9-22-2006

Provisional Peer Reviewed Toxicity Values for

Bis(2-chloroethoxy)methane
(CASRN 111-91-1)

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

Acronyms and Abbreviations

bw	body weight
cc	cubic centimeters
CD	Caesarean Delivered
CERCLA	Comprehensive Environmental Response, Compensation and Liability Act of 1980
CNS	central nervous system
cu.m	cubic meter
DWEL	Drinking Water Equivalent Level
FEL	frank-effect level
FIFRA	Federal Insecticide, Fungicide, and Rodenticide Act
g	grams
GI	gastrointestinal
HEC	human equivalent concentration
Hgb	hemoglobin
i.m.	intramuscular
i.p.	intraperitoneal
IRIS	Integrated Risk Information System
IUR	inhalation unit risk
i.v.	intravenous
kg	kilogram
L	liter
LEL	lowest-effect level
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
m	meter
MCL	maximum contaminant level
MCLG	maximum contaminant level goal
MF	modifying factor
mg	milligram
mg/kg	milligrams per kilogram
mg/L	milligrams per liter
MRL	minimal risk level
MTD	maximum tolerated dose
MTL	median threshold limit
NAAQS	National Ambient Air Quality Standards
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
PBPK	physiologically based pharmacokinetic
ppb	parts per billion
ppm	parts per million
PPRTV	Provisional Peer Reviewed Toxicity Value
RBC	red blood cell(s)
RCRA	Resource Conservation and Recovery Act
RDDR	Regional deposited dose ratio (for the indicated lung region)
REL	relative exposure level
RfC	inhalation reference concentration
RfD	oral reference dose
RGDR	Regional gas dose ratio (for the indicated lung region)
s.c.	subcutaneous
SCE	sister chromatid exchange
SDWA	Safe Drinking Water Act
sq.cm.	square centimeters
TSCA	Toxic Substances Control Act
UF	uncertainty factor
µg	microgram
µmol	micromoles
VOC	volatile organic compound

PROVISIONAL TOXICITY VALUES FOR BIS(2-CHLOROETHOXY)METHANE (CASRN 111-91-1)

Background

On December 5, 2003, the U.S. Environmental Protection Agency's (EPA's) 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. EPA's Integrated Risk Information System (IRIS).

2. Provisional Peer-Reviewed Toxicity Values (PPRTV) used in 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 EPA's Integrated Risk Information System (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 EPA IRIS Program. All provisional toxicity values receive internal review by two EPA scientists and external peer review by three independently selected scientific experts. PPRTVs differ from IRIS values in that PPRTVs do not receive the multi-program consensus review provided for IRIS values. This is because IRIS values are generally intended to be used in all EPA programs, while PPRTVs are developed specifically for the Superfund Program.

Because science and available information evolve, PPRTVs are initially derived with a three-year life-cycle. However, EPA Regions or the EPA Headquarters Superfund Program sometimes request that a frequently used PPRTV be reassessed. 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 manuscripts 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 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 manuscript and understand the strengths and limitations of the derived provisional values. PPRTVs are developed by the EPA Office of Research and Development's National Center for Environmental Assessment, Superfund Health Risk Technical Support Center for OSRTI. Other 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 EPA Office of Research and Development's National Center for Environmental Assessment, Superfund Health Risk Technical Support Center (513-569-7300), or OSRTI.

INTRODUCTION

Neither subchronic nor chronic RfDs or RfCs for bis(2-chloroethoxy)methane (BCM) are available on IRIS (U.S. EPA, 2006a), the HEAST (U.S. EPA, 1997) or the Drinking Water Standards and Health Advisories list (U.S. EPA, 2004). A carcinogenicity assessment for bis(2-chloroethoxy)methane is available on IRIS (U.S. EPA, 2006a) that includes a classification of Group D, not classifiable as to human carcinogenicity, based on no human or animal data. The CARA list (U.S. EPA, 1991a, 1994) includes no documents for this chemical. The toxicity of bis(2-chloroethoxy)methane has not been reviewed by ATSDR (2006), IARC (2006), or WHO (2006). ACGIH (2006), NIOSH (2006) and OSHA (2006) have not established occupational exposure limits for this compound. The NTP (2006) Management Status Report provided no relevant information. A technical report on haloethers prepared for EPA in 1975 (Durkin et al., 1975) and the Ambient Water Quality Criteria Document for Chloroalkyl Ethers (U.S. EPA, 1980) were reviewed for pertinent information. Literature searches were conducted from 1965 to September 2002 in TOXLINE, CANCERLIT, MEDLINE, GENETOX, HSDB, EMIC/EMICBACK, DART/ETICBACK, RTECS and TSCATS for relevant studies. During April 2004, these databases were again searched for relevant studies; none were identified that would change the conclusions of the risk estimate.

REVIEW OF PERTINENT DATA

Bis (2-chloroethoxy)methane is a synthetic organic chemical used as a solvent and as a reactant in the manufacture of polysulfide elastomers. More than 95% of polysulfide elastomers are made from bis (2-chloroethoxy) methane starting material and sodium polysulfide. The resulting products are used as heat- and solvent resistant sealants. The U.S. Food and Drug Administration has approved bis(2-chloroethoxy)methane for use in the manufacture of resins approved for direct contact with food packaging materials. Bis (2-chloroethoxy)methane is on the U.S. EPA's 1990 High Production Volume chemical list (U.S. EPA, 2006b); in 1977, 10 to 50 million pounds of bis(2-chloroethoxy)methane were produced in the U.S. (HSDB, 2005).

Human Studies

Oral Exposure. No reports were located regarding the subchronic or chronic toxicity or carcinogenicity of bis(2-chloroethoxy)methane in humans by oral exposure.

Inhalation Exposure. No reports were located regarding the subchronic or chronic toxicity or carcinogenicity of bis(2-chloroethoxy)methane in humans by inhalation exposure.

Animal Studies

Oral Exposure. Non-fasted Sprague-Dawley rats (10/sex/dose group) were treated with oral doses of 0, 10, 20, 40, 80, or 120 mg/kg-day of bis(2-chloroethoxy)methane by daily gavage in corn oil for 90 days (Bio/Dynamics, 1990a). Physical observations, body weight and food consumption measurements were recorded weekly. Hematology and clinical chemistry evaluations were performed after one month of treatment and at termination. Ophthalmoscopic examinations took place at termination. Complete gross post-mortem examination was conducted on all animals. The control and high-dose groups received comprehensive histopathological examinations, while only the kidneys, liver, lungs and gross lesions were examined in the intermediate dose groups.

Of the 20 rats that received the highest dose (120 mg/kg-day), all ten males and seven of ten females died or were killed in moribund condition prior to completion of the study (Bio/Dynamics, 1990a). One death occurred after a single dose and seven more occurred during the first week. Subsequent deaths occurred as late as day 76 of the study. These deaths were considered by the researchers to be chemical-related; myocardial degeneration seen by microscopic examination in all 120 mg/kg-day rats that died after day 14 of the study was considered by the researchers to be a possible cause of death. One female in the 80 mg/kg-day dose group died on day 78; a death that was also considered to be chemical-related by the investigators in part because microscopic examination revealed myocardial degeneration similar to that seen in the 120 mg/kg-day animals that died. One female in the 40 mg/kg-day group died due to gavage error, but no chemical-related deaths were observed in the 10, 20 or 40 mg/kg-day dose groups. Rats killed in moribund condition and some of those that died exhibited emaciation, poor food consumption, hypothermia, lethargy/prostration, dyspnea, gasping, moist

rales, ataxia, abnormal posture, slight tremors, salivation, and brown-yellow stains on the snout, paws, ventral surface and anogenital area. Clinical signs were unremarkable in rats that survived the experiment. In male rats treated with the highest dose of bis(2-chloroethoxy)methane, body weight was significantly reduced by 17-18 % after weeks 1 (n=6 survivors) and 2 (n=4 survivors), and 7 to 21% thereafter (n=1 or 2 survivors). Mean body weights of males receiving 80 mg/kg-day were slightly lower than control during the second two months of the study: 6% deficit at week 5 and 10% deficit at week 12 (differences from control not statistically significant). Mean body weights for males in the lower dose groups were similar to controls, and no effect on body weight was evident in females at any dose level. Food consumption was reduced in the high-dose male group during the first 2 weeks of the study, but was similar to controls subsequently in the 2 survivors of this group. Food consumption was similar to or higher than controls in all other test groups. Ophthalmological examinations were unremarkable.

No statistically significant changes in hematological parameters were observed (Bio/Dynamics, 1990a). Exposure to 120 mg/kg-day induced statistically significant alterations in several clinical chemistry parameters in both males and females. The alterations that were considered to be indications of an effect of exposure to 120 mg/kg-day of bis(2-chloroethoxy)methane were: 1) slight elevations in serum aspartate aminotransferase (AST) at one month in both males and females, with a marked, statistically significant elevation in AST among high-dose females at study termination (no high-dose males survived to study termination), 2) a statistically significant elevation in serum alkaline phosphatase in males at one month and nonsignificant elevations in females at both one and three months, and 3) increased blood urea nitrogen (BUN) in females at 1 month (nonsignificant) and 3 months (statistically significant). In the 80 mg/kg-day group, there was a slight, statistically significant increase in serum alanine aminotransferase (ALT) among male rats at 3 months. No changes in clinical chemistry parameters were observed among male or female rats receiving 10, 20, or 40 mg/kg-day of bis(2-chloroethoxy)methane compared to controls.

Absolute and relative liver weights were statistically significantly increased in a dose-related fashion in female rats treated with 80 or 120 mg/kg-day (Bio/Dynamics, 1990a). Liver weight measurements were not available for males in the 120 mg/kg-day dose group due to early mortality; the only statistically significant change in males was a small increase in relative liver weight at 40 mg/kg-day. Histopathologic examination of the liver revealed a dose-related increased incidence of minimal-to-slight hypertrophy of the centrilobular hepatocytes in males treated with 20, 40, or 80 mg/kg-day (0/10, 0/10, 3/10, 4/10, 6/10, and 0/10 in the 0, 10, 20, 40, 80, and 120 mg/kg-day groups, respectively). The difference from controls was statistically significant in the 40 and 80 mg/kg-day groups (Fisher exact test conducted for this assessment). The lesion was not observed in males of the 120 mg/kg-day group, but rats in this group all died early. Liver lesions were not found in female rats. Mean adrenal weights (absolute and relative) were reduced relative to control among male rats receiving 20, 40, or 80 mg/kg-day. This effect on adrenal weight, however, was not observed among females and adrenal morphology was normal; thus, the toxicological significance of this effect on the adrenal gland is uncertain. Significant increases in relative kidney and testes weights in male rats at 80 mg/kg-day were considered by the researchers to be secondary to reduced body weight in this group. Kidney lesions, seen only in male rats, were increased incidences of minimal to moderate tubular nephrosis, accompanied in some cases by birefringent intracytoplasmic inclusions in the

convoluted tubular epithelium, and hyaline droplets in the epithelial cytoplasm of the proximal convoluted tubules. The incidence and severity of the renal lesions increased with dose, with the 10 mg/kg-day group being similar to controls and the 80 mg/kg-day group showing the most pronounced effects.

Other organs affected by the 120 mg/kg-day dose were the heart (myocardial degeneration), brain and spinal cord (vacuolization, gliosis), spleen, bone marrow, and thymus (atrophy, hypocellularity), and epididymides (oligospermia, degenerated seminal product); however, these organs were not systematically examined in rats receiving lower doses (Bio/Dynamics, 1990a). Of particular interest is the heart. Postmortem examination revealed slight-to-moderate degeneration of the myocardium in all high-dose animals that died after 2 weeks of exposure to bis(2-chloroethoxy)methane. The overall incidence of myocardial degeneration was 6/10 males and 6/10 females at 120 mg/kg-day (versus 0/10 for controls of each sex). The authors speculated that myocardial degeneration was a possible cause of death. Despite the prevalence of this effect among high-dose rats of both sexes, and absence among controls, the authors did not conduct histopathological examinations of the hearts of rats receiving lower doses, aside from one female from the 80 mg/kg-day group (the female that was found dead on day 78) and one female from the 40 mg/kg-day group that died accidentally in week 5. Histological examination revealed myocardial degeneration in the 80 mg/kg-day female, but not the 40 mg/kg-day female.

The renal effects seen in male rats are consistent with the pattern of early stages of alpha-$_{2u}$ globulin-associated rat nephrotoxicity, as established by the Risk Assessment Forum (U.S. EPA, 1991b), wherein the Agency concluded these renal effects are not appropriate as a critical effect for human health risk assessment. This study identified a LOAEL of 20 mg/kg-day based on liver lesions (hypertrophy of the centrilobular hepatocytes) in males rats and a NOAEL of 10 mg/kg-day following subchronic oral administration of bis(2-chloroethoxy)methane.

More recently, the general toxicity of BCM was evaluated in mice (Battelle, 2002a) and rats (Battelle, 2002b) exposed to BCM (in 95% ethanol) dermally for 5 days per week for 90 days. Applied doses for rats and mice were 0, 50, 100, 200, 400 and 600 mg/kg. Duration adjusted doses were 0, 36, 71, 143, 286 and 429 mg/kg. Available reports do not indicate whether the dose site was occluded. For all rats, the 600 mg/kg dose was lethal, and observations consistent with heart failure were noted in some rats in the 400 and 600 mg/kg dose groups. BCM was lethal in two of 10 female rats receiving 400 mg/kg. Selected organs were histologically examined at sacrifice. Hematology and clinical chemistries were not altered. Histopathic cardiomyopathy was considered the most toxicologically significant finding, and a dose-dependent increase in severity was noted in the 400 and 600 mg/kg dose groups. In male rats, histologic alterations were noted in the glandular stomach, mesenteric lymph nodes, spleen, thymus, Harderian gland and olfactory epithelium, but only in high dose animals. Findings in female rats differed only in that spleen, Harderian gland and olfactory epithelium were affected at 400 mg/kg and renal tubular (cortex) damage was noted in high dose females.

In mice, Battelle (2002b) reported no findings of lethality in males, but BCM was lethal to 3/10 female mice receiving 600 mg/kg. Erythrocyte-related parameters (RBC, hemoglobin, hematocrit) were significantly reduced in male mice at and above 200 mg/kg and both absolute

and relative kidney weights were increased at 400 and 600 mg/kg. In female mice, absolute liver weight was increased and myocardial vacuolization were observed at 400 mg/kg. At 600 mg/kg, additional findings included histopathic alterations in heart and liver, erosion and inflammation of the stomach and duodenum, and reductions in erythrocyte parameters. Dunnick et al (2004a) also reported the results from this study and noted an increased (2/10) incidence of myocyte cytoplasmic vacuolization in female rats exposed to 200 mg/kg, with incidences of 5/10 and 8/10 in the two higher doses, respectively.

From these studies, a dermally applied, duration adjusted LOAEL of 71 mg/kg-day is indicated for decreased hemoglobin content in male mice and increased incidence of myocyte cytoplasmic vacuolization in female rats. Correspondingly, the NOAEL values would be 36 mg/kg-day. Special considerations and information must be available to translate this dermally applied dose to a corresponding internal dose. Some pertinent information describe the distribution and elimination of 14C from a 14C-labelled BCM dermal administration study (Mathews and Jeffcoat, 2002). In those studies, BCM was dermally applied. *Ex vivo* studies with excised skin demonstrated a loss of 85% of the applied dose within one hour of application. Absent a capacity of absorption and removal from the site, these results indicate that up to 85% of the administered dose may be lost to volatilization within the first hour of application. Results from dermal studies in rats exposed to 10 and 0.1 mg/kg with and without dose site appliances (covers) demonstrated that dermal absorption resulted in a total absorbed dose of approximately 15% of the administered dose with appliances and approximately 40 to 44% of applied dose without appliance, seemingly indicative of additional ingestion via grooming (Mathews and Jeffcoat, 2002). In mice with the dermal appliance, these samples accounted for approximately 9 and 18% of a dermally applied dose of 0.1 or 10 mg/kg, with dose site accounting for approximately 1% of the administered dose. Mice administered BCM without the site-protective appliance absorbed 13 and 21% of applied doses of 0.1 and 10 mg/kg, respectively. The pattern of tissue distribution, extent of urinary elimination and other pharmacokinetic information, demonstrated for total radiolabel derived from 14C-labelled BCM, demonstrate appreciable similarity between dermal and oral exposures. While these data indicate dermal absorption, potential and undescribed differences in the metabolism of orally and dermally exposed animals exist and complicate the development of a dermal correction factor, especially so in light of studies that seem to indicate thiodiglycolic acid as the potentially bioactive (toxic) metabolite (Mathews and Jeffcoat, 2002). This metabolite is common to other cardiotoxic compounds, as well. Without further adjustment, the dermally applied, duration adjusted NOAEL values indicated by Battelle (2002a,b) and quantified by Dunnick et al (2004a) are higher than NOAEL value (10 mg/kg-day) for liver lesions developed from orally administration studies (Bio/Dynamics, 1990a).

In a range-finding study for the oral subchronic study (Bio/Dynamics, 1990a), non-fasted Sprague-Dawley rats (5/sex/dose group) were treated with 0, 20, 40, 50, 60, 80, or 100 mg/kg-day of bis(2-chloroethoxy)methane by daily gavage in corn oil for two weeks (Bio/Dynamics, 1990b). When no signs of toxicity were noted after one week of dosing, the 20 and 40 mg/kg-day doses were increased to 150 and 200 mg/kg-day, respectively, for the second week of treatment and satellite groups of 5 rats/sex/group were started on doses of 120 or 160 mg/kg-day. Animals were observed twice daily for mortality and gross toxicity. Physical examinations and body weight and food consumption measurements were performed weekly. Blood was collected

from all rats surviving to study termination for hematology and clinical chemistry evaluations. Complete gross postmortem examinations were performed on all animals. The brain, heart, liver, kidneys, adrenals, and gonads were weighed for animals killed at terminal sacrifice. Histopathology was not performed.

Doses of 120 mg/kg-day and above clearly produced treatment-related mortality (7/10-10/10 dead after 1-9 doses) (Bio/Dynamics, 1990b). The only deaths in the lower dose groups were single deaths in the 60 and 80 mg/kg-day groups (1/5 females and 0/5 males died in each group after 16 doses) that may also have been due to treatment. Findings in rats that died or were sacrificed moribund included clinical signs (lethargy, tremor, dyspnea, irregular gait, yellow or brown staining of the anogenital area, salivation, moist rales, hypothermia, and general poor condition in some rats just prior to death), antemortem weight loss, hematological changes (increased hemoglobin, hematocrit, and red blood cell count in males, but not females), and serum chemistry changes (increases in serum markers for hepatotoxicity and nephrotoxicity, including ALT, AST, alkaline phosphatase, BUN, and glucose). Due to the high mortality in the $\geq$120 mg/kg-day dose groups, meaningful comparisons based on group means were not possible for these groups. Among the 20-100 mg/kg-day groups, there were no significant differences from controls for food intake or body weight, and no clinical signs were observed. The only significant hematology finding was an increase in red blood cell count in females, but not males, at 100 mg/kg-day. Blood urea nitrogen was significantly increased in the 50, 80, and 100 mg/kg-day female groups, and non-significantly increased in the 60 mg/kg-day female group. The magnitude of the change from controls was small for this parameter ($\approx$20%) and did not increase with dose. No other serum chemistry changes were seen in females or males. Absolute and relative liver weights were significantly increased in females in the 80 and 100 mg/kg-day groups (by 21-27%, a moderate change for this parameter). No other significant organ weight changes were found. Gross postmortem examination revealed no abnormalities attributable to bis(2-chloroethoxy)methane. The results of this study support the finding of the subchronic study that the liver is an important target for bis(2-chloroethoxy)methane.

In a short term study to characterize and examine the short-term time course of BCM-induced cardiotoxicity, rats were exposed dermally for up to 12 days to 400 and 600 mg/kg BCM in 95% ethanol (Dunnick et al, 2004b). Within two days of exposure to 600 mg/kg, most but not all cardiac myocytes examined showed toxic effects. Mitochondrial alterations were the most prominent, but other alterations included distention of the sarcoplasmic reticulum, myofibrillary degeneration and occasional Z-banding misalignments. Severe disintegration of mitochondria and the presence of megamitochondria were observed. Swelling of the sarcoplasmic reticulum was presented as a sign of cellular injury due to loss of membrane function in maintaining water balance. The authors noted in animals surviving to day 16 a "resolvement of the manifestations of the lesions".

Inhalation Exposure. No reports were located regarding the subchronic or chronic toxicity of bis(2-chloroethoxy)methane in animals by inhalation exposure.

Other Studies

Toxicokinetics. The disposition of BCM was investigated in rats and mice by Research Triangle Institute (RTI) under contract to NIEHS (Mathews and Jeffcoat, 2002). In that study, male and female F-344 rats and male and female B6C3F1 mice received 14-C-labeled BCM via the oral, intravenous (i.v.) and dermal routes. BCM appeared poorly absorbed via dermal application, potentially due to volatility, and so will not be further presented here. Initial 72-hr studies characterized the tissue distribution and elimination of a 10 mg/kg gavage (water vehicle) dose of BCM. Parent BCM and 14C-CO2 were quantified in expired air, and total 14C was quantified in urine, feces and tissues from male and female mice and male rats. The routes, rates and extent of elimination appeared similar in male and female mice, with combined urinary and fecal elimination accounting for 60-74% of the dose at 14 hours and with urine accounting for 50-60% and approximately 25% of the dose excreted in urine and feces, respectively, at 72 hours. Approximately 10-12% was excreted as 14C-CO2 in breath, cumulative to 72 hours; less than 0.12% was excreted as BCM in breath of male mice. Cumulative elimination via all routes accounted for greater than 90% of dose in each sex.

Tissue distribution in male and female mice was similar, and body burdens approximated less than 1% of the administered dose. After 24 hours, 14C in blood was unextractable. At 72 hours, blood concentrations of 14C (in BCM equivalents) were 162 ng/gram, and 118 ng/gram for male and female mice, respectively. For males, tissues with 14C concentrations higher (ratios of tissue:blood concentrations in parentheses) than blood included liver (2.88), kidney (2.48), thymus (1.72), skin (1.33), lung (1.31), spleen (1.29), and adipose (1.18). For female mice, tissues with 14C concentrations higher than blood included liver (3.10), thymus (2.72), kidney (2.61), adipose (1.86), ovaries (1.82), lung (1.56), spleen (1.41), and skin (1.05). Heart tissue contained concentrations of 14C approximating 85% that of blood for both sexes.

As in mice, BCM was rapidly eliminated from orally-exposed male rats, but higher rates and extent of elimination occurred via the urine; this route accounted for more than 50% of the dose at 8 hours, and for 90% of the dose at 72 hours. Feces accounted for approximately 0.4% of the dose at 72 hours. Exhalation of 14C-CO2 accounted for approximately 7% of the dose, and exhaled BCM accounted for less than 0.2% of the administered dose. Less than 2.5% of the dose's 14C equivalent was retained in the body at 72 hours. Blood concentrations of 14C (in BCM equivalents) were 390 ng/gram. For male rats, tissues with 14C concentrations higher (ratios of tissue:blood concentrations in parentheses) than blood included liver (1.74) and thymus (1.69). While higher blood concentrations in the rat may lead to speculation that species differences in the apparent concentrations of BCM in blood may shift the pattern of blood:tissue distribution, most rat solid tissues also contained higher concentrations of BCM equivalents than their mouse counterparts.

Male mice were dosed with 1.0 mg/kg, i.v. and female mice were dosed i.v. with BCM at 0.1 and 1.0 mg/kg. For all mice, urinary and fecal elimination was characterized for 72 hours; tissue distribution was evaluated for male mice. Combined urinary and fecal elimination for all dose groups approximated 85 to 95% at 72 hours, with urine accounting for 65 to 72% of the administered dose. Sex-dependent differences in the fraction exhaled seemed evident for the 1 mg/kg mice. This route accounted for nearly 10% of the dose in males and approximately 5% of

dose in females. Males eliminated nearly twice as much of the dose unchanged in expired air than did females, and approximately three-fold more of the dose as CO2 than did females. In females administered an i.v. dose of 0.1 mg/kg, a slightly lower fraction of the dose was eliminated in urine and feces, and a slightly higher fraction of the dose was eliminated as expired CO2 when compared to females administered 1.0 mg/kg via i.v.

In male mice administered 1.0 mg/kg BCM i.v., at 72 hours, approximately 4% of the administered dose was retained in the body. Blood concentrations were approximately 19 ng equivalents/gram, and tissues with 14C concentrations higher (ratios of tissue:blood concentrations in parentheses) than blood included kidney (2.33), liver (1.94), adipose (1.45), thymus (1.24), and lung (1.08). Heart contained approximately 71% the concentration of BCM equivalents as blood.

In male and female rats administered 1.0 mg/kg BCM i.v., the time course profile demonstrated rapid and marked decline of BCM, where levels circulating dose approximated 2% of administered dose within 15 minutes. BCM equivalents demonstrated a biphasic decline with the terminal slope appearing largely defined by the proportion of unextractable 14C residues. For example, for males and females, total BCM equivalents decreased from 362 to 67 and from 283 to 45 ng equivalents/ gram blood between 15 minutes and 24 hours, respectively. During this time the percentage of blood 14C present as unextractable fraction increased from approximately 20% to approximately 75% for males and from approximately 22% to approximately 95% for females. Similar results were demonstrated in male mice administered 1.0 mg/kg BCM. In addition to blood, liver and thymus tissues were analyzed for extractable radioactivity. At 15 minutes post-dosing, less than 30% of the total radioactivity in liver was extractable, with the majority of extracted radiolabel represented by parent compound. At 8 hours post-dosing, less that 5% of the total radioactivity present in liver tissue was extractable, and virtually no parent BCM was demonstrated. Results in thymic tissue were qualitatively the same: at 8 hours approximately 85% of the total radioactivity was extractable, and approximately 45% of the extractable radioactivity represented parent BCM; at 8 hours post-dosing extractable radiolabel in thymus represented approximately 10% of total radioactivity, with parent BCM levels approaching zero. The high level of binding early in the time profile (15 minutes) seems inconsistent with incorporation of radiolabelled moiety into protein.

Mathews and Jeffcoat (2002) also reported the results of investigations of BCM metabolism. The results of an experiment in which cytochrome P4502E1 was inhibited demonstrated no change in the blood concentration-time profile in male mice administered 1.0 mg/kg BCM. Urine collected from rats administered 10 and 0.1 mg/kg BCM orally and male rats administered 1.0 mg/kg i.v. demonstrated three distinct peaks, accounting for 80 to 88% of urinary 14C, when analyzed by high performance liquid chromatography, and none of these peaks was altered when urine was incubated with sulfatase, acylase and beta-glucoronidase. One of these metabolites co-eluted with thiodiglycolic acid; subsequent gas chromatography/mass spectrometric analysis confirmed that metabolite as thiodiglycolic acid. This metabolite accounted for 49 to 51% of recovered 14C. The peak that co-eluted with the sulfoxide of thiodiglycolic acid accounted for 25-31% of urinary radiolabel. Combined recoveries for these two peaks accounted for between 74 and 82% of urinary radiolabel. With the preponderance of

radiolabel eliminated in urine, these data support thiodiglycolic acid as the major metabolite of BCM.

Oral-Dermal Dose Comparison. Data from 90-day studies conducted via the dermal route of exposure in rats (Battelle, 2002a) and mice (Battelle, 2002b) offer additional insights on the dose-response relationship for several toxicities. However, in order for advantage to be made from these results, some measure of absorbed, rather than applied dose is required. The relationship between dermally applied and absorbed dose can be developed from information from a distribution study also recently available (Mathews and Jeffcoat, 2002). Twenty-four hours after administration, 15.73 and 15.44% of dermally applied doses were absorbed by rats, and urinary elimination accounted for 91.1 and 88.1% of the absorbed dose in male rats receiving dermal doses of 0.1 and 10 mg/kg. The absorbed dose from these two exposures corrects to 0.016 and 1.5 mg/kg. In mice, 24 hours after application, approximately 9% of a 0.1 mg/kg dose was absorbed, and urinary elimination accounted for 95.5% of the absorbed dose. In mice dermally exposed to 10 mg/kg, approximately 18% of the dose was absorbed, and urinary elimination accounted for 68.8% of the absorbed dose. In mice, the study authors noted the confounding issue of cross contamination of urine and feces occurring in the metabolism cage. Combined urinary and fecal elimination in 10 mg/kg-dosed mice accounted for 78.3% of the absorbed dose. While the fraction of absorbed 14C eliminated in urine is similar between oral and dermal exposures, there is no information on the comparative metabolism of 14C-labelled BCM, and there is evidence that the thiodiglycolic acid metabolite may be responsible for the noted cardiotoxicity.

DERIVATION OF PROVISIONAL SUBCHRONIC AND CHRONIC RfDs FOR BIS(2-CHLOROETHOXY)METHANE

No pertinent data regarding the oral toxicity of bis(2-chloroethoxy)methane in humans are available. Only one subchronic study of oral administration of bis(2-chloroethoxy)methane to rats was located: a 90-day oral gavage study conducted by Bio/Dynamics (1990a), wherein Sprague-Dawley rats (10/sex/dose group) received oral doses of 0, 10, 20, 40, 80, or 120 mg/kg-day of bis(2-chloroethoxy)methane in corn oil. Subchronic and chronic oral RfDs for bis(2-chloroethoxy)methane can be derived using a NOAEL/LOAEL approach, based on liver lesions (centrilobular hepatocellular hypertrophy) in male rats receiving 20 mg/kg-day or more of bis(2-chloroethoxy)methane. This study identified a NOAEL of 10 mg/kg-day for the critical effect. The finding that the liver is a sensitive target for bis(2-chloroethoxy)methane is supported by the short-term range-finding study (Bio/Dynamics, 1990b).

To the rat NOAEL of 10 mg/kg-day for liver lesions established by Bio/Dynamics (1990a), a combined uncertainty factor of 300 was applied. The uncertainty factors included a 10 for interspecies extrapolation, a 10 for human variability, and a 3 for database deficiencies (including lack of reproductive and developmental toxicity tests), resulting in a combined uncertainty factor of 300. A provisional **subchronic oral RfD of 0.03 mg/kg-day** was calculated as follows:

p-sRfD = NOAEL / UF
= 10 mg/kg-day / 300
= 0.03 mg/kg-day or 3E-2 mg/kg-day

A provisional chronic oral RfD can also be derived by dividing the NOAEL of 10 mg/kg-day established by Bio/Dynamics (1990a) by a combined uncertainty factor of 3000. The uncertainty factors included a 10 for extrapolation from a subchronic study, a 10 for interspecies extrapolation, a 10 for human variability, and a 3 for database deficiencies, resulting in a combined uncertainty factor of 3000. A provisional **chronic oral RfD of 0.003 mg/kg-day** was calculated as follows:

p-RfD = NOAEL / UF
= 10 mg/kg-day / 3000
= 0.003 mg/kg-day or 3E-3 mg/kg-day

Confidence in the principal study is low. The principal study examined a number of relevant endpoints; however, the study used only minimally adequate group sizes, failed to conduct histopathology on all tissues at lower exposure doses, and appears to have used a dose that was too high based on the range-finding study (Bio/Dynamics, 1990b). Confidence in the database is also low: the database is lacking human data, supporting subchronic or chronic animal studies, and studies of developmental, reproductive, or neurological effects of exposure to bis(2-chloroethoxy)methane. Reflecting low confidence in the principal study and low confidence in the database, confidence in the provisional RfD is low.

FEASIBILITY OF DERIVING PROVISIONAL SUBCHRONIC AND CHRONIC RfCs FOR BIS(2-CHLOROETHOXY)METHANE

Derivation of a provisional subchronic or chronic RfC for bis(2-chloroethoxy)methane is precluded by the absence of inhalation toxicity data.

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