence of carcinogenicity was observed for female B6C3F1 mice, since dibromochloromethane caused an increased incidence of hepatocellular adenomas and an increased combined incidence of hepatocellular adenomas or carcinomas."

Daniel et al. (1990) investigated the effects of dibromochloromethane in male and female Sprague-Dawley rats (10/sex/dose) exposed by daily oral gavage for 90 days, at the following doses: 0, 50, 100 and 200 mg/kg. Survival rates were unaffected in both sexes. Body weight was significantly reduced in males (31.5%) and females (13.3%) exposed to the highest dose, 200 mg/kg, compared to control groups (corn oil). Food intake was also significantly depressed in males exposed to the 200-mg/kg dose. In males, the following organ weights were significantly decreased at 200-mg/kg dose relative to controls (approximate percentages): brain (5.9%), heart (25%), liver (16.5%), and thymus (34%). At 200 mg/kg, the following organ weight/bw ratios were significantly increased: brain, spleen, and testes. At 50 and 100 mg/kg, kidney and lung weight/bw were also significantly increased. Liver lesions, including vacuolization, necrosis and inflammation exhibited a dose-dependent increase in severity. At the 200-mg/kg dose, the kidneys exhibited significant degeneration of proximal tubular cells, granularization and microvacuolization.

No significant hematological alterations in males were noted. Biochemistry profiles in males were significantly altered as follows (approximate percentages): (1) creatinine was increased by 17% at 100 mg/kg and by 33.3% at 200 mg/kg, (2) alkaline phosphatase was increased by 72.8% at 200 mg/kg, and (3) alanine transferase was increased by 37% at 100 mg/kg and 157% at 200 mg/kg. Elevated alanine transferase and alkaline phosphatase activities are indicative of hepatotoxicity. Increased serum creatinine and decreased potassium are suggestive of nephrotoxicity.

In females, the following organ weights showed significant differences relative to control organ weights (approximate percentages): (1) brain (-5.3% at 100 mg/kg and 200 mg/kg), (2) kidney (+16.0% at 100 mg/kg and +24.7% at 200 mg/kg), (3) liver (+31.1% at 200 mg/kg), and (4) thymus (-23.7% at 100 mg/kg and -39.5% at 200 mg/kg). Organ weight/bw ratios were significantly increased in females, too, at 200 mg/kg for the brain and spleen, but decreased for the thymus. Kidney and liver weights/bw ratios were also increased at 50 mg/kg and 100 mg/kg. Liver and kidney lesions were significant only at the highest dose, exhibiting characterizations noted in male liver and kidney lesions (above). Significant biochemical changes include the following: (1) creatinine (+17.0% at 100 mg/kg), (2) alkaline phosphatase (+96.6% at 200 mg/kg), (3) calcium (+5.2% at 100 mg/kg), (4) total protein (+7.5% at 100 mg/kg) and (5) albumin (+10.2% at 50 mg/kg and 100 mg/kg).

The hepatotoxicity and nephrotoxicity indicated by the serum biochemistry profiles are supported by the histopathological evaluation of liver and kidney. Table 5 presents the incidences of noncancer liver and kidney effects. Male rats exhibited liver centrilobular lipidosis in all dose groups. Liver centrilobular necrosis and kidney cortex tubular degeneration were observed only in the 100- or 200-mg/kg-day dose groups of males. Female rats exhibited liver centrilobular lipidosis and necrosis only in the 100- and 200-mg/kg-day groups. Based on the observation of liver centrilobular lipidosis in all dose groups of male rats, a LOAEL of 50 mg/kg-day is provided.

Table 5. Incidences of Noncancer Liver and Kidney Effects in Rats Treated for 90 Days

	0 mg/kg-day	50 mg/kg-day	100 mg/kg-day	200 mg/kg-day
Males				
Liver				
Chronic focal inflammation	10/10	9/10	7/10	5/10
Centrilobular lipidosis	0/10	9/10	9/10	10/10
Centrilobular necrosis	0/10	0/10	1/10	9/10
Chronic centrilobular necrosis	0/10	0/10	0/10	4/10
Kidney				
Tubular degeneration, cortex	0/10	0/10	4/10	10/10
Tubular mineralization, cortex	4/10	2/10	3/10	0/10
Tubular mineralization, medulla	4/10	0/10	2/10	4/10
Progressive nephropathy, chronic	3/10	4/10	1/10	0/10
Females				
Liver				
Chronic focal inflammation	9/10	7/10	5/10	5/10
Centrilobular lipidosis	0/10	0/10	1/10	9/10
Centrilobular necrosis	0/10	0/10	1/10	9/10
Chronic centrilobular necrosis	0/10	0/10	0/10	2/10
Kidney				
Tubular degeneration, cortex	1/10	5/10	9/10	10/10
Tubular mineralization, cortex	3/10	4/10	3/10	0/10
Tubular mineralization, medulla	3/10	1/10	3/10	3/10
Progressive nephropathy, chronic	1/10	0/10	0/10	0/10

Chu et al. (1982) exposed male and female Sprague Dawley rats to dibromochloromethane in drinking water (20/sex/group) at 0, 5, 500 or 2500 ppm for 90 days. Two control groups were established for this study: (1) tap water only and (2) 1% emulphor in tap water. At completion, 10 rats/sex/group were sacrificed and liver and kidney histologies were assessed. The remaining 10 rats/sex/group were retained on drinking water only, for an additional 90 days, to evaluate the potential recovery of toxicities induced by the initial exposures. Table 6 shows the intake estimates of dibromochloromethane based on measurements of daily water intake and known levels of dibromochloromethane in the drinking water. Survival rates of males were significantly reduced at 2500 ppm (data not provided) while female rats were not affected. There was no significant change in body weight relative to the control groups. Food intake was significantly depressed at 2500 ppm/day among male rats during both the initial and recovery 90-day periods; females showed a significant reduction in food intake at exposure of 2500 ppm/day and only during the recovery phase. There was a

general dose-dependent increase in the severity of liver lesions in both males and females, which were negligible at the end of the recovery phase. Males showed a significant increase in lymphocyte count (4.1–4.5%) relative to both controls at 500 ppm/daily during initial 90-day exposure period. At the end of the 90-day recovery period, lymphocyte counts returned to levels comparable to controls.

Table 6. Approximate Daily Intake of Dibromochloromethane

	Daily intake in mg/rat/day				
Dose	**0 ppm**	**5 ppm**	**50 ppm**	**500 ppm**	**2500 ppm**
Male	-	0.14 mg/rat/day	1.6 mg/rat/day	13 mg/rat/day	49 mg/rat/day
Female	-	0.13 mg/rat/day	1.2 mg/rat/day	9.8 mg/rat/day	32 mg/rat/day

Aida et al. (1992) evaluated the toxicity of microencapsulated dibromochloromethane administered to Wistar rats in the diet. Groups of 7 male rats/dose were provided a diet containing 0.020%, 0.062% and 0.185% dibromochloromethane; groups of 7 female rats/dose were provided a diet containing 0.038%, 0.113% and 0.338% dibromochloromethane. Microencapsulation of dibromochloroethane was selected as the vehicle of exposure because (1) this chemical is sparingly soluble in drinking water and (2) the authors desired to evaluate its oral toxicity. Hepatic lesions, such as vacuolization and liver cell swelling, were observed in both sexes of the middle- and high-dose groups. Single-cell necrosis was observed in both sexes. Relative and absolute liver weights were statistically significantly increased in the high-dose male rats and in the mid- and high-dose female rats. Female rats also exhibited significantly decreased body weight in the high-dose group. The NOAEL in this study was determined as 18.3 mg/kg dibromochloromethane based on the absence of effects in the low-dose (0.020%) group of male rats. This study demonstrates that the liver is the target organ in rats.

Munson et al. (1982) studied the subchronic toxicity of selected halomethanes that are drinking water contaminants—including dibromochloromethane at doses of 50 and 250 mg/kg-day for 14 days in CD-1 male and female mice that were performed using doses of 1/10th the LD50 for the compounds. Observations include a reduction in body weight in males at the high dose. Liver weights were increased at 125 and 250 mg/kg-day when expressed as percent of body weight. Spleen and thymus values decreased significantly at the high dose. Only fibrinogen was affected with a decrease occurring at the high-dose only. Clinical chemistry alterations were increases in SGPT and SGOT and a decrease in serum glucose—all in the high dose group. Both humeral and cell-mediated immunity were affected. The number of AFC was significantly reduced when expressed as total cells or as AFC/106 spleen cells. This was noted at the high dose, as was a reduction in hemagglutination titer. Cell-mediated immunity, as measured by the popliteal lymph node stimulation index, was depressed at the high dose. It should be noted that even though the changes were significant only at the 250-mg/kg-day level, the decreasing trend can be observed with the lower doses. No significant body weight change was observed in females. The major organ weight change in females was an increase in liver size in the intermediate- and high-dose groups. The ability of the liver to metabolize hexobarbital was impaired at the intermediate and high dose as evidenced by increased hexobarbital sleeping times. The only hematological change was a slight decrease in fibrinogen at the high dose. Altered clinical chemistry parameters were increases in SGPT and SGOT and a

decrease in serum glucose, all occurring at 250 mg/kg-day. A decrease in AFC/spleen and AFC/106 spleen cells was noted in the intermediate- and high-dose groups, whereas a reduction of hemagglutination titer occurred only at the high dose. With regard to exposure to trichloromethane (assumed to have similar mode of action), the authors observed that a 90-day exposure did not exacerbate the changes seen to occur as a result of 14-day administration, and were, in fact, less severe. The observation was supported by a tolerance experiment that revealed compensatory mechanisms being activated during the subchronic 90-day exposure.

Since these doses were considerably higher than the NTP (1985) study, they are not considered further for use in risk assessment or in the derivation of provisional toxicity values.

Da Silva et al. (2000) evaluated the blood kinetics of simultaneous dosing of binary mixtures of trihalomethanes (chloroform, bromoform, bromodichloromethane and dibromochloromethane at 0.25 nmol/kg) by gavage to male Sprague-Dawley rats. The authors concluded that at the dose level investigated, every binary combination, when orally administered, resulted in a significant modulation of their pharmacokinetics. The authors suggested that this is probably the consequence of a mutual metabolic inhibition between the trihalmethanes. Because these compounds may occur together in the environment, some consideration should be given to the interactive effects on toxicity.

Potential short term exposure effects of dibromochloromethane on mutagenicity, nephrotoxicity and serum testosterone levels were investigated by Potter et al. (1996). Male F344/N rats (4 rats/dose) were administered 0.75 mmol/kg or 1.5 mmol/kg of dibromochloromethane (purity not indicated, in 4% emulphor) by oral gavage for 7 consecutive days. Rats were sacrificed at 1, 3 and 7 days. Rats were infused with 5'-bromodeoxyuridine 3 days prior to sacrifice—except those sacrificed on days 1 and 3. The kidney/body weight ratio (day 7) was significantly increased (17.2%) in rats exposed to 1.5 mmol/kg bw dibromochloromethane. Hyaline droplets in the kidney were significantly reduced at both doses and on days 3 and 7 of sacrifice. DNA strand break rates were not affected by dibromochloromethane exposures. Based on wide variability, cell proliferation based on [^{3}H] incorporation in renal cells did not show statistically significant differences between exposed and control groups. There is a general dose-dependent decrease in mean testosterone levels, which is significant at the high dose of 1.5 mmol/kg (approximately 2.5-fold). Decreased serum testosterone was hypothesized to account for the decreased hyaline droplet formation.

Coffin et al. (2000) administered dibromochloromethane to female B6C3F1 mice (6–10/dose) by oral gavage, for 11 days (2 days off between days 5–8) at 100 and 300 mg/kg (doses of 0.48 and 1.44 mmol/kg, respectively), or for 11 consecutive days at 790 mg/L in drinking water (0.82 mmol/kg dose). There was a dose-dependent increase in liver/body weight ratios in mice that were administered dibromochloromethane by oral gavage but not in drinking water. Liver toxicity was based on a grading system of 1–4 by degree of severity/characterization. The 100-mg/kg gavage dose caused mid-lobular ballooning of hepatocytes (Grade 1); the 300 mg/kg gavage dose caused mid-lobular ballooning hepatocytes extending to the central vein (Grade 2) Liver toxicity of dibromochloromethane administered in drinking water was similar to that observed with the 10-mg/kg gavage dose. Proliferating cell nuclear antigen-labeling index (PCNA-LI) was increased in a dose-response manner when mice were treated by gavage. When administered in drinking water, PCNA-LI was increased similarly to the 100-mg/kg gavage dose. There was also a dose-dependent decrease in DNA

methylation in the promoter region of the *c-myc* gene when mice were treated by gavage; dibromochloromethane treatment by drinking water produced similar results on DNA methylation as the gavage route of exposure. The authors concluded that (1) dibromochloromethane, administered by gavage, enhanced cell proliferation, and decreased methylation of the *c-myc* gene and that (2) this conclusion was consistent with its carcinogenic activity. The authors also speculate that the more modest effects caused by drinking water administration is evidence that the carcinogenic activity of dibromochloromethane is dependent on the rate of delivery (i.e. rapid delivery by oral gavage being more efficacious than slower delivery via drinking water).

A two-generational murine reproduction on the effects of dibromochloromethane commissioned by U.S. EPA was conducted by Borzelleca and Carchman (1982). Doses of 0.1, 1.0 and 4.0 mg/mL (approximately 685 mg/kg) dibromochloromethane in drinking water were administered to ICR Swiss Albino mice beginning at 7 weeks of age and continued for two generations. There were 9–10 males/dose in the F_0 generation, 30 females/dose in the F_0 generation, and 4 males in the 4 mg/mL dose in the F_{1b} generation. Body weights were significantly reduced in both the F_0 and F_1 generations of males and females exposed to 4.0 mg/mL dose. Survival was significantly reduced among females in the F_{1b} generation (48.3%). In both generations, all treated animals showed a dose-dependent increase in liver size and severity of hepatotoxic morphology (characterizations not indicated). Significant decreases were observed in litter size in all generations at the highest dose of dibromochloromethane. Fertility was reduced in the F_{1c} and F_{2a} generations at the 4.0 mg/mL dose of dibromochloromethane. The gestational index was reduced at the 4.0 mg/mL dose in the F_{1a}, F_{1b} and F_{1c} generations.

Hewitt et al. (1983) investigated the effects of acetone potentiation of hepatotoxicity and nephrotoxicity by dibromochloroethane using adult male Sprague-Dawley rats. Rats (6/dose) were administered by oral gavage the following doses: 0, 0.10, 0.20, 0.25, 0.50, 0.75, 1.00, 2.00 or 2.50 mL/kg (concentration not provided). An LD_{50} post 24 hours of exposure was determined to be 1.08 mL/kg. Liver injury was observed at 1.0 mL/kg dose based on significantly elevated levels of glutamic-pyruvic transaminase (GPT) activity (5.0-fold) and ornithine carbamoyl transferase (OCT) activity (17.8-fold). No nephrotoxicity was observed. In a second series of experiments, rats were pretreated orally with 15 mmol/kg acetone (<99% purity, diluted in H20). After 18 hours of pretreatment, the rats were given 0.25, 0.50 or 1.00 mL/kg dibromochloromethane (in corn oil) or corn oil (10 mL/kg). Acetone-pretreatment induced significant potentiation of dibromochloromethane effects at 0.50 mL/kg and 1.00 mL/kg doses on (1) GPT activity (5-fold and 7-fold higher, respectively) and (2) OCT activity (14-fold higher at 0.50 mL/kg). Pretreatment with acetone significantly increased (1) liver/body weight ratio at 0.50 and 1.00 mL/kg doses (29.3% and 19.3%, respectively), (2) GPT activity at 0.25, 0.50 and 1.00 mL/kg doses (33.4-fold, 11.6-fold and 33.6-fold increases, respectively), and (3) OCT activity at 0.25 and 1.00 mL/kg doses (59.0-fold and 30.7-fold increases, respectively). No potentiation of nephrotoxicity was observed.

Other Studies

Dibromochloromethane is positive in mutagenicity and genotoxicity studies. Although NTP (1985) reported that dibromochloromethane was negative in Ames mutagenicity assays, another report (Simmon et al., 1977) using a dessicator method to address the volatility of dibromochloromethane described positive results. Some assays of sister chromatid exchange and

chromosome aberration assays are positive while others report negative results. Mouse lymphoma forward mutation assays are positive in two studies. However, unscheduled DNA synthesis in rat liver using thymidine incorporation is negative. Because IRIS already provides a detailed discussion on these, no further information is presented here.

Mink et al. (1986) performed toxicokinetic metabolic studies of dibromochloromethane intake using male Sprague-Dawley rats (n = 6) and B6C3F$_1$ mice (n = 20). ^{14}C-dibromochloromethane was given by oral gavage at doses of 100 mg/kg or 150 mg/kg to rats and mice, respectively. The rats and mice were kept in glass chambers connected to a series of chamber traps containing xylene/2-methoyethanol and ethanolamine/2-methoxyethanol to collect expired ^{14}C-dibromochloromethane or ^{14}C-carbon monoxide (CO). Expired air, urine samples and feces were collected over intervals during an 8-hour period. In a second experiment, urine samples were taken at intervals throughout a period of 36 hours for mice and 48 hours for rats. At 8 hours**,** the total recovery of ^{14}C was 70.3% and 91.63% for rats and mice, respectively, based on detection of ^{14}C radiolabel in expired CO_2, parent dibromochloromethane, urine, and in all organs. The majority of dichloromethane was eliminated through expiration. In rats, 18.2% was expired as ^{14}C-CO, ~48.1% was expired as unmetabolized dibromochloromethane, 1.1% was expelled in urine (^{14}C), and 1.4% was found retained in organs (^{14}C). In mice, 71.58% was expired as ^{14}C-CO, 12.31% was expired as the parent compound, 1.90% was expelled in urine (^{14}C), and 5.02% was found retained in organs (^{14}C). The half-life of dibromochloromethane is 1.2 hours and 2.5 hours in rats and mice, respectively. The urine contained <10% of total radioactivity at 36 or 48 hours postexposure.

The data suggest that clearance of dichloromethane is predominantly through expired air in both species. Species comparisons demonstrate that mice clear dichloromethane in expired air as CO ~3.8-fold more and as parent compound 4-fold less, as compared to rats. Mice retain nearly 5-fold more dichloromethane in organs, while both species excrete (^{14}C detection) the putative parent compound at similar levels. Mink et al (1986) authors suggest that mice clear dibromochloromethane faster than rats. The high recovery of unmetabolized parent compound suggests limited metabolism of dibromochloromethane.

Pankow et al. (1997) investigated the metabolism of dibromochloromethane to its oxidized metabolites, bromide and carbon monoxide in male Wistar Unilever rats. Dibromochloromethane was given either acutely (30/dose) or for 7 days (3/dose) by oral gavage at the following doses: 0.4, 0.8, 1.6 and 3.1 mmol/kg. Physiological saline was given to control groups. Table 7 provides detection of dibromochloromethane in the blood postacute exposures. No significant differences in the dibromochloromethane levels were seen between 1.6 and 3.1 mmol/kg bw doses after 1.5 hours. There was a significant dose-dependent increase in bromide detection following acute exposure to dibromochloromethane (Table 8). Accumulation of bromide levels in the blood post-7-day exposure was significantly greater than in acute exposures, with a peak ~5-fold difference at 12 hours (blood collection: 0, 3, 6, 12, 24, 48 and 72 hours). Carbon monoxide retention was also significantly higher between 0–3 hours for repeated exposure of dibromochloromethane compared to acute exposures.

Table 7. Detection of Dibromochloromethane in Blood after Various Acute Dose Exposures to Dibromochloromethane

Time postexposure (hours)	Dibromochloromethane (mmol/L) in blood			
	0.4 mmol/kg	0.8 mmol/kg	1.6 mmol/kg	3.1 mmol/kg
1.5	-	0.16 ± 0.12	0.65 ± 0.40	0.43 ± 0.09
3	03 ± 0.1	0.16 ± 0.04	0.49 ± 0.27	0.65 ± 0.06
6	Not detected	0.06 ± 0.04	0.43 ± 0.27	-
9	Not detected	Not detected	0.29 ± 0.18	-

Table 8. Detection of Bromide Levels 24 hours Post Acute Exposure to Dibromochloromethane

	Dibromochloromethane (mmol/L) in blood				
	0.0 mmol/kg	0.4 mmol/kg	0.8 mmol/kg	1.6 mmol/kg	3.1 mmol/kg
Bromide (mmol/L)	0.0	2.04 ± 0.16	3.57 ± 0.43	5.14 ± 1.22	~7.0 ± 0.9

GSH activity in the liver of rats acutely exposed to 0.8 mmol/kg of dibromochloromethane was decreased by buthionine sulphoximine pretreatment (i.p. administration of 4 mmol/kg) which depletes GSH. GSH levels were decreased by 30% by buthionine sulphoximine, but formation of carbon monoxide and bromide were not significantly affected. Pretreatment with butylated hydroxyanisole (5 x 5.5 mmol/kg, administered daily/5 days by oral gavage) resulted in increased GSH levels (31%) and significantly higher rates of carbon monoxide and bromide formation (approximately 1–2-fold). Enhancement of cytochrome P450 enzymes, as measured by levels of liver *p*-nitrophenyl hydroxylase activity was significantly increased in rats pre-treated by i.p. for 4 days with phenobarbitol, isoniazid (360 µmol/kg), or *m*-xylene, followed by an acute treatment with 0.8 mmol/kg dibromochloromethane. Blood levels of carbon monoxide and bromide were significantly increased initially, over a 24-hour period. Conversely, carbon monoxide and bromide levels were significantly reduced throughout the 24 hour assessment period, in rats dually treated with the cytochrome P450 inhibitor, diethyldithiocarbamate (i.p. daily/4 days/64 µmol/kg) and 0.8 mmol/kg dibromochloromethane.

Pankow et al (1997) concluded that oxidation of dibromochloromethane to bromide and carbon monoxide is mediated primarily by cytochrome P450 enzymes (CYP2E1, CYP2B1 and CYP2B2). Repeated exposures to dibromochloromethane result in an accumulation of bromide.

DERIVATION OF A PROVISIONAL SUBCHRONIC RfD FOR DIBROMOCHLOROMETHANE

IRIS (U.S. EPA, 1991) derived a chronic oral RfD for dibromochloromethane based on hepatic lesions observed in male rats in a 13-week NTP (1985) study.

The subchronic study (NTP, 1985) was utilized by IRIS to develop a chronic oral RfD of 2×10^{-2} mg/kg-day. IRIS considered the 60-mg/kg-day dose as a LOAEL (10/10 responding vs. a control of 4/10 [$p = .005$, Fisher Exact Test]) and selected the next lowest level (30 mg/kg-day) as a NOAEL (8/10 responding). IRIS corrected the time duration of gavage (5 days/7days) to get a POD of 21.4 mg/kg-day. The application of a composite UF of 1000 (10 intrahuman, 10 interhuman, 10 extrapolation to chronic period) obtains an RfD of 2×10^{-2} mg/kg-day.

The NOAEL of 30 mg/kg-day is selected as the POD for the derivation of the subchronic p-RfD. This value was duration adjusted for continuous exposure resulting in a POD_{ADJ} of 21.4 mg/kg-day. Applying a composite uncertainty factor of 300 (10 for intraspecies, 10 for interspecies, 3 for database correction since a developmental study is not available) is calculated below:

Subchronic p-RfD = POD_{ADJ}/UF
= 21.4/300 = 0.071 mg/kg-day (rounded to)
= **7×10^{-2} mg/kg-day**

The NTP (1985) subchronic bioassays utilized standards for numbers of animals of both sexes and two species; multiple endpoints, including complete histopathology. Confidence in the chosen study is medium. The database is given medium confidence. Medium confidence in the p-RfD follows.

FEASIBILITY OF PROVISIONAL SUBCHRONIC AND CHRONIC RfCs FOR DIBROMOCHLOROMETHANE

Subchronic and chronic p-RfCs cannot be derived for dibromochloromethane because no toxicology information from the inhalation route of exposure is available. A route-to-route extrapolation could not be performed because of the lack of information on the absorption, metabolism and distribution of pentachloroethane following inhalation exposure.

FEASIBILITY CARCINOGENICITY ASSESSMENT FOR DIBROMOCHLOROMETHANE

IRIS (U.S. EPA, 1992) classified dibromochloromethane as a possible human carcinogen (group C). U.S. EPA derived an oral cancer slope factor for dibromochloromethane from the NTP (1985) cancer bioassay of female mice. Female mice exhibited dose-related increases in hepatocellular adenoma and carcinoma (combined). Using a linearized multi-stage procedure of extra risk, an OSF of 8.4×10^{-2} per mg/kg-day was derived. No IUR is developed because of lack of useful inhalation data.

REFERENCES

Aida, Y, K. Takada, O. Uchida et al. 1992. Toxicities of microencapsulated tribromomethane, dibromochloromethane and bromodichloromethane administered in the diet to Wistar rats for one month. J. Toxicol. Sci. 17(3):119–133.

ATSDR (Agency for Toxic Substances and Disease Registry). 2005. Toxicological profile for bromoform and dibromochloromethane; Public Health Service, U.S. Department of Health and Human Services, Atlanta, GA. Online. http://www.atsdr.cdc.gov/toxprofiles.

Borzelleca, J.F. and R.A. Carchman. 1982. Effects of selected organic drinking water contaminants on male reproduction. Office of Research and Development, U.S. Environmental Protection Agency, Research Triangle Park, NC; EPA600/1-82-009. Available from: National Technical Information Service, Springfield, VA; PB82-259847.

Chu, I., D.C. Villeneuve, V.E. Secours et al. 1982. Trihalomethanes: 2. Reversibility of toxicological changes produced by chloroform, bromodichloromethane, chlorodibromomethane and bromoform in rats. J. Environ. Sci. Health Part B Pestic. Food Contam. Agric. Wastes. 17(3):225-240.

Coffin, J.C., R. Ge, S. Yang et al. 2000. Effect of trihalomethanes on cell proliferation and DNA methylation in female B6C3F1 mouse liver. Toxicol. Sci. 58(2):243-252.

Daniel, F.B., M. Robinson, L.W. Condie et al. 1990. Ninety-day oral toxicity study of dibromochloromethane in Sprague-Dawley rats. Drug Chem. Toxicol. 13(2-3):135-154.

Da Silva, M.L., et al. 2000. Evaluation of the Pharmocokinetic Interactions between Orally Administered Trihalomethanes in the Rat. J. Toxicol. Environ. Health. Part A, 60:343-353.

Hewitt, W.R., E.M. Brown and G.L. Plaa. 1983. Acetone-induced potentiation of trihalomethane toxicity in male rats. Toxicol. Lett. (Amst) 16(3-4):285-296.

IARC (International Agency for Research on Cancer). 1991. Chlorinated drinking-water; chlorination by-products; some other halogenated compounds; cobalt and cobalt compounds. IARC monographs on the evaluation of carcinogenic risks to humans. Vol. 52. Lyon, France: International Agency for Research on Cancer, p. 243-268.

IARC (International Agency for Research on Cancer). 1999. Re-evaluation of some organic chemicals, hydrazine and hydrogen peroxide. IARC monographs on the evaluation of carcinogenic risks to humans. Vol. 71. Lyon, France: International Agency for Research on Cancer, p. 1331-1338.

Mink, F.L., T.J. Brown and J. Rickabaugh. 1986. Absorption distribution and excretion of carbon-14 trihalomethanes in mice and rats. Bull. Environ. Contam. Toxicol. 37(5):752-758.

Munson, Alber E. et al. 1982. Toxicology of Organic Drinking Water Contaminants: Trichloromethane, Bromodichloromethane, Dibromochloromethand and Tribromomethane. Environ. Health Perspect. Vol. 46, p. 117-126.

NTP (National Toxicology Program). 1985. Toxicology and carcinogenesis studies of chlorodibromomethane in F344/N rats and B6C3F1 mice (gavage studies). National Institute of Environmental Health Sciences, Public Health Service, U.S. Department of Health and Human Services, Research Triangle Park, NC; NTP TR 282. Online. http://ntp-server.niehs.nih.gov.

Pankow, D., B. Damme, U. Wunscher et al. 1997. Chlorodibromomethane metabolism to bromide and carbon monoxide in rats. Arch. Toxicol. 71(4):203-210.

Potter, C.L., L.W. Chang, A.B. DeAngelo et al. 1996. Effects of four trihalomethanes on DNA strand breaks, renal hyaline droplet formation and serum testosterone in male F-344 rats. Cancer Lett. 106(2):235-242.

Simmon, V.F., K. Kauhanen and R.G. Tardiff. 1977. Mutagenic activity of chemicals identified in drinking water. Dev. Toxicol. Environ. Sci. 2:249-258.

U.S. EPA. 1991. Integrated Risk Information System (IRIS). National Center for Environmental Assessment, Office of Research and Development, Washington, DC. Online. http://www.epa.gov/iris. Accessed June 2009.

U.S. EPA. 1992. Integrated Risk Information System (IRIS). National Center for Environmental Assessment, Office of Research and Development, Washington, DC. Online. http://www.epa.gov/iris. Accessed June 2009.

U.S. EPA. 1997. Health Effects Assessment Summary Tables. FY-1997 Update. Prepared by the Office of Research and Development, National Center for Environmental Assessment, Cincinnati OH for the Office of Emergency and Remedial Response, Washington, DC. July. EPA/540/R-97/036. NTIS PB97-921199.

U.S. EPA. 2009. Integrated Risk Information System (IRIS). National Center for Environmental Assessment, Office of Research and Development, Washington, DC. Online. http://www.epa.gov/iris.