EVIDENCE ON THE CARCINOGENICITY OF

TECHNICAL GRADE BIS(2-CHLORO-1-METHYLETHYL) ETHER

FINAL

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Reproductive and Cancer Hazard Assessment Section
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PREFACE

The Safe Drinking Water and Toxic Enforcement Act of 1986 (Proposition 65, California Health and Safety Code 25249.5 *et seq*.) requires that the Governor cause to be published a list of those chemicals "known to the state" to cause cancer or reproductive toxicity. The Act specifies that "a chemical is known to the state to cause cancer or reproductive toxicity...if in the opinion of the state's qualified experts the chemical has been clearly shown through scientifically valid testing according to generally accepted principles to cause cancer or reproductive toxicity." The lead agency for implementing Proposition 65 is the Office of Environmental Health Hazard Assessment (OEHHA) of the California Environmental Protection Agency. The "state's qualified experts" regarding findings of carcinogenicity are identified as the members of the Carcinogen Identification Committee of the OEHHA Science Advisory Board (22 CCR 12301).

Bis(2-chloro-1-methylethyl) ether was assigned a final priority of 'high' carcinogenicity concern and placed on the Final Candidate list of chemicals for Committee review on June 12, 1998. A public request for information relevant to the assessment of the evidence on the carcinogenicity of this chemical was announced in the *California Regulatory Notice Register* on June 12, 1998. This document reviews the available evidence on the carcinogenic potential of bis(2-chloro-1-methylethyl) ether. It was released as the draft document *Evidence on the Carcinogenicity of Technical Grade Bis(2-chloro-1-methylethyl) ether* in July 1999.

At their October 7, 1999, meeting the Committee found, by a vote of six in favor and one against, that technical grade bis(2-chloro-1-methylethyl) ether had been "clearly shown through scientifically valid testing according to generally accepted principles to cause cancer." *Bis(2-chloro-1-methylethyl) ether, technical grade* was added to the list of chemicals known to the State to cause cancer, for purposes of Proposition 65, effective October 29, 1999.

The following document is the final version of the document that was discussed by the Committee at their October 1999 meeting.

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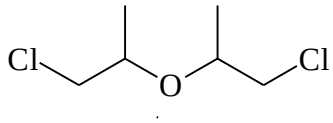
1 EXECUTIVE SUMMARY

Bis(2-chloro-1-methylethyl) ether (BCMEE) is a β-haloether with numerous industrial uses and is primarily produced as a by-product from the manufacture of propylene glycol and propylene oxide. Its generation as a by-product results in the most significant potential for environmental contamination. Technical grade BCMEE [~70% bis(2-chloro-1-methylethyl) ether and ~30% 2-chloro-1-methyl-(2-chloropropyl) ether] induced tumors of the liver in male mice and of the lung in female mice. Some evidence suggested the induction of lung tumors in male mice as well, and there was some suggestion of forestomach effects related to compound administration. Since currently available evidence cannot eliminate the possibility that other components of technical grade BCMEE are not also active, conclusions regarding the evidence of carcinogenicity are limited to the technical grade material. BCMEE also has demonstrated genotoxic potential in a number of short-term tests *in vitro*. In support of the concern for the carcinogenicity of BCMEE, haloethers with structural homology to BCMEE have shown evidence of carcinogenicity.

There is evidence for the carcinogenicity of *technical grade bis(2-chloro-1-methylethyl) ether*, with the development of lung tumors in male and female mice, and liver tumors in male mice. Further evidence of carcinogenic potential is provided by genotoxicity in several short-term tests, and by strong chemical structural analogies with known carcinogens.

2 INTRODUCTION

2.1 Identity of Technical grade Bis(2-chloro-1-methylethyl) ether



Molecular Formula: $C_6H_{12}Cl_2O$

Chemical Class: β-haloether

Molecular Weight: 171.07

CAS Registry No.: Not applicable for technical grade preparation; primary components are the structural isomers bis(2-chloro-1-methylethyl) ether (CAS Reg. No. 108-60-1) and 2-chloro-1-methylethyl(2-chloropropyl) ether (CAS Reg. No. 83270-31-9)

Synonyms: Bis-chloroisopropyl ether; 2,2'-oxybis[1-chloropropane]; β,β'-dichloro-diisopropyl ether; DCIP; Nemamorte®; Pichloram

BCMEE is a colorless liquid with a melting point of -97°C, a boiling point of 187.4°C, and a water solubility of <0.1 mg/mL at 22°C (Chemfinder WebServer, 1997). A vapor pressure of 1 mm Hg at 29.6°C has been reported (IARC, 1986).

2.2 Occurrence and Use

The primary occurrence of BCMEE is as a by-product from the manufacture of propylene oxide and propylene glycol (NCI, 1979). BCMEE has numerous industrial uses and is a component of numerous chemical mixtures. BCMEE is used in paint and varnish removers, spotting agents, cleaning solutions, and as a soap adjuvant in the textile industry. BCMEE is also an intermediate in manufacture of certain dyes, resins, and pharmaceuticals. In Japan, BCMEE has been used as the active ingredient in a nematocide (Nemamorte®; Mitsumori *et al.*, 1979; NTP, 1982).

The production of BCMEE as a by-product of the production of propylene glycol and propylene oxide has resulted in demonstrated environmental contamination. Waste stream effluent from production facilities has been shown to contain measurable amounts of BCMEE and the levels persist at considerable distances downstream in rivers.

BCMEE is a chemical for which reporting is required in the U.S. EPA's Toxic Release Inventory (TRI) program. Data from the 1996 TRI database show two Dow Chemical Company facilities (one each in Louisiana and Texas) which released a total of 4100 pounds of BCMEE, predominantly as stack air emissions (U.S. EPA, 1999). Projected

releases for 1997 and 1998 were quantitatively similar. Total production-related waste of BCMEE totaled ~22 million pounds, with approximately 60% of this recycled on-site and the balance treated on-site.

BCMEE has been considered 'practically nonbiodegradable' as indicated by studies showing no increased oxygen uptake into a sample of Ohio River water incubated with BCMEE for 5 days at 20°C (NCI, 1979; citing Kleopfer and Fairless, 1972).

3 DATA ON BIS(2-CHLORO-1-METHYLETHYL) ETHER CARCINOGENICITY

Three series of carcinogenicity studies have been reported, in each of which the compound was administered orally either by gavage ($B6C3F_1$ mice, Fischer 344 rats) or in the diet (ICR mice). BCMEE has also been tested for genetic toxicity in several assays in *Salmonella*, *Drosophila* and mammalian cells in culture.

3.1 Epidemiological Studies of Carcinogenicity in Humans

No data on long-term/cancer effects of human exposure to BCMEE were found in an earlier search by the International Agency for Research on Cancer (IARC, 1986) or more recently by OEHHA.

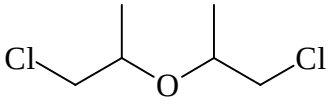
3.2 Carcinogenicity Studies in Animals

In a series of carcinogenicity studies, both rats and mice received BCMEE orally either by gavage or in the diet by NCI (1979), Mitsumori *et al.* (1979), and NTP (1982). In the NTP studies, increased incidences of hepatocellular carcinomas were observed among male mice and combined lung adenomas and carcinomas were observed in male and female mice treated by oral gavage.

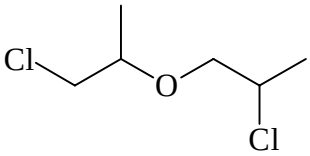
3.2.1 Oral Exposure Studies

Mouse Gavage Exposure: NTP, 1982

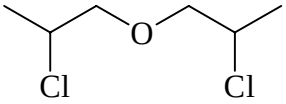
Male and female $B6C3F_1$ mice (50/group) received technical grade BCMEE by oral gavage five days/week for 103 weeks (NTP, 1982). Animals received 0, 100, or 200 mg/kg_{bw} of technical grade BCMEE dissolved in corn oil. Two different lots of the test chemical were used in the study. The first lot was used for the first 94 weeks and consisted of a mixture of 69.4% BCMEE, 28.5% 2-chloro-1-methylethyl(2-chloropropyl) ether [mixed isomer] and 2.1% bis(2-chloropropyl) ether. The second lot, used for the final nine weeks of the study, contained 71.5% BCMEE, 26% of the mixed isomer, and 2.6% bis(2-chloro-*n*-propyl) ether. The chemical structures of the components of the test material are shown below.



BCMEE



2-Chloro-1-methylethyl (2-chloropropyl) ether



Bis(2-chloro-*n*-propyl) ether

No evidence of treatment-related changes in mean body weight gains, body weights, or clinical signs were reported at the end of the study, although high-dose female mice showed generally lower body weights at the end of the study. No significant changes in survival were reported between treated and control groups. Notable tumor incidences are reported in Table 1.

Briefly, among male mice statistically significant increases in the incidences of carcinomas and combined adenomas and carcinomas of the liver were observed in both low- and high-dose groups. Adenomas and combined adenomas and carcinomas of the alveolar/bronchiolar regions of the lung were significantly increased among males in the low-dose group. There were also significant dose-related trends for these tumors in both the liver and the lung by the Cochran-Armitage trend test. Among female mice, significant increases in the incidences of adenomas and combined adenomas and carcinomas of the alveolar/bronchiolar regions of the lung were observed in the high-dose group.

A finding of squamous cell papillomas in the stomach or forestomach of treated mice (two in males, two in females) plus a single squamous cell carcinoma in a different female mouse is suggestive of a treatment-related effect because of the rarity of stomach tumors in untreated animals. NTP reports that among corn oil treated vehicle control animals in the Bioassay Program, 1/362 female mice and 0/365 male mice developed stomach tumors. Thus, in female mice a comparison of the historical incidence with the combined papilloma/carcinoma incidence (3/49) would produce a statistically significant increase in incidence (p=0.006, by the Fisher Exact test).

No information has been located concerning the carcinogenicity of the other compounds present in the test material [2-chloro-1-methylethyl(2-chloropropyl) ether and bis(2-chloro-*n*-propyl) ether].

NTP (1982) found as follows: "Under the conditions of this bioassay, bis(2-chloro-1-methylethyl) ether — containing 2-chloro-1-methylethyl(2-chloropropyl)ether — was carcinogenic for B6C3F_1 mice, causing increased incidences of alveolar/bronchiolar adenomas in males and females and hepatocellular carcinomas in males. In addition, the occurrence of a low incidence of squamous cell papillomas or carcinomas in the stomach or forestomach of females (a rare tumor in B6C3F_1 mice) was probably associated with the administration of bis(2-chloro-1-methylethyl) ether."

Table 1: Tumors in B6C3F$_1$ mice receiving technical grade bis(2-chloro-1-methylethyl) ether at 0, 100, or 200 mg/kg$_{bw}$ by gavage for 103 weeks (NTP, 1982).

Tumor Site and Type		Dose, mg/kg$_{bw}$[a]		
		0	100	200
Males				
Lung: Alveolar/ Bronchiolar	Adenoma	5/50[b]	13/50[c]	11/50[d]
	Carcinoma	1/50	2/50	2/50
	Adenoma or carcinoma	6/50	15/50[c]	13/50[d]
Liver	Adenoma	8/50	10/50	13/50
	Carcinoma	5/50	13/50[c]	17/50[c,d]
	Adenoma or carcinoma	13/50	23/50[c]	27/50[c,d]
	Metastases to lung	1/50	4/50	3/50
Stomach / Forestomach	Squamous-cell papilloma	0/49	1/50	1/50
Females				
Lung: Alveolar/ Bronchiolar	Adenoma	1/50	4/50	8/50[c,d]
	Carcinoma	0/50	0/50	2/50
	Adenoma or carcinoma	1/50	4/50	10/50[c,d]
Stomach / Forestomach	Squamous-cell papilloma	0/50	0/49	2/49
	Squamous-cell carcinoma	0/50	0/49	1/49

[a] Dose of BCMEE administered.
[b] Number of lesion-bearing animals/total examined.
[c] Significantly increased incidence relative to control group by the Fisher Exact test ($p < 0.05$).
[d] Significant dose-related trend by the Cochran-Armitage trend test ($p < 0.05$).

Rat Gavage Exposure: NCI, 1979

Male and female Fischer F344 rats (50/group) received technical grade BCMEE by oral gavage five days/week for 103 weeks (NCI, 1979). Three batches of the test compound, which were determined to be mixtures of iso- and *n*-propyl ether isomers, were used in the studies. One batch (Lot No. 7, MC&B Manufacturing Chemists) was used during the first 46 weeks of the studies, a second batch (Lot No. PB41576, Pfaltz and Bauer, Inc.) during weeks 47 through 83 of the studies, and a third batch (Lot No. I62976, ICN

Pharmaceuticals; also used in NTP's $B6C3F_1$ mouse studies) from week 84 through the end of the study. Only the third batch was analyzed for composition, although the other two were thought to be consistent in terms of isomer content based upon similar nuclear magnetic resonance spectra. The third batch was found to contain 69.4% BCMEE, 28.5% 2-chloro-1-methylethyl(2-chloropropyl) ether, and 2.1% bis(2-chloro-*n*-propyl) ether.

Rats were treated with technical grade BCMEE at doses of 0, 100, or 200 mg/kg_{bw}-day in corn oil. Vehicle as well as untreated control groups were included in the study. Dose-related decreases in body weight were observed for both male and female rats throughout the study. Survival was also significantly affected by the administration of the test compound. Among high-dose male and female rats almost no animals survived to the end of the study and in these groups sufficient numbers were not considered to have survived for the observation of late appearing tumors. Among males, 56% of the high-dose and 92% of the low-dose rats survived to 78 weeks, whereas among females, 50% of the high-dose and 88% of the low-dose rats survived to 78 weeks. Hyperkeratosis of the esophagus was increased among high-dose male and female rats (40/49 vs. 9/50 vehicle controls, males; 31/48 vs. 13/40 vehicle controls, females) and acanthosis of the esophagus was increased among high-dose females (5/48 vs. 1/50 vehicle controls). Aspiration pneumonia showed a dose-related increase in both low- and high-dose male and female rats. No significantly increased incidences of tumors were found in either sex, although the studies' strength may have been limited by the loss of animals in the high-dose groups.

Overall, no treatment related carcinogenic effect was observed in rats treated with BCMEE by oral gavage (NCI, 1979). These studies may have been limited by excessive mortality among both sexes of animals in the high-dose group, although NCI concluded that "under the conditions of this bioassay, the technical grade test material, bis(2-chloro-1-methylethyl) ether, was not carcinogenic for F344 rats of either sex."

Mouse Dietary Exposure: Mitsumori et al., 1979

ICR mice (56/sex/group) were fed diet containing 0, 80, 400, 2000 or 10,000 ppm BCMEE for 104 weeks (Mitsumori *et al.*, 1979). These doses were calculated by the authors to be equivalent to 0, 8.4, 40, 198, and 927 mg/kg_{bw}-day for males and 0, 7.6, 36, 194, and 961 mg/kg_{bw}-day for females based on average daily food intake. Interim sacrifices of six or seven animals of each sex and dose group were conducted at 13, 26, 52, and 78 weeks. The test compound was stated to be 98.5% pure, but no components other than BCMEE were identified. High-dose male and female mice showed significantly decreased food consumption and weight gain. The authors attributed a slightly increased mortality among female mice in the high-dose group to undernutrition from reduced food intake. No significant increases in tumor incidences were reported among the BCMEE treated animals relative to the control groups.

Because of the scheduled interim kill groups (seven animals/sex/group, except for the 78 week kill which was six animals/sex/group) and some increase in mortality, only a limited number of animals were available for evaluation at the end of the study (males: 8,

5, 8, 5, and 6 for increasing dose groups, including controls; females: 5, 9, 9, 7, and 1). Animals alive at 52 weeks numbered 39, 37, 41, 37, and 28 for male mice and 38, 35, 41, 37, and 20 for female mice in increasing dose groups.

Overall, feed studies in male and female ICR mice with a higher grade material than that used in the NCI/NTP studies did not produce evidence for the carcinogenicity of BCMEE (Mitsumori *et al.*, 1979). However, because the scheduled sacrifices left few surviving mice at the end of the studies, it is not clear the studies have adequate power to detect a carcinogenic effect. The IARC Working Group reviewing BCMEE noted that the study was of limited sensitivity for the detection of carcinogenic effects (IARC, 1986).

Discussion of Carcinogenicity Studies in Animals

In summary, in male mice technical grade BCMEE induced adenomas and carcinomas of the liver and adenomas of the lung, and in female mice technical grade BCMEE induced adenomas and carcinomas of the lung. Another bioassay series with a higher grade test material in a different strain of mice showed no carcinogenic effect although the study design may have limited the ability to detect such an effect. Long-term bioassays in rats showed no carcinogenic effect, although late-developing tumors may not have been detected because of high mortality in the animals at the end of the study.

3.3 Other Relevant Data

In addition to the reported animal bioassays, additional evidence related to the possible carcinogenicity of BCMEE is available. This includes studies of genetic toxicity, observations of the pharmacokinetics and metabolism, and structure-activity comparisons.

3.3.1 Genetic Toxicology

BCMEE [purity not stated] was found to be mutagenic to *Salmonella typhimurium* strain TA100 when tested in a dessicator, both with and without metabolic activation by liver homogenates from mice and hamsters pretreated with Arochlor 1254, and from humans (Simmon, 1978; Simmon and Tardiff, 1978). The test materials were "of the highest available purity" from commercial suppliers. Addition of Arochlor 1254-induced rodent liver homogenates increased mutagenic activity of BCMEE above that seen in the absence of metabolic activation. Human liver homogenate increased mutagenic activity more than uninduced rat homogenate.

In another study, the addition of Arochlor-induced hamster liver S9 homogenate slightly increased BCMEE's mutagenic activity above that seen in the absence of metabolic activation in *Salmonella typhimurium* strains TA100 and TA1535 using a preincubation protocol (Mortelmans *et al.*, 1986).

Another testing of BCMEE in *Salmonella typhimurium* strains TA1535, TA1537, TA1538, TA98, and TA100 and *Escherichia coli* WP2 *hcr* failed to demonstrate

mutagenicity (Moriya *et al.*, 1983), however, the testing conditions appeared not to have prevented volatilization of the test material. The test materials were "standard materials obtainable in Japan." NTP testing of BCMEE in *Salmonella typhimurium* strains TA100 and TA1535 produced weakly positive but nonreproducible responses (NTP, 1982).

Treatment of *Drosophila melanogaster* with BCMEE [34.3% analyzed purity, supplied by Aldrich A2279, Batch 07] in feed (250 or 283 ppm in 10% ethanol) or by injection (1600 ppm in 5% ethanol) produced no significant increase in sex-linked recessive lethal mutations (Valencia *et al.*, 1985). A subsequent re-evaluation of the original data characterized the findings as "equivocal" (Mason *et al.*, 1992).

In each of the remaining genotoxicity studies described in this section, the test material was obtained from the NTP Chemical Repository maintained by Radian Corporation (Austin, TX). The purity of the material was not stated and it is unclear which lot was made available for these studies. Presumably, technical grade preparations as described in the bioassays conducted by NTP/NCI were provided.

No induction of unscheduled DNA synthesis (UDS) was reported in mouse hepatocytes treated *in vivo* with technical grade BCMEE, however, a significant increase in S-phase synthesis was observed in male but not female mouse hepatocytes following treatment with up to 300 mg/kg_{bw} BCMEE (Mirsalis, 1987). Toxicity was observed at the administered dose of 400 mg/kg_{bw} in male mice (Mirsalis *et al.*, 1989).

A mutagenic response from treatment with technical grade BCMEE was observed in the L5178Y tk^+/tk^- mouse lymphoma assay without metabolic activation (McGregor *et al.*, 1988). Although a non-significant response was observed for the positive control included in the experiment, the authors did not consider this a "crucial" defect of the study since a reproducible positive response was observed with the test material.

In a test for chromosomal aberrations in Chinese hamster ovary (CHO) cells, technical grade BCMEE tested positive with an "extremely high frequency" of aberrations only in the presence of a metabolic activation system (S9 homogenate from livers from Arochlor-induced Sprague-Dawley rats) (Galloway *et al.*, 1987). Increased sister chromatid exchange was observed in BCMEE treated CHO cells both with and without the metabolic activation system. Cellular toxicity was also observed at the doses at which the genotoxic effects were observed.

Technical grade BCMEE was evaluated in the prophage-induction assay (Microscreen) for its ability to induce DNA damage in *Escherichia coli* (DeMarini and Brooks, 1992). BCMEE was not found to induce prophage lambda in duplicate experiments either with or without S9 metabolic activation.

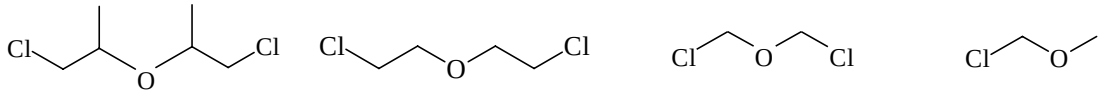
No information has been located specifically concerning the genetic toxicity of the other compounds present in technical grade BCMEE [2-chloro-1-methylethyl(2-chloropropyl) ether and bis(2-chloro-*n*-propyl) ether].

In summary, data on the genetic toxicity of technical grade BCMEE suggest that it has the potential to cause mutational changes in *Salmonella* reverse mutation assays and mouse lymphoma cell assays. Chromosomal aberrations and increased sister-chromatid exchange were observed in cultured hamster cells.

3.3.2 Structure-Activity Comparisons

BCMEE is a β-haloether and bears strong structural resemblance to another β-haloether, bis(2-chloroethyl) ether (BCEE). BCEE has been shown to induce liver tumors in two strains of mice and is a direct-acting mutagen (U.S. EPA, 1994). BCEE is listed as "causing cancer" under Proposition 65 (see structures below).

BCMEE is also homologous to the α-haloether bis(chloromethyl) ether (BCME), a potent respiratory carcinogen in mice, rats and humans (U.S. EPA, 1991). BCME is listed as "causing cancer" under Proposition 65. BCME is known to be an alkylating agent and is a more potent carcinogen than the β-haloether BCEE. The more reactive nature of this compound may, in part, explain its higher carcinogenic potency. Exposure to technical grade chloromethyl methyl ether (CMME), which contains BCME, is associated with lung cancer in humans, and is also listed as "causing cancer" under Proposition 65.



BCMEE **BCEE** **BCME** **CMME**

These compounds are also similar to BCMEE's structural isomer, 2-chloro-1-methylethyl(2-chloropropyl) ether, which is present in significant amounts in preparations of technical grade BCMEE (structure shown in description of 1982 NTP study, p. 4). This compound, a β-haloether, also bears strong structural resemblance to the carcinogen BCEE.

3.3.3 Pharmacokinetics and Metabolism

Studies of the pharmacokinetics and metabolism of BCMEE are somewhat limited. Available information includes a qualitative and quantitative comparison of absorption, distribution and metabolism between rats and monkeys treated once parenterally with several doses of radiolabeled BCMEE (Smith *et al.*, 1977). A more detailed evaluation of the metabolism of BCMEE in rats treated orally with BCMEE was also conducted (Lingg *et al.*, 1982). No studies concerning the metabolism of BCMEE in mice were identified in the literature. The overall evidence indicates essentially two metabolic routes: ether cleavage and oxidative dechlorination. Mutagenic and carcinogenic metabolic products have been identified including propylene oxide and 1-chloro-2-propanol.

CD rats and rhesus monkeys were parenterally administered BCMEE labeled with ^{14}C at the β-position at doses ranging from 0.2 $\mu g/kg_{bw}$ to 300 mg/kg_{bw} (Smith *et al.*, 1977).

The test material was considered to be of >95% purity. Qualitative differences were observed between the rat and the monkey with respect to several parameters. Monkeys achieved high peak blood levels of BCMEE whereas rat levels remained low; monkeys had high levels in the liver one week following administration whereas rats had only low levels; rats had moderate to high levels of urinary and fecal excretion whereas in monkeys these routes of elimination were low. Among the excreted products in the rat were unmetabolized BCMEE (1% in urine and expired air), 1-chloro-2-propanol (0.1-1.0% in urine), propylene oxide (detected in urine), 2-(1-methyl-2-chloroethoxy) propionic acid (detected in urine), and carbon dioxide (detected in expired air).

Available evidence from rats indicates that BCMEE undergoes both ether cleavage and oxidative dechlorination (Lingg *et al.*, 1982). Urinary metabolites of 1-^{14}C-BCMEE following peroral administration of a single dose of 90 mg/kg$_{bw}$ included 2-(2-chloro-1-methylethoxy) propanoic acid as an oxidation product and N-acetyl-S-(2-hydroxypropyl)-L-cysteine as a product of ether cleavage. The radiolabeled test compound had a reported purity of >98% as determined by gas chromatography, with impurities of 1% dichloropropene and 1% 1,3-dichloropropan-2-ol by gas chromatography/mass spectroscopy. The radiolabeled compound was diluted with unlabeled BCMEE containing 27% chloroisopropyl-2-chloropropyl ether and 3% bis(2-chloropropyl) ether. Lingg *et al.* (1982) speculated that reactive intermediates generated by ether cleavage of haloethers may be alkylating agents. A quantitative comparison of metabolites of the β-haloethers BCMEE and BCEE indicated that BCEE is more readily cleaved at the ether bond than BCMEE and suggested that the methyl group in the alpha position lends some stability to the ether bond. Since BCEE is considered a more potent carcinogen than BCMEE, the difference in susceptibility to ether cleavage may provide an explanation for the difference in potency. The authors also considered the possibility that stomach acid may hydrolyze these haloethers leading to the production of alkylating agents. The identification of a metabolite of BCEE directly substituted at a chlorine (N-acetyl-S-[2-(-chloroethoxy)-ethyl]-L-cysteine) indicates there is also a potential for direct alkylating activity. However, there is no evidence that this metabolic path occurs with the branched chain β-haloether BCMEE.

3.3.4 Pathology

The lung tumors observed in the NTP (1982) studies in B6C3F$_1$ mice were considered to meet standard criteria for bronchiolar/alveolar adenomas and carcinomas. These tumors were described as "solid cells filling adjacent alveoli or, in some cases, cells having a columnar to papillary nature. The cells were uniform with moderate cytoplasm and round to oval nuclei. The tumors are not unlike the morphology of bronchiolar/alveolar tumors in control mice."

The tumors observed in the NTP (1982) studies in mouse liver were considered by the authors to meet standard criteria for hepatocellular adenomas and carcinomas. It is generally considered that these tumor phenotypes are related in origin, and that the adenomas may progress to carcinomas.

3.4 Mechanism

Based on several results in tests *in vivo* and *in vitro*, BCMEE appears to be genotoxic, probably after metabolic activation. One metabolite of BCMEE identified in the urine of rodents, propylene oxide, is listed as "causing cancer" under Proposition 65 and is mutagenic (NTP, 1985). Another urinary metabolite, 1-chloro-2-propanol, has been found to be genotoxic in numerous *in vitro* tests in bacteria and mammalian cells (NTP, 1998). Two-year drinking water bioassays with technical grade 1-chloro-2-propanol in male and female rats and mice, however, produced no evidence of carcinogenicity in either species (NTP, 1998). A genotoxic mechanism mediated by a metabolite may, therefore, be responsible for the observed carcinogenic activity of BCMEE. Another possibility is that BCMEE, like BCEE, is a direct-acting mutagen, although this has not been confirmed experimentally.

4 OTHER REVIEWS

The International Agency for Research on Cancer (IARC) has classified BCMEE in Group 3, unclassifiable as to its carcinogenicity to humans, based on inadequate evidence in humans and limited evidence in animals (IARC, 1986). IARC has recently re-examined the carcinogenicity of a number of compounds, and in the case of BCMEE, reaffirmed its classification as Group 3 (IARC, 1999). In this recent update, it was reported in the "Genetic and related effects" section that "some experiments reported in the previous monograph (IARC, 1986) were not, in fact, done with this compound" (IARC, 1999). A re-examination of this statement by IARC staff suggests that this conclusion is in error and the studies in question were likely done with the appropriate test compound (McGregor, 1999). Since IARC's monograph addresses the carcinogenicity of BCMEE itself, rather than the technical grade preparation, its conclusions may have been based on purity concerns over the material used in the NTP/NCI bioassays. Structural relationships of BCMEE to known carcinogens did not appear to play a role in IARC's evaluation.

5 SUMMARY AND CONCLUSIONS

5.1 Summary of Evidence

In male mice technical grade BCMEE induced adenomas and carcinomas of the liver and adenomas of the lung, and in female mice technical grade BCMEE induced adenomas and carcinomas of the lung. A small number of rare tumors of the stomach and forestomach among the BCMEE-treated mice were also suggestive of a treatment-related effect. Another series of bioassays in a different strain of mice showed no carcinogenic effect, although the study design may have limited the ability to detect such an effect. Long-term bioassays in rats showed no carcinogenic effect, although late-developing

tumors may not have been detected because of high mortality in the animals during the latter part of the study. Data on the genetic toxicity of BCMEE suggests that it has the potential to cause mutational changes in *Salmonella typhimurium* reverse mutation assays and mouse lymphoma cell assays and chromosomal aberrations and sister chromatid exchange in cultured hamster cells. BCMEE shows structural homologies with other haloethers (BCEE and BCME) that are carcinogenic in both humans and experimental animals.

5.2 Conclusion

There is evidence for the carcinogenicity of technical grade BCMEE, with the development of liver and lung tumors in male mice and lung tumors in female mice treated with BCMEE by oral gavage. Further evidence includes observations of genotoxicity in several short-term tests in bacterial and mammalian cells, and by strong chemical structural analogies with several known carcinogens.

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