FINAL

Report on Carcinogens Background Document for

2,2-bis(Bromomethyl)-1,3-propanediol (Technical Grade)

Meeting of the
NTP Board of Scientific Counselors
Report on Carcinogens Subcommittee

Prepared for the:
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Public Health Services
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Criteria for Listing Agents, Substances or Mixtures in the Report on Carcinogens

US Department of Health and Human Services
National Toxicology Program

Known to be Human Carcinogens:

There is sufficient evidence of carcinogenicity from studies in humans which indicates a causal relationship between exposure to the agent, substance or mixture and human cancer.

Reasonably Anticipated to be Human Carcinogens:

There is limited evidence of carcinogenicity from studies in humans which indicates that causal interpretation is credible but that alternative explanations such as chance, bias or confounding factors could not adequately be excluded; or

There is sufficient evidence of carcinogenicity from studies in experimental animals which indicates there is an increased incidence of malignant and/or a combination of malignant and benign tumors: (1) in multiple species, or at multiple tissue sites, or (2) by multiple routes of exposure, or (3) to an unusual degree with regard to incidence, site or type of tumor or age at onset; or

There is less than sufficient evidence of carcinogenicity in humans or laboratory animals, however; the agent, substance or mixture belongs to a well defined, structurally-related class of substances whose members are listed in a previous Report on Carcinogens as either a *known to be human carcinogen, or reasonably anticipated to be human carcinogen* or there is convincing relevant information that the agent acts through mechanisms indicating it would likely cause cancer in humans.

Conclusions regarding carcinogenicity in humans or experimental animals are based on scientific judgment, with consideration given to all relevant information. Relevant information includes, but is not limited to dose response, route of exposure, chemical structure, metabolism, pharmacokinetics, sensitive sub populations, genetic effects, or other data relating to mechanism of action or factors that may be unique to a given substance. For example, there may be substances for which there is evidence of carcinogenicity in laboratory animals but there are compelling data indicating that the agent acts through mechanisms which do not operate in humans and would therefore not reasonably be anticipated to cause cancer in humans.

Summary Statement

Technical grade 2,2-bis(bromomethyl)-1,3-propanediol (~79% 2,2-bis[bromomethyl]-1,3-propanediol, ~7% 2,2-bis[hydroxymethyl]-1-bromo-3-hydroxypropane, ~7% 2,2-bis[bromomethyl]-1-bromo-3-hydroxypropane, ~0.2% pentaerythritol, and ~8% dimers and structural isomers)(BBMP)

Carcinogenicity

The flame retardant 2,2-bis(bromomethyl)-1,3-propanediol, technical grade (BBMP) is *reasonably anticipated to be a human carcinogen* based on evidence of tumor induction at multiple organ sites in rats and mice. Two-year dietary studies of the flame retardant BBMP showed a significant increase in the incidence of neoplasms of the skin, subcutaneous tissue, mammary gland, Zymbal gland, oral cavity, esophagus, forestomach, small and large intestines, mesothelium, urinary bladder, lung, thyroid gland, and seminal vesicle and in the incidence of mononuclear cell leukemia in male F344 rats; in the incidence of neoplasms of the oral cavity, esophagus, mammary gland, and thyroid gland in female F344 rats; in the incidence of neoplasms of the Harderian gland, lung, and kidney in male $B6C3F_1$ mice; and in the incidence of neoplasms of the Harderian gland, lung, and subcutaneous tissue in female $B6C3F_1$ mice (NTP 1996; Dunnick *et al.* 1997).

In a stop-exposure study, BBMP was administered in the feed to male F344 rats for three months, followed by maintenance on control diet for up to two years. Neoplasms were observed at the same sites as in the two-year continuous-exposure study of male F344 rats. The incidences of neoplasms in the stop-exposure study were greater than in the continuous-exposure study for the oral cavity, forestomach, small intestine, large intestine, lung, Zymbal gland, thyroid gland, and mesothelium and were considered to be related to treatment (NTP 1996; Dunnick *et al.* 1997).

No case reports or epidemiological studies of the occurrence of human cancer and exposure to BBMP were available.

Other Information Relating to Carcinogenesis or Possible Mechanisms of Carcinogenesis

BBMP was shown to be mutagenic *in vitro* and *in vivo,* but special conditions were required to induce mutagenicity. BBMP is mutagenic in *Salmonella typhimurium* strains TA100 and TA1535 only when tested in the presence of 30% S9 liver homogenate from induced hamsters (Zeiger *et al*. 1992). In cultured CHO cells, BBMP induced chromosomal aberrations (CA) only in the presence of S9; no induction of sister chromatid exchanges (SCE) was observed with or without S9 mix. *In vivo* exposure to BBMP induced significant increases in the frequencies of micronucleated erythrocytes in male and female mice under varying conditions (MacGregor *et al*. 1990, cited in NTP 1996).

No data are available that would suggest that mechanisms thought to account for tumor induction by BBMP in experimental animals would not also operate in humans.

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1 Introduction

The flame retardant 2,2-bis(bromomethyl)-1,3-propanediol (BBMP) was nominated for listing in the Report on Carcinogens by the NIEHS Report on Carcinogens (RoC) Review Group (RG1) based on the results of a dosed-feed study reported in a 1996 National Toxicology Program (NTP) bioassay technical report that indicated clear evidence of carcinogenicity in rats and mice.

1.1 Chemical identification

The flame retardant BBMP (FR-1138) is a technical-grade mixture of 78.6% 2,2-bis(bromomethyl)-1,3-propanediol, 6.6% 2,2-bis(hydroxymethyl)-1-bromo-3-hydroxypropane, 6.9% 2,2-bis(bromomethyl)-1-bromo-3-hydroxypropane, 0.2% pentaerythritol, and 7.7% dimers and structural isomers (NTP 1996). 2,2-bis(Bromomethyl)-1,3-propanediol, the major component of BBMP ($C_5H_{10}Br_2O_2$, mol wt 261.94, CASRN 3296-90-0) also is known by the following names:

dibromoneopentyl glycol
bisbromomethylpropanediol
bis(bromomethyl) propanol
dibromopentaerythritol
pentaerythritol dibromide
pentaerythritol dibromohydrin
dibromohydrin pentaerythritol
1,3-dibromo-2,2-dimethylolpropane.

BBMP is a white solid with a slight musty odor. It is used as a flame retardant for epoxy, polyester, and urethane foams. It also is used as a chemical intermediate for pentaerythritol. The structure of BBMP is illustrated in Figure 1-1.

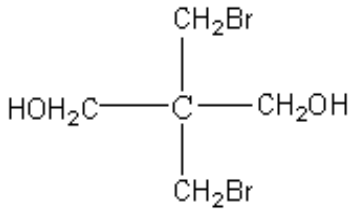

Figure 1-1. Structure of BBMP

1.2 Physical-chemical properties

The RTECS number for BBMP is TY3195500, and its physical and chemical properties are summarized in Table 1-1.

Table 1-1. Physical and chemical properties of BBMP

Property	Information	Reference
Molecular weight	261.94	Budavari *et al*. (1996)
Physical state	off-white powder	Budavari *et al*. (1996)
Odor	mild musty odor	NTP (1996)
Melting point (^{o}C) at 750 mm	109 - 110	Chemfinder (1999)
Boiling point (^{o}C) at 750 mm	235	Budavari *et al*. (1996)
Flash point (^{o}C)	nonflammable	MRI (1978)
Specific gravity	2.2	Dow Chemical (1975)
Solubility in: Water Dimethylsulfoxide 95% Ethanol Acetone Methanol Toluene	 < 1 mg/mL at 19°C ≥ 100 mg/mL at 21°C ≥ 100 mg/mL at 21°C ≥ 100 mg/mL at 21°C ≥ 102.4 g/100g at 20°C 0.6 g/100g at 25°C	 Radian (1991) Radian (1991) Radian (1991) Radian (1991) Miller (1977) Miller (1977)

BBMP is unique in that the aliphatic neopentyl structure contains no hydrogen atoms on the carbon atom adjacent to the carbon bonded to bromine. This results in the compound's being very resistant to dehydrobromination. The remaining hydroxyl groups are reactive sites that allow for polymerization. These –OH groups readily react with organic acids or epoxides to form esters and with isocyanates to form urethanes. BBMP also can react with aldehydes and ketones to form cyclic acetals or ketals, or with phosphorous oxyhalides to form cyclic phosphates or phosphites (Larsen 1969; Larsen and Weaver 1973, both cited in NTP 1996).

The physical and chemical properties of the other components of the flame retardant BBMP (FR-1138), (2,2-bis(hydroxymethyl)-1-bromo-3-hydroxypropane, 2,2-bis(bromomethyl)-1-bromo-3-hydroxypropane, and pentaerythritol) are summarized in Tables 1-2, 1-3, and 1-4. The chemical structures for these components are illustrated in Figures 1-2, 1-3, and 1-4, respectively.

Table 1-2. Physical and chemical properties of 2,2-bis(hydroxymethyl)-1-bromo-3-hydroxypropane

Property	Information	Reference
Molecular weight	199.04	Chemfinder (1999)
Physical state	np	–
Melting point (^{o}C) at 750 mm	109 - 110	Chemfinder (1999)
Boiling point (^{o}C) at 750 mm	235	Budavari *et al*. (1996)
Flash point (^{o}C)	nonflammable	MRI (1978)
Specific Gravity	2.2	Dow Chemical (1975)

Property	Information	Reference
Solubility in:		
Water	< 1 mg/mL at 19°C	Radian (1991)
Dimethylsulfoxide	≥ 100 mg/mL at 21°C	Radian (1991)
95% Ethanol	≥ 100 mg/mL at 21°C	Radian (1991)
Acetone	≥ 100 mg/mL at 21°C	Radian (1991)
Methanol	≥ 102.4 g/100g at 20°C	Miller (1977)
Toluene	0.6 g/100g at 25°C	Miller (1977)

np: not published

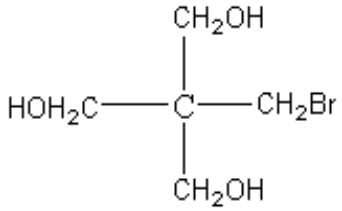

Figure 1-2. Structure of 2,2-bis(hydroxymethyl)-1-bromo-3-hydroxypropane

Table 1-3. Physical and chemical properties of 2,2-bis(bromomethyl)-1-bromo-3-hydroxypropane

Property	Information	Reference
Molecular weight	324.84	Chemfinder (1999)
Physical state	white solid	Budavari *et al*. (1996)
Solubility in:		
Water	< 0.1 g/100mL at 21.5°C	Chemfinder (1999)

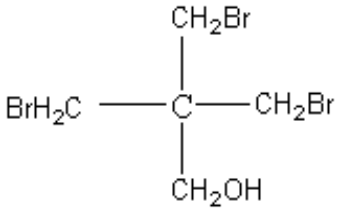

Figure 1-3. Structure of 2,2-bis(bromomethyl)-1-bromo-3-hydroxypropane

Table 1-4. Physical and chemical properties of pentaerythritol

Property	Information	Reference
Molecular weight	136.15	Chemfinder (1999)
Physical state	colorless to white crystalline powder	HSDB (1992)
Melting point (^{o}C) at 750 mm	255 - 259	Chemfinder (1999)
Boiling point (^{o}C) at 30 mm	276	Chemfinder (1999)
Flash point (^{o}C)	240	Chemfinder (1999)
Specific gravity	1.396	Chemfinder (1999)

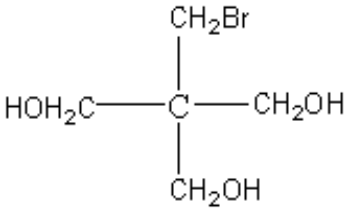

Figure 1-4. Structure of pentaerythritol

2 Human Exposure

2.1 Use

BBMP is used as a flame retardant in unsaturated polyester resins, for molded products, and in the production of rigid polyurethane foam. BBMP also is used to produce the flame retardant, FR-1138. It also is used as a chemical intermediate for pentaerythritol ethers and other derivatives used as flame retardants (Radian 1991; HSDB 1998; NTP 1996).

2.2 Production

Dow Chemical Co., of Midland, MI, was the largest producer of BBMP until the late 1980s. The current producer of BBMP is Albemarle Co., of Baton Rouge, LA. It was estimated that U.S. production in 1977 and 1979 was greater than 2.27×10^6 g (5000 pounds) (SRI 1977, 1979; cited by HSDB 1998). In 1983, U.S. Environmental Protection Agency (EPA) estimated BBMP production to be 3 to 4 million pounds per year (U.S. EPA 1983, cited in NTP 1996). U.S. EPA listed BBMP in the high production volume (HPV) program chemical list, identifying BBMP as being manufactured in or imported into the United States in amounts equal to or greater than 1 million pounds per year. The 1990 HPV list identified BBMP manufacture and importation at 2.21 to 2.95×10^6 lb/yr (U.S. EPA 1990).

2.3 Environmental occurrence

BBMP is found in nature only when it is released into the environment by industry (HSDB 1998). BBMP may enter the environment as fugitive dust and through wastewater (NTP 1996). BBMP was not identified as being released by industry into the environment through the Toxic Release Inventory (TRI 1996).

2.4 Environmental fate

BBMP is expected to remain in water for long periods of time (NTP 1996). No other environmental fate data could be found for BBMP.

2.5 Environmental exposure

The primary modes of potential human exposure to BBMP are inhalation, oral, and dermal contact. Consumer exposure may occur as a result of releases from products containing BBMP.

2.6 Occupational exposure

Occupational exposure to BBMP may occur in industries where it is used as a flame retardant in unsaturated polyester resins, in molded products, and in rigid polyurethane foam (NTP 1996). The National Institute of Occupational Safety and Health (NIOSH) did not survey BBMP to determine occupational exposure (NIOSH 1995, cited in NTP 1996).

2.7 Regulations

The U.S. EPA regulates BBMP under the Toxic Substances Control Act (TSCA). Table 2-1 summarizes the U.S. EPA health and safety data reporting regulations.

Table 2-1. U.S. EPA Regulations

U.S. EPA Regulations	
Regulatory action	Effect of regulation and other comments
40 CFR 716 – PART 716 – HEALTH AND SAFETY DATA REPORTING. Promulgated: 51 FR 32726, 09/15/86. U.S. Codes: 15 U.S.C. 2607(d). 2,2-Bis(bromomethyl)-1,3-propanediol has an effective date of 6/1/87 and a sunset date of 12/19/95.	This subpart sets forth requirements for the submission of lists and copies of health and safety studies on chemical substances and mixtures selected for priority consideration for testing rules under section 4(a) of TSCA and on other chemical substances and mixtures for which U.S. EPA requires health and safety information in fulfilling the purposes of TSCA.

Source: The regulations in this table have been updated through the 1998 Code of Federal Regulations 40 CFR, July 1, 1996; 21 CFR, April 1, 1996; 29 CFR, July 1, 1996.

3 Human Cancer Studies

There were no case reports or epidemiological studies on the occurrence of human cancer and exposure to BBMP or FR-1138.

4 Studies of Cancer in Experimental Animals

4.1 Carcinogenesis studies of BBMP

4.1.1 Carcinogenicity studies in rats

In a study conducted by the Dow Chemical Co., BBMP (FR-1138) was administered in the diet to male and female Sprague-Dawley rats for two years. The test substance (FR-1138) contained 80% 2,2-bis[bromomethyl]-1,3-propanediol, 8% 3-bromo-2,2-bis(bromomethyl)propanol, and 6% 2-(bromomethyl)-2-(hydroxymethyl)-1,3-propanediol. Dietary concentrations of BBMP were sufficient to deliver daily doses of 0, 5, or 100 mg/kg/day. These doses are equivalent to 0, 0.2, and 2.9%, respectively, of the BBMP oral LD_{50} in male rats. The number of rats used for the 0, 5, or 100 mg/kg/day doses were 48, 50, or 50, respectively, for females and 50 for controls and per dose group for males. BBMP administration had no effect on food consumption, weight gain, clinical signs, or survival of the rats, suggesting that the animals may have been able to tolerate higher doses. At the termination of the study, representative samples of all major organs of all surviving rats were necropsied. Upon statistical analysis of tumors found in both control and treated groups of rats, it was concluded that no treatment-related neoplasms were observed in rats of either sex (Keyes *et al.* 1980).

In a range-finding study conducted by the NTP, technical grade BBMP, as the commercial flame retardant FR-1138 (78.6% 2,2-bis[bromomethyl]-1,3-propanediol, 6.6% 2,2-bis[hydroxymethyl]-1-bromo-3-hydroxypropane, 6.9% 2,2-bis[bromomethyl]-1-bromo-3-hydroxypropane, 0.2% pentaerythritol, and 7.7% dimers and structural isomers), was administered in the diet at concentrations of 0, 1,250, 2,500, 5,000, 10,000, or 20,000 ppm to F344/N rats (10/sex) for 13 weeks.. Based upon food consumption, average daily doses of BBMP in the 13-week study were 100, 200, 400, 800, and 1700 mg/kg-body weight for males, and 100, 200, 400, 800, and 1,630 mg/kg-body weight for females. All rats survived to the end of the 13-week study. Male and female rats fed diets containing BBMP at concentrations ≥ 10,000 ppm exhibited dose-related decrements in body weight gain. BBMP-associated microscopic lesions of the kidney and urinary bladder were observed in rats of both sexes in the 13-week study. Nine of 10 male rats administered BBMP at 20,000 ppm for 13 weeks had hyperplasia of the transitional cells of the urinary bladder, whereas none of the 10 high-dose female rats had a similar change. Papillary degeneration of the kidney was observed in 3/10, 6/10, and 8/10 male rats at BBMP concentrations of 5,000, 10,000, or 20,000 ppm, respectively. One of 10 female rats in the high-dose group had papillary degeneration of the kidney. Based on the body-weight effects and the urinary bladder and/or kidney lesions in rats in the 13-week studies, the high dose selected for the two-year NTP rat study was 10,000 ppm (Elwell *et al.* 1989, cited in NTP 1996).

In the NTP two-year cancer bioassay, groups of F344/N rats (60/sex) received BBMP in feed at concentrations of 2,500, 5,000, or 10,000 ppm for 104 to 105 weeks. The control group for male rats contained 70 animals, and the control group for female rats contained 60 animals. Up to 10 male and female rats from each group were evaluated at 15 months. A 3-month “stop exposure study” was also conducted in male rats. For this study, an

additional group of 70 male rats received 20,000 ppm BBMP in feed for 3 months and then control diet for the remainder of the 2-year dosing period. At 3 months, 10 male rats, each, from the control and 20,000-ppm groups, were evaluated for histopathologic lesions. No neoplastic lesions were observed at this interim sacrifice (data not shown in this report). Based upon food consumption, average daily doses of BBMP in the two-year study were 100, 200, and 430 mg/kg, for males, and 115, 230, and 460 mg/kg, for females, in the continuous exposure study. In the stop-exposure study, male rats received an average daily dose of 800 mg/kg for 13 weeks followed by standard diet only (NTP 1996).

After three weeks of the two-year study, the mean body weights of rats 10,000-ppm groups were approximately 10% and 4% lower than those of the control group for males and females, respectively. The mean body weights of male and female rats administered BBMP at 10,000 ppm remained lower than those of the controls throughout most of the study. After three weeks, mean body weights of males in the 20,000-ppm group were 20% lower than those of controls, and this decrement persisted until the dosed feed was replaced by standard diet (after 13 weeks). Mean body weights for the 20,000-ppm group remained consistently 5% to 15% lower than those of controls for the duration of the study. The survival of male and female rats in the 2,500-ppm groups (20 male and 27 female rats survived) was similar to that of controls (26 male and 36 female rats survived). However, tumor development caused early deaths in the higher-dose groups. Survival of animals in the 5,000-ppm groups (13 male and 23 female rats survived) or 10,000-ppm groups (1 male and 5 female rats survived) was significantly less than that of the controls. None of the animals in the stop-exposure group (20,000-ppm) survived to the scheduled termination of the study. Neoplastic lesions were not observed in control or 20,000-ppm rats evaluated at three months. A few neoplasms were seen in male and female rats at 15 months, but there was no clear treatment-related neoplastic response at that time (data not shown). Due to the marked decrease in survivability of the treated animals, the tumor incidences were partially evaluated using Life Table analysis.

Administration of BBMP for two years caused increased incidences of neoplasms in multiple organs of rats of both sexes, with males exhibiting a wider array of affected organs than females. Treatment-associated neoplasms, appearing exclusively in male rats, are summarized in Table 4-1.

Table 4-1. Treatment-related neoplasms and proliferative, nonneoplastic lesions in male F344/N rats administered BBMP in the diet for up to two years

Tumor type	Dietary concentration of BBMP (ppm)				
	0	2,500	5,000	10,000	20,000[a]
	Tumor response/No. examined				
Skin[b]					
Squamous cell papilloma	1/51	0/53	2/51	5/55	11/59**
Keratoacanthoma	3/51	5/53	11/51**	16/55**	10/59**
Squamous cell carcinoma	0/51	0/53	0/51	0/55	1/59
Trichoepithelioma	0/51	0/53	0/51	1/55	1/59
Sebaceous gland adenoma	0/51	1/53	0/51	2/55	2/59
Basal cell adenoma	0/51	1/53	0/51	3/55**	6/59**
Basal cell carcinoma	0/51	0/53	2/51	2/55	0/59
All skin tumors combined	4/51	6/53	14/51**	24/55**	21/59**
Subcutaneous tissue[c]					
Fibroma	2/51	8/53*	11/51**	15/55**	7/59**
Fibrosarcoma/sarcoma	0/51	1/53	2/51	3/55**	3/59
Fibroma, fibrosarcoma, or sarcoma	2/51	9/53*	13/51**	16/55**	10/59**
Zymbal gland[c]					
Adenoma	0/51	0/53	1/51	3/55*	2/60
Carcinoma	2/51	1/53	3/51	2/55	15/60**
Adenoma/carcinoma	2/51	1/53	4/51	5/55	15/60**
Forestomach[b]					
Squamous cell papilloma	0/51	0/53	0/51	1/55	5/60*
Small intestine[b]					
Adenoma	0/51	0/53	0/51	0/53	1/59
Carcinoma	0/51	0/53	0/51	2/53	4/59
Adenoma/carcinoma	0/51	0/53	0/51	2/53	5/59*
Large intestine[b]					
Adenoma	0/51	0/53	3/51	4/55	10/59**
Carcinoma	0/51	0/53	0/51	0/55	2/59
Adenoma/carcinoma	0/51	0/53	3/51	4/55	11/59**
Peritoneum[b]					
Malignant mesothelioma	0/51	3/53	8/51**	9/55**	26/60**
Urinary bladder					
Transitional cell hyperplasia	0/51	0/53	1/51	3/55	10/59**
Transitional cell papilloma	0/51	0/53	1/51	2/55	1/59
Transitional cell carcinoma	0/51	0/53	0/51	1/55	1/59
Transitional cell papilloma/carcinoma	0/51	0/53	1/51	3/55	2/59

Tumor type	Dietary concentration of BBMP (ppm)				
	0	2,500	5,000	10,000	20,000[a]
	Tumor response/No. examined				
Lung[b]					
Alveolar/bronchiolar adenoma	1/51	0/53	3/51	1/55	4/60
Alveolar/bronchiolar carcinoma	0/51	1/53	0/51	3/55*	3/60
Alveolar/bronchiolar adenoma/carcinoma	1/51	1/53	3/51	4/55*	7/60*
Squamous cell carcinoma	0/51	0/53	0/51	0/55	3/60
Seminal vesicle[b]					
Hyperplasia	1/51	6/53	4/51	16/55**	33/60**
Adenoma/carcinoma	0/51	0/53	0/51	0/55	2/60
Hematopoietic system[c]					
Mononuclear cell leukemia	27/51	29/53	40/51**	34/55**	25/60**
Pancreas[b]					
Acinar cell focal hyperplasia	3/51	9/53*	12/51*	14/53**	27/59**
Acinar cell adenoma	1/51	2/53	4/51*	3/53	3/59

Source: NTP (1996)
[a] Dosing was terminated after 13 weeks for stop-exposure portion of the two-year study.
[b] Statistical significance by logistic regression test: *$P < 0.05$; **$P < 0.01$ vs. controls.
[c] Statistical significance by Life Tables Analysis: *$P < 0.05$; **$P < 0.01$ vs. controls.

In addition to a wide variety of tissues and organs exhibiting proliferative changes in male rats in the continuous-feeding portion of the study, the presence of a high incidence of neoplasms in animals in the stop-exposure portion of the study is noteworthy. Male rats receiving BBMP at 20,000 ppm for only 13 weeks, then fed standard diet for the remainder of the study generally exhibited essentially the same pattern of proliferative changes as male rats in the continuous-feeding study.

The magnitude of the proliferative response was generally similar between the stop- and continuous-exposure groups. In the case of the Zymbal gland, however, the stop-exposure group had approximately three times as many tumors as either the 5,000- or 10,000-ppm continuous-exposure groups. Further, the Zymbal gland lesions in the stop-exposure group were nearly all malignant, whereas in males in the 10,000-ppm continuous-exposure group, the only statistically significant increase in Zymbal gland tumor incidence was in adenomas. The stop-exposure group also had a higher incidence of malignant mesotheliomas (26/60, 43%) than the continuous-exposure groups (controls, 0; 2,500 ppm, 3/53, 6%; 5,000 ppm, 8/51, 16%; 10,000 ppm, 9/55, 16%).

The presence of a statistically significant incidence of alveolar/bronchiolar carcinomas in male rats in the 10,000-ppm continuous-exposure group (3/55, 5%) is noteworthy, as this is a rare tumor in F344/N rats. The historical control incidences of alveolar/bronchiolar carcinomas in untreated-control F344/N male rats in the NTP database is only 12/1,350 (0.9%). Animals in the stop-exposure group exhibited a full continuum of alveolar/bronchiolar changes (i.e., hyperplasia, adenoma, and carcinoma). Furthermore,

three animals in the stop-exposure group had squamous cell carcinoma of the lung, a lesion that has never been observed in untreated F344/N rats in the NTP database.

Proliferative changes in the oral mucosa (squamous-cell papillomas) increased in a dose-related manner, and the incidence in the stop-exposure group was similar to that in the 10,000-ppm continuous-exposure group. Although such changes did not appear in the esophagus of rats in the stop-exposure group, forestomach papillomas, as well as adenomas and carcinomas of the small and large intestines, were significantly increased. Clearly, in the stop-exposure group, cellular changes leading to the development of papillomas of the oral cavity and alimentary canal occurred early and persisted until manifestation of the neoplasms closer to the conclusion of the experiment. There was no evidence of BBMP-associated changes in the oral cavity and alimentary canal of animals sacrificed at the 13-week interim sacrifice.

BBMP administered at 20,000 ppm caused the early deaths of all treated male rats. These early deaths were attributed primarily to the carcinogenic effects of the chemical. Of the 59 animals in the two-year group of the stop-exposure study, 55 (93%) were sacrificed in moribund condition due to development of tumors, and several animals in that group had highly aggressive, life-threatening malignancies.

The administration of BBMP caused increased incidences of several neoplasms in both male and female rats. The incidences of these tumors are summarized in Table 4-2.

Table 4-2. Treatment-related neoplasms and nonneoplastic lesions in male and female F344/N rats administered BBMP in the diet for up to two years

Tumor type	Dietary concentration of BBMP(ppm)				
	0	2,500	5,000	10,000	20,000[a]
	Tumor response/No. examined[b]				
Mammary Gland					
Males					
Fibroadenoma	0/51	4/53*	6/51**	6/55**	5/60**
Adenoma	0/51	0/53	1/51	1/55	0/60
Adenoma/fibrosarcoma	0/51	4/53*	7/51**	7/55**	5/60**
Females					
Fibroadenoma (single or multiple)	25/50	45/51**	46/53**	45/52**	—
Fibroadenoma (multiple)	6/50	37/51**	40/53**	37/52**	—
Adenoma	0/50	2/51	0/53	0/52	—
Carcinoma	4/50	4/51	3/53	4/52	—
All tumors combined	27/50	47/51**	47/53**	47/52**	—
Oral Cavity					
Males					
Squamous cell papilloma	0/51	4/53*	8/51**	10/55**	12/60**
Squamous cell carcinoma	0/51	0/53	1/51	0/55	2/60
Squamous cell papilloma/carcinoma	0/51	4/53*	9/51**	10/55**	13/60**
Females					
Squamous cell papilloma	2/50	2/51	4/53	5/52	—
Squamous cell carcinoma	0/50	1/51	1/53	1/52	—
Squamous cell papilloma/carcinoma	2/50	3/51	5/53	6/52	—
Esophagus					
Males					
Squamous cell papilloma	0/51	0/53	1/51	5/55*	0/60
Squamous cell carcinoma	0/51	0/53	0/51	1/55	0/60
Females					
Squamous cell papilloma	0/50	0/51	1/53	10/52**	—
Kidney					
Males					
Papillary epithelial hyperplasia	10/51	20/53**	25/51**	47/55**	21/59*
Transitional cell hyperplasia	0/51	0/53	0/51	4/55	4/59
Transitional cell carcinoma	0/51	0/53	0/51	0/55	1/59
Renal tubule adenoma	0/51	0/53	1/51	3/55**	1/59
Females					
Papillary epithelial hyperplasia	0/50	1/51	1/53	7/52**	—
Renal tubule adenoma	0/50	1/51	0/53	0/52	—

Tumor type	Dietary concentration of BBMP(ppm)				
	0	2,500	5,000	10,000	20,000[a]
	Tumor response/No. examined[b]				
Thyroid					
Males					
Follicular cell hyperplasia	1/51	0/53	2/51	5/55	6/59*
Follicular cell adenoma	0/51	1/53	2/51	2/55	7/59*
Follicular cell carcinoma	0/51	1/53	4/51*	1/55	2/59
Adenoma/carcinoma	0/51	2/53	6/51*	3/55	9/59**
Females					
Follicular cell adenoma	0/50	0/51	2/53	3/52*	—
Follicular cell carcinoma	0/50	0/51	0/53	1/52	—
Adenoma/carcinoma	0/50	0/51	2/53	4/52**	—

Source: NTP (1996)

[a] Dosing was terminated after 13 weeks for the stop-exposure portion of the two-year study.

[b] Statistical significance by logistic regression test: *$P < 0.05$; **$P < 0.01$ vs. controls.

—, No data.

The variety of tissues and organs showing proliferative effects in response to BBMP was clearly greater in males than in females. In addition, the significantly increased incidences of neoplasms in females tended to be restricted to benign tumors. Explanations for these sex-associated differences are not apparent. The NTP (1996) concluded that under the conditions of this two-year dietary bioassay, BBMP showed *clear evidence of carcinogenic activity* in male and female F344/N rats, based on increased incidences of neoplasms of the skin, subcutaneous tissue, mammary gland, Zymbal gland, oral cavity, esophagus, forestomach, small and large intestines, mesothelium, urinary bladder, lung, thyroid gland, and seminal vesicle and increased incidence of mononuclear cell leukemia in male rats and increased incidences of neoplasms of the oral cavity, esophagus, mammary gland, and thyroid gland in female rats.

4.1.2 Carcinogenicity studies in mice

Technical grade BBMP, as the commercial flame retardant FR-1138 [78.6% 2,2-bis(bromomethyl)-1,3-propanediol, 6.6% 2,2-bis(hydroxymethyl)-1-bromo-3-hydroxypropane, 6.9% 2,2-bis(bromomethyl)-1-bromo-3-hydroxypropane, 0.2% pentaerythritol, and 7.7% dimers and structural isomers], was administered in the diet to male and female $B6C3F_1$ mice for 13 weeks in a range-finding study for a cancer bioassay (Elwell *et al.* 1989, cited by NTP 1996). During the 13-week study, groups of 10 mice of each sex were fed diets containing BBMP at concentrations of 0, 625, 1,250, 2,500, 5,000, or 10,000 ppm. Based upon food consumption, male mice received average daily doses of 100, 200, 500, 1300, or 3000 mg/kg-body weight while females received 140, 300, 600, 1200, or 2900 mg/kg-body weight. Five male and four female mice died during the study. All BBMP-dosed female mice and male mice receiving BBMP at concentrations` ≥ 1,250 ppm BBMP exhibited significantly reduced body weight gains. In the 13-week study, BBMP-associated microscopic lesions of the kidney and urinary bladder were observed in mice of both sexes. Urinary bladder transitional cell hyperplasia

was observed in 4/10 and 7/8 male mice in the 5,000-ppm and 10,000-ppm groups, respectively, and in almost all female mice in the 5,000-ppm or 10,000-ppm groups (10/10 and 9/10, respectively). In both male and female mice, the prominent renal lesion was papillary necrosis with evidence of tubular cell regeneration. These effects were more prevalent in males (4 –to 9 of 10 animals in the 2,500, 5,000, or 10,000-ppm groups)and were noted only among high-dose females with 4/10 having tubular cell regeneration and 2/10 having papillary necrosis.

In the two-year cancer bioassay by the NTP, groups of B6C3F_1 mice (60/sex) received diets containing BBMP at concentrations of 0, 312, 625, or 1,250 ppm for 104 to 105 weeks (NTP 1996). Based upon food consumption, average daily doses of BBMP were 35, 70, or 140 mg/kg and 40, 80, or 170 mg/kg for females. Dietary concentrations for the two-year feeding studies were based upon effects on body weight gains and treatment-associated kidney/urinary bladder pathology observed during the 13-week range-finding studies. Survival rates of male and female mice in the 312- or 625-ppm groups were similar to those of the controls. Survival of animals in the 1,250-ppm group was significantly reduced, and females were more severely affected than males. Despite reduced survival at high dietary concentrations of BBMP, mean body weight gains and weight maintenance by exposed animals were similar to those of controls. Reduced survival among high-dose females was related to an increase in the number of rats sacrificed in moribund condition. Of the high-dose females, 29/60 (48%) of the animals were sacrificed in moribund condition, whereas 9/60 (15%), 14/60 (23%), and 14/60 (23%) were sacrificed in the control, low-dose, and mid-dose groups, respectively.

Although a few neoplasms were seen in male and female mice at 15 months, but no clear treatment-related neoplastic response was seen at that time (data not shown). However, administration of BBMP to male and female mice for 2 years increased the incidences of neoplasms in the Harderian gland, lung, and forestomach. While incidences of total Harderian gland tumors (adenomas plus carcinomas) were significantly increased in low-dose females and in mid- and high-dose males and females, the increases in low-dose females and males were generally attributable to changes in the incidence of adenomas. For high-dose female mice, a statistically significant increase in incidence of carcinomas was observed. Additionally, in the case of lung tumors, high-dose males exhibited a significantly increased incidence of alveolar/bronchiolar adenomas and carcinomas and a significant increase in adenoma multiplicity. Mid- and high-dose female mice had significantly increased incidences of alveolar/bronchiolar hyperplasia, adenomas, or carcinomas (combined).

Both male and female mice had increased incidences of squamous-cell tumors of the forestomach; however no tumors of the oral cavity or the intestinal tract were observed. These tumor responses are summarized in Table 4-3.

Table 4-3. Increased incidences of neoplasms and proliferative, nonneoplastic lesions in both male and female B6C3F$_1$ mice administered BBMP in the diet for up to two years

	Dietary concentration of BBMP(ppm)			
	0	313	625	1,250
Tumor type	Tumor response/No. examined[a]			
Harderian gland				
Males				
Adenoma	3/50	6/51	12/50**	18/49**
Carcinoma	1/50	1/51	4/50	4/49
Adenoma/carcinoma	4/50	7/51	16/50**	22/49**
Females				
Adenoma	2/52	6/50	8/51*	15/50**
Carcinoma	1/52	6/50	5/51	7/50*
Adenoma/carcinoma	3/52	12/50**	13/51**	19/50**
Lung				
Males				
Alveolar/bronchiolar adenoma	12/50	4/51	12/50	21/49*
Alveolar/bronchiolar adenoma (multiple)	0/50	0/51	4/50	10/49**
Alveolar/bronchiolar carcinoma	3/50	7/51	8/50	11/49**
Alveolar/bronchiolar adenoma/carcinoma	15/50	11/51	16/50	25/49*
Females				
Alveolar/bronchiolar hyperplasia	1/52	3/50	8/51**	15/50**
Alveolar/bronchiolar adenoma	3/52	3/50	9/51*	17/50**
Alveolar/bronchiolar carcinoma	2/52	2/50	6/51	5/50
Alveolar/bronchiolar adenoma/carcinoma	5/52	5/50	15/51*	19/50**
Forestomach				
Males				
Squamous cell papilloma	0/50	3/51	2/50	2/49
Squamous cell carcinoma	0/50	0/51	1/50	2/49
Squamous cell papilloma/carcinoma	0/50	3/51	3/50	4/49*
Females				
Squamous cell papilloma	0/52	1/50	5/51*	3/50*

Source: NTP (1996)

[a]Statistical significance by logistic regression test: *$P < 0.05$, **$P < 0.01$ vs. controls.

In addition to the neoplasms common to the two sexes, male and female mice exhibited proliferative changes appearing in only one sex. These changes are summarized in Table 4-4.

Table 4-4. Other neoplasms observed in B6C3F$_1$ mice of one sex administered BBMP in the diet for up to two years

	Dietary concentration (ppm)			
	0	312	625	1,250
Tumor type	Tumor response/No. examined[a]			
Males				
Kidney				
Renal tubular adenoma	0/49	0/51	3/50	2/49
Females				
Skin: subcutaneous tissue				
Fibrosarcoma	0/52	0/50	0/51	1/50
Sarcoma	0/52	1/50	4/51	11/50**
Fibrosarcoma/sarcoma	0/52	1/50	4/51	12/50**
Mammary gland				
Carcinoma	0/52	0/50	1/51	3/50
All organs				
Hemangioma/hemangiosarcoma	1/52	2/50	0/51	5/50*

Source: NTP (1996)
[a]Statistical significance by logistic regression test: *$P < 0.05$, **$P < 0.01$ vs. controls.

Although the increase was not statistically significant, the appearance of renal tubule adenomas in the kidneys of mid- and high-dose male mice is noteworthy because of the rarity of occurrence of this neoplasm. In the NTP database, the incidence of renal tubule adenoma was 3/1,466 (0.2%) in untreated control male B6C3F$_1$ mice. In the two-study, 5/99 (5%) males in the mid- and high-dose groups had this tumor.

The statistically significant increase in incidence of skin tumors noted in high-dose female mice was driven by an increased incidence of sarcomas. Like renal tubule adenomas in male mice, subcutaneous sarcomas are extremely rare in untreated female B6C3F$_1$ mice. The NTP database includes only 3/1,470 (0.2%) in untreated females with this tumor.

The NTP (1996) concluded that under the conditions of this two-year dietary bioassay, BBMP showed *clear evidence of carcinogenic activity* in male and female B6C3F$_1$ mice based on increased incidences of neoplasms of the Harderian gland, lung, and kidney in males; and increased incidences of neoplasms of the Harderian gland, lung, and subcutaneous tissue in females.

4.2 Summary

BBMP shows *clear evidence of carcinogenic activity* in F344 rats and B6C3F$_1$ mice of both sexes based on increased incidences of neoplasms in the tissues and organs of both species. In rats, these tissues and organs include the skin, subcutaneous tissue, mammary gland, Zymbal gland, oral cavity, esophagus, forestomach, small and large intestines,

mesothelium, urinary bladder, lung, thyroid gland, seminal vesicle and peripheral blood in males; and the oral cavity, esophagus, mammary gland, and thyroid gland in females. In mice, these tissues and organs include the Harderian gland, lung, and kidney in males; and Harderian gland, lung, and subcutaneous tissue in females. Based on the results of the stop-exposure studies, in which administration of BBMP to the male rats was stopped at 13 weeks and the animals were observed for an additional 100 weeks, BBMP induced early preneoplastic changes that developed to benign and malignant tumors in both species. The variety of tumors attributable to the stop-exposure administration of BBMP was nearly identical to that attributable to the continuous administration of the chemical. The target sites for the tumors differed between the sexes.

5 Genotoxicity

5.1 Prokaryotic systems

5.1.1 Induction of mutation in Salmonella typhimurium

The NTP (1996) conducted two studies to test the mutagenicity of technical grade BBMP in *Salmonella typhimurium* assays. In the first assay (Mortelmans *et al*. 1986), BBMP was tested for mutagenicity at concentrations ranging from 10 to 10,000 µg/plate in various *S. typhimurium* strains without metabolic activation or with metabolic activation by 10% S9 liver homogenate from Aroclor 1254-induced rats or hamsters. BBMP was nonmutagenic in all *S. typhimurium* strains tested with or without, metabolic activation (Mortelmans *et al*. 1986, cited in NTP 1996).

In the second assay (Zeiger *et al*. 1992, cited in NTP 1996), the mutagenicity of BBMP (analyzed purity of 84%) at (concentrations ranging from 10 to 6,666 µg/plate was tested) in *S. typhimurium* strains TA98 and TA100 without metabolic activation or with activation by 30% hamster- or rat- liver S9. BBMP was mutagenic in strain TA100 only in the presence of 30% liver S9 from Aroclor-1254-induced male Syrian hamsters. BBMP was not mutagenic in either *S. typhimurium* strain without metabolic activation, or with 30% rat-liver S9 metabolic activation.

5.2 Mammalian systems *in vitro*

5.2.1 Sister chromatid exchanges

The ability of BBMP to induce sister chromatid exchanges (SCE) was studied *in vitro* in Chinese hamster ovary (CHO) cells (Galloway *et al*. 1987, cited in NTP 1996). The results of this study are summarized in Table 5-1. BBMP was tested in CHO cells at doses ranging from 16.7 to 500 µg/mL in the absence of liver S9 metabolic activation and 800 to 1,200 µg/mL in the presence of liver S9 metabolic activation. No significant increase in the SCE frequency was observed in cultured CHO cells with or without S9 mix; small increases were seen in the presence of S9 but these results were judged equivocal.

5.2.2 Chromosomal aberrations

Galloway *et al*. (1987, cited in NTP 1996) investigated the induction of chromosomal aberrations (CA) in CHO cells exposed to BBMP. A concentration-related increase in CA was observed in CHO cells treated with BBMP at concentrations ranging from 400 to 700 µg/mL in the presence of rat-liver S9 metabolic activation (Table 5-2). The aberrations observed were considered unusual by the researchers because the majority of the breaks were preferentially located in the heterochromatic region of the long arm of the X chromosome. Without metabolic activation, no increase in CA was observed.

Table 5-1. Induction of SCE in CHO cells by BBMP

Conc. (µg/mL)	Total cells	No. of chromo-somes	No. of SCE	SCE/chromo-somes	SCE/ cell	Time in BrdU[a] (h)	Relative change of SCE/chromo-some[b](%)
-S9							
Summary; Negative							
0[f]	50	1,038	496	0.47	9.9	26.3	—
16.7	50	1,041	485	0.46	9.7	26.3	-2.50
16.7	50	1,041	485	0.46	9.7	26.3	-2.50
50	50	1,042	498	0.47	10.0	26.3	0.02
167	50	1,050	545	0.51	10.9	33.5[c]	8.62
500	0	—	—	—	—	33.5[c]	—
				$P = 0.077$[d]			
				$P = 0.077$[d]			
+S9							
Summary; Equivocal							
0[f]	50	1,050	496	0.47	9.9	25.5	—
800	50	1,048	556	0.53	11.1	25.5	12.31
800	50	1,048	556	0.53	11.1	25.5	12.31
1,000	50	1,047	590	0.56	11.8	25.5	19.29
1,200[e]	50	1,046	574	0.54	11.5	25.5	16.17
				$P = 0.004$			

Source: Study performed at Litton Bionetics, Inc. The detailed protocol and these data are presented in Galloway *et al.* (1987, cited in NTP 1996).

[a] BrdU = bromodeoxyuridine.

[b] SCE/chromosome in treated cells versus SCE/chromosome in solvent-control cells (dimethylsulfoxide).

[c] Because of chemical-induced cell cycle delay, incubation time was extended to provide sufficient cells for scoring.

[d] Significance of relative SCE/chromosome tested by the linear regression trend test vs. log of the dose.

[e] Marked toxicity noted at this dose level.

[f] Dimethylsulfoxide – negative control.

—, No data.

Table 5-2. Induction of chromosomal aberrations (CA) in CHO cells by BBMP

-S9					+S9				
Dose (µg/mL)	Total cells	No. of CA	CA/ cell	Cells with CA (%)	Dose (µg/mL)	Total cells	No. of CA	CA/ cell	Cells with CA (%)
Harvest time: 20.5 h[a]					Harvest time: 10.5 h				
Summary: Negative					Summary: Positive				
0[b]	100	1	0.01	1.0	0[b]	100	5	0.05	5.0
400	100	1	0.01	1.0	600	100	8	0.08	4.0
500	100	2	0.02	2.0	800	100	24	0.24	22.0[d]
600	100	0	0.00	0.0	1000	100	17	0.17	16.0[d]
700	0				1200	0			
				$P = 0.833$[c]					$P \leq 0.001$

Source: Study performed at Litton Bionetics, Inc. The detailed protocol and these data are presented in Galloway *et al.* (1987, cited in NTP 1996).

[a] Because of significant chemical-induced cycle delay, incubation time prior to addition of Colcemid was lengthened to provide sufficient metaphase cells at harvest.

[b] Dimethylsulfoxide – negative control.

[c] Significance of percent cells with aberrations tested by the linear regression trend test vs. log of the dose.

[d] Positive: $P < 0.05$.

5.3 Mammalian systems *in vivo*

5.3.1 Mouse bone marrow micronucleus test

The genotoxicity of BBMP was evaluated *in vivo* in male and female mice exposed to BBMP in feed at concentrations ranging from 625 to 10,000 ppm for 13 weeks (MacGregor *et al*. 1990, cited in NTP 1996). BBMP caused significant increases in micronucleated normochromatic erythrocytes (NCEs) in peripheral blood samples obtained from male mice exposed at the two highest BBMP concentrations, 5,000 and 10,000 ppm, and from female mice exposed at the three highest BBMP concentrations, 2,500, 5,000, and 10,000 ppm (Table 5-3).

Table 5-3. Frequency of micronucleated NCEs in mouse peripheral blood following dietary exposure to BBMP for 13 weeks[a]

Dose (ppm)	Micronucleated NCEs/1000 cells (mean ± SE)	Number of mice	Micronucleated NCEs/1000 cells (mean ± SE)	Number of mice
	Male		Female	
0	2.36 ± 0.17	10	1.46 ± 0.26	9
625	2.28 ± 0.29	8	1.86 ± 0.30	9
1,250	2.55 ± 0.18	10	1.86 ± 0.22	9
2,500	2.98 ± 0.21	10	2.72 ± 0.32[b]	9
5,000	3.80 ± 0.19[b]	10	4.26 ± 0.47[b]	9
10,000	9.30 ± 1.26[b]	7	11.81 ± 0.54[b]	9
	$P < 0.001$[c]		$P < 0.001$[c]	

Source: MacGregor *et al.* (1990, cited in NTP 1996).
[a] Ten thousand NCEs scored per animal. 0 ppm is the control.
[b] Significant response by pairwise comparison to control.
[c] Trend test.

Equivocal results were obtained in a mouse bone marrow micronucleus test where male mice received three gavage doses of BBMP (100 to 400 mg/kg) at 24-hour intervals (Table 5-4). The results of the initial trial were negative, but the second trial revealed a clear dose-related increase in micronucleated polychromatic erythrocytes (PCEs) (NTP 1996).

Table 5-4. Frequency of micronuclei in bone marrow cells of mice treated with BBMP by gavage[a]

Dose (mg/kg)	Micronucleated cells/1000 PCEs (mean ± SE)	Micronucleated cells/1000 PCEs (mean ± SE)
	Trial 1—Negative	Trial 2—Positive
0	1.4 ± 0.6	1.5 ± 0.5
100	0.7 ± 0.4	2.3 ± 0.3
200	2.5 ± 0.5	2.6 ± 0.7
300	2.0 ± 0.7	—
400[b]	1.2 ± 1.2	4.8 ± 1.2[d]
	$P = 0.220$[c]	$P = 0.000$[c]

Source: Study performed at Environmental Health Research and Testing Inc (NTP 1996).
[a] Two thousand PCEs scored per animal.
[b] Only 2 mice survived in this dose group.
[c] Trend test.
[d] Significantly different ($P < 0.008$) from control.
—, No data.

The mouse micronucleus test was repeated with male and female mice administered a single intraperitoneal injection (150, 300, or 600 mg/kg) of BBMP. Bone marrow samples were taken 48 hours after dosing. Although male mice showed a two-fold increase in the frequency of micronucleated PCEs, neither the trend test nor pairwise analyses gave statistically significant results. The response in females was stronger (similar to that seen in the 13-week dietary study, Table 5-3) and was considered to be positive evidence of the ability of BBMP to induce micronuclei in bone marrow cells of female mice (NTP 1996). Results of this mouse micronucleus study are summarized in Table 5-5.

Table 5-5. Frequency of micronuclei in bone marrow cells of mice treated with BBMP by intraperitoneal injection[a]

Dose (mg/kg)	Number of mice	Micronucleated cells/1000 PCEs (mean ± SE)	Number of mice	Micronucleated cells/1000 PCEs (mean ± SE)
	Male		Female	
0	4	1.5 ± 0.3	4	2.0 ± 0.4
150	4	3.2 ± 0.8[b]	4	2.7 ± 1.1
300	4	3.0 ± 0.7[b]	3	3.6 ± 0.9[b]
600	3	3.0 ± 1.0[b]	4	5.2 ± 0.5[b]
		$P = 0.150$[c]		$P = 0.003$[c]

Source: NTP (1996)
[a] One thousand PCEs scored per animal.
[b] Significantly different ($P < 0.008$) from control.
[c] Trend test.

5.4 Summary

BBMP was to be mutagenic in several *in vitro* and *in vivo* systems, but specific conditions of metabolic activation were required to observe mutagenicity. BBMP was mutagenic in *S. typhimurium* strains TA100 and TA1535 only in the presence of 30% liver S9 from induced hamsters. In cultured CHO cells, BBMP induced CA only with S9 metabolic activation; no induction of SCE was observed with or without activation. *In vivo* exposure to BBMP induced significant increases in the frequency of micronucleated erythrocytes in male and female mice under various treatment protocols.

6 Other Relevant Data

6.1 Absorption, distribution, metabolism, and excretion of BBMP

BBMP was not detected in tissues of rats orally administered 5 or 100 mg/kg/day BBMP (as the flame retardant FR-1138) in a lifetime oncogenicity study. However, there was a statistically increased level of bromide (< 10-fold over controls) in the liver, kidney, fat, and serum of male rats that received the 100 mg/kg/day dose. The concentration of bromide in the liver was comparable to the concentration in serum although kidney levels exceeded serum levels in a ratio of 2:3 (Keyes *et al.* 1980).

Male F344 rats received single doses of BBMP at 150, 300, or 600 mg/kg by gavage or 15 mg/kg by intravenous (i.v.) injection into the caudal vein. Doses were prepared by diluting [^{14}C]-BBMP (uniformly labeled) with unlabeled BBMP in ethanol, emulphor, and water at a ratio of 1:1:3 by volume, to administer 25 to 50 μCi/kg-body weight. BBMP was rapidly, and nearly completely, absorbed from the gastrointestinal tract of the rats. BBMP was rapidly excreted in the urine of the rats as the glucuronide conjugate, with < 10% of the total dose being excreted in feces and none being detected as exhaled volatiles or CO_2. The ^{14}C in bile consisted of > 99% of the same glucuronide conjugate. The amount of excreted BBMP was determined by analysis for ^{14}C in urine, feces, and tissue. The relative amounts of BBMP and radiolabeled metabolites in rat urine, plasma, and bile were analyzed via high-performance liquid chromatography. The major metabolite derived from BBMP in rat urine was identified as a glucuronide conjugate of BBMP (Sanders *et al.* 1995 [abstract]).

The absorption, tissue distribution, metabolism, and excretion of BBMP in B6C3F$_1$ mice has been studied. Mice received BBMP at either 150 mg/kg by gavage or 15 mg/kg by i.v. injection (N = 4/group). Doses were prepared by diluting [^{14}C]-BBMP (uniformly labeled) with unlabeled BBMP in ethanol, emulphor, and water at a ratio of 1:1:3 by volume, to administer 25 to 50 μCi/kg-body weight. BBMP was rapidly, and nearly completely, absorbed from the gastrointestinal tract of the mice and rapidly excreted in the urine as the glucuronide conjugate, with <10% of the total dose being excreted in feces and none being detected as exhaled volatiles or CO_2. The ^{14}C in bile consisted of >99% of the same glucuronide conjugate. The amount of excreted BBMP was determined by analysis for ^{14}C in urine, feces, and tissue. The relative amounts of BBMP and radiolabeled metabolites in mouse urine were analyzed via high-performance liquid chromatography (Sanders *et al.* 1995 [abstract]).

6.2 Summary

BBMP undergoes rapid conjugation and excretion following absorption from the gut in rats and mice. BBMP did not form reactive metabolites or accumulate in the tissues of either species. However, exposure of rats to BBMP significantly increased bromide concentrations in the liver, kidney, fat, and serum of the exposed rats.

7 References

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RESULTS OF A TWO-YEAR TOXICITY AND ONCONGENIC STUDY OF RATS INGESTING DIETS CONTAINING DIBROMONEOPENTYL GLYCOL (FR-1138)

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ABSTRACT: FR-1138 is being developed as a reactive flame retardant for unsaturated polyester resins and polyurethane foams. FR-1138 primarily consists of 80% dibromoneopentyl glycol, 8% tribromoneopentyl alcohol and 6% monobromoneopentyl triol. A lifetime toxicity dietary study of FR-1138 was conducted in rats to assess the potential for chronic toxicity and possible oncogenesis.

Rats ingesting the lower dietary level of 5 mg FR-1138/kg/day had no adverse effects related to the lifetime treatment. Rats ingesting a higher dietary level of 100 mg FR-1138/kg/day had some evidence of toxicity, including degenerative changes in the liver, eye and possibly thyroid gland; however, there was no oncogenic response, even when FR-1138 was administered at a sufficiently high dosage to induce some toxicity.

Analysis of selected tissues during the course of the study indicated a slight increase in bromide content in the tissues of rats ingesting the higher dose level of 100 mg FR-1138/kg/day. At the lower dose level of 5 mg FR-1138/kg/day, there was only a marginal increase in bromide content of some of the tissues, with most values in the same range as the controls.

INTRODUCTION

DIBROMONEOPENTYL GLYCOL (FR-1138) is being developed as a flame-retardant chemical. The primary market is in unsaturated polyester resins. It is also used as a reactive flame retardant for flexible and rigid polyurethane foams, and polymeric plasticizers. It differs from other flame retardants proposed for similar applications because it is bonded to the polyester polymers by ester linkage.

The acute oral LD_{50} in male rats was found to be 3458 mg/kg, with 95% confidence limits of 2810—4257 mg/kg (unpublished data, the Dow Chemical Com-

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pany, Midland, MI). Other acute toxicity studies showed that it was nonirritating to the eye and intact skin, not likely to be absorbed in toxic amounts, and slightly irritating to abraded skin.

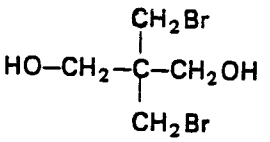
In a 30-day toxicity study conducted in this laboratory (unpublished data, The Dow Chemical Company, Midland, MI), male and female Sprague-Dawley Spartan substrain rats were given dose levels of 0, 10, 30, 100, or 300 mg/kg/day. The parameters evaluated included appearance, demeanor, body weight, food consumption, hematology, urinalysis, and clinical chemistry parameters including serum levels of urea nitrogen, alkaline phosphatase and glutamic pyruvic transaminase, organ weight and organ-to-body weight ratios, and gross and histopathologic examination. Effects attributed to treatment included a trend towards decreased body weight of male rats ingesting 300 mg/kg/day, and a statistical increase in liver weight of both males and females given the 300 mg/kg/day dose level. At 100 mg/kg/day, there was a trend towards increased liver weights. No untoward effects were observed at the lower dose levels of 30 and 10 mg FR-1138/kg/day.

Since the toxicologic effects associated with lifetime ingestion of FR-1138 have not been evaluated, the study reported herein was conducted to assess the toxicological and carcinogenic potential of FR-1138 when administered to rats in the diet throughout their lifetime. In addition, liver, kidney, fat and serum were saved for analysis for bromine content to determine if chronic ingestion of the test material is associated with an elevation of bromine levels in these tissues of the body.

EXPERIMENTAL

Test Material

The test material used in this study was supplied by the Halogens Research Laboratory, The Dow Chemical Company, Midland, MI. The analytical description of this lot of FR-1138 indicates that it contained approximately 6% monobromoneopentyl triol, 80% dibromoneopentyl glycol, 8% tribromoneopentyl alcohol and 3% other impurities. This analysis represents a recovery of approximately 97% of the original sample and was considered to be typical of normal production material. The chemical formula for dibromoneopentyl glycol (FR-1138) is as follows:



Experimental Design

Groups of male and female Sprague-Dawley (Spartan substrain) SPF-derived rats (Spartan Research Animals, Haslett, MI), 7–8 weeks of age were housed in suspended wire-bottom cages with 2 rats/cage, food (Purina Laboratory Chow, Ralston-

Purina Company, St. Louis, MO), and water were accessible at all times. The rats were randomized into test groups using a table of random numbers and were allowed to acclimate for a period of one week prior to placement on diets containing the test material. Individual rats were identified by metal ear tags and rats of each dose level were also identified by toe-clipping. Three groups consisting of 49–50 rats of each sex, plus 5/sex/group for the one year interim kill, and 10/sex/group for tissue analysis, were placed on diets supplying 0, 5 or 100 mg FR-1138/kg/day.

Diet Preparation and Analysis

The test diets were prepared by mixing FR-1138 with ground Purina Laboratory Chow to make a 5% premix. The concentration of the test material was adjusted on a weekly basis for the first 3 months, and quarterly thereafter to maintain the designated dose levels on a mg/kg of body weight/day basis according to the mean food consumption and body weight data. Control rats were supplied with untreated ground laboratory chow. Diet samples collected on 8 different dates during the study were submitted for neutron activation analysis for bromide.

Body Weight-Food Consumption

Body weights were recorded weekly for the first three months of the study on the first 20 rats/sex/dose level, and on all rats at monthly intervals. Food consumption was measured on 20 rats/sex/dose level during each week for the first three months and during one week of each month throughout the remainder of the study.

Clinical Observations

Rats were observed for general health status and possible toxicologic response during the study with observations recorded at least weekly for the first year of the study, and almost daily for the second year of the study.

Hematological determinations were conducted on blood samples collected from the tail veins from 10 rats/sex/group after 90–91 days and 356–357 days of treatment. Also, blood samples were collected from 6–10 rats/sex/group after 713–714 days, and additional bleedings of 10/group of the female rats after 725 and 731 days. Hematological parameters monitored included total erythrocyte count (RBC), total and differential leukocyte counts (WBC), packed cell volume (PCV), and hemoglobin (Hgb) concentration using automated (Coulter Counter Model ZBI and Hemoglobinometer, Coulter Electronics, Hialeah, FL), or manual procedures.

Urine samples were collected from identical numbers of rats as had been used for hematological determinations after 90–91, 356–357 and 713–714 days of treatment. Parameters measured (Ames Multilabstix, Ames Company, Elkhart, IN, and

TS Meter, AO Optical, Buffalo, NY) included urine specific gravity, pH, and the presence or absence of glucose, protein, ketones, bilirubin, occult blood and urobilinogen.

Clinical chemistry parameters were monitored from blood samples collected by orbital eye puncture after 94 days of treatment from 10 rats/sex/group. Blood samples from the rats killed after 1 year were collected by decapitation from 5 rats/sex/group. At the termination of the study after two years, blood samples were collected from a maximum of 10 rats/sex/group. Clinical chemistry parameters included blood urea nitrogen (BUN), serum glutamic pyruvic transaminase (SGPT), and serum alkaline phosphatase (AP). In addition, glucose values were measured on serum samples from rats which were part of the two-year terminal kill. Automated procedures (Technicon AutoAnalyzer, Technicon Corporation, Rye, NY (94 Days), Centrifichem System 400, Methods File, Union Carbide Corporation, Rye, NY (1-year and 2-year), were used for these determinations.

Bromide Analysis on Selected Tissues

Samples of adipose tissue, liver, kidney and serum were collected and the weights of liver and kidneys were determined from 3 or 4 rats/sex/group on days 10, 31 and 90 of the study. Tissues for analysis were also saved from 3 rats/sex/group at the 1-year interim kill and at termination of the study. Neutron activation analysis (Radio Chemical Research, The Dow Chemical Company, Midland, MI) was used to detect total bromide content of these tissue specimens.

Necropsy Examination

An interim necropsy examination was conducted after 1 year of treatment on 5 rats/sex/group that had been predesignated for this purpose. All rats were deprived of food overnight prior to killing by decapitation. The eyes of all rats were examined by gently pressing a glass slide against the cornea under bright fluorescent illumination. Any observations on the eyes were recorded as part of the gross necropsy observation records. The eyes from 5 rats/sex/group were preserved in Zenker's fixative. A complete gross pathologic examination was performed by a veterinary pathologist. Representative sections of all major organs and tissues were preserved in formalin fixative. These tissues included liver, kidneys, heart, pancreas, spleen, brain (cerebrum, cerebellum and brain stem), vertebrae with spinal cord, peripheral (sciatic) nerve, pituitary gland, stomach, small intestine, large intestine, cecum, mesenteric lymph node(s), skeletal (thigh) muscle, salivary gland, testes, epididymides, accessory male sex glands, urinary bladder, urerus, ovary trachea, esophagus, aorta, thoracic lymph node(s), thymus, lungs, integument, thyroid gland, parathyroid glands, adipose tissue, adrenal gland(s), sternum, tongue, mandible and any other grossly observed lesion. The weights of the liver, kidneys, brain, heart and testes (males) were recorded on all 5 rats/sex/group.

All rats which died or were culled during the course of the two-year study were

also subjected to a gross pathologic examination. Representative portions of all major organs and tissues as listed above, plus mandible, skull (including nasal turbinates, ear canal) along with any gross lesions suggestive of a significant pathologic process or tumor formation were collected from each rat and preserved in formalin fixative.

Terminal necropsy examination was conducted on all survivors at the end of 2 years of treatment, using procedures similar to those described for the 1-year interim necropsy to weigh and preserve in fixative all those organs and tissues listed above. Also, at the time of terminal necropsy, a femoral bone marrow smear was prepared from all female rats, and filed for future reference, if indicated.

Histologic Examination of Tissues

Histologic examination was conducted on paraffin embedded sections of tissues which were stained with hematoxylin and eosin (H&E). All rats from the control and treated groups killed after 1 year of treatment were subjected to histologic examination of an extensive list of tissues. All rats from all treatment and control groups of the 2-year study, regardless of whether they died, or were culled during the study, or were killed at the termination, were subjected to histologic examination of H&E stained sections of an extensive list of tissues from all organ systems of the body. Additional sections of livers from male and female rats from the control and treated groups of the terminal kill were stained with Oil Red O, for lipid content.

Statistical Evaluation of Data

Hematology, urinary and clinical chemistry parameters, body weights, organ weights, and organ/body weight ratio data were statistically analyzed by a one-way analysis of variance followed by Dunnett's Test, $p<0.05$ [1]. Food consumption data were analyzed using the sequential outlier's test [2], $P = 0.02$ (two-sided) to identify outlying data points. These data points were not included in the subsequent analysis of data using a one-way analysis of variance followed by Dunnett's Test, $p<0.05$. Data on mortality, palpable masses, gross pathology, histopathology and tumor incidences of the rats of the 2-year study were analyzed using Fisher's Exact Probability Test, $p<0.05$, one-sided test [3]. For gross pathology observations, statistical evaluation of the cumulative data for the entire study compared the data of each of the treatment groups against the data of the control group of that sex. The data were visually inspected, and those cases suggestive of a possible statistical difference from control were analyzed. For histopathology observations and tumor incidence, this statistical evaluation compared the cumulative data of each dose group against the data of the control group of that sex. The exact number of tissues examined was used as the total group size in each analysis performed. The data were visually inspected and those cases suggestive of a possible statistical difference from control were analyzed.

RESULTS AND DISCUSSION

Due to the voluminous nature of the data generated during this two-year toxicity study, most of the actual data can not be included in this publication; however, these are on file with the authors.

Dietary Content of FR-1138

Results of the analyses of diet samples collected on 8 different dates during the study, overall showed generally good agreement during the course of the study between the analytically-observed dietary content and the desired nominal content for each sex and dose level.

The results of 8 different analyses conducted on samples collected during months 5, 8, 13, 17, 18, 19, 20, and 22 indicated the following analytical content of diets (expressed as percentage of analytically observed/desired nominal content of test material):

Control	Group	None Detected
5 mg/kg/day	Female	95.5 ± 24.7%
5 mg/kg/day	Male	113.0 ± 46.0%
100 mg/kg/day	Female	97.8 ± 7.2%
100 mg/kg/day	Male	97.8 ± 4.4%

Body Weights

Although body weight data indicated a trend towards lower body weights of both groups of males given FR-1138 after 100 days on test, the only statistical decrease in body weight was noted on Day 272 in the group of males ingesting 100 mg of FR-1138/kg/day (Summary Table 1). In view of the relatively higher than normal male control body weights noted in the study when compared to historical control data from two previous 2-year dietary studies [4, 5], using rats of the same age and strain, this isolated statistical decrease was probably not related to treatment with 100 mg of FR-1138/kg/day. Female rats given 5 or 100 mg of FR-1138/kg/day showed no difference in body weights when compared to the controls.

Food Consumption

There were no consistent deviations in the food consumption of males or females at either of the 2 dose levels during the course of the 2-year study. The sporadic cases in which there was a statistical increase or decrease between the control and various treatment groups followed no consistent trend, and were considered of no toxicological significance.

Cumulative Mortality

Male rats given 5 or 100 mg/kg/day showed no differences in mortality when

Table 1. Summary of Major Observations in Male and Female Rats Maintained on Diets Containing FR-1138 (Dibromoneopentyl Glycol) For Up to Two Years. (Including selected data extracted from entire data base for the purpose of depicting effects considered related to treatment.)

Sex	Males			Females		
Dose (mg/kg/day)	0	5	100	0	5	100
Number of Rats in 2-Year Study	50	50	50	48	50	50
Number of Rats Killed After 1 Year	5	5	5	5	5	5
PARAMETER						
BODY WEIGHT (grams)	616±59	598±58	591[a]±53	340±33	337±23	337±25
FOOD CONSUMPTION	-	-	-	-	-	-
MORTALITY	-	-	-	-	-	-
PALPABLE MASSES	-	-	-	-	-	-
CLINICAL OBSERVATIONS	-	-	-	-	-	-
HEMATOLOGY (RBC, PCV, Hgb, WBC, differential WBC)	-	-	-	-	-	-
URINALYSIS (specific gravity, pH, protein, glucose, ketones, bilirubin, occult blood, urobilonogen)	-	-	-	-	-	-
CLINICAL CHEMISTRY (SGPT, BUN, AP, glucose)	-	-	-	-	-	-
ORGAN WEIGHTS						
Brain, heart, liver, testes	-	-	-	-	-	-
Kidneys (90-Day Kill Only) (gram/100 gram body weight)	0.65±0.03	0.67±0.02	0.70±0.03	0.64±0.04	0.68±0.02	0.71[a]±0.04
Liver (1-Year Kill Only) (gram/100 gram body weight)	2.74±0.41	3.11±0.25	3.30[a]±0.34	2.63±0.20	2.70±0.50	2.39±0.12
TISSUES FOR BROMINE ANALYSIS (elevated at all time periods measured) (ppm)						
Liver (1 year)	4.6±0.6	6.0±0.3	20[a]±3	4.7±0.6	4.6±0.3	10.1[a]±0.9
Kidney (1 year)	10±2	12±1	66[a]±34	10±1	12[a]±1	22[a]±1
Serum (1 year)	10±2	13±1	51[a]±5	10±0	10±2	27[a]±3
Fat (1 year)	2.7±0.3	2.7±0.5	8.3±0.7	4.2±1.5	3.2±0.4	6.6[a]±0.3

[a]Statistically different from control values(s) when analyzed using analysis of variance and Dunnett's Test, $p<0.05$ or Fisher's Exact Probability Test, $p<0.05$.

– Indicates parameter was considered unaffected at any level of treatment. For parameters with multiple observations, only selected representative values are listed in summary table.

Table 1. (Concluded).

GROSS AND HISTOPATHOLOGIC EXAMINATION (No. effected)						
1-YEAR KILL						
Liver - Increased hepatocellular centrilobular cytoplasmic homogeneity	0	0	5	0	0	0
2-YEAR STUDY						
Thyroid						
Thyroid retention cyst formation	1	0	7[a]	1	1	2
Liver						
Increased hepatocellular cytoplasmic homogeneity	0	0	0	1	0	14[a]
Several foci of hepatocellular alteration	12	8	18	7	7	13
Single area of hepatocellular alteration	5	4	5	5	9	13[a]
Eyes						
Bilateral diffuse opacity of lenses	0	0	2	0	0	6[a]
Bilateral lenticular degeneration, anterior cortex - moderate	0	0	0	0	0	6[a]
Bilateral lenticular degeneration, posterior cortex - moderate	0	0	0	0	1	3
Basophilic staining material, posterior cortex	0	0	0	0	0	5
Tumors (all organ systems)	-	-	-	-	-	-

[a]Statistically different from control value(s) when analyzed using analysis of variance and Dunnett's Test, $p<0.05$ or Fisher's Exact Probability Test, $p<0.05$.

— Indicates parameter was considered unaffected at any level of treatment. For parameters with multiple observations, only selected representative values are listed in summary table.

compared to that of the control group. Female rats receiving 100 mg/kg/day had statistically increased mortality rates for months 16 and 17. In view of the isolated occurrence, this observation was considered to be of questionable toxicological significance. Mortality data on female rats given 5 mg/kg/day showed no differences from control data.

Palpable Masses

There were no statistical differences between control and treated rats over the course of the two-year study and, thus, the incidence of palpable masses was considered to be unaffected by treatment. After approximately 1 year, the females given the higher dose level had a slight transient upward trend in the number of rats with palpable masses when compared to concurrent controls. However, this apparent transient upward trend was not considered related to treatment in view of the somewhat lower incidence noted in this concurrent control group when compared to historical control data.

Clinical Observations

There were no changes in appearance or demeanor of the rats during the course of the study that could be attributed to ingestion of FR-1138 in the diet.

Hematology

Repetitive hematological parameters monitored after approximately 90–91, 356–357, 713–714, 725, and 731 days showed no effects which were considered to be related to treatment. Isolated occurrences of statistical differences between the treated and control groups were encountered; however, due to either the lack of a dose response and/or comparison with historical control data, these were not considered to be of any toxicological significance.

Urinalysis

Routine examination of urinary parameters after approximately 90–91, 356–357 and 713–714 days revealed no observations for either males or females given 5 or 100 mg/kg/day which were considered to be the result of treatment.

Clinical Chemistry

Analyses of serum samples from rats killed after 94 days, 1 year, 2 years, measuring levels of BUN, SGPT, and AP showed no alterations considered to be the result of treatment. There was a statistical decrease in SGPT of male rats given 100 mg/kg/day. The slight decrease in SGPT was not considered to be of any toxicological significance. Glucose values of serum samples from rats terminated after two years showed no effects which could be considered to be the result of treatment.

female rats given 5 mg/kg/day. Microscopically, the group of female rats given 100 mg/kg/day showed six of eleven rats with bilateral lenticular degeneration of the anterior cortex graded as moderate in degree (Figure 2). In addition, some of the rats of this high dose level showed bilateral degeneration of the posterior cortex and basophilic staining material within this region of the lens (Figure 3). These changes in the posterior cortex of the lens were probably also a result of treatment with 100 mg/kg/day of FR-1138.

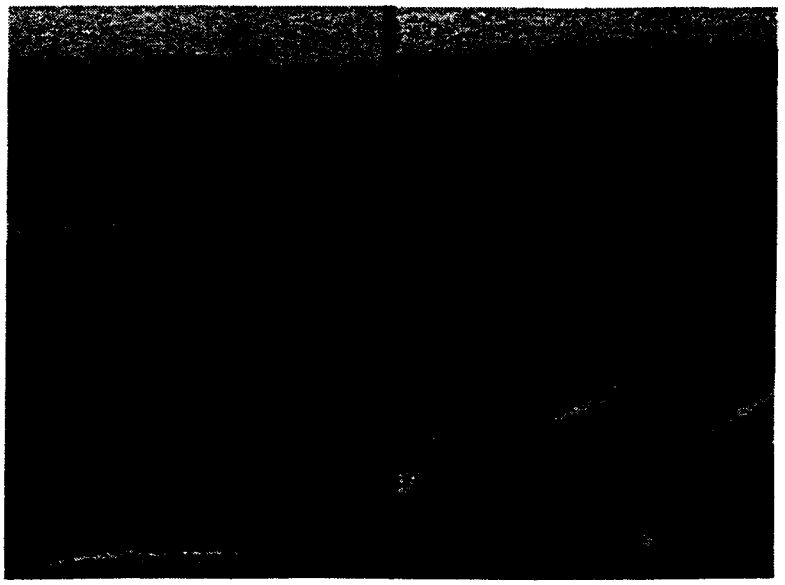


Figure 2. Note degeneration of anterior lenticular fibers of lens of female rat given 100 mg/kg/day of FR-1138 for 2 years (B) compared to control (A). Hematoxylin and eosin x 400.

These lenticular changes were similar in morphology and location to those experimentally induced by a number of agents, including galactose, xylose, lactose, corticosteroids or catecholamines [7]. The formation of cataracts, by high levels of the three sugars listed previously, had been proposed to be related to the formation and accumulation of sugar alcohols in the lens. In the lens, the enzyme aldose reductase converts sugar to the sugar alcohol form. The enzyme polyol dehydrogenase metabolizes some sugar alcohols, but not others including galactose. This explains the more severe cataractogenic activity of this sugar. When sugar alcohol concentrations increase within the lens, an osmotic change results with increased amounts of water being drawn into the lens to maintain an osmotic equilibrium. The increased fluid levels in the lens cause swelling and eventual rupture of the lens fibers leaving areas of degenerate protein. Once these degenerate fibers occur, they will persist and only progress if continued high sugar alcohol levels are maintained [7].

Organ Weights

Female rats given 100 mg/kg/day which were killed after 90 days showed a statistical increase in the relative kidney weight (Summary Table 1). This was not evident at any of the other time periods monitored. Due to its isolated occurrence, this alteration may or may not have been related to treatment. However, if it was related to treatment, it was only a transient phenomenon and was not seen after one or two years. Male rats given 100 mg/kg/day killed after 1 year of treatment showed a statistically significant increase in relative liver weights. This was considered to be the result of treatment. Male rats given 5 mg/kg/day which were killed after one year showed a statistical increase in the relative heart weight; this was not considered of any toxicological significance due to its isolated occurrence, lack of dose response, and absence at the termination of the 2-year study. There were no alterations or consistent trends in the weights of brain, or testes when compared to control values at any of the time periods.

Tissue Levels of Bromide

Results of tissue analysis for bromide content of male rats are summarized in Figure 1. Rats ingesting the high dose level of 100 mg/kg/day of FR-1138 had statistically increased level of bromide in liver, kidney, fat and serum. However these increased bromide levels, less than a 10-fold increase over the controls, were achieved relatively early and appeared to plateau during the remainder of the study, with the possible exception of the kidney in both sexes and fat in females, in which there was a slight upward trend which appeared to continue to some degree throughout the duration of the study. The concentration of bromide in the liver and fat never exceeded that in the serum any time during the study at either dose level of FR-1138 or in either sex. The concentration in the kidney exceeded that in the serum only in the rats killed at one and two years. The highest kidney to serum ratio, 2.3, was seen in the 100 mg/kg/day males at the termination of the study. In the group of rats ingesting 5 mg/kg/day of FR-1138, there was only a marginal increase in bromide content of some of the tissues measured, with most values in the same range as the controls. Overall, these data are interpreted to indicate that the compound FR-1138 does not have significant potential to bioaccumulate in mammalian tissues.

Gross and Histopathologic Observations

The voluminous data compiled during gross necropsy and histopathological examination of rats from the 1-year interim kill, spontaneous deaths and termination after 2 years are on file with the authors. Examples of the major effects considered to be related to treatment have been included in the attached Summary Table 1, and these pathology results are discussed below.

After 1 year of treatment, there were no gross observations which were consid-

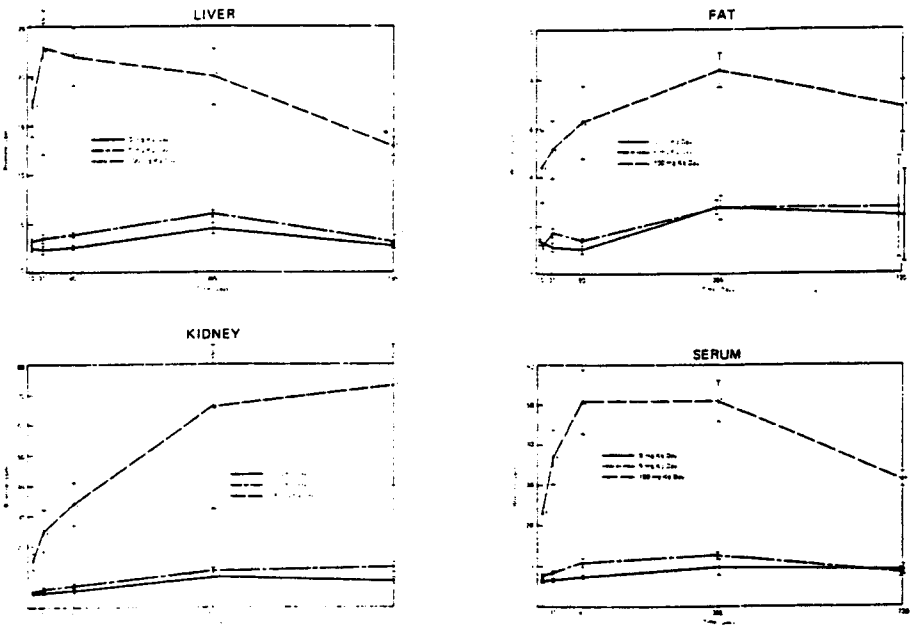


*Figure 1. Bromine levels (PPM) in male rats maintained on diets containing dibromoneopentyl glycol (FR-1138) for up to two years. (*Statistically different from control mean using analysis of variance and Dunnett's Test, $p<0.05$.*

ered to be related to treatment. Histopathologic examination indicated a slight liver alteration which was only seen in the group of male rats given 100 mg/kg/day. All of the rats within this group showed an increased centrilobular homogeneity of the hepatocellular cytoplasm. All other histopathologic observations were considered to be spontaneous in nature and unrelated to treatment.

The gross necropsy and histopathologic observations for rats that died, were culled or survived to the termination as part of the 2-year study indicated certain observations which were considered to be related to treatment. Histopathologic examination showed a statistical increase in the incidence of thyroid retention cyst formation in the group of male rats given 100 mg/kg/day. This observation may or may not have been the result of treatment, but there was no increase in follicular hypertrophy or hyperplasia. Male rats given 5 mg/kg/day showed no evidence of this alteration. A previously published subchronic study of sodium bromine attributed thyroid hyperplasia to treatment with relatively high levels of sodium bromide [6].

Gross necropsy and histopathologic examination of the lenses of eyes of female rats which were terminated after receiving 100 mg/kg/day for two years, indicated alterations which were considered to be the result of treatment. Gross necropsy examination revealed six of eleven female rats given 100 mg/kg/day with bilateral diffuse opacity of the lenses. This was not noted in the controls or the group of

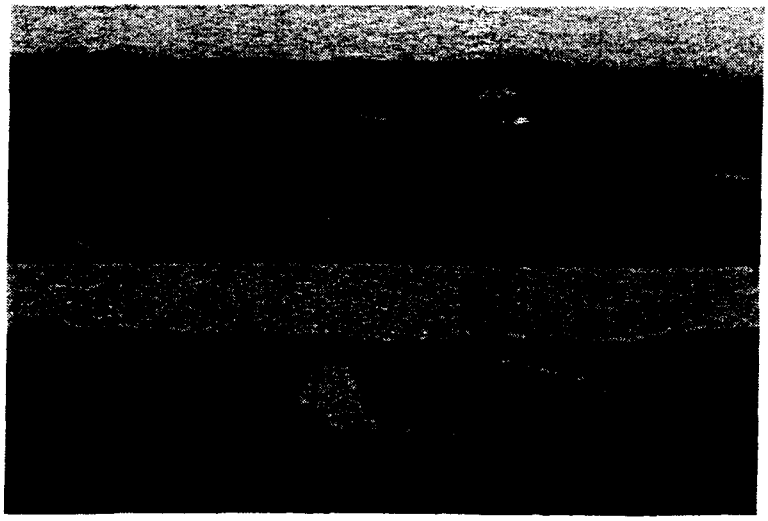


Figure 3. Note degeneration of posterior lenticular fibers and accumulation of basophilic staining material in lens of female rat given 100 mg/kg/day of FR-1138 for 2 years (B) compared to control (A). Hematoxylin and eosin x 400.

One possible explanation of the alterations noted in the lenses of female rats given 100 mg FR-1138/kg/day could be that metabolism of dibromoneopentyl glycol eliminated bromine and left a compound structurally similar to a sugar alcohol. This might explain the mechanism by which the lenticular degeneration occurred in this study.

Gross necropsy observations of female rats given 100 mg/kg/day showed a statistical increase in the number of rats bearing 3 subcutaneous masses in the mammary or midcervical region. This observation is not considered to be related to treatment due to the somewhat lower incidence of this observation in this control group when compared to historical control data [4, 5] and the absence of any increase in the number of rats of this group bearing 1, 2, 4, 5 or 6 subcutaneous masses in the mammary or midcervical region.

The livers of female rats given 100 mg/kg/day showed no gross evidence of toxicity; however, upon microscopic examination there were several observations noted primarily in the group of rats terminated after 2 years which were considered to be related to treatment. These included hepatocellular degenerative changes consisting of increased eosinophilic cytoplasmic homogeneity (Figure 4) which was accompanied by a slight increase in the incidence of livers having several foci or a single area of hepatocellular alteration. The trend towards a slight increased incidence of individual hepatocellular necrosis noted in this group may or may not have

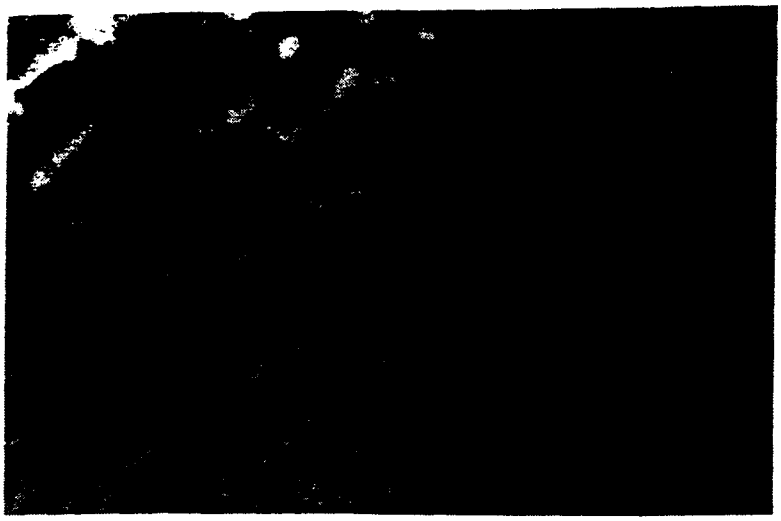


Figure 4. Note increased eosinophilic homogeneity of hepatocellular cytoplasm of female rat given 100 mg/kg/day of FR-1138 for 2 years (B) compared to control (A). Hematoxylin and eosin x 400.

been the result of treatment. Oil Red O stain for lipid revealed no discernible difference between control and treated groups.

Gross and histopathologic examination of tissues of the urinary tract, cardiovascular system, respiratory tract, reproductive system, endocrine organs (except thyroid), gastrointestinal tract, tongue, mesenteric tissue, pancreas, salivary glands, musculoskeletal system, lymphoreticular system, central nervous system, peripheral nerve, subcutaneous tissues, integument and mammary glands revealed various geriatric and inflammatory processes in all control and experimental groups which were of the type and severity historically encountered in rats of this strain and age, and were considered as spontaneous in origin and unrelated to ingestion of these dose levels of FR-1138.

Tumor Incidence

Tumor incidence rates for male and female rats are tabulated in Tables 2 and 3, respectively. These data are listed for each of the sequential 6-month periods as well as the terminal kill. The total results for each category of tumor type are also included. The spectrum of tumors that have been historically observed in rats of this strain were noted in the liver, nasal turbinates/hard palate, lungs, pancreas, kidney, urinary bladder, testes, ovary, uterus, musculoskeletal tissue, oral cavity, tongue, salivary glands, stomach, small intestine, large intestine, subcutaneous tis-

Table 2. Tumor Incidence in Male Rats Maintained on Diets Containing FR-1138 (Dibromoneopentyl Glycol) For Up to Two Years.

Time Interval	Months 13-18			Months 19-24			Terminal Kill			Total		
Dose Level (mg/kg/day)	0	5	100	0	5	100	0	5	100	0	5	100
Number of Rats Examined	13	12	12	25	26	26	9	7	6	50	50	50
Tumor/Tumor-like Lesions												
Hepatocellular hyperplastic nodule(s)	0	0	0	0	0	0	1	0	0	1	0	0
Hepatocellular carcinoma(s)	0	0	0	1	1	0	0	0	0	1	1	0
Renal tubular adenocarcinoma	0	0	0	1	0	1	0	0	0	1	0	1
Osteoma of nasal turbinates	0	0	0	0	1	0	0	0	0	0	1	0
Interstitial cell tumor of testis	0	0	0	1	0	1	0	0	3	1	0	4
Interstitial cell tumor - malignant of testis	0	0	0	1	0	0	0	0	0	1	0	0
Total interstitial cell tumors of testis	0	0	0	1	0	1	0	0	3	2	0	4
Stratified squamous cell carcinoma of tongue	1	0	0	0	0	2	0	0	0	1	0	2
Squamous papilloma of tongue	0	0	0	0	1	0	0	0	0	0	1	0
Squamous polyp of stomach	1	0	0	0	0	1	0	0	0	1	0	1
Mucocystadenocarcinoma of stomach	0	0	0	0	1	0	0	0	0	0	1	0
Squamous cell carcinoma of stomach	0	0	0	0	0	1	0	0	0	0	0	1
Mucocystadenocarcinoma of small intestine	1	0	0	0	1	1	0	0	0	1	1	1
Lymphosarcoma of small intestine	0	0	0	0	1	1	0	0	0	0	1	1
Leiomyosarcoma of small intestine	0	0	0	0	1	1	0	0	0	0	1	1
Mucocystadenoma of small intestine	0	0	0	0	1	1	0	0	0	0	1	1
Lymphosarcoma of large intestine with metastasis	0	0	0	0	1	1	0	0	0	0	1	1
Intraabdominal malignant neoplasm with metastasis	0	0	0	0	1	1	0	0	0	0	1	1
Astrocytoma of brain	0	0	0	0	1	0	0	0	0	0	1	0
Granular cell myoblastoma of brain	0	0	0	1	1	0	0	0	0	1	1	0
Malignant tumor of pineal gland	0	0	0	0	0	0	0	0	0	0	0	0
Mixed glioma of spinal cord	0	0	0	0	0	0	0	0	0	0	0	0
Oligodendroglioma of spinal cord	0	0	0	0	0	0	0	0	0	0	0	0
Malignant schwannoma involving epididymis	0	0	0	0	0	0	0	0	0	0	0	0
Subauricular malignant schwannoma	0	0	1	0	0	1	0	0	0	0	0	1
Generalized lymphosarcoma with widespread metastasis	0	0	0	0	0	0	0	0	0	0	0	0
Myelomonocytic leukemia (generalized)	0	0	1	0	0	1	0	0	0	0	0	1
Hemangioma of mesenteric lymph node	0	0	0	0	2	0	0	0	0	0	2	0
				1	1	1	0	0	0	1	1	1
					0	0				1	0	0

During Months 1-6, there was only one tumor (subcutaneous carcinosarcoma) noted in the control group. Tumors occurring during Months 7-12 included one adrenal pheochromocytoma - unilateral (control), one subcutaneous fibrosarcoma with metastasis (control), one mixed glioma of the spinal cord (5 mg/kg/day), one adrenal pheochromocytoma - unilateral (5 mg/kg/day), one mammary gland fibroadenoma/adenofibroma (5 mg/kg/day), one pancreatic islet cell adenoma (100 mg/kg/day), and one mammary gland fibroadenoma/adenofibroma (100 mg/kg/day). All of these tumors have been included in the above total tabulation.

There were no statistical differences from control values when analyzed using Fisher's Exact Probability Test, $p<0.05$.

Table 2. (Continued).

Time Interval	Months 13-18			Months 19-24			Terminal Kill			Total		
Dose Level (mg/kg/day)	0	5	100	0	5	100	0	5	100	0	5	100
Number of Rats Examined	13	12	12	25	26	26	9	7	6	50	50	50
Tumor/Tumor-like Lesions (cont'd)												
Pituitary adenoma formation(s)	1	0	0	2	6	6	0	2	2	3	8	8
Pituitary adenocarcinoma without extension or invasion of brain	0	0	0	0	0	0	0	1	1	0	1	1
Pituitary adenocarcinoma with invasion of brain	0	1	0	0	0	0	0	0	0	0	1	0
Unclassified tumor of pituitary	1	0	0	0	0	0	0	0	0	1	0	0
Adrenal adenoma - unilateral	0	0	1	3	0	2	0	1	1	3	1	4
Adrenal adenoma - bilateral	0	0	1	0	0	0	0	0	0	0	0	1
Adrenal adenocarcinoma	1	0	0	0	0	1	0	0	0	1	0	1
Adrenal adenocarcinoma with metastasis	1	0	0	0	0	0	0	0	0	1	0	0
Adrenal pheochromocytoma - unilateral	0	1	1	8	5	7	1	1	0	10	8	8
Adrenal pheochromocytoma - bilateral	0	1	0	1	0	2	0	0	0	1	1	2
Adrenal pheochromocytoma - malignant, no metastasis - unilateral	0	0	0	1	1	0	1	0	2	2	1	2
Adrenal pheochromocytoma - malignant, metastasis - unilateral	0	0	0	0	0	0	1	0	0	1	0	0
Thyroid interfollicular C-cell adenoma(s)	0	0	1	1	1	2	2	2	1	3	3	4
Thyroid interfollicular C-cell adenocarcinoma without metastasis	0	0	0	3	1	1	2	0	0	5	1	1
Pancreatic acinar adenoma	2/1*	2/2	5/3	16/6	15/11	28/7	2/2	6/4	3/1	20/9	23/17	36/11
Pancreatic acinar adenocarcinoma	0	0	0	1/1	0	0	0	0	0	1/1	0	0
Pancreatic islet cell adenoma	0	0	0	4/3	4/4	4/4	1/1	1/1	0/0	5/4	5/5	5/5
Pancreatic islet cell adenocarcinoma	0	0	0	0	0	2/2	0	0	0	0	0	2/2
Subcutaneous fibroma	0	0	1/1	1/1	2/2	5/4	1/1	0/0	2/2	2/2	2/2	8/7
Subcutaneous fibrosarcoma with metastasis	0	0	0	0	0	0	0	0	0	1/1	0	0
Subcutaneous carcinosarcoma	0	0	0	0	0	0	0	0	0	1/1	0	0
Subcutaneous malignant fibrous histiocytoma with metastasis	0	0	0	0	0	1/1	0	0	0	0	0	1/1
Subcutaneous neurofibrosarcoma with metastasis	0	0	0	0	0	1/1	0	0	0	0	0	1/1

*In those cases where multiple tumors of the same classification occurred in individual rats, data entry listed as total number of this tumor/number of rats bearing this type tumor.

During Months 1-6, there was only one tumor (subcutaneous carcinosarcoma) noted in the control group. Tumors occurring during Months 7-12 included one adrenal pheochromocytoma - unilateral (control), one subcutaneous fibrosarcoma with metastasis (control), one mixed glioma of the spinal cord (5 mg/kg/day), one adrenal pheochromocytoma - unilateral (5 mg/kg/day), one mammary gland fibroadenoma/adenofibroma (5 mg/kg/day), one pancreatic islet cell adenoma (100 mg/kg/day), and one mammary gland fibroadenoma/adenofibroma (100 mg/kg/day). All of these tumors have been included in the above total tabulation.

There were no statistical differences from control values when analyzed using Fisher's Exact Probability Test, $p<0.05$.

Table 2. (Concluded).

Time Interval	Months 13-18			Months 19-24			Terminal Kill			Total		
Dose Level (mg/kg/day)	0	5	100	0	5	100	0	5	100	0	5	100
Number of Rats Examined	13	12	12	25	26	26	9	7	6	50	50	50
Tumor/Tumor-like Lesions (cont'd)												
Mammary gland fibroadenoma/adenofibroma	0	1/1	0	0	1/1	0	1/1	0	0	1/1	3/1	1/1
Mammary gland adenocarcinoma without metastasis	0	0	0	0	1/1	0	0	0	0	0	1/1	0
Squamous epithelial polyp	0	0	0	0	0	0	1/1	0	0	1/1	0	0
Squamous cell carcinoma	0	1/1	0	0	0	1/1	0	0	0	0	1/1	1/1
Squamous papilloma	1/1	1/1	0	0	0	0	0	0	2/2	1/1	1/1	2/2
Basal cell carcinoma	0	0	0	0	0	0	1/1	0	0	1/1	0	0
Keratoacanthoma	0	0	0	0	0	0	1/1	0	0	1/1	0	0
Squamous cell carcinoma of prepuce	0	0	0	0	0	1/1	0	0	0	0	0	1/1
Sebaceous cystadenoma	0	0	0	0	0	1/1	0	0	0	0	0	1/1
Basal/squamous cell carcinoma	0	0	0	0	0	0	0	0	1/1	0	0	1/1
Hemangiopericytoma	0	0	0	0	1/1	0	0	0	0	0	1/1	0
Zymbal gland carcinoma	0	0	1/1	1/1	1/1	1/1	0	0	0	1/1	1/1	2/2
Rhabdomyosarcoma with metastasis	0	1	0	0	0	0	0	0	0	0	1	0
Chondrosarcoma of mandible	0	0	0	1	0	0	0	0	0	1	0	0

During Months 1-6, there was only one tumor (subcutaneous carcinosarcoma) noted in the control group. Tumors occurring during Months 7-12 included one adrenal pheochromocytoma - unilateral (control), one subcutaneous fibrosarcoma with metastasis (control), one mixed glioma of the spinal cord (5 mg/kg/day), one adrenal pheochromocytoma - unilateral (5 mg/kg/day), one mammary gland fibroadenoma/adenofibroma (5 mg/kg/day), one pancreatic islet cell adenoma (100 mg/kg/day), and one mammary gland fibroadenoma/adenofibroma (100 mg/kg/day). All of these tumors have been included in the above total tabulation.

There were o statistical differences from control values when analyzed using Fisher's Exact Probability Test, $p<0.05$.

Table 3. Tumor Incidence in Female Rats Maintained on Diets Containing FR-1138 (Dibromoneopentyl Glycol) For Up to Two Years.

Time Interval	Months 13-18			Months 19-24			Terminal Kill			Total		
Dose Level (mg/kg/day)	0	5	100	0	5	100	0	5	100	0	5	100
Number of Rats Examined	6	8	8	28	23	27	12	19	11	48	50	50
Tumor/Tumor-like Lesions												
Hepatocellular hyperplastic nodule(s)	0	0	0	1	0	1	2	0	2	3	0	3
Hepatocellular carcinoma(s)	0	0	0	1	0	0	0	0	0	1	0	0
Squamous cell carcinoma of hard palate	0	0	0	0	0	1	0	0	0	0	0	1
Keratinizing squamous cell carcinoma of lung	0	0	0	0	0	1	0	0	0	0	0	1
Granulosal cell neoplasm of ovary - unilateral	0	0	0	0	0	2	1	0	0	1	0	2
Granulosal cell neoplasm of ovaries - bilateral	0	0	0	1	0	0	0	0	0	1	0	0
Adenomatous polyp of uterus	0	0	0	0	0	0	0	0	1/1	0	0	1/1
Stromal polyp of uterus	0	0	1/1*	3/3	2/2	5/5	2/2	9/5	4/2	5/5	11/7	10/8
Cystic polyp of uterus	0	0	0	1/1	0	0	0	0	0	1/1	0	0
Adenoma or papillary adenoma of uterus	0	0	0	1	0	0	0	1	0	1	1	0
Malignant schwannoma of vagina	0	0	0	0	2	0	0	0	0	0	2	0
Malignant schwannoma of uterus with metastasis	1	0	0	0	1	0	0	0	1	2	1	1
Myxosarcoma of uterus	0	0	0	1	0	0	0	0	0	1	0	0
Hemangioma of uterus	0	0	0	1	0	0	0	0	0	1	0	0
Sclerosing carcinoma of uterus with extension	0	0	0	0	0	1	0	0	0	0	0	1
Fibrosarcoma of vagina	0	0	0	0	0	0	0	0	1	0	0	1
Malignant schwannoma of vagina or proximal uterus with local invasion	0	0	1	0	0	0	0	0	0	0	0	1
Stratified squamous cell carcinoma of tongue	0	0	0	0	0	0	1	0	0	1	0	0
Squamous polyp of stomach	0	0	0	1	0	0	3	2	0	4	2	0
Squamous papilloma of stomach	0	0	0	0	1	0	0	0	0	0	1	0
Leiomyoma of small intestine	0	0	0	0	1	0	0	0	0	0	1	0
Leiomyosarcoma of small intestine	0	0	0	0	0	1	0	0	0	0	0	1
Adenocarcinoma of large intestine	0	0	0	1	0	0	0	0	0	1	0	0
Intrasbdominal carcinosarcoma	0	0	0	0	1	0	0	0	0	0	1	0
Glioma of spinal cord	0	0	0	1	0	0	0	0	0	1	0	0

*In those cases where multiple tumors of the same classification occurred in individual rats, data entry listed as total number of this type tumor/number of rats bearing this type tumor.

Only one tumor occurred during Months 1-6, a mammary gland adenocarcinoma without metastasis (100 mg/kg/day). Tumors occurring during Months 7-12 included one malignant schwannoma of the uterus (control), one mammary gland fibroadenoma/adenofibroma (control), one mammary gland fibroadenoma/adenofibroma (100 mg/kg/day), and one generalized myeloid leukemia (control). These tumors are included in the above total tabulation.

There were no statistical differences from control values when analyzed using Fisher's Exact Probability Test, $p<0.05$.

Table 3. (Continued).

Time Interval	Months 13-18			Months 19-24			Terminal Kill			Total		
Dose Level (mg/kg/day)	0	5	100	0	5	100	0	5	100	0	5	100
Number of Rats Examined	6	8	8	28	23	27	12	19	11	48	50	50
Tumor/Tumor-like Lesions (cont'd)												
Benign schwannoma of abdominal peripheral nerve	0	0	0	0	0	0	0	0	1	0	0	1
Neurofibrosarcoma of cranial cavity	0	0	0	0	1	0	0	0	0	0	1	0
Generalized lymphosarcoma	0	0	0	0	0	0	1	0	1	1	0	1
Generalized myelomonocytic leukemia	1	0	0	1	0	0	0	0	0	2	0	0
Thymic lymphosarcoma	0	0	0	0	0	1	0	0	0	0	0	1
Thymic myxosarcoma	0	0	0	0	0	0	0	0	1	0	0	1
Pituitary adenoma formation(s)	1	0	5	13	13	8	6	11	3	20	24	16
Pituitary adenocarcinoma without extension or invasion of brain	1	2	0	4	3	4	1	3	3	6	8	7
Pituitary adenocarcinoma with invasion of brain	0	1	0	3	1	2	0	0	0	3	2	2
Unclassified tumor of pituitary	0	0	0	1	0	0	0	0	0	1	0	0
Adrenal adenoma - unilateral	0	0	0	2	1	0	1	1	1	3	2	1
Adrenal adenocarcinoma	0	0	0	0	0	0	0	0	1	0	0	1
Adrenal pheochromocytoma - unilateral	0	0	0	1	1	1	0	2	1	1	3	2
Adrenal pheochromocytoma - bilateral	0	0	0	0	0	1	1	1	1	1	1	2
Adrenal pheochromocytoma, malignant, metastasis - unilateral	0	0	0	0	0	0	0	1	0	0	1	0
Thyroid interfollicular C-cell adenoma(s)	0	0	0	3	0	4	1	4	2	4	4	6
Thyroid interfollicular C-cell adenocarcinoma without metastasis	0	1	0	1	1	0	0	1	1	1	3	1
Thyroid interfollicular C-cell adenocarcinoma with metastasis	0	0	0	0	0	0	0	1	0	0	1	0
Parathyroid adenoma	0	0	0	0	1	0	0	0	0	0	1	0
Pancreatic acinar adenoma(s)	1/1	0	0	0	0	0	0	0	0	1/1	0	0
Pancreatic islet cell adenoma(s)	0	0	0	1/1*	0	3/3	4/4	1/1	2/2	5/5	1/1	5/5
Subcutaneous neurofibrosarcoma with metastasis	0	1/1	0	0	0	0	0	0	0	0	1/1	0

*In those cases where multiple tumors of the same classification occurred in individual rats, data entry listed as total number of this type tumor/number of rats bearing this type tumor.

Only one tumor occurred during Months 1-6, a mammary gland adenocarcinoma without metastasis (100 mg/kg/day). Tumors occurring during Months 7-12 included one malignant schwannoma of the uterus (control), one mammary gland fibroadenoma/adenofibroma (control), one mammary gland fibroadenoma/adenofibroma (100 mg/kg/day), and one generalized myeloid leukemia (control). These tumors are included in the above total tabulation.

There were no statistical differences from control values when analyzed using Fisher's Exact Probability Test, $p<0.05$.

Table 3. (Concluded).

Time Interval	Months 13-18			Months 19-24			Terminal Kill			Total		
Dose Level (mg/kg/day)	0	5	100	0	5	100	0	5	100	0	5	100
Number of Rats Examined	6	8	8	28	23	27	12	19	11	48	50	50
Tumor/Tumor-like Lesions (cont'd)												
Subcutaneous undifferentiated sarcoma with metastasis	0	0	0	1/1	0	0	0	0	0	1/1	0	0
Mammary gland fibroadenoma/adenofibroma	3/3*	2/2	8/5	43/21	36/17	63/26	17/9	23/14	22/8	64/34	61/33	94/40
Mammary gland adenoma	0	1/1	1/1	1/1	1/1	2/2	3/2	4/2	4/2	4/3	6/4	7/5
Mammary gland adenocarcinoma without metastasis	1/1	0	1/1	5/5	2/2	0	0	2/2	0	6/6	4/4	2/2
Mammary gland adenocarcinoma with metastasis	0	0	0	0	1/1	0	0	0	0	0	1/1	0
Mammary gland cystadenoma	1/1	0	0	3/1	1/1	0	0	3/2	2/2	4/2	4/3	2/2
Mammary gland fibroma	0	1/1	0	1/1	0	3/3	0	1/1	0	1/1	2/2	3/3
Mammary gland cystfibroadenoma/cystadenofibroma	0	0	0	3/2	3/3	10/8	4/3	7/5	1/1	7/5	10/8	11/9
Trichoepithelioma	0	0	0	0	1/1	0	0	0	0	0	1/1	0
Fibroadenoma	0	0	0	0	0	1/1	0	0	0	0	0	1/1
Zymbal gland carcinoma	0	0	0	0	1/1	0	0	0	0	0	1/1	0
Generalized myeloid leukemia	0	0	0	0	0	0	0	0	0	1	0	0

*In those cases where multiple tumors of the same classification occurred in individual rats, data entry listed as total number of this type tumor/number of rats bearing this type tumor.

Only one tumor occurred during Months 1-6, a mammary gland adenocarcinoma without metastasis (100 mg/kg/day). Tumors occurring during Months 7-12 included one malignant schwannoma of the uterus (control), one mammary gland fibroadenoma/adenofibroma (control), one mammary gland fibroadenoma/adenofibroma (100 mg/kg/day), and one generalized myeloid leukemia (control). These tumors are included in the above total tabulation.

There were no statistical differences from control values when analyzed using Fisher's Exact Probability Test, $p<0.05$.

sues, integument, mammary gland, ear canal, brain, peripheral nerves, pituitary gland, cranial cavity, adrenal glands, eye, lymph nodes, thymus, spleen, mesentery, thyroid and parathyroid glands. Upon statistical anslyses of these tumor data, the incidence rates for all categories of tumor types occurring in any of the groups given 5 or 100 mg FR-1138/kg/day were comparable to the control group.

Thus, none of the tumors listed above were considered related to treatment with either of those dose levels of FR-1138. Table 4 summarizes the total numbers of tumors per group, the average number of tumors per rat, and the time of necropsy of rats with tumors for each group. None of these parameters were considered to be affected by any of these dose levels of FR-1138 when compared to the control data and historical control data [5].

Table 4. Tumor Incidence Data in Male and Female Rats Maintained on Diets Containing FR-1138 (Dibromoneopentyl Glycol) For Up to Two Years.

Sex	Historical	Males			Historical	Females		
Dose in mg/kg/day	Control	0	5	100	Control	0	5	100
Number of Rats in Group	Data	50	50	50	Data	48	50	50
TOTAL NUMBER OF TUMORS IN GROUP		83	79	112		162	163	193
AVERAGE NUMBER OF TUMORS PER RAT	2.0	1.7	1.6	2.2	3.6	3.4	3.3	3.9
NUMBER OF TUMOR BEARING RATS/NUMBER OF RATS IN GROUP								
(These data listed in time intervals in months):								
1-6		1/1	0/0	0/1		0/0	0/0	1/2
7-12		1/2	3/5	2/5		2/2	0/0	1/2
13-18		6/13	5/12	9/12		5/6	7/8	8/8
19-24		24/25	25/26	22/26		28/28	23/23	27/27
Terminal Kill		7/9	7/7	6/6		12/12	19/19	11/11
Total[a]		39/50	40/50	39/50		47/48	49/50	48/50
Percentage	88%	78%	80%	78%	97%	98%	98%	96%

[a]No statistical differences from control data when analyzed using Fisher's Exact Probability Test, $p<0.05$.

KEY WORD INDEX

Carcinogenicity
Chronic Toxicity
Dibromoneopentyl Glycol
Eye (lens) Toxicity
Flame Retardant
FR-1138
Liver Toxicity
Monobromoneopentyl Triol
Oncogenicity
Thyroid Gland Toxicity
Tribromoneopentyl Alcohol

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