NTP TECHNICAL REPORT

ON THE

TOXICOLOGY AND CARCINOGENESIS

STUDIES OF 1-BROMOPROPANE
(CAS NO. 106-94-5)

IN F344/N RATS AND B6C3F1 MICE
(INHALATION STUDIES)

NATIONAL TOXICOLOGY PROGRAM
P.O. Box 12233
Research triangle Park, NC 27709

August 2011

NTP TR 564

NIH Publication No. 11-5906

National Institutes of Health
Public Health Service
U.S. DEPARTMENT OF HEALTH AND HUMAN SERVICES

FOREWORD

The National Toxicology Program (NTP) is an interagency program within the Public Health Service (PHS) of the Department of Health and Human Services (HHS) and is headquartered at the National Institute of Environmental Health Sciences of the National Institutes of Health (NIEHS/NIH). Three agencies contribute resources to the program: NIEHS/NIH, the National Institute for Occupational Safety and Health of the Centers for Disease Control and Prevention (NIOSH/CDC), and the National Center for Toxicological Research of the Food and Drug Administration (NCTR/FDA). Established in 1978, the NTP is charged with coordinating toxicological testing activities, strengthening the science base in toxicology, developing and validating improved testing methods, and providing information about potentially toxic substances to health regulatory and research agencies, scientific and medical communities, and the public.

The Technical Report series began in 1976 with carcinogenesis studies conducted by the National Cancer Institute. In 1981, this bioassay program was transferred to the NTP. The studies described in the Technical Report series are designed and conducted to characterize and evaluate the toxicologic potential, including carcinogenic activity, of selected substances in laboratory animals (usually two species, rats and mice). Substances selected for NTP toxicity and carcinogenicity studies are chosen primarily on the basis of human exposure, level of production, and chemical structure. The interpretive conclusions presented in NTP Technical Reports are based only on the results of these NTP studies. Extrapolation of these results to other species, including characterization of hazards and risks to humans, requires analyses beyond the intent of these reports. Selection *per se* is not an indicator of a substance's carcinogenic potential.

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CONTRIBUTORS

National Toxicology Program
Evaluated and interpreted results and reported findings

D.L. Morgan, Ph.D., Study Scientist
M.F. Cesta, D.V.M., Study Pathologist
J.B. Bishop, Ph.D.
J.R. Bucher, Ph.D.
R.S. Chhabra, Ph.D.
P.M. Foster, Ph.D.
D. Germolec, Ph.D.
R.A. Herbert, D.V.M., Ph.D.
M.J. Hooth, Ph.D.
A.P. King-Herbert, D.V.M.
G.E. Kissling, Ph.D.
D.E. Malarkey, D.V.M., Ph.D.
J.H. Roycroft, Ph.D.
C.S. Smith, Ph.D.
G.S. Travlos, D.V.M.
S. Waidyanatha, Ph.D.
N.J. Walker, Ph.D.
K.L. Witt, M.S.

Battelle Toxicology Northwest
Conducted studies and evaluated pathology findings

J.A. Dill, Ph.D., Principal Investigator
S.L. Grumbein, D.V.M., Ph.D.
S.J. Harbo, D.V.M.
B.K. Hayden
R.A. Renne, D.V.M.

Experimental Pathology Laboratories, Inc.
Provided pathology review

M.H. Hamlin, II, D.V.M., Principal Investigator
E.T. Adams, D.V.M., Ph.D.
R.A. Miller, D.V.M., Ph.D.
J.C. Peckham, D.V.M., M.S., Ph.D.
C.C. Shackelford, D.V.M., M.S., Ph.D.

TherImmune Research Corporation
Provided SMVCE analysis

G.W. Wolfe, Ph.D., Principal Investigator
Y. Wang, M.S.

Dynamac Corporation
Prepared quality assessment audits

S. Brecher, Ph.D., Principal Investigator
S. Iyer, B.S.
V.S. Tharakan, D.V.M.

NTP Pathology Working Group
Evaluated slides and contributed to pathology report on 2-year rats (December 11, 2007)

G.D. Hill, D.V.M., Ph.D., Coordinator
ILS, Inc.
E.T. Adams, D.V.M., Ph.D.
Experimental Pathology Laboratories, Inc.
M.F. Cesta, D.V.M.
National Toxicology Program
S.A. Elmore, D.V.M., M.S.
National Toxicology Program
B. Hamilton, D.V.M., Ph.D.
GlaxoSmithKline
R.A. Herbert, D.V.M., Ph.D.
National Toxicology Program
R.R. Maronpot, D.V.M.
Consultant
R.A. Miller, D.V.M., Ph.D.
Experimental Pathology Laboratories, Inc.
A. Nyska, D.V.M.
National Toxicology Program
B.P. Singh, B.V.Sc., M.S.
National Toxicology Program

NTP Pathology Working Group

Evaluated slides and contributed to pathology report on 2-year mice (February 5, 2008)

K.R. Hobbie, D.V.M., Coordinator
ILS, Inc.
M.F. Cesta, D.V.M.
National Toxicology Program
S.A. Elmore, D.V.M., M.S.
National Toxicology Program
S.L. Grumbein, D.V.M., Ph.D.
Battelle Toxicology Northwest
B. Hamilton, D.V.M., Ph.D.
GlaxoSmithKline
D.E. Malarkey, D.V.M., Ph.D.
National Toxicology Program
R.R. Maronpot, D.V.M.
Consultant
R.A. Miller, D.V.M., Ph.D.
Experimental Pathology Laboratories, Inc.
J.C. Peckham, D.V.M., M.S., Ph.D.
Experimental Pathology Laboratories, Inc.
C.C. Shackelford, D.V.M., M.S., Ph.D.
Experimental Pathology Laboratories, Inc.

SRA International, Inc.

Provided statistical analyses

P.W. Crockett, Ph.D., Principal Investigator
L.J. Betz, M.S.
K.P. McGowan, M.B.A.

Biotechnical Services, Inc.

Prepared Technical Report

S.R. Gunnels, M.A., Principal Investigator
L.M. Harper, B.S.
P.C. Rathman, B.S.E.
D.C. Serbus, Ph.D.

CONTENTS

SUMMARY

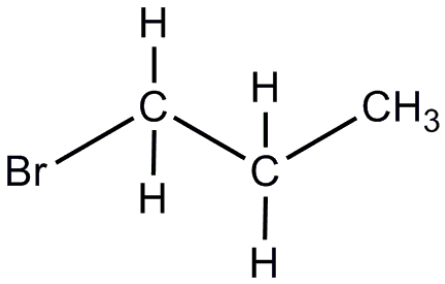
Background
1-Bromopropane is used for degreasing and as a solvent for adhesive resins. We studied 1-bromopropane to determine if it caused cancer in rats or mice.

Methods
We exposed groups of 50 male and female rats and mice to air containing 1-bromopropane six hours per day, five days a week for two years. Rats were exposed to concentrations of 125, 250, or 500 parts per million (ppm) of 1-bromopropane in air, and mice were exposed to concentrations of 62.5, 125, or 250 ppm. Similar groups of 50 animals were exposed to clean air in the same inhalation chambers as the treated control groups six hours per day. Tissues from more than 40 sites were examined for every animal.

Results
A few rare tumors of large intestine were seen in exposed male and female rats, and a variety of skin tumors were seen in exposed male rats and to a lesser extent in exposed female rats. Male rats also had slightly increased incidences of malignant mesotheliomas and of pancreatic islet adenoma and carcinoma. Female mice had markedly increased incidences of adenomas and carcinomas of the lung. Male and female rats and mice exposed to 1-bromopropane had hyperplasia of the nose. Some exposed male and female rats also had inflammation of the larynx, and most exposed male and female mice had regeneration of bronchioles of the lung.

Conclusions
We conclude that 1-bromopropane caused cancer of the large intestine in male and female rats and of the lung in female mice. 1-Bromopropane caused tumors of the skin in male rats and possibly in female rats also. Malignant mesotheliomas and pancreatic islet adenomas and carcinomas in male rats also were possibly associated with exposure to 1-bromopropane. Rats and mice of both sexes exposed to 1-bromopropane had hyperplasia and inflammation of the nose and other effects in the upper respiratory system (inflammation of the larynx in rats, regeneration of lung tissue in mice).

ABSTRACT



1-BROMOPROPANE

CAS No. 106-94-5

Chemical Formula: C_3H_7Br Molecular Weight: 122.99

Synonyms: 1-BP; propyl bromide; n-BP; N-propyl bromide

In the early to mid 1990s, 1-bromopropane was used primarily as an intermediate in the production of pesticides, quaternary ammonium compounds, flavors and fragrances, pharmaceuticals, and other chemicals in well-controlled, closed processes. In the mid to late 1990s, it was introduced as a less toxic replacement for methylene chloride in emissive applications such as vapor and immersion degreasing operations and critical cleaning of electronics and metals. 1-Bromopropane was also introduced as a nonflammable, nontoxic, fast-drying, and inexpensive solvent for adhesive resins, and has been marketed as a replacement for ozone depleting refrigerants. 1-Bromopropane was nominated for study by the Occupational Safety and Health Administration based on the potential for widespread occupational and environmental exposure and a lack of toxicity and carcinogenicity data. Male and female F344/N rats and B6C3F1 mice were exposed to 1-bromopropane (99% or greater pure) by inhalation for 2 weeks, 3 months, or 2 years. Genetic toxicology studies were conducted in *Salmonella typhimurium* and *Escherichia coli* and mouse peripheral blood.

2-WEEK STUDY IN RATS

Groups of five male and five female rats were exposed to 1-bromopropane vapor at concentrations of 0, 125, 250, 500, 1,000, or 2,000 ppm, 6 hours plus T_{90} (12 minutes) per day, 5 days per week for 16 days. All rats survived to the end of the study except one 500 ppm male. Mean body weights of 2,000 ppm rats were significantly less than those of the chamber controls. The absolute kidney weight of 1,000 ppm males, relative kidney weights of all exposed groups of males, and absolute and relative kidney weights of all exposed groups of females were significantly increased. The absolute and relative liver weights of 1,000 ppm males, relative liver weights of 500 and 2,000 ppm males, and absolute and relative liver weights of 500 ppm or greater females were significantly increased. Nasal

lesions included suppurative inflammation in males exposed to 500 ppm or greater, respiratory epithelial necrosis in 1,000 and 2,000 ppm males, and respiratory epithelial regeneration in 1,000 and 2,000 ppm females.

2-Week Study in Mice

Groups of five male and five female mice were exposed to 1-bromopropane vapor at concentrations of 0, 125, 250, 500, 1,000, or 2,000 ppm, 6 hours plus T_{90} (12 minutes) per day, 5 days per week for 17 days. All 2,000 ppm males, two 2,000 ppm females, four 500 ppm males, one 1,000 ppm male, and one 1,000 ppm female died early. The mean body weight gain of 1,000 ppm males was significantly less than that of the chamber controls. Abnormal breathing, lethargy, and eye discharge were observed primarily during week 1 in groups exposed to 500 ppm or greater. Liver weights of 1,000 ppm males and of females exposed to 500 ppm or greater were significantly increased. Kidney weights of 1,000 and 2,000 ppm females were significantly increased. Microscopic lesions related to 1-bromopropane exposure occurred in the lung, liver, and nose of males and females and were primarily seen in mice exposed to 500 ppm or greater.

3-Month Study in Rats

Groups of 10 male and 10 female rats were exposed to 1-bromopropane vapor at concentrations of 0, 62.5, 125, 250, 500, or 1,000 ppm, 6 hours plus T_{90} (10 minutes) per day, 5 days per week for 14 weeks. Additional clinical pathology groups of 10 male and 10 female rats were exposed to the same concentrations for 23 days. All rats survived to the end of the study. Mean body weights of 1,000 ppm males were significantly less than those of the chamber controls. The increases in sorbitol dehydrogenase activities in 500 ppm males and 1,000 ppm males and females were consistent with the histopathologic evidence of mild hepatotoxicity caused by 1-bromopropane. Liver weights of males exposed to 250 ppm or greater and of females exposed to 125 ppm or greater were significantly increased. Spleen and kidney weights of 1,000 ppm females were significantly increased. Exposure concentration-related decreases of 28% in sperm motility and 37% in sperm counts were seen in the 1,000 ppm group of male rats. Female rats in all three exposure groups evaluated exhibited altered estrous cycles, spending significantly more time in extended estrus and less time in extended diestrus. The incidences of cytoplasmic vacuolization of the liver were significantly increased in males exposed to 250 ppm or greater and in females exposed to 500 ppm or greater. Hepatocyte degeneration was also observed in 1,000 ppm females.

3-Month Study in Mice

Groups of 10 male and 10 female mice were exposed to 1-bromopropane vapor at concentrations of 0, 62.5, 125, 250, or 500 ppm, 6 hours plus T_{90} (10 minutes) per day, 5 days per week for 14 weeks. One 250 ppm male and four males and five females in the 500 ppm groups died early. Mean body weights of exposed groups were similar to those of the chamber controls. Lethargy was observed in males and females exposed to 500 ppm, and abnormal breathing was observed in moribund mice. The kidney, liver, and lung weights of 500 ppm females were significantly greater than those of the chamber controls. The kidney weights of 500 ppm males were significantly decreased. Sperm counts in the 500 ppm group of male mice were 28% less than that in the chamber controls. Female mice exhibited altered estrous cycles, with females in the 500 ppm group spending significantly more time in extended diestrus and those in the 250 ppm group spending significantly more time in extended estrus compared to the chamber controls. Nonneoplastic lesions were observed in the nose, larynx, trachea, lung, and liver of 500 ppm males and females and in the adrenal cortex of 500 ppm females.

2-Year Study in Rats

Groups of 50 male and 50 female rats were exposed to 1-bromopropane vapor at concentrations of 0, 125, 250, or 500 ppm, 6 hours plus T_{90} (10 minutes) per day, 5 days per week for 105 weeks. Survival of 500 ppm males was significantly less than that of the chamber control group. Mean body weights of exposed groups were similar to those of the chamber controls.

Increased incidences of macroscopic, soft, pale-yellow to green, variably sized nodules were seen predominantly in the nose and skin of exposed rats. The number of animals with multiple masses was increased in the 500 ppm groups. In most cases, these lesions were microscopically shown to be suppurative inflammation, many with Splendore-Hoeppli material.

The incidence of adenoma of the large intestine (colon or rectum) was significantly greater in 500 ppm females than in the chamber control group. The incidence of adenoma of the large intestine in 250 ppm males exceeded the historical control ranges for inhalation studies and all routes.

The incidences of keratoacanthoma, basal cell adenoma, basal cell carcinoma, or squamous cell carcinoma (combined) were significantly greater in all exposed groups of males than in the chamber control group and exceeded the historical control range for inhalation studies. The incidences of keratoacanthoma and of keratoacanthoma or squamous cell carcinoma (combined) in 250 and 500 ppm males were also significantly increased and exceeded the historical control ranges for inhalation studies. In 500 ppm females, the incidence of squamous cell papilloma, keratoacanthoma, basal cell adenoma, or basal cell carcinoma (combined) exceeded the historical control range for inhalation studies.

The incidence of malignant mesothelioma was significantly greater in 500 ppm males than in the chamber control group.

The incidences of pancreatic islet adenoma in all exposed groups of males and of pancreatic islet adenoma or carcinoma (combined) in 125 and 250 ppm males were significantly increased.

Treatment-related nonneoplastic lesions were observed in the respiratory system of exposed male and female rats. In the nose, the incidences of suppurative chronic inflammation, chronic active inflammation, glandular hyperplasia, respiratory epithelial hyperplasia (females), and respiratory metaplasia of the olfactory epithelium (females) were increased in all exposed groups. In the larynx, the incidences of chronic active inflammation and squamous metaplasia (except 125 ppm females) were increased in all exposed groups, and the incidences of suppurative chronic inflammation were increased in the 500 ppm groups. Also, chronic inflammation of the lung was observed in the 500 ppm females. In the trachea, there were increased incidences of chronic active inflammation in all exposed groups of females and 500 ppm males, and the incidence of epithelial hyperplasia was increased in 500 ppm females.

2-YEAR STUDY IN MICE

Groups of 50 male and 50 female mice were exposed to 1-bromopropane vapor at concentrations of 0, 62.5, 125, or 250 ppm, 6 hours plus T_{90} (10 minutes) per day, 5 days per week for 105 weeks. Survival of exposed groups was similar to that of the chamber controls. Mean body weights of all exposed groups were similar to those of the chamber controls throughout the study.

In the females, there were increased incidences of alveolar/bronchiolar adenoma, alveolar/bronchiolar carcinoma, and alveolar/bronchiolar adenoma or carcinoma (combined); the incidences of alveolar/bronchiolar adenoma or carcinoma (combined) were significantly increased in all exposed groups of females. There were significantly increased incidences of cytoplasmic vacuolization of the bronchiolar epithelium in all exposed male groups and regeneration of the bronchiolar epithelium in all exposed groups of males and females.

In the nose, there were significantly increased incidences of cytoplasmic vacuolization of the respiratory epithelium in all exposed groups of males and in 125 and 250 ppm females. There were significantly increased incidences of respiratory epithelial hyperplasia in all exposed female groups and in 62.5 and 250 ppm males. There were significantly increased incidences of respiratory metaplasia of olfactory epithelium in 62.5 and 125 ppm males and 125 and 250 ppm females.

There were significantly increased incidences of cytoplasmic vacuolization of respiratory epithelium in the larynx and trachea of all exposed male groups and in the trachea of 62.5 and 125 ppm females.

GENETIC TOXICOLOGY

1-Bromopropane was not mutagenic in either of two independent bacterial mutagenicity assays, each conducted with and without induced rat liver activation enzymes. Bacterial strains tested included *Salmonella typhimurium* strains TA97, TA98, TA100, and TA1535, and *Escherichia coli* strain WP2 *uvrA*/pKM101. In addition, no increases in the frequencies of micronucleated normochromatic erythrocytes were seen in male or female B6C3F1 mice exposed for 3 months to 62.5 to 500 ppm 1-bromopropane via inhalation.

CONCLUSIONS

Under the conditions of these 2-year inhalation studies, there was *some evidence of carcinogenic activity** of 1-bromopropane in male F344/N rats based on the occurrence of rare adenomas of the large intestine and increased incidences of epithelial neoplasms of the skin (keratoacanthoma, squamous cell carcinoma, and basal cell neoplasms). Increased incidences of malignant mesothelioma and pancreatic islet adenoma and carcinoma (combined) may also have been related to 1-bromopropane exposure. There was *clear evidence of carcinogenic activity* of 1-bromopropane in female F344/N rats based on increased incidences of adenoma of the large intestine. Increased incidences of skin

neoplasms may also have been related to 1-bromopropane exposure. There was *no evidence of carcinogenic activity* of 1-bromopropane in male B6C3F1 mice exposed to concentrations of 62.5, 125, or 250 ppm 1-bromopropane. There was *clear evidence of carcinogenic activity* of 1-bromopropane in female B6C3F1 mice based on increased incidences of alveolar/bronchiolar neoplasms.

Exposure to 1-bromopropane resulted in increased incidences of nonneoplastic lesions in the nose of rats and mice, the larynx of rats and male mice, and the trachea and lung of female rats and male and female mice. Suppurative inflammatory lesions with Splendore-Hoeppli material were present primarily in the nose and skin of male and female rats exposed to 1-bromopropane.

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* Explanation of Levels of Evidence of Carcinogenic Activity is on page 13. A summary of the Technical Reports Review Subcommittee comments and the public discussion on this Technical Report appears on page 15.

Summary of the 2-Year Carcinogenesis and Genetic Toxicology Studies of 1-Bromopropane

	Male F344/N Rats	Female F344/N Rats	Male B6C3F1 Mice	Female B6C3F1 Mice
Concentrations in air	0, 125, 250, or 500 ppm	0, 125, 250, or 500 ppm	0, 62.5, 125, or 250 ppm	0, 62.5, 125, or 250 ppm
Body weights	Exposed groups similar to the chamber control group	Exposed groups similar to the chamber control group	Exposed groups similar to the chamber control group	Exposed groups similar to the chamber control group
Survival rates	23/50, 26/50, 18/50, 13/50	34/50, 33/50, 30/50, 24/50	37/50, 33/50, 32/50, 36/50	36/50, 40/50, 37/50, 42/50
Nonneoplastic effects	Nose: inflammation, suppurative, chronic (0/50, 1/48, 2/48, 7/50); inflammation, chronic active (29/50, 33/48, 34/48, 35/50); glands, hyperplasia (5/50, 14/48, 14/48, 15/50) Larynx: inflammation, chronic active (21/50, 28/50, 31/50, 26/50)	Nose: inflammation, suppurative, chronic (0/50, 1/50, 3/49, 7/50); inflammation, chronic active (24/50, 37/50, 37/49, 36/50); glands, hyperplasia (6/50, 23/50, 28/49, 30/50); respiratory epithelium, hyperplasia (5/50, 13/50, 9/49, 18/50); olfactory epithelium, metaplasia, respiratory (3/50, 4/50, 6/49, 9/50) Larynx: inflammation, chronic active (18/50, 25/50, 30/50, 32/50); metaplasia, squamous (3/50, 2/50, 6/50, 21/50) Lung: inflammation, suppurative, chronic (0/50, 0/50, 0/50, 4/50) Trachea: inflammation, chronic active (0/50, 4/50, 1/50, 6/50); epithelium, hyperplasia (0/50, 0/50, 0/50, 4/50)	Lung: bronchiole, vacuolization cytoplasmic (0/50, 18/50, 19/49, 17/49); bronchiole, regeneration (1/50, 44/50, 38/49, 47/49) Nose: respiratory epithelium, vacuolization cytoplasmic (0/50, 12/50, 19/50, 20/50); respiratory epithelium, hyperplasia (16/50, 29/50, 23/50, 26/50); olfactory epithelium, metaplasia, respiratory (0/50, 7/50, 6/50, 3/50) Larynx: vacuolization cytoplasmic (0/48, 5/50, 10/48, 11/50) Trachea: vacuolization cytoplasmic (0/49, 15/50, 24/47, 24/50)	Lung: bronchiole, regeneration (0/50, 45/50, 43/50, 49/50) Nose: respiratory epithelium, vacuolization cytoplasmic (0/50, 3/50, 5/50, 8/50); respiratory epithelium, hyperplasia (11/50, 25/50, 28/50, 27/50); olfactory epithelium, metaplasia, respiratory (0/50, 4/50, 5/50, 14/50) Trachea: vacuolization cytoplasmic (0/50, 8/49, 7/50, 4/50)
Neoplastic effects	Large intestine: adenoma (0/50, 0/50, 2/50, 1/50) Skin: keratoacanthoma (0/50, 3/50, 6/50, 6/50); keratoacanthoma or squamous cell carcinoma (1/50, 4/50, 6/50, 8/50); keratoacanthoma, basal cell adenoma, basal cell carcinoma, or squamous cell carcinoma (1/50, 7/50, 9/50, 10/50)	Large intestine: adenoma (0/50, 1/50, 2/50, 5/50)	None	Lung: alveolar/bronchiolar adenoma (1/50, 6/50, 4/50, 10/50); alveolar/bronchiolar carcinoma (0/50, 7/50, 5/50, 4/50); alveolar/bronchiolar adenoma or carcinoma (1/50, 9/50, 8/50, 14/50)
Equivocal findings	Malignant mesothelioma: (0/50, 2/50, 2/50, 4/50) Pancreatic islets: adenoma (0/50, 5/50, 4/50, 5/50); adenoma or carcinoma (3/50, 10/50, 9/50, 8/50)	Skin: squamous cell papilloma, keratoacanthoma, basal cell adenoma, or basal cell carcinoma (1/50, 1/50, 1/50, 4/50)	None	None
Level of evidence of carcinogenic activity	Some evidence	Clear evidence	No evidence	Clear evidence

Summary of the 2-Year Carcinogenesis and Genetic Toxicology Studies of 1-Bromopropane

Genetic toxicology	
Bacterial gene mutations:	Negative in *Salmonella typhimurium* strains TA97, TA98, TA100, and TA1535 with and without S9; and in *Escherichia coli* WP2 *uvrA*/pKM101 with and without S9
Micronucleated erythrocytes	
Mouse peripheral blood *in vivo:*	Negative

EXPLANATION OF LEVELS OF EVIDENCE OF CARCINOGENIC ACTIVITY

The National Toxicology Program describes the results of individual experiments on a chemical agent and notes the strength of the evidence for conclusions regarding each study. Negative results, in which the study animals do not have a greater incidence of neoplasia than control animals, do not necessarily mean that a chemical is not a carcinogen, inasmuch as the experiments are conducted under a limited set of conditions. Positive results demonstrate that a chemical is carcinogenic for laboratory animals under the conditions of the study and indicate that exposure to the chemical has the potential for hazard to humans. Other organizations, such as the International Agency for Research on Cancer, assign a strength of evidence for conclusions based on an examination of all available evidence, including animal studies such as those conducted by the NTP, epidemiologic studies, and estimates of exposure. Thus, the actual determination of risk to humans from chemicals found to be carcinogenic in laboratory animals requires a wider analysis that extends beyond the purview of these studies.

Five categories of evidence of carcinogenic activity are used in the Technical Report series to summarize the strength of evidence observed in each experiment: two categories for positive results **(clear evidence and some evidence)**; one category for uncertain findings **(equivocal evidence)**; one category for no observable effects **(no evidence)**; and one category for experiments that cannot be evaluated because of major flaws **(inadequate study)**. These categories of interpretative conclusions were first adopted in June 1983 and then revised on March 1986 for use in the Technical Report series to incorporate more specifically the concept of actual weight of evidence of carcinogenic activity. For each separate experiment (male rats, female rats, male mice, female mice), one of the following five categories is selected to describe the findings. These categories refer to the strength of the experimental evidence and not to potency or mechanism.

- **Clear evidence** of carcinogenic activity is demonstrated by studies that are interpreted as showing a dose-related (i) increase of malignant neoplasms, (ii) increase of a combination of malignant and benign neoplasms, or (iii) marked increase of benign neoplasms if there is an indication from this or other studies of the ability of such tumors to progress to malignancy.
- **Some evidence** of carcinogenic activity is demonstrated by studies that are interpreted as showing a chemical-related increased incidence of neoplasms (malignant, benign, or combined) in which the strength of the response is less than that required for clear evidence.
- **Equivocal evidence** of carcinogenic activity is demonstrated by studies that are interpreted as showing a marginal increase of neoplasms that may be chemical related.
- **No evidence** of carcinogenic activity is demonstrated by studies that are interpreted as showing no chemical-related increases in malignant or benign neoplasms
- **Inadequate study** of carcinogenic activity is demonstrated by studies that, because of major qualitative or quantitative limitations, cannot be interpreted as valid for showing either the presence or absence of carcinogenic activity.

For studies showing multiple chemical-related neoplastic effects that if considered individually would be assigned to different levels of evidence categories, the following convention has been adopted to convey completely the study results. In a study with clear evidence of carcinogenic activity at some tissue sites, other responses that alone might be deemed some evidence are indicated as “were also related” to chemical exposure. In studies with clear or some evidence of carcinogenic activity, other responses that alone might be termed equivocal evidence are indicated as “may have been” related to chemical exposure.

When a conclusion statement for a particular experiment is selected, consideration must be given to key factors that would extend the actual boundary of an individual category of evidence. Such consideration should allow for incorporation of scientific experience and current understanding of long-term carcinogenesis studies in laboratory animals, especially for those evaluations that may be on the borderline between two adjacent levels. These considerations should include:

- adequacy of the experimental design and conduct;
- occurrence of common versus uncommon neoplasia;
- progression (or lack thereof) from benign to malignant neoplasia as well as from preneoplastic to neoplastic lesions;
- some benign neoplasms have the capacity to regress but others (of the same morphologic type) progress. At present, it is impossible to identify the difference. Therefore, where progression is known to be a possibility, the most prudent course is to assume that benign neoplasms of those types have the potential to become malignant;
- combining benign and malignant tumor incidence known or thought to represent stages of progression in the same organ or tissue;
- latency in tumor induction;
- multiplicity in site-specific neoplasia;
- metastases;
- supporting information from proliferative lesions (hyperplasia) in the same site of neoplasia or other experiments (same lesion in another sex or species);
- presence or absence of dose relationships;
- statistical significance of the observed tumor increase;
- concurrent control tumor incidence as well as the historical control rate and variability for a specific neoplasm;
- survival-adjusted analyses and false positive or false negative concerns;
- structure-activity correlations; and
- in some cases, genetic toxicology.

NATIONAL TOXICOLOGY PROGRAM BOARD OF SCIENTIFIC COUNSELORS
TECHNICAL REPORTS REVIEW SUBCOMMITTEE

The members of the Technical Reports Review Subcommittee who evaluated the draft NTP Technical Report on 1-bromopropane on November 19, 2009, are listed below. Subcommittee members serve as independent scientists, not as representatives of any institution, company, or governmental agency. In this capacity, subcommittee members have five major responsibilities in reviewing the NTP studies:

- to ascertain that all relevant literature data have been adequately cited and interpreted,
- to determine if the design and conditions of the NTP studies were appropriate,
- to ensure that the Technical Report presents the experimental results and conclusions fully and clearly,
- to judge the significance of the experimental results by scientific criteria, and
- to assess the evaluation of the evidence of carcinogenic activity and other observed toxic responses.

Raymond F. Novak, Ph.D., Chairperson
Children's Hospital of Michigan
Wayne State University School of Medicine
Detroit, MI

Tracie E. Bunton, D.V.M., Ph.D.
Toxicology Consultant
Eicarte LLC
Gettysburg, PA

Russell C. Cattley, V.M.D., Ph.D.
Amgen
Thousand Oaks, CA

David A. Eastmond, Ph.D., Principal Reviewer
Department of Cell Biology and Neuroscience
University of California
Riverside, CA

Stephen W. Looney, Ph.D.
Department of Biostatistics
Medical College of Georgia
Augusta, GA

Mitzi Nagarkatti, Ph.D.
Department of Pathology, Microbiology, and Immunology
University of South Carolina School of Medicine
Columbia, SC

Michael V. Pino, D.V.M., Ph.D., Principal Reviewer
Drug Safety Evaluation
Sanofi-aventis
Alfortville, France

Kenneth M. Portier, Ph.D., Principal Reviewer
American Cancer Society
Atlanta, GA

Jim E. Riviere, D.V.M., Ph.D.
College of Veterinary Medicine
North Carolina State University
Raleigh, NC

James L. Sherley, M.D., Ph.D.
Programs in Regenerative Biology and Cancer
Boston Biomedical Research Institute
Watertown, MA

Justin G. Teeguarden, Ph.D.
Pacific Northwest National Laboratory
Richland, WA

SUMMARY OF TECHNICAL REPORTS REVIEW SUBCOMMITTEE COMMENTS

On November 19, 2009, the draft Technical Report on the toxicology and carcinogenesis studies of 1-bromopropane received public review by the National Toxicology Program's Board of Scientific Counselors' Technical Reports Review Subcommittee. The review meeting was held at the National Institute of Environmental Health Sciences, Research Triangle Park, NC.

Dr. D.L. Morgan, NIEHS, introduced the toxicology and carcinogenesis studies of 1-bromopropane by reviewing the chemical's extensive use as a solvent, the rationale for and design of the inhalation studies, the observed toxicity and body weight effects in the short-term studies, and the observed neoplasms and nonneoplastic lesions in the long-term studies. The proposed conclusions were *some evidence of carcinogenic activity* of 1-bromopropane in male rats, *clear evidence of carcinogenic activity* of 1-bromopropane in female rats, *no evidence of carcinogenic activity* of 1-bromopropane in male mice, and *clear evidence of carcinogenic activity* of 1-bromopropane in female mice.

Dr. Pino, the first principal reviewer, had no scientific criticisms. He suggested that basal cell carcinomas and adenomas be listed in the results table for rats. He also suggested that pancreatic carcinomas and adenomas be added to the conclusion statement for male rats. He asked for clarification of the time and extent of the Splendore-Hoeppli inflammatory lesions and suggested specifying the types of skin tumors in rats. He also asked if some decreased tumor incidences should be discussed as in some other reports.

Dr. Morgan agreed to include mention of the basal cell carcinomas in rat skin in the results section. He noted that the pancreatic carcinomas had not been included because they did not increase with dose and were not considered treatment related. Dr. Morgan said the decreased incidences of skin sarcomas and liver adenomas and carcinomas in female rats could be included under other findings in the results section.

Dr. Eastmond, the second principal reviewer, felt the study was well conducted and the report well written. He felt the proposed conclusion of *some evidence* for the intestinal tumors was fair even without statistical significance, given the rarity of those tumors. He suggested some caveats in reporting the results of the mutagenicity tests for this volatile chemical. He asked for clarification of the T_{90} period at the start of the animal exposure period in the inhalation chambers and inquired if the pathology slides were coded during the initial diagnoses.

Dr. Morgan said more description would be provided about the mutagenicity test methodology and the chamber startup periods. Dr. M.F. Cesta, NIEHS, said slides are not coded during the initial readings, though they are in subsequent reviews.

Dr. Portier, the third principal reviewer, agreed with the proposed conclusions.

Dr. Cattley expressed a preference for specifying the skin neoplasm types in the conclusion statement.

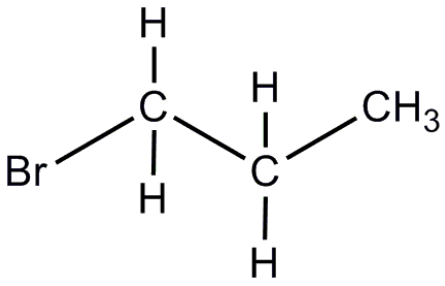
Dr. Nagarkatti asked if any further studies were performed to determine if the chemical produced any sensitization leading to development of the Splendore-Hoeppli bodies. Dr. Morgan said that while no other studies were performed, a number of other brominated compounds were immunosuppressive.

Dr. D. Lynch, NIOSH, commented that the present data would be helpful in the current development of exposure guidelines.

Dr. Pino moved that the conclusion in male rats specify adenomas and carcinomas (combined) for pancreatic lesions, and that the skin tumors be identified as epithelial tumors. Dr. Cattley added that the types of skin lesions should be specified, and Dr. Pino identified keratoacanthomas, squamous cell carcinoma, or basal cell neoplasms to be added parenthetically as the skin neoplasms for male rats.

Dr. Portier moved and Dr. Nagarkatti seconded that the conclusions be accepted as modified. Dr. Teeguarden was recused from the peer review and vote because of a conflict of interest and the motion was approved unanimously with nine votes.

INTRODUCTION



1-BROMOPROPANE

CAS No. 106-94-5

Chemical Formula: C_3H_7Br Molecular Weight: 122.99

Synonyms: 1-BP; propyl bromide; n-BP; N-propyl bromide

CHEMICAL AND PHYSICAL PROPERTIES

1-Bromopropane is a colorless to pale yellow liquid with a strong, characteristic odor. The boiling point is 71° C and the vapor pressure is 110.8 mm Hg at 20° C (UNEP, 2001; *Merck*, 2006). 1-Bromopropane is less flammable than many other halogenated alkanes at room temperature. Thermal decomposition of 1-bromopropane produces hydrogen bromide. 1-Bromopropane reacts with oxidizing agents to produce hazardous flammable compounds and can react with water to form acids. The 1-bromopropane used for industrial applications and in most commercial products is stabilized to inhibit hydrolysis, and formulated to improve performance and enhance the useable life of the product (UNEP, 2001; HSDB, 2009).

PRODUCTION, USE, AND HUMAN EXPOSURE

Industrial production of 1-bromopropane generally involves reacting propanol with an excess of hydrogen bromide gas. The principal product is 1-bromopropane with small amounts of 2-bromopropane (isopropyl bromide) and other by-products. Various modifications of the synthetic method are used to increase the purity of the 1-bromopropane, and distillation procedures are often used to remove most of the by-products (UNEP, 2001).

In 2006, worldwide annual production capacity of 1-bromopropane was estimated at greater than 20,000 metric tons (44 million pounds/year), of which 5,000 metric tons were thought to be used as a pharmaceutical intermediate or process agent. United States production was estimated at approximately 5,000 metric tons (11 million pounds/year) and growing at a rate of 15% to 20% per year (UNEP, 2006).

In the early to mid 1990s, 1-bromopropane was used primarily as an intermediate in the production of pesticides, quaternary ammonium compounds, flavors and fragrances, pharmaceuticals, and other chemicals in well-controlled, closed processes (Hanley *et al*., 2006). In the mid to late 1990s, 1-bromopropane was introduced as a less toxic replacement for methylene chloride

in emissive applications such as vapor and immersion degreasing operations and critical cleaning of electronics and metals. 1-Bromopropane has a relatively high vapor pressure and is well suited for use in cleaning equipment where it is repeatedly vaporized. Because it is relatively nonflammable, 1-bromopropane can be used safely in metal cleaning processes where heating is required. 1-Bromopropane was also introduced as a nonflammable, nontoxic, fast-drying, and inexpensive solvent for adhesive resins. Aerosol-applied adhesives containing 1-bromopropane were used extensively by foam fabricating companies (Hanley *et al*., 2006). Because 1-bromopropane has a relatively short atmospheric half-life (16 days), it is considered to have a relatively low ozone depletion potential and has been marketed as a replacement for ozone depleting refrigerants (e.g., chlorofluorohydrocarbons and hydrochlorofluorocarbons) (*Fed. Regist.*, 2000).

Emissive use of 1-bromopropane leads to substantial dermal and inhalation exposure of workers. Currently, there are no data available for dermal exposure to 1-bromopropane. The National Institute for Occupational Safety and Health (NIOSH) obtained personal breathing zone exposure data for workers at three foam fabrication plants that used spray adhesives containing 1-bromopropane. In these three plants, 8-hour time-weighted average (TWA) exposures ranged from 18 to 381 ppm (mean = 142 ppm) (NIOSH, 2002, 2003). In two of these plants the 8-hour TWA exposure concentrations of 1-bromopropane were significantly lowered (1.2 to 58 ppm; mean = 19 ppm) after improvements were made in ventilation (NIOSH, 2002).

ABSORPTION, DISTRIBUTION, METABOLISM, AND EXCRETION

Experimental Animals

Rats (strain not specified) dosed intraperitoneally with [^{14}C]-1-bromopropane exhaled unchanged 1-bromopropane and [^{14}C]-carbon dioxide (Jones and Walsh, 1979). The metabolites, *N*-acetyl-*S*-(2-hydroxypropyl)cysteine, *N*-acetyl-*S*-propylcysteine, and the corresponding *S*-oxide [*N*-acetyl-3-(propylsulfinyl)alanine] were identified in the urine (Barnsley *et al*., 1964, 1966; Barnsley, 1966; Jones and Walsh, 1979). Lee *et al.* (2007a) observed *S*-propyl glutathione in livers of BALB/c mice following a single oral dose of 1,000 mg/kg 1-bromopropane, with maximum levels of *S*-propyl glutathione detected 6 hours after administration. Exposure to 1-bromopropane has been shown to deplete glutathione in rat brain in a dose-dependent manner, independent of duration of exposure (Wang *et al*., 2003). In male Wistar rats exposed to 700 or 1,500 ppm 1-bromopropane vapor, 6 hours a day, 5 days a week, for 3 or more weeks, 1-bromopropane in blood decreased rapidly to the detection limit within 0.7 hours after exposure ceased. On the other hand, bromide ions persisted longer in both blood and urine; the biological half-life of bromide ion was 4.7 to 15.0 days in blood and 5.0 to 7.5 days in urine (Ishidao *et al*., 2002).

The National Toxicology Program (NTP) examined the disposition and metabolism of 1-bromopropane and the factors influencing them in male F344/N rats and B6C3F1 mice following inhalation exposure (800 ppm) or intravenous administration (5, 20, or 100 mg/kg) (Garner *et al*., 2006). [1,2,3-^{13}C]-1-bromopropane and [^{14}C]-1-bromopropane were co-administered to enable characterization of urinary metabolites using nuclear magnetic resonance spectroscopy and liquid chromatography coupled with either tandem mass spectrometry or radiochromatography. By 4 hours following intravenous administration, rats and mice exhaled a majority of the administered [^{14}C]-1-bromopropane dose as either volatile organic compounds (rats, 50% to 71%; mice, 39% to 48%) or carbon dioxide (rats, 10% to 30%; mice, 19% to 26%). The radioactivity was also excreted in urine (rats, 13% to 19%; mice, 13% to 23%) and feces (rats, < 2 %; mice, 4%) or retained in tissues and carcass (rats, ≤ 6%; mice, < 4%). *N*-acetyl-*S*-propyl-cysteine, *N*-acetyl-*S*-(2-hydroxypropyl)cysteine, *N*-acetyl-3-(propylsulfinyl)alanine, 1-bromo-2-hydroxypropane-*O*-glucuronide, *N*-acetyl-*S*-(2-oxopropyl)cysteine, and *N*-acetyl-3-[(2-oxopropyl)sulfinyl]alanine were the urinary metabolites characterized in rats and mice following both inhalation exposure and intravenous administration.

In rats, but not in mice, the route of elimination and the metabolite distribution changed significantly as the dose increased, with the percentage of dose excreted as volatile organic compounds increasing significantly between the mid- and high-dose groups (Garner *et al*., 2006). Concomitantly, the percentage of the dose exhaled as carbon dioxide decreased. An investigation of the molar ratio of exhaled carbon dioxide to the total released bromide showed that the proportion of 1-bromopropane metabolized via oxidation to pathways dependent on glutathione conjugation with bromine replacement decreased with increasing dose in rats. As the dose increased, the relative proportion of *N*-acetyl-*S*-propylcysteine and *N*-acetyl-*S*-(2-hydroxypropyl)-cysteine shifted such that the *N*-acetyl-*S*-propylcysteine peak predominated at 100 mg/kg accounting for more than 80% of the urinary radioactivity and indicating saturation of oxidation pathways. Rats pretreated with 1-aminobenzotriazole, a potent inhibitor of cytochrome

P450, had decreased total radioactivity excreted in urine, exhaled as carbon dioxide, or retained in liver with a concomitant increase in radioactivity in expired volatile organic compounds. The number of urinary metabolites was reduced from 10 to one with *N*-acetyl-*S*-propylcysteine accounting for more than 90% of the total urinary radioactivity in the pretreated rats. These data demonstrate a role for P450 and glutathione in the dose-dependent metabolism and disposition of 1-bromopropane in the rat.

Garner *et al.* (2007) also investigated the contribution of cytochrome P450 2E1 (CYP2E1) to 1-bromopropane metabolism using CYP2E1 knockout and wild type mice. The mercapturic acid of 1-bromo-2-hydroxypropane, *N*-acetyl-*S*-(2-hydroxypropyl)cysteine, was observed as the major urinary metabolite in wild type mice; the products of direct conjugation of 1-bromopropane with glutathione were reportedly insignificant. The ratio of glutathione conjugation to 2-hydroxylation was increased fivefold in the CYP2E1 knockout mice relative to wild type mice. In Sprague-Dawley rats exposed to as much as 1,800 ppm 1-bromopropane/kg (6 hours a day, 5 days a week, for 8 weeks), Kim *et al.* (1999) showed that 1-bromopropane primarily induced CYP2E1 as the major form of CYP and that glutathione *S*-transferase (GST) enzymes played important roles in the metabolism. Sex difference in the metabolic mechanism in the rat liver was also proposed (Kim *et al*., 1999). These results suggest that CYP2E1 is a major metabolic enzyme of 1-bromopropane although other P450s may be involved. Based on these findings, Garner *et al*. (2006) proposed a scheme for the metabolism of 1-bromopropane in rodents (Figure 1).

Humans

N-acetyl-*S*-propylcysteine was detected in the urine of occupationally exposed workers (Ichihara *et al*., 2004a). In another study, workers exposed to 1-bromopropane at two facilities using 1-bromopropane adhesives showed a significant association of 48-hour urinary bromide ion concentration with 1-bromopropane exposure measured in the breathing zone (Hanley *et al*., 2006). These data demonstrate that urinary elimination is an important excretion pathway of 1-bromopropane in humans.

GENERAL TOXICITY

Experimental Animals

The acute toxicity of 1-bromopropane has been investigated in rats by several routes of administration. The inhalation LC_{50} for 1-bromopropane was reported as 7,000 ppm for 4 hours in Wistar rats (ACGIH, 2005). For non-inhalation routes, the LD_{50} was reported as 2.9 g/kg in rats and 2.5 g/kg in mice by intraperitoneal injection (*Patty's*, 2001). The LD_{50} for oral and dermal routes of administration in rats is greater than 2,000 mg/kg (ACGIH, 2005).

The LC_{50} for 2-bromopropane, a structurally related brominated alkane is reported as 31,171 ppm for a 4-hour inhalation exposure in mice (ACGIH, 2005). The LD_{50} for intraperitoneal injection of 2-bromopropane in mice is 4.8 g/kg (Lewis, 1996). In rats, the LD_{50} for oral administration of 2-bromopropane is greater than 2,000 mg/kg (ACGIH, 2005)

REPRODUCTIVE AND DEVELOPMENTAL TOXICITY

Experimental Animals

1-Bromopropane was shown to cause developmental toxicity in rat pups whose dams were exposed during the period of *in utero* development (Huntingdon Life Sciences, 2001). Decreased fetal weights and increased incidences of skeletal variations were observed in pups of dams exposed to 1-bromopropane at 500 ppm or greater for 6 hours/day during gestation days 6 through 19.

A two-generation reproductive toxicity study (WIL Research Laboratories, 2001) showed exposure of rats to 250 ppm or greater altered numerous reproductive endpoints in both females and males. These included decreased sperm motility and percent normal sperm in males and decreased litter size, decreased numbers of implantation sites, increased ovarian follicular cysts, and increased estrous cycle length in females. Another study in Wistar rats (Ichihara *et al*., 2000a) comparing reproductive toxicities of 1- and 2-bromopropane

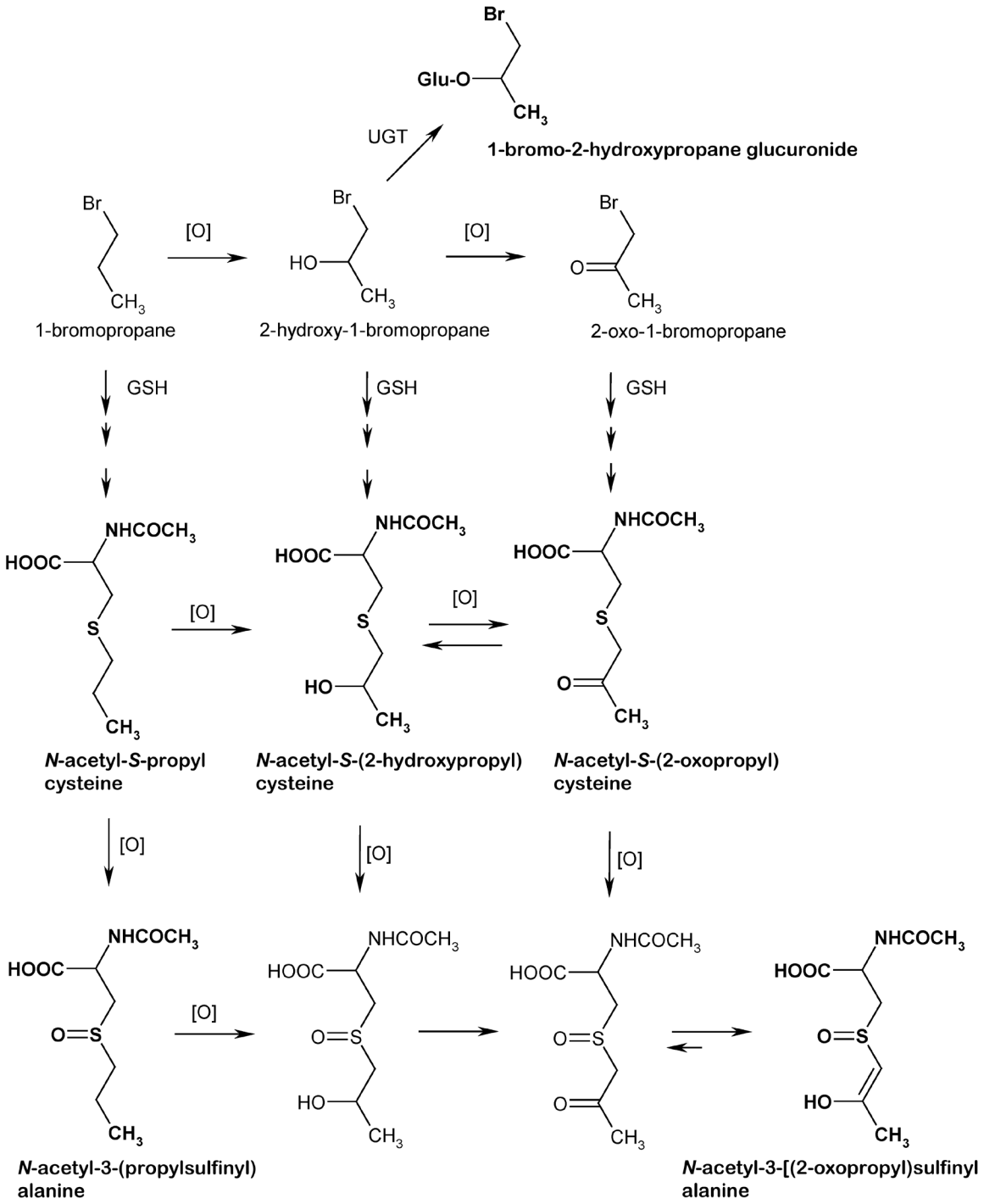


FIGURE 1
Proposed Metabolism of 1-Bromopropane in Rodents
Metabolites shown in bold font were characterized in urine from rodents exposed to 1-bromopropane (Garner *et al*., 2006)

demonstrated that exposure to 400 or 800 ppm 1-bromopropane for 12 weeks (8 hours/day, 7 days per week) inhibited sperm count and sperm motility; it was less toxic than 2-bromopropane. Yamada *et al*. (2003) reported that female Wistar rats exposed to 400 ppm 1-bromopropane 8 hours/day for 12 weeks had a significant increase in the number of irregular estrous cycles with extended diestrus. Histological examination of the ovary showed a significant reduction in the number of normal antral follicles, and a decrease in the number of normal growing follicles in the 400 ppm group.

Humans

The NTP Center for the Evaluation of Risks to Human Reproduction (CERHR) evaluated the potential for 1-bromopropane to produce adverse reproductive and developmental effects in humans (NTP, 2003). CERHR concluded that there was convincing evidence for reproductive and developmental toxicity in experimental animals. Evidence in humans was limited, but in the monograph, note was made of a new case that was not available to the expert panel indicating positive findings in women (altered menstruation) occupationally exposed to 1-bromopropane. The overall NTP conclusion was that "there is serious concern for reproductive and developmental effects of 1-bromopropane at the upper end of the human occupational exposure range (18 to 381 ppm)." "Serious concern" is the highest level of NTP conclusion regarding the possibilities that human development and reproduction might be adversely affected.

NEUROTOXICITY

Experimental Animals

In an animal study designed to evaluate the neurotoxic effects of 2-bromopropane on rat tail nerve, 1-bromopropane was used as a negative control (Yu *et al*., 1998). Wistar rats were exposed to air, 100 or 1,000 ppm 2-bromopropane or 1,000 ppm 1-bromopropane 8 hours/day, 7 days per week for up to 7 weeks. Unexpectedly, 1-bromopropane was found to be more neurotoxic than 2-bromopropane. Electrophysiological changes (decreased maximum motor nerve conduction velocity and increased distal latency) and histopathologic changes in peripheral nerves were detected in rats exposed to 1,000 ppm 1-bromopropane for 4 weeks. The exposures were discontinued after 5 weeks because of hindlimb paralysis in exposed rats. No changes were observed in rats exposed to the same concentration of 2-bromopropane. Histopathologic changes were also observed in axons in the gracile nucleus and in Purkinje cells in the cerebellum, suggesting that 1-bromopropane may also be toxic to the CNS. Ohnishi *et al*. (1999) also reported that exposure to 1,500 ppm 1-bromopropane 6 hours/day, 5 days per week for 4 weeks resulted in degeneration of Purkinje cells in the rat cerebellum. In a subsequent inhalation study, male Wistar rats exposed to 800 ppm 1-bromopropane showed myelin degeneration in the peripheral nerves, pre-terminal axon swelling in the gracile nucleus in the medulla oblongata, weight loss in the cerebrum, limb muscle weakness, distal latency prolongation, and decrease of motor nerve conduction velocity (Ichihara *et al*., 2000b).

Humans

The first case of 1-bromopropane toxicity in humans was reported by Sclar (1999). A worker developed weakness in the proximal extremities and right hand after using an industrial metal cleaning and degreasing solvent that contained 1-bromopropane (95.5% by weight) to strip metal for about 2 months. Nerve conduction studies indicated a primary symmetric demyelinating polyneuropathy, and magnetic resonance imaging scans revealed evidence of central nervous system (CNS) involvement. Subsequently, neurological signs of toxicity were reported in three workers at a foam fabrication factory where they used a spray adhesive containing 1-bromopropane (Ichihara *et al*., 2004b). The workers complained of numbness in the feet, thighs, and buttocks, and reduced sensations of vibration in the feet suggesting peripheral neuropathy. Other symptoms such as headache, dizziness, and memory loss suggested adverse effects on the CNS. NIOSH reported that four additional workers in a different cushion manufacturing company were hospitalized about a year earlier with similar neurological symptoms (NIOSH, 2002). All four workers had elevated serum bromide levels; however, exposure-related effects on blood cell counts, male reproductive functions, or nerve conduction velocity were not detected. NIOSH conducted health hazard evaluations at another foam cushion manufacturing plant where 1-bromopropane was used in the spray adhesive (NIOSH, 2003). Workers complained of headaches, painful tingling in the hands or feet, tremor, and feeling drunk when not drinking, indicating possible CNS and peripheral neurotoxicity.

In 1995, a new cleaning solvent containing about 97% 2-bromopropane (isopropyl bromide) was introduced in a Korean electronics manufacturing company to replace Freon (CFC-113). Workers exposed to 2-bromopropane presented with symptoms such as hand numbness, suggesting that 2-bromopropane exposure caused polyneuropathy (Kim *et al*., 1996; Park *et al*., 1997).

IMMUNOTOXICITY

1-Bromopropane was demonstrated to cause immunosuppression after oral administration to mice. A single treatment of female BALB/c mice with 1-bromopropane by gavage significantly suppressed the antibody response to T-cell dependent antigen and the production of splenic intracellular IL-2 in response to conconavalin A (Lee *et al*., 2007a). Similarly, oral treatment with 1,3-dibromopropane significantly suppressed the antibody response to T-dependent antigen in a dose-dependent manner. Mice treated with 1,3-dibromopropane for 7 consecutive days suppressed the antibody response to T-dependent antigen sheep red blood cells (SRBC) (Lee *et al*., 2007b). The structurally related chemical 2-bromopropane was first indicated to be immunotoxic in a study designed to evaluate reproductive toxicity in male rats. In this study, leukocyte numbers were significantly decreased in rats treated with 2-bromopropane concentrations that caused reproductive toxicity (Yu *et al*., 1997). In a subsequent study (Jeong *et al*., 2002), 2-bromopropane was shown to cause immunosuppression in rats following oral exposure to 1,000 mg/kg of 2-bromopropane for 28 consecutive days. 2-Bromopropane treatment significantly reduced numbers of leukocytes, spleen cells, and thymic cells and suppressed the antibody response to SRBCs.

As part of the inhalation studies reported in this Technical Report, the potential immunotoxicity of 1-bromopropane was investigated because of the earlier reports of immunosuppression following oral exposure to 1-bromopropane (Lee *et al*., 2007a) and the immunotoxicity of structurally related chemicals. In an NTP study, 1-bromopropane was found to cause some evidence of immunosuppression in F344/N rats and B6C3F1 mice after inhalation exposure for 4 and 10 weeks (Anderson *et al*., 2010). Significant decreases in total spleen cells and T-cells were detected after exposure for 4 weeks to 1-bromopropane concentrations ranging from 125 to 1,000 ppm. In addition, significant decreases in the IgM response to SRBCs were observed in both species after exposure to 1-bromopropane.

CARCINOGENICITY

There have been no carcinogenicity studies of 1-bromopropane in experimental animals or epidemiology studies in humans. However, NTP studies in rodents have demonstrated that several structurally related brominated hydrocarbons are potent multisite carcinogens. Inhalation exposure of F344/N rats to 1,2-dibromoethane (ethylene dibromide) caused carcinomas and adenocarcinomas of the nasal cavity and hemangiosarcomas of the circulatory system in males and females, mesotheliomas of the tunica vaginalis and adenomatous polyps of the nasal cavity in males, and fibroadenomas of the mammary gland and alveolar/bronchiolar adenomas and carcinomas in females (NTP, 1982a). 1,2-Dibromoethane was also carcinogenic in B6C3F1 mice, causing alveolar/bronchiolar adenomas and carcinomas in males and females; and hemangiosarcomas of the circulatory system, fibrosarcomas in subcutaneous tissue, carcinomas of the nasal cavity, and adenocarcinomas of the mammary gland in females. Inhalation exposure to 1,2-dibromo-3-chloropropane caused carcinomas, squamous cell carcinomas, and adenocarcinomas of the nasal cavity and squamous cell papillomas of the tongue in male and female F344/N rats (NTP, 1982b). In B6C3F1 mice, 1,2-dibromo-3-chloropropane caused adenocarcinomas of the nasal cavity and papillary carcinomas in females and squamous cell carcinomas of the nasal cavity and alveolar/bronchiolar adenomas and carcinomas in males and females. In F344/N rats and B6C3F1 mice, dermal application of 2,3-dibromo-1-propanol caused tumors at multiple sites (NTP, 1993).

GENETIC TOXICITY

Published data on the genotoxicity of 1-bromopropane suggest that, when tested appropriately to control for volatility, the compound demonstrates direct-acting mutagenicity in bacteria, and it has a weak potential for induction of chromosomal damage *in vitro* and *in vivo* in some mammalian test systems. Barber *et al*. (1981) reported that 1-bromopropane, tested over a concentration range of 1.1 to 20.3 µM, was mutagenic in *Salmonella typhimurium* strains TA100 and TA1535, with or without induced rat liver S9 activation enzymes; the lowest effective concentration was 4.9 µM, and potency of response, which directly correlated with concentration, was unaffected by the addition of S9, thus indicating that 1-bromopropane is a direct-acting mutagen in *Salmonella*. Barber *et al*. (1981) used a closed system to control for the volatility of 1-bromopropane. Yu *et al*. (2008) conducted a dominant lethal assay in male ICR mice administered 1-bromopropane (300 or 600 mg/kg per day) by gavage for 10 days and mated once weekly for 6 weeks to undosed female mice. Females were sacrificed and uterine contents were examined on gestation days 15 through 17. No significant treatment-related increases in pre- or post-implantation losses, resorptions, nonviable fetuses, or other fertility endpoints were seen at either dose at any mating time point. The authors concluded that under these experimental conditions, 1-bromopropane did not

induce dominant lethal mutations in germ cells of male mice. Negative results were also reported from an earlier dominant lethal study conducted in male Sprague-Dawley rats treated with 400 mg/kg per day 1-bromopropane by gavage for 5 consecutive days and mated to undosed females once weekly for 8 successive weeks (Saito-Suzuki *et al*., 1982); females were sacrificed 13 to 14 days after mating and uterine contents showed no significant treatment-related changes in most indicators of fertility or embryonic deaths compared with the vehicle controls. The one exception was a marginally elevated frequency of dead implants observed at mating week 8, in the absence of a significant dominant lethal mutation index. In a parallel investigation at the same laboratory, male Sprague-Dawley rats were treated with 200 mg/kg per day 1,2-dibromopropane for 5 days and mated to undosed females for 8 weeks (Saito-Suzuki *et al*., 1982). Examination of uterine contents at gestation day 13 or 14 revealed small increases in dead implants and in the dominant lethal mutation index in the week-1 mating group, but no other indications of adverse effects were observed for 1,2-dibromopropane. In contrast, 1,2,3-tribromopropane (50 mg/kg per day for 5 days) induced a significant increase in dominant lethal mutations in male Sprague-Dawley rats in post-meiotic sperm (primarily early spermatids, mating weeks 5 and 6) (Saito-Suzuki *et al*., 1982).

NIOSH assessed DNA-damage levels, measured by the alkaline single cell gel electrophoresis (comet) assay in peripheral leukocytes of 64 workers from two facilities where spray adhesives containing 1-bromopropane were used (Toraason *et al*., 2006). Workers were divided into two groups based on 1-bromopropane exposure level (high or low); bromide levels in blood were used as an internal confirmation of exposure, along with personal breathing zone air samples that were used to calculate TWAs of 1-bromopropane concentrations. Although the high exposure group showed significantly increased levels of urine and serum bromide compared with the low exposure group, no significant differences were seen in the levels of DNA damage between the two exposure groups. Overall DNA damage measurements made at the end of the work week in one facility showed a small but not statistically significant increase, the result of a significant ($P<0.05$) increase in the lowest exposure group only, while end-of-week DNA damage measures at the second facility were actually lower in both exposure groups than at the beginning of the work week. Polymorphisms in GST genes GSTM1 and GSTT1 were considered in the data analysis process along with other factors associated with DNA damage induction such as smoking, age, and gender. In addition to the *in vivo* assessments of DNA damage in exposed workers, whole blood cultures from an unexposed individual were treated *in vitro* with either 1-bromopropane or 2-bromopropane, and evaluated for DNA damage using the comet assay (Toraason *et al*., 2006). Both 1-bromopropane and 2-bromopropane induced significant increases in DNA damage at the highest concentration tested, 1 mM; in addition, both compounds increased the number of apoptotic cells (cells with diffuse DNA) at 0.1 mM, a 10-fold lower concentration than required to induce significant levels of DNA damage.

REGULATORY STANDARDS

The ozone-depleting solvents methyl chloroform and CFC-113 were phased out of production in the United States beginning in 1996 under the United States Environmental Protection Agency (USEPA) regulations and under the Montreal Protocol on Substances that Deplete the Ozone Layer. In the late 1990s, 1-bromopropane was introduced as a replacement for methyl chloroform and CFC-113 for cleaning metal parts and electronics and as a nontoxic replacement for methylene chloride and methyl chloroform adhesives. As the United States Occupational Safety and Health Administration (OSHA) began phasing in a more restrictive exposure standard for methylene chloride, the use of 1-bromopropane in adhesives increased. In April 2000, OSHA's final phase for instituting a workplace exposure standard for methylene chloride took full effect, leading companies to switch to 1-bromopropane rather than install ventilation or other controls needed to reduce employee methylene chloride exposures to the requisite 25 ppm level. Because 1-bromopropane was not regulated to protect workers, consumers, or the environment, the increased production and use was a concern.

Several companies petitioned the USEPA's Significant New Alternatives Policy (SNAP) program to accept the use of 1-bromopropane as an alternative to ozone-depleting solvents (*Fed. Regist.*, 1999). In 2007, the USEPA issued a final ruling, finding 1-bromopropane acceptable as a substitute for methyl chloroform and for CFC-113 in metals, electronics and precision cleaning (*Fed. Regist.*, 2007a). This rule did not address the use of 1-bromopropane as an aerosol solvent or as a carrier solvent in adhesives or coatings. The USEPA proposed a new rule to prohibit the use of 1-bromopropane as a carrier solvent in adhesives and as a solvent in aerosol cans, and restrict the use of 1-bromopropane to that of a carrier solvent in coatings (*Fed. Regist.*, 2007b). The

American Conference of Governmental Industrial Hygienists (2009) has recommended a threshold limit value 8-hour time-weighted average for 1-bromopropane of 10 ppm.

STUDY RATIONALE

1-Bromopropane was nominated by OSHA for evaluation of toxicokinetics, genetic toxicity, reproductive and developmental toxicity, neurotoxicity, and carcinogenicity. The nomination was based on the potential for widespread occupational and environmental exposure to 1-bromopropane and a lack of toxicity and carcinogenicity data. Because previous short-term studies demonstrated that 1-bromopropane is a neurotoxicant and a reproductive toxicant in animals (Yu *et al*., 1998; Ichihara *et al*, 2000a), the main focus of the current studies was to evaluate the chronic toxicity and carcinogenicity of 1-bromopropane.

The 2-week, 3-month, and 2-year studies were conducted in male and female F344/N rats and B6C3F1 mice to evaluate the toxicity and carcinogenicity of 1-bromopropane in association with inhalation exposure. Additional studies were conducted to evaluate the absorption, distribution, metabolism, and excretion of 1-bromopropane and its potential immunotoxicity (Garner *et al*., 2006; Anderson *et al*., 2010).

MATERIALS AND METHODS

PROCUREMENT AND CHARACTERIZATION OF 1-BROMOPROPANE

1-Bromopropane was obtained in 55-gallon metal drums from Diaz Chemical Corporation (Holley, NY) in one lot (106-015) and from Albemarle PCC (Thann, France) in one lot (1581313004). Lot 106-015 was used in the 2-week and 3-month studies, and lot 1581313004 was used during the 2-year studies. Identity and purity analyses were conducted by the study laboratory at Battelle Toxicology Northwest (Richland, WA), and by Chemir/Polytech Laboratories, Inc. (Maryland Heights, MO) (Appendix I). Reports on analyses performed in support of the 1-bromopropane studies are on file at the National Institute of Environmental Health Sciences.

Both lots of the chemical, a colorless to pale yellow liquid with a strong, characteristic odor, were identified as 1-bromopropane by Chemir/Polytech Laboratories, Inc., by infrared and ^{1}H-nuclear magnetic resonance spectroscopy.

For lot 106-015, Karl Fischer titration indicated a water content of 39 ppm. Elemental analyses for carbon, hydrogen, nitrogen, and sulfur were in agreement with the theoretical values for 1-bromopropane; residual HBr was determined to be 1.3 ppm. Gas chromatography (GC) indicated one major peak and three impurities with areas exceeding 0.1% of the total peak area. Using prepared standards, the impurities were identified as 1-propanol (0.14%), 2-bromopropane (0.11%), and di-*n*-propyl ether (0.74%). Measured purity of the bulk chemical was consistent throughout the sampled metal drum, and the overall purity of lot 106-015 was determined to be approximately 99%.

For lot 1581313004, Karl Fischer titration indicated a water content of 119 ppm. Elemental analyses for carbon, hydrogen, nitrogen, and sulfur were in agreement with the theoretical values for 1-bromopropane; residual HBr was determined to be 61.6 ppm. GC detected one major peak and no impurities with areas greater than 0.1% of the total peak area. Three impurities identified with prepared standards were 1-propanol (0.03%), 2-bromopropane (0.02%), and di-*n*-propyl ether (0.02%). Measured purity of the bulk chemical was consistent throughout the sampled metal drums, and the overall purity of lot 1581313004 was determined to be 99.9% or greater.

To ensure stability, the bulk chemical was stored at controlled room temperature in the original shipping containers (55-gallon metal drums). Periodic reanalyses of the bulk chemical were performed during the 2-week, 3-month, and 2-year studies by the study laboratory using GC, and no degradation of the bulk chemical was detected.

VAPOR GENERATION AND EXPOSURE SYSTEM

A preheater was necessary for the 2-week and 3-month studies. 1-Bromopropane was pumped through a preheater and into a heated glass column filled with glass beads that increased the surface area for vaporization. Heated nitrogen entered the column from below and assisted in vaporizing the chemical while conveying it into a short distribution manifold. Concentration in the manifold was determined by the chemical pump rate and nitrogen flow rate. The pressure in the distribution manifold was kept fixed to ensure constant flow through the manifold and into all chambers as the flow of vapor to each chamber was adjusted.

Metering valves at the manifold controlled flow to each chamber through individual Teflon® delivery lines that carried the vapor from the manifold to three-way exposure valves at the chamber inlets. The exposure valves diverted vapor delivery to exposure chamber exhaust until the generation system was stable and exposures were ready to proceed. To initiate exposure, the chamber exposure valves were rotated to allow the 1-bromopropane vapor to flow to each exposure chamber inlet duct where it was further diluted with filtered, conditioned air to achieve the desired exposure concentration.

The study laboratory designed the inhalation exposure chamber (Harford Systems Division of Lab Products, Inc., Aberdeen, MD) so that uniform vapor concentrations could be maintained throughout the chamber with the catch pans in place. The total active mixing volume of each chamber was 1.7 m^3. A small particle detector was used with and without animals in the exposure chambers to ensure that 1-bromopropane vapor, and not aerosol, was produced. No particle counts above the minimum resolvable level (approximately 200 particles per cm^3) were detected.

VAPOR CONCENTRATION MONITORING

Chamber and room concentrations of 1-bromopropane were monitored by an on-line gas chromatograph. Samples were drawn from each exposure chamber approximately every 20 (2-week and 3-month studies) or 30 (2-year studies) minutes during each 6-hour exposure period using Hastelloy®-C stream-select and gas-sampling valves in a separate, heated valve oven. The sample lines composing each sample loop were made from Teflon® tubing and were connected to the exposure chamber relative humidity sampling lines at a location close to the gas chromatograph. A vacuum regulator maintained a constant vacuum in the sample loop to compensate for variations in sample line pressure. An in-line flow meter between the vacuum regulator and the gas chromatograph allowed digital measurement of sample flow. Summaries of the chamber vapor concentrations are given in Tables I2 through I4.

The on-line gas chromatograph was checked throughout the day for instrument drift against an on-line standard vapor of 1-bromopropane in nitrogen supplied by a standard generator. The on-line gas chromatograph was recalibrated as required to meet acceptance criteria. Calibration was performed by comparing chamber concentration data to data from grab samples that were collected with activated coconut charcoal gas sampling tubes, extracted with methylene chloride containing 1-bromobutane as an internal standard, and analyzed using an off-line gas chromatograph. Known volumes of chamber atmosphere were sampled at a constant flow rate ensured by a calibrated critical orifice. The off-line gas chromatograph was calibrated with gravimetrically prepared standards of 1-bromopropane containing 1-bromobutane as an internal standard in methylene chloride.

CHAMBER ATMOSPHERE CHARACTERIZATION

Buildup and decay rates for chamber vapor concentrations were determined with and without animals present in the chambers. At a chamber airflow rate of 15 air changes per hour, the theoretical value for the time to achieve 90% of the target concentration after the beginning of vapor generation (T_{90}) and the time for the chamber concentration to decay to 10% of the target concentration after vapor generation was terminated (T_{10}) was approximately 12.5 minutes. A T_{90} value of 12 minutes was selected for the 2-week studies, and a T_{90} value of 10 minutes was selected for the 3-month and 2-year studies.

Evaluations of chamber uniformity and persistence and monitoring for 1-bromopropane degradation impurities were conducted periodically throughout the studies by GC. Chamber uniformity was maintained and no degradation was detected.

2-WEEK STUDIES

Male and female F344/N rats and B6C3F1 mice were obtained from Taconic Farms, Inc. (Germantown, NY). On receipt, the rats and mice were approximately 4 weeks old. Animals were quarantined for 13 days and were approximately 5 to 6 weeks old on the first day of the studies. Before the studies began, 10 female rats and five male and five female mice were randomly selected for parasite evaluation and gross observation for evidence of disease. At terminal sacrifice, serologic analyses were performed on five male and five female chamber control rats and mice using the protocols of the NTP Sentinel Animal Program (Appendix K).

Groups of five male and five female rats and mice were exposed to 1-bromopropane vapor at concentrations of 0, 125, 250, 500, 1,000, or 2,000 ppm, 6 hours plus T_{90} (12 minutes) per day, 5 days per week for 16 (rats) or 17 (mice) days. Feed was available *ad libitum* except during exposure periods; water was available *ad libitum*. Rats and mice were housed individually. Clinical findings were recorded twice daily on exposure days for rats and mice. The animals were weighed initially, on days 6 and 13, and at the end of the studies. Details of the study design and animal maintenance are summarized in Table 1.

Necropsies were performed on all animals. The heart, right kidney, liver, lung, right testis, and thymus were weighed. Tissues for microscopic examination were fixed and preserved in 10% neutral buffered formalin (except eyes were first fixed in Davidson's solution), processed and trimmed, embedded in paraffin, sectioned to a thickness of 4 to 6 µm, and stained with hematoxylin and eosin. Histopathologic examinations were performed on all chamber control and 2,000 ppm animals. The lung and nose of rats and mice; the mediastinal lymph node, sciatic nerve, and spinal cord of rats; and the liver of mice were examined to a no-effect level. Table 1 lists the tissues and organs examined.

3-MONTH STUDIES

The 3-month studies were conducted to evaluate the cumulative toxic effects of repeated exposure to 1-bromopropane and to determine the appropriate exposure concentrations to be used in the 2-year studies.

Male and female F344/N rats and B6C3F1 mice were obtained from Taconic Farms, Inc. (Germantown, NY). On receipt, the rats and mice were approximately 3 to 4 weeks old. Animals were quarantined for 12 (female rats and male and female mice) or 13 (male rats) days and were approximately 5 to 6 weeks old on the first day of the studies. Before the studies began, five male and five female rats and mice were randomly selected for parasite evaluation and gross observation for evidence of disease. Serological analyses were performed on five male and five female sentinel rats and mice during week 1 and five male and five female chamber control rats and mice at the end of the studies using the protocols of the NTP Sentinel Animal Program (Appendix K).

Groups of 10 male and 10 female rats and mice were exposed to 1-bromopropane vapor at concentrations of 0, 62.5, 125, 250, 500, or 1,000 ppm (rats only), 6 hours plus T_{90} (10 minutes) per day, 5 days per week for 14 weeks. Additional clinical pathology groups of 10 male and 10 female rats were exposed to the same concentrations for 23 days. One additional exposure day was scheduled during the last exposure week to give the rats at least 2 consecutive days of exposure before terminal sacrifice. Feed was available *ad libitum* except during exposure periods; water was available *ad libitum*. Rats and mice were housed individually. Clinical findings were recorded on day 9 (male rats) or 10 and then weekly. Core study animals were weighed initially, on day 9 (male rats) or 10, weekly thereafter, and at the end of the studies. Details of the study design and animal maintenance are summarized in Table 1.

Animals were anesthetized with carbon dioxide, and blood was collected from the retroorbital sinus of clinical pathology rats on days 3 and 23 and from core study rats at the end of the study for hematology and clinical chemistry analyses; blood was collected from the retroorbital sinus of mice at the end of the study for hematology analyses. Blood samples for hematology analyses were placed in tubes containing potassium EDTA. Erythrocyte, leukocyte, and platelet counts; hemoglobin; packed red cell volume; mean cell volume; mean cell hemoglobin; and mean cell hemoglobin concentration were determined using an Abbott Cell-Dyn 3700 hematology analyzer (Abbott Diagnostic Systems, Abbott Park, IL). Manual hematocrit values were determined using a microcentrifuge (Heraeus Haemofuge, Germany) and a Damon/IEC capillary reader (International Equipment Company, Needham Heights, MA) for comparison to Cell-Dyn values for packed cell volume. Blood smears for rats and mice were stained with Romanowsky-type aqueous stain in a Wescor 7100 Aerospray Slide Stainer (Wescor, Inc., Logan, UT). Leukocyte differential counts for rats and mice were based on classifying a minimum of 100 white cells. Reticulocytes were stained with New Methylene Blue and enumerated as a reticulocyte:erythrocyte ratio using the Miller disc method (Brecher and Schneiderman, 1950). Blood samples for clinical chemistry analyses were placed in tubes containing separator gel and allowed to clot. After clot retraction occurred, the samples were centrifuged, and the serum was aliquoted for assay of serum chemistry analytes using a Roche Hitachi 912 (Roche Diagnostic Corporation, Indianapolis, IN). Table 1 lists the parameters measured.

At the end of the 3-month studies, samples were collected for sperm motility and vaginal cytology evaluations on rats and mice exposed to 0, 125 (mice), 250, 500, or 1,000 (rats) ppm. The parameters evaluated are listed in Table 1. For 12 consecutive days prior to scheduled terminal sacrifice, the vaginal vaults of the females were moistened with saline, if necessary, and samples of vaginal fluid and cells were stained. Relative numbers of leukocytes, nucleated epithelial cells, and large squamous epithelial cells were determined and used to ascertain estrous cycle stage (i.e., diestrus, proestrus, estrus, and metestrus). Male animals were evaluated for sperm count and motility. The left testis and left epididymis were isolated and weighed. The tail of the epididymis (cauda epididymis) was then removed from the epididymal body (corpus epididymis) and

weighed. Test yolk (rats) or modified Tyrode's buffer (mice) was applied to slides and a small incision was made at the distal border of the cauda epididymis. The sperm effluxing from the incision were dispersed in the buffer on the slides, and the numbers of motile and nonmotile spermatozoa were counted for five fields per slide by two observers. Following completion of sperm motility estimates, each left cauda epididymis was placed in buffered saline solution. Caudae were finely minced, and the tissue was incubated in the saline solution and then heat fixed at 65° C. Sperm density was then determined microscopically with the aid of a hemacytometer. To quantify spermatogenesis, the testicular spermatid head count was determined by removing the tunica albuginea and homogenizing the left testis in phosphate-buffered saline containing 10% dimethyl sulfoxide. Homogenization-resistant spermatid nuclei were counted with a hemacytometer.

Necropsies were performed on all core study animals. The heart, right kidney, liver, lung, spleen, right testis, and thymus were weighed. Tissues for microscopic examination were fixed and preserved in 10% neutral buffered formalin (eyes were fixed in Davidson's solution for up to 3 days and then transferred to 10% neutral buffered formalin), processed and trimmed, embedded in paraffin, sectioned to a thickness of 4 to 6 µm, and stained with hematoxylin and eosin. Complete histopathologic examinations were performed on all chamber control, 250 and 500 ppm mice, and 1,000 ppm rats. The larynx, liver, lung, and nose of rats and mice, the prostate gland of male rats, the trachea of mice, and the adrenal gland and kidney of female mice were examined in all groups; the remaining tissues were examined to a no-effect level in the remaining exposure groups. Table 1 lists the tissues and organs routinely examined.

After a review of the laboratory reports and selected histopathology slides by a quality assessment pathologist, the findings and reviewed slides were submitted to a NTP Pathology Working Group (PWG) coordinator for a second independent review. Any inconsistencies in the diagnoses made by the laboratory and quality assessment pathologists were resolved by the NTP pathology peer review process. Final diagnoses for reviewed lesions represent a consensus between the laboratory pathologist, NTP pathologist, reviewing pathologist(s), if any, and the PWG coordinator. Details of these review procedures have been described, in part, by Maronpot and Boorman (1982) and Boorman *et al.* (1985).

2-YEAR STUDIES

Study Design

Groups of 50 male and 50 female rats and mice were exposed to 1-bromopropane vapor at concentrations of 0, 62.5 (mice only), 125, 250, or 500 (rats only) ppm, 6 hours plus T_{90} (10 minutes) per day, 5 days per week for up to 105 weeks.

Source and Specification of Animals

Male and female F344/N rats and B6C3F1 mice were obtained from Taconic Farms, Inc. (Germantown, NY), for use in the 2-year studies. Rats were quarantined for 13 days and mice were quarantined for 11 days before the beginning of the studies. Five male and five female rats and 10 male mice were randomly selected for parasite evaluation and gross observation of disease. Rats and mice were approximately 5 to 6 weeks old at the beginning of the studies. The health of the animals was monitored during the studies according to the protocols of the NTP Sentinel Animal Program (Appendix K).

Animal Maintenance

Rats and mice were housed individually. Feed was available *ad libitum* except during exposure periods; water was available *ad libitum*. Cages, racks, and chambers were changed weekly. Cages were rotated weekly in chambers. Further details of animal maintenance are given in Table 1. Information on feed composition and contaminants is provided in Appendix J.

Clinical Examinations and Pathology

All animals were observed twice daily. Clinical findings were recorded for all animals every 4 weeks through week 93, every 2 weeks thereafter, and at the end of the studies. Rats and mice were weighed initially, weekly for the first 13 weeks, then every 4 weeks through week 93, every 2 weeks thereafter, and at the end of the studies.

Complete necropsies and microscopic examinations were performed on all rats and mice. At necropsy, all organs and tissues were examined for grossly visible lesions, and all major tissues were fixed and preserved in 10% neutral buffered formalin (eyes were fixed in Davidson's solution for up to 72 hours and then transferred to 10% neutral buffered formalin), processed and trimmed, embedded in paraffin, sectioned to a thickness of 4 to 6 µm, and stained with hematoxylin and eosin for microscopic examination. For all paired organs (e.g., adrenal gland, kidney, ovary), samples from each

organ were examined. Tissues examined microscopically are listed in Table 1.

Microscopic evaluations were completed by the study laboratory pathologist, and the pathology data were entered into the Toxicology Data Management System. The reports, slides, paraffin blocks, residual wet tissues, and pathology data were sent to the NTP Archives for inventory, slide/block match, wet tissue audit, and storage. The slides, individual animal data records, and pathology tables were evaluated by an independent quality assessment laboratory. The individual animal records and tables were compared for accuracy, the slide and tissue counts were verified, and the histotechnique was evaluated. For the 2-year studies, a quality assessment pathologist evaluated slides from all tumors and all potential target organs, which included the larynx, nose, and trachea of rats and mice and the lung of mice.

The quality assessment report and the reviewed slides were submitted to the NTP PWG coordinator, who reviewed the selected tissues and addressed any inconsistencies in the diagnoses made by the laboratory and quality assessment pathologists. Representative histopathology slides containing examples of lesions related to chemical administration, examples of disagreements in diagnoses between the laboratory and quality assessment pathologists, or lesions of general interest were presented by the coordinator to the PWG for review. The PWG consisted of the quality assessment pathologist and other pathologists experienced in rodent toxicologic pathology. This group examined the tissues without any knowledge of dose groups. When the PWG consensus differed from the opinion of the laboratory pathologist, the diagnosis was changed. Final diagnoses for reviewed lesions represent a consensus between the laboratory pathologist, reviewing pathologist(s), and the PWG. Details of these review procedures have been described, in part, by Maronpot and Boorman (1982) and Boorman *et al.* (1985). For subsequent analyses of the pathology data, the decision of whether to evaluate the diagnosed lesions for each tissue type separately or combined was generally based on the guidelines of McConnell *et al.* (1986).

Three standard sections are taken through the nose in NTP studies, and these are referred to as Levels I, II, and III. Proceeding from anterior to posterior, Level I is taken immediately posterior to the upper incisor teeth; Level II is taken through the level of the incisive papilla anterior to the first palatal ridge; and Level III is taken through the middle of the second molar teeth (Figure 2). The mucosa of the nasal passages in Levels I and II is lined by respiratory and transitional epithelium, except for the ventral meatus of Levels I and II (squamous epithelium) and the dorsal meatus of Level II (olfactory epithelium). Level III is lined almost entirely by olfactory epithelium, except for the ventral meatus, which is lined by respiratory epithelium.

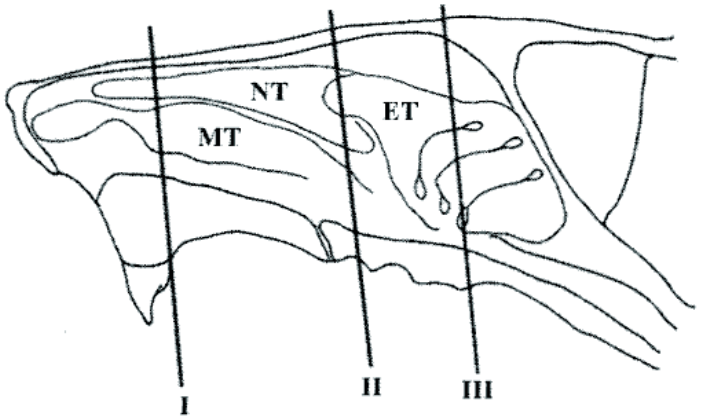


FIGURE 2
Rodent Nasal Cavity Diagram Illustrating the Levels of Sections
Level I: Immediately posterior to incisor teeth
Level II: At incisive papilla anterior to first palatal ridge
Level III: Section taken through the second molar
MT=maxilloturbinate; NT=nasoturbinate; ET=ethmoid turbinates

TABLE 1
Experimental Design and Materials and Methods in the Inhalation Studies of 1-Bromopropane

2-Week Studies	3-Month Studies	2-Year Studies
Study Laboratory Battelle Toxicology Northwest (Richland, WA)	Battelle Toxicology Northwest (Richland, WA)	Battelle Toxicology Northwest (Richland, WA)
Strain and Species F344/N rats B6C3F1 mice	F344/N rats B6C3F1 mice	F344/N rats B6C3F1 mice
Animal Source Taconic Farms, Inc. (Germantown, NY)	Taconic Farms, Inc. (Germantown, NY)	Taconic Farms, Inc. (Germantown, NY)
Time Held Before Studies 13 days	Rats: 12 (females) or 13 (males) days Mice: 12 days	Rats: 13 days Mice: 11 days
Average Age When Studies Began 5 to 6 weeks	5 to 6 weeks	5 to 6 weeks
Date of First Exposure July 15, 2002	Rats: November 11 (females) or 12 (males), 2002 Mice: November 11, 2002	Rats: July 14, 2003 Mice: July 21, 2003
Duration of Exposure 6 hours plus T_{90} (12 minutes) per day, 5 days per week, for 16 (rats) or 17 (mice) days	6 hours plus T_{90} (10 minutes) per day, 5 days per week, for 14 weeks	6 hours plus T_{90} (10 minutes) per day, 5 days per week, for 105 weeks
Date of Last Exposure Rats: July 30, 2002 Mice: July 31, 2002	Rats: February 10 (females) or 11 (males), 2003 Mice: February 12 (males) or 13 (females), 2003	Rats: July 13, 2005 Mice: July 21, 2005
Necropsy Dates Rats: July 31, 2002 Mice: August 1, 2002	Rats: February 11 (females) or 12 (males), 2003 Mice: February 13 (males) or 14 (females), 2003	Rats: July 11-14, 2005 Mice: July 18-22, 2005
Average Age at Necropsy 8 to 9 weeks	18 to 19 weeks	Rats: 110 weeks Mice: 109 to 111 weeks
Size of Study Groups 5 males and 5 females	10 males and 10 females	50 males and 50 females
Method of Distribution Animals were distributed randomly into groups of approximately equal initial mean body weights.	Same as 2-week studies	Same as 2-week studies
Animals per Cage 1	1	1
Method of Animal Identification Tail tattoo	Tail tattoo	Tail tattoo

TABLE 1
Experimental Design and Materials and Methods in the Inhalation Studies of 1-Bromopropane

2-Week Studies	3-Month Studies	2-Year Studies
Diet NTP-2000 irradiated pelleted diet (Zeigler Brothers, Inc., Gardners, PA); available *ad libitum*, except during exposure periods; changed weekly	Same as 2-week studies	Same as 2-week studies
Water Tap water (Richland, WA, municipal supply) via automatic watering system (Edstrom Industries, Waterford, WI); available *ad libitum*	Same as 2-week studies	Same as 2-week studies
Cages Stainless steel, wire bottom (Lab Products, Inc., Seaford, DE); changed weekly, rotated daily	Same as 2-week studies, except rotated weekly	Same as 3-month studies
Cageboard Untreated paper cage pan liner (Sheperd Specialty Papers, Kalamazoo, MI); changed daily	Same as 2-week studies	Same as 2-week studies
Chamber Air Supply Filters Single HEPA (open stock), charcoal (RSE, Inc., New Baltimore, MI), Purafil (Environmental Systems, Lynnwood, WA), all new at study start	Same as 2-week studies, except not changed after 2-week studies	Same as 2-week studies, except single HEPA changed annually
Chambers Stainless steel, excreta pan at each of six levels (Lab Products, Inc., Seaford, DE); chambers changed weekly; excreta pans changed daily	Same as 2-week studies	Same as 2-week studies
Chamber Environment Temperature: 72° ± 3° F Relative humidity: 50% ± 15% Room fluorescent light: 12 hours/day Chamber air changes: 15 ± 2/hour	Temperature: 72° ± 3° F Relative humidity: 50% ± 15% Room fluorescent light: 12 hours/day Chamber air changes: 15 ± 2/hour	Temperature: 72° ± 3° F Relative humidity: 50% ± 15% Room fluorescent light: 12 hours/day Chamber air changes: 15 ± 2/hour
Exposure Concentrations 0, 125, 250, 500, 1,000, or 2,000 ppm	Rats: 0, 62.5, 125, 250, 500, or 1,000 ppm Mice: 0, 62.5, 125, 250, or 500 ppm	Rats: 0, 125, 250, or 500 ppm Mice: 0, 62.5, 125, or 250 ppm
Type and Frequency of Observation Observed twice daily; animals were weighed initially, on days 6 and 13, and at the end of the studies; clinical findings were recorded twice daily on exposure days.	Observed twice daily; core study animals were weighed initially, on days 9 (male rats) or 10, weekly thereafter, and at the end of the studies. Clinical findings were recorded on day 9 (male rats) or 10 and then weekly.	Observed twice daily; animals were weighed initially, weekly for 13 weeks, every 4 weeks through week 93, every 2 weeks thereafter, and at the end of the studies; clinical findings were recorded every 4 weeks through week 93, every 2 weeks thereafter, and at the end of the studies.

TABLE 1
Experimental Design and Materials and Methods in the Inhalation Studies of 1-Bromopropane

2-Week Studies	3-Month Studies	2-Year Studies
Method of Sacrifice		
Carbon dioxide asphyxiation	Same as 2-week studies	Same as 2-week studies
Necropsy		
Necropsies were performed on all animals. Organs weighed were heart, right kidney, liver, lung, right testis, and thymus.	Necropsies were performed on core study animals. Organs weighed were heart, right kidney, liver, lung, spleen, right testis, and thymus.	Necropsies were performed on all animals.
Clinical Pathology		
None	Blood was collected from the retroorbital sinus of clinical pathology rats on days 3 and 23 and from core study rats at the end of the studies for hematology and clinical chemistry. Blood was collected from the retroorbital sinus of mice at the end of the study for hematology. ***Hematology:*** hematocrit; packed red cell volume; hemoglobin; erythrocyte, nucleated erythrocyte, reticulocyte, and platelet counts; mean cell volume; mean cell hemoglobin; mean cell hemoglobin concentration; and leukocyte count and differentials ***Clinical chemistry:*** urea nitrogen, creatinine, total protein, albumin, globulin, alanine aminotransferase, alkaline phosphatase, creatine kinase, sorbitol dehydrogenase, and bile acids	None
Histopathology		
Histopathology was performed on 0 and 2,000 ppm rats and mice. In addition to gross lesions and tissue masses, the following tissues were examined: lung, mediastinal lymph node, nose, sciatic nerve, and spinal cord of rats and liver, lung, and nose of mice. These tissues were examined to a no-effect level in the remaining exposure groups.	Complete histopathology was performed on 0 and 1,000 ppm core study rats and 0, 250, and 500 ppm mice. In addition to gross lesions and tissue masses, the following tissues were examined to a no-effect level: adrenal gland, bone with marrow, brain, clitoral gland, esophagus, eyes, gallbladder (mice), Harderian gland, heart and aorta, large intestine (cecum, colon, rectum), small intestine (duodenum, jejunum, ileum), kidney, larynx, liver, lung, lymph nodes (mandibular, mesenteric, bronchial, mediastinal), mammary gland, nose, ovary, pancreas, parathyroid gland, pituitary gland, preputial gland, prostate gland, salivary gland, skin, spleen, stomach (forestomach and glandular), testis with epididymis and seminal vesicle, thymus, thyroid gland, trachea, urinary bladder, and uterus. In addition, the liver, larynx, lung, and nose of rats and mice; the trachea of mice; the prostate gland of male rats; and the adrenal gland and kidney of female mice were examined in the remaining groups.	Complete histopathology was performed on all rats and mice. In addition to gross lesions and tissue masses, the following tissues were examined: adrenal gland, bone with marrow, brain, clitoral gland, esophagus, eye, gallbladder (mice only), Harderian gland, heart with aorta, large intestine (cecum, colon, rectum), small intestine (duodenum, jejunum, ileum), kidney, larynx, liver, lung, lymph nodes (mandibular, mesenteric, bronchial, mediastinal), mammary gland, nose, ovary, pancreas, pancreatic islets, parathyroid gland, pituitary gland, preputial gland, prostate gland, salivary gland, skin, spleen, stomach (forestomach and glandular), testis with epididymis and seminal vesicle, thymus, thyroid gland, trachea, urinary bladder, and uterus.

TABLE 1
Experimental Design and Materials and Methods in the Inhalation Studies of 1-Bromopropane

2-Week Studies	3-Month Studies	2-Year Studies
Sperm Motility and Vaginal Cytology		
None	At the end of the studies, sperm samples were collected from male animals in the 0, 125 (mice), 250, 500, and 1,000 (rats) ppm groups for sperm motility evaluations. The following parameters were evaluated: spermatid heads per testis and per gram testis, spermatid counts, and epididymal spermatozoal motility and concentration. The left cauda, left epididymis, and left testis were weighed. Vaginal samples were collected for up to 12 consecutive days prior to the end of the studies from females exposed to 0, 125 (mice), 250, 500, or 1,000 (rats) ppm for vaginal cytology evaluations. The percentage of time spent in the various estrous cycle stages and estrous cycle length were evaluated.	None

STATISTICAL METHODS

Survival Analyses

The probability of survival was estimated by the product-limit procedure of Kaplan and Meier (1958) and is presented in the form of graphs. Animals found dead of other than natural causes were censored; animals dying from natural causes were not censored. Statistical analyses for possible dose-related effects on survival used Cox's (1972) method for testing two groups for equality and Tarone's (1975) life table test to identify dose-related trends. All reported P values for the survival analyses are two sided.

Calculation of Incidence

The incidences of neoplasms or nonneoplastic lesions are presented in Tables A1, A4, B1, B4, C1, C3, D1, and D4 as the numbers of animals bearing such lesions at a specific anatomic site and the numbers of animals with that site examined microscopically. For calculation of statistical significance, the incidences of most neoplasms (Tables A2, B2, C2, and D2) and all nonneoplastic lesions are given as the numbers of animals affected at each site examined microscopically. However, when macroscopic examination was required to detect neoplasms in certain tissues (e.g., mesentery, pleura, peripheral nerve, skeletal muscle, tongue, tooth, and Zymbal's gland) before microscopic evaluation, the denominators consist of the number of animals that had a gross abnormality. When neoplasms had multiple potential sites of occurrence (e.g., leukemia or lymphoma), the denominators consist of the number of animals on which a necropsy was performed. Tables A2, B2, C2, and D2 also give the survival-adjusted neoplasm rate for each group and each site-specific neoplasm. This survival-adjusted rate (based on the Poly-3 method described below) accounts for differential mortality by assigning a reduced risk of neoplasm, proportional to the third power of the fraction of time on study, only to site-specific, lesion-free animals that do not reach terminal sacrifice.

Analysis of Neoplasm and Nonneoplastic Lesion Incidences

The Poly-k test (Bailer and Portier, 1988; Portier and Bailer, 1989; Piegorsch and Bailer, 1997) was used to assess neoplasm and nonneoplastic lesion prevalence. This test is a survival-adjusted quantal-response procedure that modifies the Cochran-Armitage linear trend test to take survival differences into account. More specifically, this method modifies the denominator in the quantal estimate of lesion incidence to approximate more closely the total number of animal years at risk. For analysis of a given site, each animal is assigned a risk weight. This value is one if the animal had a lesion at that site or if it survived until terminal sacrifice; if the animal died prior to terminal sacrifice and did not have

a lesion at that site, its risk weight is the fraction of the entire study time that it survived, raised to the kth power.

This method yields a lesion prevalence rate that depends only upon the choice of a shape parameter for a Weibull hazard function describing cumulative lesion incidence over time (Bailer and Portier, 1988). Unless otherwise specified, a value of k=3 was used in the analysis of site-specific lesions. This value was recommended by Bailer and Portier (1988) following an evaluation of neoplasm onset time distributions for a variety of site-specific neoplasms in control F344 rats and B6C3F1 mice (Portier *et al.*, 1986). Bailer and Portier (1988) showed that the Poly-3 test gave valid results if the true value of k was anywhere in the range from 1 to 5. A further advantage of the Poly-3 method is that it does not require lesion lethality assumptions. Variation introduced by the use of risk weights, which reflect differential mortality, was accommodated by adjusting the variance of the Poly-3 statistic as recommended by Bieler and Williams (1993).

Tests of significance included pairwise comparisons of each exposed group with controls and a test for an overall exposure-related trend. Continuity-corrected Poly-3 tests were used in the analysis of lesion incidence, and reported P values are one sided. The significance of lower incidences or decreasing trends in lesions is represented as 1–P with the letter N added (e.g., P=0.99 is presented as P=0.01N).

Analysis of Continuous Variables

Two approaches were employed to assess the significance of pairwise comparisons between exposed and control groups in the analysis of continuous variables. Organ and body weight data, which historically have approximately normal distributions, were analyzed with the parametric multiple comparison procedures of Dunnett (1955) and Williams (1971, 1972). Hematology, clinical chemistry, spermatid, and epididymal spermatozoal data, which have typically skewed distributions, were analyzed using the nonparametric multiple comparison methods of Shirley (1977) (as modified by Williams, 1986) and Dunn (1964). Jonckheere's test (Jonckheere, 1954) was used to assess the significance of the dose-related trends and to determine whether a trend-sensitive test (Williams' or Shirley's test) was more appropriate for pairwise comparisons than a test that does not assume a monotonic dose-related trend (Dunnett's or Dunn's test). Prior to statistical analysis, extreme values identified by the outlier test of Dixon and Massey (1957) were examined by NTP personnel, and implausible values were eliminated from the analysis. Because vaginal cytology data are proportions (the proportion of the observation period that an animal was in a given estrous stage), an arcsine transformation was used to bring the data into closer conformance with a normality assumption. Treatment effects were investigated by applying a multivariate analysis of variance (Morrison, 1976) to the transformed data to test for simultaneous equality of measurements across exposure concentrations. Proportions of regular cycling females in each exposed group were compared to the control group using the Fisher exact test (Gart *et al.*, 1979). Tests for extended periods of estrus and diestrus were constructed based on a Markov chain model proposed by Girard and Sager (1987). For each exposure group, a transition probability matrix was estimated for transitions among the proestrus, estrus, metestrus, and diestrus stages, with provision for extended stays within estrus and diestrus. Equality of transition matrices among exposure groups and between the control group and each exposed group was tested using chi-square statistics.

Historical Control Data

The concurrent control group represents the most valid comparison to the treated groups and is the only control group analyzed statistically in NTP bioassays. However, historical control data are often helpful in interpreting potential treatment-related effects, particularly for uncommon or rare neoplasm types. For meaningful comparisons, the conditions for studies in the historical database must be generally similar. One significant factor affecting the background incidence of neoplasms at a variety of sites is diet. In 1995, the NTP incorporated a new diet (NTP-2000) that contains less protein and more fiber and fat than the NIH-07 diet previously used in toxicity and carcinogenicity studies (Rao, 1996, 1997). The NTP historical database contains all studies that use the NTP-2000 diet with histopathology findings completed within the most recent 5-year period. A second potential source of variability is route of administration. In general, the historical database for a given study will include studies using the same route of administration, and the overall incidences of neoplasms for all routes of administration are included for comparison, including the present study.

QUALITY ASSURANCE METHODS

The 3-month and 2-year studies were conducted in compliance with Food and Drug Administration Good Laboratory Practice Regulations (21 CFR, Part 58). In

addition, as records from the 2-year studies were submitted to the NTP Archives, these studies were audited retrospectively by an independent quality assurance contractor. Separate audits covered completeness and accuracy of the pathology data, pathology specimens, final pathology tables, and a draft of this NTP Technical Report. Audit procedures and findings are presented in the reports and are on file at NIEHS. The audit findings were reviewed and assessed by NTP staff, and all comments were resolved or otherwise addressed during the preparation of this Technical Report.

GENETIC TOXICOLOGY

The genetic toxicity of 1-bromopropane was assessed by testing the ability of the chemical to induce mutations in various strains of *Salmonella typhimurium* and *Escherichia coli* and increases in the frequency of micronucleated erythrocytes in mouse peripheral blood. Micronuclei (literally "small nuclei" or Howell-Jolly bodies) are biomarkers of induced structural or numerical chromosomal alterations and are formed when acentric fragments or whole chromosomes fail to incorporate into either of two daughter nuclei during cell division (Schmid, 1975; Heddle *et al.*, 1983). The protocols for these studies and the results are given in Appendix E.

The genetic toxicity studies have evolved from an earlier effort by the NTP to develop a comprehensive database permitting a critical anticipation of a chemical's carcinogenicity in experimental animals based on numerous considerations, including the molecular structure of the chemical and its observed effects in short-term *in vitro* and *in vivo* genetic toxicity tests (structure-activity relationships). The short-term tests were originally developed to clarify proposed mechanisms of chemical-induced DNA damage based on the relationship between electrophilicity and mutagenicity (Miller and Miller, 1977) and the somatic mutation theory of cancer (Straus, 1981; Crawford, 1985). However, it should be noted that not all cancers arise through genotoxic mechanisms.

DNA reactivity combined with *Salmonella* mutagenicity is highly correlated with induction of carcinogenicity in multiple species/sexes of rodents and at multiple tissue sites (Ashby and Tennant, 1991). A positive response in the *Salmonella* test was shown to be the most predictive *in vitro* indicator for rodent carcinogenicity (89% of the *Salmonella* mutagens are rodent carcinogens) (Tennant *et al.*, 1987; Zeiger *et al.*, 1990). Additionally, no battery of tests that included the *Salmonella* test improved the predictivity of the *Salmonella* test alone. However, these other tests can provide useful information on the types of DNA and chromosomal damage induced by the chemical under investigation.

The predictivity for carcinogenicity of a positive response in acute *in vivo* bone marrow chromosome aberration or micronucleus tests appears to be less than that in the *Salmonella* test (Shelby *et al.*, 1993; Shelby and Witt, 1995). However, clearly positive results in long-term peripheral blood micronucleus tests have high predictivity for rodent carcinogenicity; a weak response in one sex only or negative results in both sexes in this assay do not correlate well with either negative or positive results in rodent carcinogenicity studies (Witt *et al.*, 2000). Because of the theoretical and observed associations between induced genetic damage and adverse effects in somatic and germ cells, the determination of *in vivo* genetic effects is important to the overall understanding of the risks associated with exposure to a particular chemical. Most organic chemicals that are identified by the International Agency for Research on Cancer as human carcinogens, other than hormones, are genotoxic. The vast majority of these are detected by both the *Salmonella* assay and rodent bone marrow cytogenetics tests (Shelby, 1988; Shelby and Zeiger, 1990).

RESULTS

RATS

2-WEEK STUDY

Except for one 500 ppm male that died on day 14, all rats survived to the end of the study (Table 2). Final mean body weights and body weight gains of 2,000 ppm rats were significantly less than those of the chamber controls, as was the body weight gain of 1,000 ppm males. The neurological sign of hind limb splaying was observed in some 2,000 ppm rats after 1 week of exposure to 1-bromopropane. This was a transient change and the animals showed marked improvement after a day.

The absolute weights of right kidney in 1,000 ppm males and all exposed groups of females were significantly greater than those of the chamber controls (Table G1). The relative right kidney weights were significantly increased in all exposed groups of both sexes. Absolute liver weights were significantly increased in 1,000 ppm males and 500 ppm or greater females. Relative liver weights were significantly increased in 500 ppm or greater males and all exposed groups of females.

TABLE 2
Survival and Body Weights of Rats in the 2-Week Inhalation Study of 1-Bromopropane

Concentration (ppm)	Survival[a]	Mean Body Weight[b] (g)			Final Weight Relative to Controls (%)
		Initial	Final	Change	
Male					
0	5/5	99 ± 4	169 ± 4	69 ± 3	
125	5/5	99 ± 4	167 ± 4	68 ± 1	99
250	5/5	98 ± 4	169 ± 5	70 ± 3	100
500	4/5[c]	98 ± 4	166 ± 6	69 ± 1	98
1,000	5/5	102 ± 4	164 ± 5	63 ± 2*	97
2,000	5/5	98 ± 4	123 ± 4**	25 ± 2**	73
Female					
0	5/5	87 ± 3	120 ± 2	33 ± 1	
125	5/5	88 ± 2	124 ± 3	37 ± 2	104
250	5/5	86 ± 3	120 ± 4	34 ± 3	100
500	5/5	89 ± 3	124 ± 5	35 ± 3	104
1,000	5/5	88 ± 2	119 ± 2	31 ± 1	99
2,000	5/5	89 ± 2	107 ± 2*	18 ± 1**	89

* Significantly different ($P \leq 0.05$) from the chamber control group by Williams' or Dunnett's test

** $P \leq 0.01$

[a] Number of animals surviving at 2 weeks/number initially in group

[b] Weights and weight changes are given as mean ± standard error. Subsequent calculations are based on animals surviving to the end of the study.

[c] Day of death: 14

Microscopic lesions related to 1-bromopropane exposure occurred in the nose. Incidences of minimal to mild suppurative inflammation occurred in males exposed to 500 ppm or greater, incidences of minimal respiratory epithelium necrosis occurred in 1,000 and 2,000 ppm males, and incidences of minimal respiratory epithelium regeneration occurred in females exposed to 1,000 and 2,000 ppm (Table 3).

Sciatic nerve and spinal cord were examined microscopically, but no lesions were identified.

Exposure Concentration Selection Rationale: Based on decreased body weights and signs of neurotoxicity at 2,000 ppm in the 2-week study, 1-bromopropane exposure concentrations selected for the 3-month inhalation study in rats were 62.5, 125, 250, 500, and 1,000 ppm.

TABLE 3
Incidences of Nonneoplastic Lesions in the Nose of Rats in the 2-Week Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm	1,000 ppm	2,000 ppm
Male						
Number Examined Microscopically	5	0	5	5	5	5
Respiratory Epithelium,						
Necrosis[a]	0		0	0	2 (1.0)[b]	1 (1.0)
Suppurative Inflammation	0		0	1 (1.0)	2 (1.5)	2 (1.5)
Female						
Number Examined Microscopically	5	0	0	5	5	5
Respiratory Epithelium,						
Regeneration	0			0	1 (1.0)	1 (1.0)

[a] Number of animals with lesion
[b] Average severity grade of lesions in affected animals: 1=minimal, 2=mild, 3=moderate, 4=marked

3-MONTH STUDY

All rats survived to the end of the study (Table 4). The final mean body weight and mean body weight gain of 1,000 ppm males were significantly less than those of the chamber controls (Table 4 and Figure 3). Mean body weights of exposed females were similar to those of the chamber controls.

There were no changes in hematology endpoints that were considered related to 1-bromopropane exposure (Table F1). There were early, transient decreases in albumin and total protein concentrations and alanine aminotransferase activities in most exposed groups of male and female rats. These transient decreases may have been associated with effects of 1-bromopropane on hepatic protein metabolism. Additionally, sorbitol dehydrogenase (SDH) activity was increased at day 23 and at the end of the study in 1,000 ppm females and at the end of the study in 500 and 1,000 ppm males. Increased SDH activity would be consistent with mild hepatotoxicity caused by 1-bromopropane.

The absolute and relative liver weights of males exposed to 250 ppm or greater and females exposed to 125 ppm or greater were significantly increased (Table G2). The absolute spleen weights of females exposed to 125 ppm or greater and the relative spleen weight of 1,000 ppm females were greater than those of the chamber controls. In addition, the absolute and relative right kidney weights of 1,000 ppm females were greater than those of the chamber controls.

TABLE 4
Survival and Body Weights of Rats in the 3-Month Inhalation Study of 1-Bromopropane

Concentration (ppm)	Survival[a]	Mean Body Weight[b] (g)			Final Weight Relative to Controls (%)
		Initial	Final	Change	
Male					
0	10/10	99 ± 2	345 ± 7	246 ± 6	
62.5	10/10	102 ± 2	346 ± 5	244 ± 5	100
125	10/10	100 ± 2	348 ± 6	248 ± 5	101
250	10/10	101 ± 3	350 ± 7	250 ± 6	101
500	10/10	99 ± 2	344 ± 10	245 ± 10	100
1,000	10/10	100 ± 2	305 ± 6**	205 ± 4**	88
Female					
0	10/10	88 ± 1	202 ± 3	114 ± 3	
62.5	10/10	92 ± 3	203 ± 5	111 ± 3	101
125	10/10	88 ± 2	207 ± 4	120 ± 4	103
250	10/10	91 ± 2	209 ± 2	119 ± 3	104
500	10/10	86 ± 2	205 ± 3	119 ± 3	102
1,000	10/10	88 ± 2	190 ± 4	102 ± 3	95

** Significantly different ($P \leq 0.01$) from the chamber control group by Dunnett's or Williams' test

[a] Number of animals surviving at 3 months/number initially in group

[b] Weights and weight changes are given as mean ± standard error.

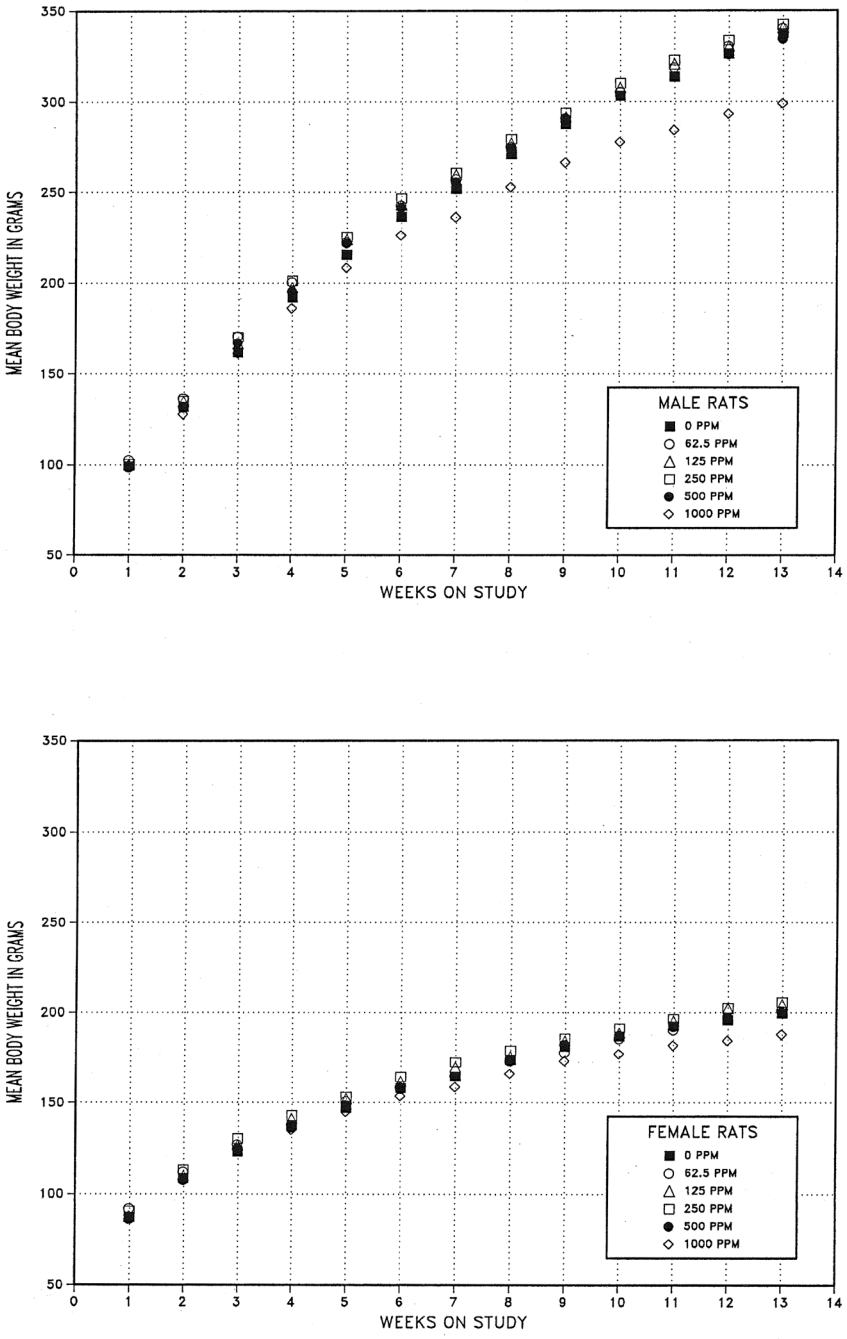


FIGURE 3
Growth Curves for Rats Exposed to 1-Bromopropane by Inhalation for 3 Months

There were significant exposure concentration-related decreases in sperm motility in male rats exposed to 250 ppm or greater (7%, 10%, and 28% in the 250, 500, and 1,000 ppm groups, respectively), and there were significant decreases in the number of sperm per gram cauda and the total sperm per cauda (25% and 37%, respectively), as well as significant decreases in the absolute weights of the cauda (14%) and left epididymis (19%) of 1,000 ppm male rats (Table H1). Female rats in each of the exposure groups evaluated differed significantly from the chamber controls in the relative amount of time spent in the various estrous cycle stages, with each exposed group spending significantly more time in extended estrus and significantly less time in extended diestrus (Table H2).

Treatment-related lesions occurred in the liver. There were significantly increased incidences of hepatocellular cytoplasmic vacuolization in males exposed to 250 ppm or greater and in females exposed to 500 or 1,000 ppm (Table 5). The incidence of hepatocellular degeneration was significantly increased in females exposed to 1,000 ppm. Hepatocellular cytoplasmic vacuolization consisted of swollen hepatocytes with centrally located nuclei and one to three variably sized vacuoles displacing the cytoplasm. Hepatocellular vacuolar degeneration consisted of a distinct population of enlarged, pale-staining, degenerative, "balloon-like" cells admixed with low numbers of necrotic hepatocytes. The lesion involved hepatocytes surrounding the central vein, but in the most severe cases, the vacuolated hepatocytes extended into the midzonal region.

The incidence of minimal suppurative inflammation of the prostate in 1,000 ppm males was increased, but the increase was not statistically significant (data not shown). The increasing trend of this lesion, however, was significant. Because this lesion is a common background finding in F344/N rats, the biological significance of the increased incidence in the 1,000 ppm males is unclear (Boorman *et al*., 1990).

Exposure Concentration Selection Rationale: Based on decreased body weights and increased incidences of liver lesions in the 3-month study, 1-bromopropane exposure concentrations selected for the 2-year inhalation study in rats were 125, 250, and 500 ppm. Although 500 ppm caused vacuolization in liver, this lesion was not considered life-threatening and the use of this concentration was not expected to compromise the 2-year study.

TABLE 5
Incidences of Nonneoplastic Lesions of the Liver in Rats in the 3-Month Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm	500 ppm	1,000 ppm
Male						
Number Examined Microscopically	10	10	10	10	10	10
Vacuolization Cytoplasmic[a]	0	0	0	5* (1.0)[b]	10**(2.2)	10**(2.8)
Female						
Number Examined Microscopically	10	10	10	10	10	10
Vacuolization Cytoplasmic	0	0	0	0	10**(1.1)	10**(2.6)
Hepatocyte, Degeneration	0	0	0	0	0	7**(1.9)

* Significantly different ($P \leq 0.05$) from the chamber control group by the Fisher exact test

** $P \leq 0.01$

[a] Number of animals with lesion

[b] Average severity grade of lesions in affected animals: 1=minimal, 2=mild, 3=moderate, 4=marked

2-YEAR STUDY

Survival

Estimates of 2-year survival probabilities for male and female rats are shown in Table 6 and in the Kaplan-Meier survival curves (Figure 4). Survival of 500 ppm males was significantly less than that of the chamber control group. The majority of the early deaths in the 500 ppm males were attributed to various types of neoplasia, none of which were treatment related. However, in nine of these males, the cause of death was attributed to inflammation in various organs, all of which contained Splendore-Hoeppli material. Survival of exposed groups of females decreased with increasing exposure concentration, but survival in each exposed group was not significantly different from that of the chamber control group.

TABLE 6
Survival of Rats in the 2-Year Inhalation Study of 1-Bromopropane

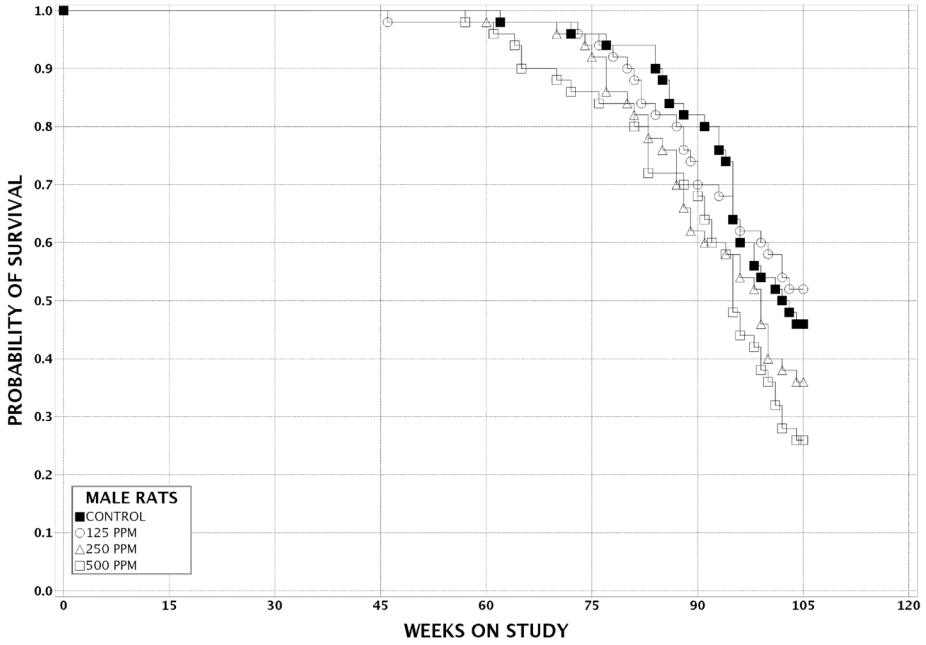
	Chamber Control	125 ppm	250 ppm	500 ppm
Male				
Animals initially in study	50	50	50	50
Moribund	24	20	28	35
Natural deaths	3	4	4	2
Animals surviving to study termination	23	26	18	13
Percent probability of survival at end of study[a]	46	52	36	26
Mean survival (days)[b]	679	671	654	639
Survival analysis[c]	P=0.009	P=0.844N	P=0.262	P=0.033
Female				
Animals initially in study	50	50	50	50
Accidental deaths[d]	0	0	0	2
Moribund	13	17	17	23
Natural deaths	3	0	3	1
Animals surviving to study termination	34	33	30	24
Percent probability of survival at end of study	68	66	60	50
Mean survival (days)	702	692	683	637
Survival analysis	P=0.028	P=0.858	P=0.419	P=0.054

[a] Kaplan-Meier determinations

[b] Mean of all deaths (uncensored, censored, and terminal sacrifice)

[c] The result of the life table trend test (Tarone, 1975) is in the chamber control column, and the results of the life table pairwise comparisons (Cox, 1972) with the chamber controls are in the exposed group columns. A lower mortality in an exposed group is indicated by **N**.

[d] Censored from survival analyses



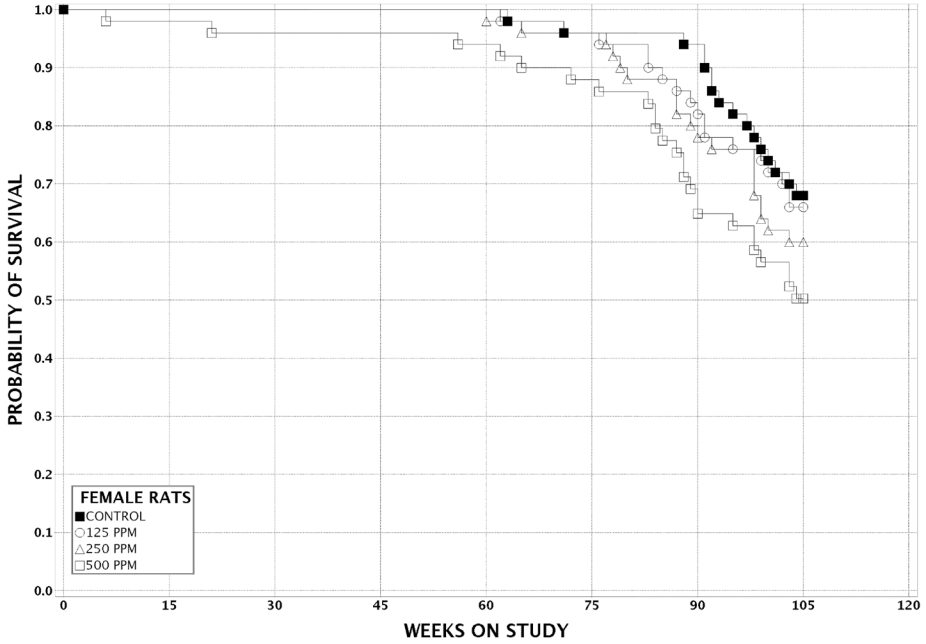
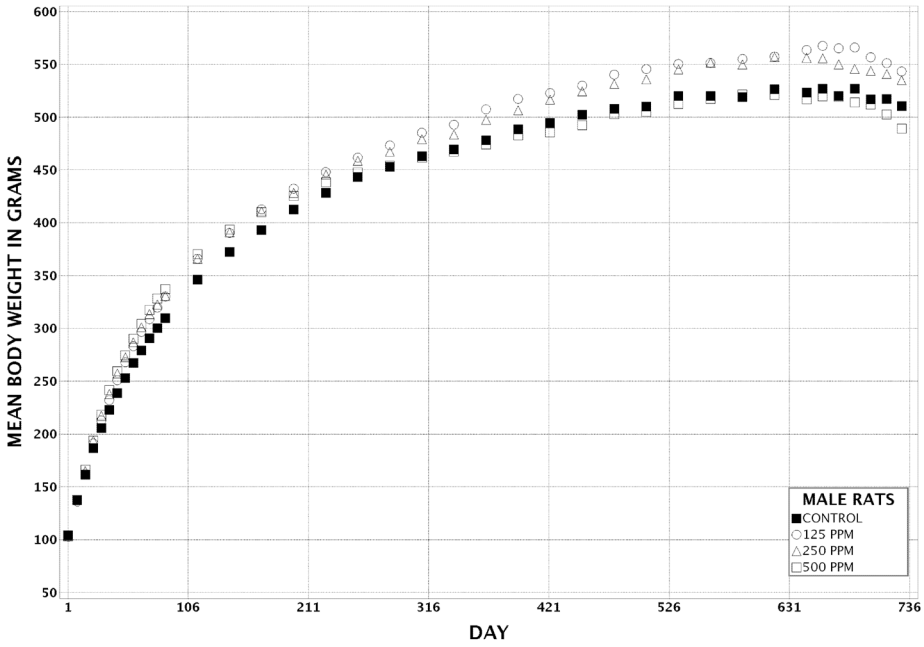


FIGURE 4
Kaplan-Meier Survival Curves for Rats
Exposed to 1-Bromopropane by Inhalation for 2 Years

Body Weights and Clinical and Macroscopic Findings

Mean body weights of exposed groups of male and female rats were similar to those of the chamber controls throughout the study (Figure 5; Tables 7 and 8). In males, clinical findings that appeared to be related to 1-bromopropane exposure included head mass (0 ppm, 1/50; 125 ppm, 2/50; 250 ppm, 5/50, 500 ppm, 9/50) and torso/ventral ulcer/abscess (2/50, 7/50, 6/50, 20/50). No clinical findings related to exposure to 1-bromopropane were observed in females. In male and female rats exposed to 1-bromopropane, there was an exposure-related increased incidence of soft, pale-yellow to green, variably sized nodules. These lesions were predominantly located in the nose and/or skin, but other sites included bone, ear, Harderian gland, larynx, lung, muscle, peritoneum, preputial gland, and prostate gland. The incidences of these lesions were greater in males (2, 3, 6, 15) than in females (1, 2, 0, 7). In addition, the number of animals with multiple masses was increased in the 500 ppm groups. In most cases, these lesions were microscopically shown to be suppurative inflammation, many with Splendore-Hoeppli material.



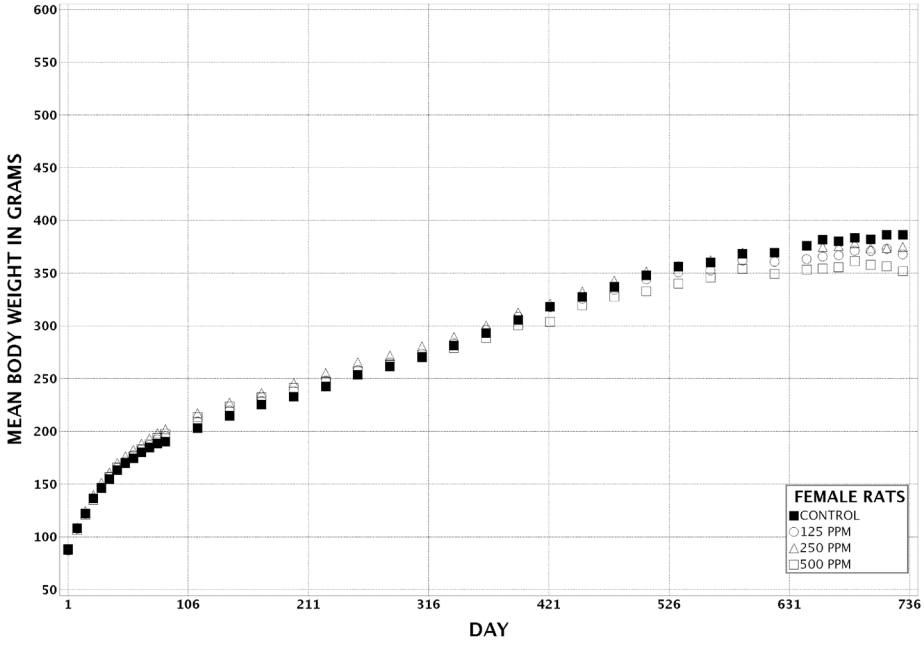


FIGURE 5
Growth Curves for Rats Exposed to 1-Bromopropane by Inhalation for 2 Years

TABLE 7
Mean Body Weights and Survival of Male Rats in the 2-Year Inhalation Study of 1-Bromopropane

Days on Study	Chamber Control		125 ppm			250 ppm			500 ppm		
	Av. Wt. (g)	No. of Survivors	Av. Wt. (g)	Wt. (% of controls)	No. of Survivors	Av. Wt. (g)	Wt. (% of controls)	No. of Survivors	Av. Wt. (g)	Wt. (% of controls)	No. of Survivors
1	104	50	103	99	50	103	99	50	104	100	50
9	137	50	136	99	50	138	100	50	138	100	50
16	162	50	163	101	50	165	102	50	166	103	50
23	187	50	191	102	50	194	104	50	194	104	50
30	206	50	213	104	50	217	106	50	218	106	50
37	223	50	232	104	50	238	107	50	242	109	50
44	239	50	251	105	50	257	108	50	260	109	50
51	253	50	268	106	50	273	108	50	275	108	50
58	267	50	283	106	50	287	107	50	290	109	50
65	279	50	297	106	50	301	108	50	304	109	50
72	291	50	309	106	50	314	108	50	318	109	50
79	300	50	320	107	50	323	108	50	328	109	50
86	310	50	330	107	50	331	107	50	337	109	50
114	346	50	366	106	50	366	106	50	370	107	50
142	373	50	390	105	50	391	105	50	394	106	50
170	393	50	413	105	50	411	104	50	410	104	50
198	413	50	432	105	50	428	104	50	426	103	50
226	428	50	448	105	50	446	104	50	438	102	50
254	443	50	462	104	50	459	104	50	448	101	50
282	453	50	473	105	50	467	103	50	454	100	50
310	463	50	485	105	50	479	103	50	462	100	50
338	470	50	493	105	49	484	103	50	467	100	50
366	478	50	508	106	49	498	104	50	474	99	50
394	489	50	517	106	49	507	104	50	483	99	49
422	495	50	523	106	49	517	104	49	486	98	48
450	502	49	530	106	49	525	104	49	493	98	46
478	508	49	540	106	49	532	105	49	503	99	45
506	510	48	546	107	48	536	105	48	505	99	43
534	520	47	551	106	47	545	105	44	513	99	42
562	520	47	551	106	44	552	106	41	517	100	40
590	519	45	555	107	41	550	106	38	522	101	36
618	527	41	557	106	37	557	106	31	521	99	35
646	523	38	564	108	35	556	106	30	517	99	30
660	527	32	568	108	33	556	106	29	520	99	25
674	520	30	565	109	31	550	106	27	520	100	22
688	527	27	566	107	30	546	104	25	514	98	19
702	517	27	557	108	29	544	105	20	512	99	16
716	517	24	551	107	27	541	105	19	503	97	14
Mean for weeks											
1-13	228		238	104		242	106		244	106	
14-52	420		440	105		437	104		430	103	
53-103	512		547	107		538	105		506	99	

TABLE 8
Mean Body Weights and Survival of Female Rats in the 2-Year Inhalation Study of 1-Bromopropane

Days on Study	Chamber Control		125 ppm			250 ppm			500 ppm		
	Av. Wt. (g)	No. of Survivors	Av. Wt. (g)	Wt. (% of controls)	No. of Survivors	Av. Wt. (g)	Wt. (% of controls)	No. of Survivors	Av. Wt. (g)	Wt. (% of controls)	No. of Survivors
1	88	50	87	99	50	88	100	50	88	99	50
9	108	50	108	100	50	109	101	50	106	98	50
16	122	50	122	100	50	125	102	50	121	99	50
23	137	50	137	100	50	140	102	50	135	99	50
30	146	50	147	100	50	151	103	50	147	100	50
37	155	50	157	102	50	161	104	50	157	101	50
44	163	50	166	101	50	170	104	50	166	102	49
51	170	50	172	101	50	177	104	50	170	100	49
58	175	50	177	102	50	183	105	50	176	101	49
65	180	50	183	101	50	188	105	50	183	102	49
72	185	50	187	102	50	194	105	50	188	102	49
79	189	50	192	102	50	198	105	50	194	103	49
86	191	50	196	103	50	202	106	50	198	104	49
114	203	50	210	103	50	217	107	50	214	105	49
142	215	50	220	102	50	227	106	50	224	104	48
170	226	50	229	102	50	237	105	50	232	103	48
198	233	50	238	102	50	246	106	50	241	104	48
226	243	50	248	102	50	256	105	50	247	102	48
254	254	50	258	102	50	266	105	50	257	101	48
282	262	50	265	101	50	272	104	50	263	101	48
310	270	50	274	101	50	281	104	50	271	100	48
338	281	50	283	101	50	290	103	50	279	99	48
366	293	50	297	101	50	300	102	50	289	98	48
394	306	50	309	101	50	313	102	50	301	98	47
422	318	50	318	100	50	321	101	49	304	96	47
450	328	49	326	99	49	333	102	49	320	98	45
478	337	49	334	99	49	343	102	48	328	97	44
506	348	48	344	99	48	352	101	48	333	96	42
534	357	48	351	98	47	357	100	47	340	95	41
562	360	48	353	98	47	362	101	44	346	96	41
590	369	48	362	98	44	370	100	44	354	96	37
618	370	47	361	98	42	370	100	40	350	95	34
646	376	43	363	97	39	376	100	38	353	94	31
660	382	41	366	96	38	375	98	38	355	93	30
674	380	41	367	97	38	376	99	38	356	94	30
688	384	38	371	97	37	379	99	32	362	94	27
702	382	36	371	97	36	374	98	31	358	94	27
716	387	35	373	97	34	374	97	31	357	92	26
Mean for weeks											
1-13	155		156	101		160	104		156	101	
14-52	243		247	102		255	105		248	102	
53-103	355		348	98		355	100		338	95	

Pathology and Statistical Analyses

This section describes the statistically significant or biologically noteworthy changes in the incidences of malignant mesothelioma and neoplasms or nonneoplastic lesions of the large intestine, skin, pancreatic islets, nose, larynx, trachea, and lung. Summaries of the incidences of neoplasms and nonneoplastic lesions, statistical analyses of primary neoplasms that occurred with an incidence of at least 5% in at least one animal group, and historical incidences for the neoplasms mentioned in this section are presented in Appendix A for male rats and Appendix B for female rats.

Large Intestine: The incidence of adenoma of the large intestine (colon or rectum) in 500 ppm females was significantly greater than that in the chamber controls (Tables 9, B1, and B2). The incidences in the 250 and 500 ppm groups of females exceeded the historical ranges for controls in inhalation studies and all routes of administration (Tables 9 and B3a). In 250 and 500 ppm males, the incidences of adenoma of the large intestine were slightly increased compared to that in the chamber controls; although the increases were not statistically significant, the incidence in the 250 ppm group exceeded the historical control ranges for inhalation studies and all routes (Tables 9 and A3a).

Large intestine adenomas were polypoid masses that protruded into the intestinal lumen (Plates 1 and 2). The epithelium lining the glands had fewer goblet cells than the epithelium from normal glands and was occasionally thickened by multiple layers of slightly enlarged neoplastic epithelial cells with basophilic cytoplasm and enlarged nuclei. There were variable numbers of mixed inflammatory cells in the stroma around the glands and in the stalk. No invasion of the submucosa layer of the large intestine was observed.

Skin: In male rats, there were exposure concentration-related increased incidences of keratoacanthoma; keratoacanthoma or squamous cell carcinoma (combined); and keratoacanthoma, basal cell adenoma, basal cell carcinoma, or squamous cell carcinoma (combined) (Tables 10, A1, and A2). The incidences of keratoacanthoma and of keratoacanthoma or squamous cell carcinoma (combined) in 250 and 500 ppm males were significantly increased and exceeded the historical control ranges for inhalation studies (Tables 10 and A3b). The incidences of keratoacanthoma, basal cell adenoma, basal cell carcinoma, or squamous cell carcinoma (combined) were significantly increased in all exposed groups of males and exceeded the historical control range for inhalation studies. The incidences of basal cell adenoma in 250 ppm males and of squamous cell carcinoma in 500 ppm males were not significantly increased, but they exceeded the respective historical control ranges for inhalation studies (Tables 10, A1, and A3b).

In female rats, there were increased incidences of squamous cell papilloma, keratoacanthoma, basal cell adenoma, or basal cell carcinoma (combined) in the 500 ppm group (Tables 10, B1, and B2). Although the increased incidences were not significant, they exceeded the respective historical control ranges for inhalation studies (Tables 10 and B3b).

Keratoacanthomas were well-demarcated, variably sized, crateriform masses in the dermis composed of squamous epithelium that formed thick folds. The center of the mass was filled with abundant keratin. Squamous cell carcinomas were characterized as masses composed of cords of pleomorphic squamous cells infiltrating the dermis and/or subcutis. There were varying amounts of keratin and fibrous connective tissue, which separated the cords of squamous cells within the neoplasms. Basal cell adenomas were well-demarcated masses composed of cords and lobules of basal cells with areas of sebaceous or squamous differentiation. Basal cell carcinomas had a similar appearance, but the cells in the carcinomas were more pleomorphic. The carcinomas were locally invasive and often contained areas of necrosis.

TABLE 9
Incidences of Adenoma of the Large Intestine in Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Male				
Colon[a]	50	50	50	50
Adenoma[b]	0	0	0	1
Rectum	50	50	50	50
Adenoma	0	0	2	0
Colon or Rectum: Adenoma[c]				
Overall rate[d]	0/50 (0%)	0/50 (0%)	2/50 (4%)	1/50 (2%)
Adjusted rate[e]	0.0%	0.0%	5.3%	2.8%
Terminal rate[f]	0/23 (0%)	0/26 (0%)	2/18 (11%)	0/13 (0%)
First incidence (days)	—[h]	—	729 (T)	682
Poly-3 test[g]	P=0.197	—[i]	P=0.216	P=0.472
Female				
Colon	50	50	50	50
Adenoma	0	1	1	1
Rectum	50	50	50	50
Adenoma	0	0	1	4
Colon or Rectum: Adenoma[j]				
Overall rate	0/50 (0%)	1/50 (2%)	2/50 (4%)	5/50 (10%)
Adjusted rate	0.0%	2.3%	4.7%	13.3%
Terminal rate	0/34 (0%)	1/33 (3%)	1/30 (3%)	4/24 (17%)
First incidence (days)	—	730 (T)	607	719
Poly-3 test	P=0.004	P=0.493	P=0.225	P=0.018

(T) Terminal sacrifice

[a] Number necropsied

[b] Number of animals with neoplasm

[c] Historical incidence for 2-year inhalation studies with chamber controls (mean ± standard deviation): 0/349; all routes: 2/1,398 (0.1% ± 0.5%), range 0%-2%

[d] Number of animals with neoplasm per number of animals necropsied

[e] Poly-3 estimated neoplasm incidence after adjustment for intercurrent mortality

[f] Observed incidence at terminal kill

[g] Beneath the chamber control incidence are the P values associated with the trend test. Beneath the exposed group incidence are the P values corresponding to pairwise comparisons between the chamber controls and that exposed group. The Poly-3 test accounts for differential mortality in animals that do not reach terminal sacrifice.

[h] Not applicable; no neoplasm in animal group

[i] Value of statistic cannot be computed.

[j] Historical incidence for inhalation studies: 0/350; all routes 3/1,350 (0.2% ± 0.6%), range 0%-2%

TABLE 10
Incidences of Neoplasms of the Skin in Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Male				
Number Necropsied	50	50	50	50
Basal Cell Adenoma[a,b]	0	1	2	1
Basal Cell Carcinoma[c]	0	2	1	2
Keratoacanthoma[d]				
Overall rate[e]	0/50 (0%)	3/50 (6%)	6/50 (12%)	6/50 (12%)
Adjusted rate[f]	0.0%	7.4%	15.4%	16.2%
Terminal rate[g]	0/23 (0%)	3/26 (12%)	2/18 (11%)	2/13 (15%)
First incidence (days)	—[i]	729 (T)	488	562
Poly-3 test[h]	P=0.008	P=0.115	P=0.012	P=0.010
Squamous Cell Carcinoma[j]	1	1	0	2
Keratoacanthoma or Squamous Cell Carcinoma[k]				
Overall rate	1/50 (2%)	4/50 (8%)	6/50 (12%)	8/50 (16%)
Adjusted rate	2.4%	9.8%	15.4%	21.4%
Terminal rate	0/23 (0%)	3/26 (12%)	2/18 (11%)	3/13 (23%)
First incidence (days)	669	711	488	562
Poly-3 test	P=0.006	P=0.171	P=0.044	P=0.009
Keratoacanthoma, Basal Cell Adenoma, Basal Cell Carcinoma, or Squamous Cell Carcinoma[l]				
Overall rate	1/50 (2%)	7/50 (14%)	9/50 (18%)	10/50 (20%)
Adjusted rate	2.4%	17.0%	22.6%	26.7%
Terminal rate	0/23 (0%)	5/26 (19%)	2/18 (11%)	4/13 (31%)
First incidence (days)	669	585	488	562
Poly-3 test	P=0.003	P=0.028	P=0.006	P=0.002
Female				
Squamous Cell Papilloma, Keratoacanthoma, Basal Cell Adenoma, or Basal Cell Carcinoma[m]				
Overall rate	1/50 (2%)	1/50 (2%)	1/50 (2%)	4/50 (8%)
Adjusted rate	2.2%	2.3%	2.4%	10.6%
Terminal rate	1/34 (3%)	1/33 (3%)	1/30 (3%)	4/24 (17%)
First incidence (days)	730 (T)	730 (T)	730 (T)	730 (T)
Poly-3 test	P=0.050	P=0.753	P=0.746	P=0.128

(T) Terminal sacrifice

[a] Number of animals with neoplasm

[b] Historical incidence for 2-year inhalation studies with chamber controls (mean ± standard deviation): 4/349 (1.2% ± 1.1%), range 0%-2%; all routes: 15/1,398 (1.1% ± 1.4%), range 0%-4%

[c] Historical incidence for inhalation studies: 4/349 (1.1% ± 2.3%), range 0%-6%; all routes: 11/1,398 (0.8% ± 1.5%), range 0%-6%

[d] Historical incidence for inhalation studies: 10/349 (2.9% ± 3.7%), range 0%-8%; all routes: 66/1,398 (4.7% ± 4.2%), range 0%-16%

[e] Number of animals with neoplasm per number of animals necropsied

[f] Poly-3 estimated neoplasm incidence after adjustment for intercurrent mortality

[g] Observed incidence at terminal kill

[h] Beneath the chamber control incidence are the P values associated with the trend test. Beneath the exposed group incidence are the P values corresponding to pairwise comparisons between the chamber controls and that exposed group. The Poly-3 test accounts for differential mortality in animals that do not reach terminal sacrifice.

[i] Not applicable; no neoplasm in animal group

[j] Historical incidence for inhalation studies: 1/349 (0.3% ± 0.8%), range 0%-2%; all routes: 8/1,398 (0.6% ± 0.9%), range 0%-2%

[k] Historical incidence for inhalation studies: 11/349 (3.2% ± 3.5%), range 0%-8%; all routes: 74/1,398 (5.3% ± 4.1%), range 0%-16%

[l] Historical incidence for inhalation studies: 19/349 (5.5% ± 4.5%), range 0%-10%; all routes: 97/1,398 (6.9% ± 4.9%), range 0%-20%

[m] Historical incidence for inhalation studies: 2/350 (0.6% ± 1.0%), range 0%-2%; all routes: 16/1,350 (1.2% ± 1.8%), range 0%-6%

Malignant Mesothelioma: The incidence of malignant mesothelioma in 500 ppm male rats was significantly greater than that in the chamber controls (Tables 11, A1, and A2). The incidence in the 500 ppm group exceeded the historical ranges for controls in inhalation studies and all routes (Tables 11 and A3c).

This neoplasm was found in the epididymis in all affected animals with other tissues variably affected, particularly the testis, which was affected in all but one animal. This suggests that the tunica vaginalis, the most common site of origin for mesotheliomas in F344/N rats (Hall, 1990), was the site of origin for all mesotheliomas in this study. These mesotheliomas were characterized by numerous, complex, papillary structures composed of pedunculated, fibrovascular stalks covered by one to several layers of cuboidal to flattened mesothelial cells (Plates 3 and 4). The stroma was prominent and often contained clusters of pleomorphic mesothelial cells that sometimes formed disorganized tubular structures. There was often extensive invasion of skeletal muscle and adipose tissue, but minimal invasion of underlying tissues at other sites.

TABLE 11
Incidences of Malignant Mesothelioma in Male Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Malignant Mesothelioma[a]				
Overall rate[b]	0/50 (0%)	2/50 (4%)	2/50 (4%)	4/50 (8%)
Adjusted rate[c]	0.0%	4.9%	5.2%	10.8%
Terminal rate[d]	0/23 (0%)	2/26 (8%)	0/18 (0%)	1/13 (8%)
First incidence (days)	—[f]	729 (T)	536	394
Poly-3 test[e]	P=0.031	P=0.233	P=0.220	P=0.046

(T) Terminal sacrifice

[a] Historical incidence for 2-year inhalation studies with chamber controls (mean ± standard deviation): 5/349 (1.4% ± 2.2%), range 0%-6%; all routes: 35/1,398 (2.5% ± 2.3%), range 0%-6%

[b] Number of animals with neoplasm per number of animals necropsied

[c] Poly-3 estimated neoplasm incidence after adjustment for intercurrent mortality

[d] Observed incidence at terminal kill

[e] Beneath the chamber control incidence are the P values associated with the trend test. Beneath the exposed group incidence are the P values corresponding to pairwise comparisons between the chamber controls and that exposed group. The Poly-3 test accounts for differential mortality in animals that do not reach terminal sacrifice.

[f] Not applicable; no neoplasm in animal group

Pancreatic Islets: In male rats, the incidences of pancreatic islet adenoma were significantly increased in all exposed groups compared to the chamber controls (Tables 12, A1, and A2). However, the incidences were within the historical control ranges for inhalation studies and for all routes (Tables 12 and A3d). The incidences of pancreatic islet carcinoma were increased in the 125 and 250 ppm groups, but these increases were not statistically significant. However, the incidence in the 125 ppm group exceeded the historical control ranges for inhalation studies and for all routes. The incidences of pancreatic islet adenoma or carcinoma (combined) were significantly increased in the 125 and 250 ppm groups, and the incidence in the 125 ppm group exceeded the historical control ranges for inhalation studies and for all routes of administration.

Adenomas were usually discrete, well-circumscribed masses of islet cells that were 1 mm in diameter or larger and compressed the surrounding acinar tissue. Occasionally, adenomas were encapsulated by a thin band of fibrous connective tissue. In some adenomas, groups of exocrine pancreatic acini were present within the adenomas at their periphery. Carcinomas, which tended to be larger than adenomas, were characterized by varying degrees of atypia and pleomorphism of the neoplastic cells, and they typically invaded the surrounding fibrous capsule or pancreatic tissue.

TABLE 12
Incidences of Pancreatic Islet Neoplasms in Male Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Adenoma[a]				
Overall rate[b]	0/50 (0%)	5/50 (10%)	4/50 (8%)	5/50 (10%)
Adjusted rate[c]	0.0%	12.2%	10.4%	13.9%
Terminal rate[d]	0/23 (0%)	4/26 (15%)	1/18 (6%)	4/13 (31%)
First incidence (days)	—[f]	666	608	697
Poly-3 test[e]	P=0.043	P=0.029	P=0.050	P=0.019
Carcinoma[g]				
Overall rate	3/50 (6%)	7/50 (14%)	5/50 (10%)	3/50 (6%)
Adjusted rate	7.2%	17.0%	13.0%	8.3%
Terminal rate	2/23 (9%)	3/26 (12%)	3/18 (17%)	2/13 (15%)
First incidence (days)	686	687	578	687
Poly-3 test	P=0.0516N	P=0.149	P=0.312	P=0.594
Adenoma or Carcinoma[h]				
Overall rate	3/50 (6%)	10/50 (20%)	9/50 (18%)	8/50 (16%)
Adjusted rate	7.2%	24.2%	23.1%	22.2%
Terminal rate	2/23 (9%)	5/26 (19%)	4/18 (22%)	6/13 (46%)
First incidence (days)	686	666	578	687
Poly-3 test	P=0.093	P=0.031	P=0.043	P=0.057

[a] Historical incidence for 2-year inhalation studies with chamber controls (mean ± standard deviation): 20/349 (5.7% ± 3.9%), range 0%-12%; all routes: 90/1,394 (6.5% ± 3.6%), range 0%-14%

[b] Number of animals with neoplasm per number of animals with pancreatic islets examined microscopically

[c] Poly-3 estimated neoplasm incidence after adjustment for intercurrent mortality

[d] Observed incidence at terminal kill

[e] Beneath the chamber control incidence are the P values associated with the trend test. Beneath the exposed group incidence are the P values corresponding to pairwise comparisons between the chamber controls and that exposed group. The Poly-3 test accounts for differential mortality in animals that do not reach terminal sacrifice. A negative trend is indicated by **N**.

[f] Not applicable; no neoplasm in animal group

[g] Historical incidence for inhalation studies: 17/349 (4.9% ± 3.3%), range 2%-10%; all routes: 29/1,394 (2.1% ± 2.6%), range 0%-10%

[h] Historical incidence for inhalation studies: 37/349 (10.6% ± 4.8%), range 6%-18%; all routes: 119/1,394 (8.6% ± 4.0%), range 0%-18%

Nose: Incidences of suppurative chronic inflammation in 500 ppm males and females were significantly greater than those in the chamber controls (Tables 13, A4, and B4). Suppurative chronic inflammation was characterized by marked numbers of neutrophils with Splendore-Hoeppli material within the nasal cavity lumen, located primarily in Levels II and III of the nasal sections only (Plates 5 and 6). The designation of chronic was used to differentiate suppurative inflammation with Splendore-Hoeppli material from suppurative inflammation without Splendore-Hoeppli material, which was observed in other organs. Occasionally, neutrophils in the affected area would invade the subjacent lamina propria. Application of special staining for bacteria indicated the presence of Giemsa-positive and Gram-negative bacteria associated with the Splendore-Hoeppli material.

Chronic active inflammation was diagnosed in many animals of both sexes, including the chamber controls (Tables 13, A4, and B4). The incidences in all exposed groups of females were significantly increased compared to that in the chamber controls. Chronic active inflammation was most commonly observed in Level II of nasal sections, and was characterized by the presence of lymphocytes and macrophages, with fewer neutrophils within or around the nasolacrimal duct, or within the lumina of submucosal glands or the lamina propria of the septum and turbinates.

The severity of epithelial hyaline droplet accumulation was increased in all exposed groups of females compared to that in the chamber controls (Table 13). Hyaline droplet accumulation was characterized by the presence of intracytoplasmic, eosinophilic globules primarily within the olfactory epithelium and to a lesser extent the respiratory epithelium. In general, this lesion was seen in many animals in the chamber control and exposed groups and it occurred most often in Levels II and III.

The incidences of glandular hyperplasia were significantly increased in all exposed groups of males and females (Tables 13, A4, and B4). Glandular hyperplasia occurred most often in Level II of the nasal section, and was at times associated with inflammation. This lesion consisted of mild to moderate proliferation of submucosal glands.

The incidences of respiratory epithelial hyperplasia were significantly increased in 125 and 500 ppm females (Tables 13 and B4). Respiratory epithelial hyperplasia was located most often along the septum in Level II and occasionally the ethmoid turbinates in Level III of the nasal sections. This lesion consisted of thickening of the respiratory epithelium by increased numbers of tall, columnar, respiratory epithelial cells.

The incidence of respiratory metaplasia of the olfactory epithelium was significantly increased in 500 ppm females (Tables 13 and B4). This lesion was primarily located in Level II at the dorsal meatus and septum but occasionally in Level III of the nasal sections. In the affected areas, the olfactory epithelium had been replaced by ciliated respiratory epithelium.

Larynx: Suppurative chronic inflammation was present in only one male and three females exposed to 500 ppm (Tables 13 and B4). This lesion was not seen in the chamber controls of either sex. Suppurative, chronic inflammation of the larynx was characterized by marked numbers of neutrophils, often degenerate, within the glands and lamina propria with intraglandular Splendore-Hoeppli material (Plate 7).

The incidences of chronic active inflammation in 250 ppm males and females and 500 ppm females were significantly increased (Tables 13, A4, and B4). This lesion consisted of minimal to moderate numbers of lymphocytes, macrophages, and neutrophils within the lamina propria and glandular lumens.

In females, the incidence of squamous metaplasia in the 500 ppm group was significantly greater than that in the chamber controls (Tables 13 and B4). Squamous metaplasia was characterized by the replacement of normal respiratory epithelium by squamous epithelial cells.

Trachea: In females, the incidences of chronic active inflammation and epithelial hyperplasia were significantly increased in the 500 ppm group (Tables 13 and B4). Both lesions also occurred in males, but the incidences were not significantly increased. Chronic active inflammation was characterized by the presence of minimal to moderate infiltrates of lymphocytes and macrophages, with fewer neutrophils in the lamina propria. Hyperplasia of the epithelium consisted of increased numbers of respiratory epithelial cells with densely packed nuclei.

Lung: Suppurative chronic inflammation occurred in a few 500 ppm males and females and in one 250 ppm male (Tables 13, A4, and B4). The lesion was morphologically similar to that seen in other organs.

TABLE 13
Incidences of Selected Nonneoplastic Lesions of the Respiratory System in Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control		125 ppm		250 ppm		500 ppm	
Male								
Nose[a]	50		48		48		50	
Inflammation, Suppurative, Chronic[b]	0		1	(4.0)[c]	2	(4.0)	7**	(4.0)
Inflammation, Chronic Active	29	(1.6)	33	(1.4)	34	(1.5)	35	(1.5)
Epithelium, Accumulation, Hyaline Droplet	44	(1.0)	39	(1.5)	36	(1.3)	44	(1.3)
Glands, Hyperplasia	5	(2.0)	14*	(2.0)	14**	(2.0)	15**	(2.0)
Larynx	50		50		50		50	
Inflammation, Suppurative, Chronic	0		0		0		1	(4.0)
Inflammation, Chronic Active	21	(1.4)	28	(1.3)	31*	(1.4)	26	(1.3)
Metaplasia, Squamous	4	(1.0)	6	(1.0)	8	(1.1)	5	(1.2)
Trachea	50		50		50		50	
Inflammation, Chronic Active	1	(2.0)	1	(1.0)	1	(1.0)	4	(1.5)
Epithelium, Hyperplasia	1	(2.0)	0		0		1	(2.0)
Lung	50		50		50		50	
Inflammation, Suppurative, Chronic	0		0		1	(4.0)	3	(4.0)
Female								
Nose	50		50		49		50	
Inflammation, Suppurative, Chronic	0		1	(4.0)	3	(4.0)	7**	(4.0)
Inflammation, Chronic Active	24	(1.3)	37**	(1.5)	37**	(1.5)	36**	(1.3)
Epithelium, Accumulation, Hyaline Droplet	48	(1.1)	48	(1.8)	48	(1.7)	47	(1.9)
Glands, Hyperplasia	6	(2.0)	23**	(2.0)	28**	(2.0)	30**	(2.0)
Respiratory Epithelium, Hyperplasia	5	(1.2)	13*	(1.3)	9	(1.7)	18**	(1.5)
Olfactory Epithelium, Metaplasia, Respiratory	3	(1.7)	4	(1.8)	6	(1.8)	9*	(2.2)
Larynx	50		50		50		50	
Inflammation, Suppurative, Chronic	0		0		0		3	(4.0)
Inflammation, Chronic Active	18	(1.1)	25	(1.5)	30**	(1.4)	32**	(1.5)
Metaplasia, Squamous	3	(1.3)	2	(1.5)	6	(1.3)	21**	(1.7)
Trachea	50		50		50		50	
Inflammation, Chronic Active	0		4	(1.0)	1	(1.0)	6**	(1.7)
Epithelium, Hyperplasia	0		0		0		4*	(1.8)
Lung	50		50		50		50	
Inflammation, Suppurative, Chronic	0		0		0		4*	(4.0)

* Significantly different ($P \leq 0.05$) from the chamber control group by the Poly-3 test

** $P \leq 0.01$

[a] Number of animals with tissue examined microscopically

[b] Number of animals with lesion

[c] Average severity grade of lesions in affected animals: 1=minimal, 2=mild, 3=moderate, 4=marked

Other Findings: Suppurative chronic inflammation with Splendore-Hoeppli material was sporadically observed in several other organs of exposed animals, including skin [all exposed groups of males (with an exposure concentration-related increase in incidences), and 125 and 500 ppm females], prostate gland (500 ppm males), preputial gland (125 ppm males), Harderian gland (500 ppm males and females), skeletal muscle (500 ppm males), bone (500 ppm males and females), ear (500 ppm females), and peritoneum (500 ppm males) (Tables A4 and B4). The designation of chronic was given to all diagnoses in which Splendore-Hoeppli material was present to distinguish it from suppurative inflammation without Splendore-Hoeppli material.

Swabs were collected from abscesses on the tail, Harderian gland, head, and salivary gland of five exposed rats. The swabs were cultured under aerobic and anaerobic conditions and bacterial isolates were identified. No bacterial growth was observed under anaerobic conditions. *Pseudomonas aeruginosa* was the primary isolate in all aerobic cultures. Splendore-Hoeppli bodies were later observed microscopically in these lesions.

MICE
2-WEEK STUDY

All 2,000 ppm males and two 2,000 ppm females, four 500 ppm males, one 1,000 ppm male, and one 1,000 ppm female died early (Table 14); all these early deaths occurred during the first week of exposure. Final mean body weights of exposed groups were similar to those of the chamber controls; the mean body weight gain of 1,000 ppm males was significantly decreased. Abnormal breathing, lethargy, and eye discharge were observed primarily during week 1 and occurred only in groups exposed to 500 ppm or greater.

In the 1,000 ppm males, the absolute and relative heart weights were significantly less than those of the chamber controls while the absolute and relative liver weights were significantly greater than those of the chamber controls (Table G3). Absolute right kidney weights were significantly increased in 250 ppm or greater females, and the relative right kidney weights were significantly increased in 1,000 and 2,000 ppm females. Absolute and relative liver weights of 500 ppm or greater females were significantly greater than those of the chamber controls. The absolute and relative thymus weights of 1,000 and 2,000 ppm females were significantly decreased.

TABLE 14
Survival and Body Weights of Mice in the 2-Week Inhalation Study of 1-Bromopropane

Concentration (ppm)	Survival[a]	Mean Body Weight[b] (g)			Final Weight Relative to Controls (%)
		Initial	Final	Change	
Male					
0	5/5	23.9 ± 0.2	28.0 ± 0.4	4.1 ± 0.3	
125	5/5	24.2 ± 0.1	28.9 ± 0.5	4.7 ± 0.5	103
250	5/5	23.7 ± 0.3	27.0 ± 0.4	3.3 ± 0.2	96
500	1/5[c]	24.0 ± 0.4	25.8	2.1	92
1,000	4/5[d]	23.9 ± 0.2	26.6 ± 0.5	2.6 ± 0.8*	95
2,000	0/5[e]	24.1 ± 0.2	—	—	
Female					
0	5/5	20.0 ± 0.5	22.9 ± 0.2	3.0 ± 0.3	
125	5/5	19.4 ± 0.4	22.2 ± 0.5	2.8 ± 0.5	97
250	5/5	20.4 ± 0.2	22.8 ± 0.2	2.5 ± 0.2	100
500	5/5	19.8 ± 0.2	23.4 ± 0.6	3.6 ± 0.5	102
1,000	4/5[f]	19.8 ± 0.3	22.8 ± 0.5	3.3 ± 0.6	99
2,000	3/5[d]	19.8 ± 0.2	22.7 ± 0.3	3.2 ± 0.2	99

* Significantly different ($P \leq 0.05$) from the chamber control group by Williams' test

[a] Number of animals surviving at 2 weeks/number initially in group

[b] Weights and weight changes are given as mean ± standard error. Subsequent calculations are based on animals surviving to the end of the study.

[c] Days of death: 4, 4, 4, 5

[d] Day of deaths: 3

[e] Days of death: 2, 2, 2, 2, 3

[f] Day of death: 5

Microscopic lesions related to 1-bromopropane exposure occurred in the lung, liver, and nose of males and females and were primarily seen in mice exposed to 500 ppm or greater (Table 15). Bronchiole necrosis occurred in the lungs of all exposed male and female mice and the severity of this lesion was increased in the 2,000 ppm groups. The lungs of exposed mice also had sporadic incidences of regeneration, cytoplasmic vacuolization, and acute inflammation involving bronchiolar epithelium. Incidences of centrilobular necrosis occurred in the liver of most males and females exposed to 500 ppm or greater. In addition, the incidences of centrilobular chronic inflammation and cytoplasmic vacuolization were significantly increased in 1,000 ppm males and females. Sporadic incidences of nasal lesions occurred in exposed groups of mice; the no-effect level for nose lesions was 250 ppm in males and 500 ppm in females.

Exposure Concentration Selection Rationale: Based on decreased survival and increased liver and kidney weights in the 2-week study, 1-bromopropane exposure concentrations selected for the 3-month inhalation study in mice were 62.5, 125, 250, and 500 ppm.

TABLE 15
Incidences of Selected Nonneoplastic Lesions in Mice in the 2-Week Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm	1,000 ppm	2,000 ppm
Male						
Lung[a]	5	5	5	5	5	5
Bronchiole, Necrosis[b]	0	5** (1.0) [c]	5** (1.0)	5** (2.0)	5** (1.4)	5** (3.4)
Bronchiole, Regeneration	0	5** (1.0)	5** (1.0)	2 (2.0)	4* (1.5)	0
Bronchiole, Vacuolization, Cytoplasmic	0	0	0	4* (3.0)	1 (3.0)	0
Perivascular, Inflammation, Acute	0	0	0	3 (2.0)	0	5** (1.4)
Liver	5	0	5	5	5	5
Centrilobular, Necrosis	0		0	5** (3.8)	5** (3.2)	5** (2.4)
Centrilobular, Inflammation, Chronic	0		0	1 (2.0)	4* (3.3)	0
Centrilobular, Vacuolization, Cytoplasmic	0		0	3 (1.7)	4* (2.0)	0
Nose	5	0	5	5	5	5
Glands, Necrosis	0		0	4* (2.8)	3 (1.7)	5** (2.6)
Inflammation, Suppurative	0		0	0	3 (1.7)	0
Olfactory Epithelium, Atrophy	0		0	0	4* (1.8)	0
Olfactory Epithelium, Necrosis	0		0	4* (2.0)	2 (2.0)	5** (3.8)
Olfactory Epithelium, Regeneration	0		0	1 (1.0)	4* (2.3)	0
Respiratory Epithelium, Necrosis	0		0	0	1 (3.0)	5** (2.8)
Respiratory Epithelium, Vacuolization, Cytoplasmic	0		0	4* (3.0)	1 (3.0)	0

TABLE 15
Incidences of Selected Nonneoplastic Lesions in Mice in the 2-Week Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm	1,000 ppm	2,000 ppm
Female						
Lung	5	5	5	5	5	5
Bronchiole, Necrosis	0	5** (1.2)	5** (1.0)	5** (1.0)	5** (1.4)	5** (2.2)
Bronchiole, Regeneration	0	5** (1.2)	5** (1.0)	5** (2.0)	4* (1.8)	3 (2.3)
Bronchiole, Vacuolization, Cytoplasmic	0	0	0	0	1 (3.0)	0
Perivascular, Inflammation, Acute	0	0	0	0	1 (1.0)	2 (4.0)
Liver	5	0	5	5	5	5
Centrilobular, Necrosis	0		0	3 (1.3)	5** (2.8)	5** (1.8)
Centrilobular, Inflammation, Chronic	0		0	3 (1.3)	4* (3.0)	3 (1.3)
Centrilobular, Vacuolization, Cytoplasmic	0		0	3 (1.7)	5** (1.4)	3 (2.0)
Nose	5	0	0	5	5	5
Glands, Necrosis	0			0	1 (3.0)	2 (3.0)
Inflammation, Suppurative	0			0	0	1 (1.0)
Olfactory Epithelium, Atrophy	0			0	4* (1.0)	3 (1.7)
Olfactory Epithelium, Necrosis	0			0	1 (3.0)	2 (4.0)
Olfactory Epithelium, Regeneration	0			0	4* (1.5)	3 (2.3)
Respiratory Epithelium, Necrosis	0			0	1 (2.0)	2 (2.5)
Respiratory Epithelium, Vacuolization, Cytoplasmic	0			0	1 (4.0)	0

* Significantly different ($P \leq 0.05$) from the chamber control group by the Fisher exact test
** $P \leq 0.01$
[a] Number of animals with tissue examined microscopically
[b] Number of animals with lesion
[c] Average severity grade of lesions in affected animals: 1=minimal, 2=mild, 3=moderate, 4=marked

3-MONTH STUDY

One 250 ppm male and four males and five females in the 500 ppm groups died early (Table 16). Final mean body weights and body weight gains of all exposed groups were similar to those of the chamber control groups (Table 16 and Figure 6). By day 3, lethargy was observed in males and females exposed to 500 ppm, and abnormal breathing was observed in moribund mice during week 1.

There were no changes in hematology endpoints that were considered to be related to 1-bromopropane exposure (Table F2).

Absolute right kidney weights of 250 and 500 ppm males, and the relative right kidney weight of 500 ppm males were significantly less than those of the chamber controls (Table G4). Relative liver weights of 250 and 500 ppm males were significantly increased. The absolute and relative right kidney, liver, and lung weights of 500 ppm females and the relative liver weight of 250 ppm females were significantly greater than those of the chamber controls.

TABLE 16
Survival and Body Weights of Mice in the 3-Month Inhalation Study of 1-Bromopropane

Concentration (ppm)	Survival[a]	Mean Body Weight[b] (g)			Final Weight Relative to Controls (%)
		Initial	Final	Change	
Male					
0	10/10	23.6 ± 0.3	39.6 ± 0.8	16.0 ± 0.7	
62.5	10/10	23.7 ± 0.2	39.0 ± 0.9	15.3 ± 0.8	98
125	10/10	23.6 ± 0.1	39.6 ± 0.6	15.9 ± 0.6	100
250	9/10[c]	23.6 ± 0.3	37.9 ± 0.7	14.4 ± 0.6	96
500	6/10[d]	23.4 ± 0.3	37.4 ± 1.4	14.2 ± 1.2	95
Female					
0	10/10	19.9 ± 0.2	30.9 ± 0.7	11.0 ± 0.5	
62.5	10/10	20.3 ± 0.3	33.0 ± 1.3	12.7 ± 1.1	107
125	10/10	20.4 ± 0.3	31.9 ± 0.8	11.5 ± 0.6	103
250	10/10	19.7 ± 0.3	30.2 ± 0.9	10.4 ± 0.9	98
500	5/10[e]	20.2 ± 0.1	31.3 ± 1.4	11.0 ± 1.5	101

[a] Number of animals surviving at 3 months/number initially in group

[b] Weights and weight changes are given as mean ± standard error. Differences from the chamber control group are not significant by Dunnett's test. Subsequent calculations are based on animals surviving to the end of the study.

[c] Week of death: 4

[d] Weeks of death: 1, 1, 1, 2

[e] Week of deaths: 1

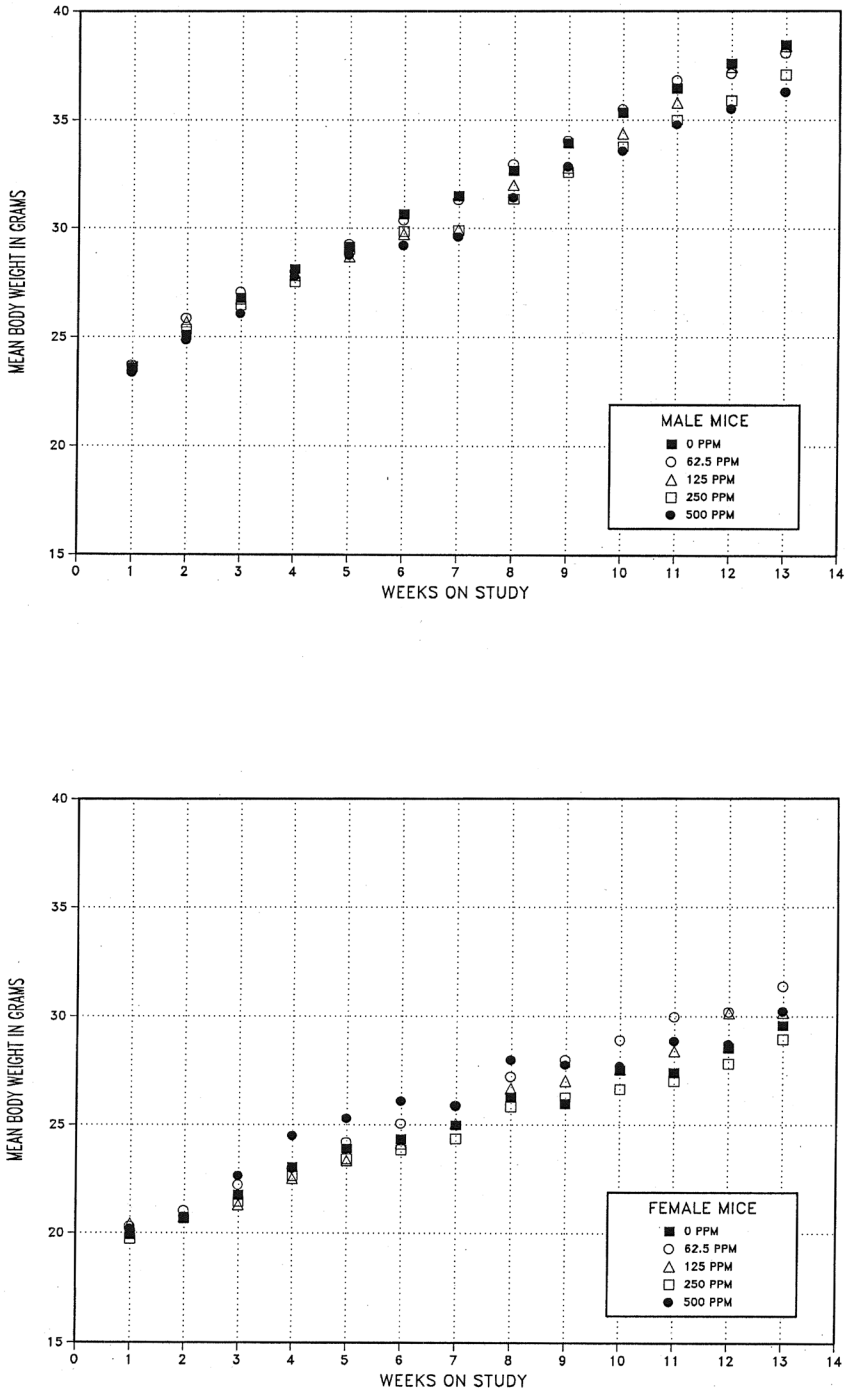


FIGURE 6
Growth Curves for Mice Exposed to 1-Bromopropane by Inhalation for 3 Months

There were slight increases in the cauda epididymis weights (9% and 17%) and significant decreases in sperm motility (4%) in 250 and 500 ppm male mice, respectively (Table H3). There was also a significant decrease in the sperm per gram cauda (28%) in 500 ppm males. Exposed groups of female mice differed significantly from the chamber controls in the relative amount of time spent in the various stages of estrous (Table H4). Female mice in the 500 ppm group spent significantly more time in extended diestrus than the chamber controls; 250 ppm females spent significantly more time in extended estrus than the chamber controls. The length of the estrous cycle of 500 ppm females was increased slightly (one half day).

Treatment-related nasal lesions were seen only in mice that died early. The incidences of cytoplasmic vacuolization of the respiratory epithelium were significantly increased in 500 ppm males and females, and the incidence of necrosis of the respiratory epithelium was significantly increased in 500 ppm females (Table 17). Olfactory epithelium necrosis and necrosis of the lateral nasal glands occurred in a few 500 ppm males. A single incidence of cytoplasmic vacuolization of the respiratory epithelium occurred in a male exposed to 250 ppm. Respiratory epithelial lesions were located in the lateral walls and turbinates of Level I. The olfactory necrosis was located in the dorsal meatus of Level II and in the dorsal meatus, septum, and turbinates of Level III. Vacuolization consisted of a single, large, clear vacuole displacing the cytoplasm. Necrosis was characterized by cells with increased cytoplasmic eosinophilia with loss of cellular detail and/or pyknotic or fragmented nuclei. Necrotic cells were frequently sloughed into the airways or missing.

Incidences of cytoplasmic vacuolization and necrosis of the respiratory epithelium lining the larynx and trachea occurred in male and female mice exposed to 500 ppm that died early and in the one early death male exposed to 250 ppm; the incidences of tracheal cytoplasmic vacuolization were significantly increased in 500 ppm males and females (Table 17). Vacuolization of the laryngeal lining epithelium also occurred in one 62.5 ppm female that survived to the end of the study. Vacuolization was characterized by cells with a large expansile clear space occupying the majority of the cytoplasm. These cells often had a pyknotic nucleus at the center, suggesting that vacuolization progressed to necrosis. These cells had often lost their cilia. Hyperplasia of the tracheal epithelium occurred in two 500 ppm males (one an early death and the other a terminal animal). Hyperplasia was characterized by increased cell numbers with loss of the normal organization of the epithelium.

In the lung, incidences of cytoplasmic vacuolization of the bronchiolar epithelium were significantly increased in 500 ppm males and females (Table 17). This lesion also occurred in the 250 ppm male that died early. The incidence of necrosis of the bronchiolar epithelium was significantly increased in 500 ppm females; this lesion also occurred in 500 ppm males and the 250 ppm male that died early. Necrosis consisted of epithelial cells with pyknotic nuclei, sloughed epithelial cells, and areas of bronchial mucosa devoid of epithelial cells or lined by attenuated epithelial cells. Cells remaining often contained colorless vacuoles displacing the cytoplasm. Regeneration of bronchiolar epithelium occurred in all groups of mice and was not treatment related. Regeneration was described as flattened and disorganized epithelium with variation in nuclear size and location. Nuclei tended to be apical rather than basilar. This lesion was graded as minimal in all cases.

In the liver, incidences of hepatocellular necrosis and hepatocellular degeneration were significantly increased in 500 ppm males and females; a single incidence of cytoplasmic vacuolization occurred in a 500 ppm male (Table 17). These lesions were also seen in the early death animals in the 500 ppm groups. Necrosis often extended beyond the centrilobular zone and was massive in some animals; it was characterized by uniform eosinophilic staining of hepatocytes and loss of cellular detail. Cytoplasmic degeneration and/or vacuolization, which were often seen peripheral to the necrosis, consisted of swollen and foamy hepatocytes with one large or many tiny clear vacuoles displacing the cytoplasm and with centrally located nuclei with condensed chromatin. Chronic inflammation occurred in 500 ppm terminal sacrifice animals of both sexes and in one male exposed to 250 ppm, and the incidences of this lesion were significantly increased in the 500 ppm groups. The inflammation consisted of lymphocytes, macrophages, large pigment-laden macrophages, and occasional multinucleated giant cells. Mineralization occurred in 500 ppm mice of both sexes that survived to the end of the study, and the incidences of this lesion were significantly increased in these groups.

Necrosis of the adrenal cortex (the zona fasciculata being most affected with extension into the zona

TABLE 17
Incidences of Selected Nonneoplastic Lesions in Mice in the 3-Month Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm	500 ppm
Male					
Nose[a]	10	10	10	10	10
Respiratory Epithelium, Vacuolization Cytoplasmic[b]	0	0	0	1 (3.0)[c]	4* (3.0)
Respiratory Epithelium, Necrosis	0	0	0	0	2 (1.5)
Olfactory Epithelium, Necrosis	0	0	0	0	2 (2.5)
Glands, Necrosis	0	0	0	0	1 (2.0)
Larynx	10	10	10	10	9
Vacuolization Cytoplasmic	0	0	0	1 (1.0)	3 (1.7)
Necrosis	0	0	0	0	2 (1.5)
Trachea	10	10	10	10	10
Vacuolization, Cytoplasmic	0	0	0	1 (3.0)	4* (1.0)
Necrosis	0	0	0	1 (2.0)	0
Hyperplasia	0	0	0	0	2 (1.0)
Lung	10	10	10	10	10
Bronchiole, Vacuolization Cytoplasmic	0	0	0	1 (3.0)	4* (2.5)
Bronchiole, Necrosis	0	0	0	1 (2.0)	3 (2.0)
Bronchiole, Regeneration	3 (1.0)	3 (1.0)	4 (1.0)	7 (1.0)	5 (1.0)
Liver	10	10	10	10	10
Necrosis	0	0	0	0	4* (4.0)
Hepatocyte, Degeneration	0	0	0	0	4* (2.5)
Vacuolization Cytoplasmic	0	0	0	0	1 (1.0)
Inflammation, Chronic	0	0	0	1 (1.0)	6** (1.3)
Mineralization	0	0	0	0	5* (1.8)

TABLE 17
Incidences of Selected Nonneoplastic Lesions in Mice in the 3-Month Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm	500 ppm
Female					
Nose	10	10	10	10	10
Respiratory Epithelium, Vacuolization Cytoplasmic	0	0	0	0	5* (3.0)
Respiratory Epithelium, Necrosis	0	0	0	0	5* (1.4)
Larynx	10	10	10	10	10
Vacuolization Cytoplasmic	0	1 (1.0)	0	0	1 (1.0)
Necrosis	0	0	0	0	1 (1.0)
Trachea	10	10	10	9	10
Vacuolization Cytoplasmic	0	0	0	0	4* (2.0)
Necrosis	0	0	0	0	2 (2.5)
Lung	10	10	10	10	10
Bronchiole, Vacuolization Cytoplasmic	0	0	0	0	5* (3.2)
Bronchiole, Necrosis	0	0	0	0	5* (2.0)
Bronchiole, Regeneration	1 (1.0)	4 (1.0)	5 (1.0)	5 (1.0)	3 (1.0)
Liver	10	10	10	10	10
Necrosis	0	0	0	0	5* (3.6)
Hepatocyte, Degeneration	0	0	0	0	5* (2.2)
Inflammation, Chronic	0	0	0	0	5* (1.8)
Mineralization	0	0	0	0	4* (1.5)
Adrenal Cortex	10	10	10	10	10
Necrosis	0	0	0	0	5* (3.4)
Inflammation, Chronic	0	0	0	0	2 (2.0)

* Significantly different ($P \leq 0.05$) from the chamber control group by the Fisher exact test
** $P \leq 0.01$
[a] Number of animals with tissue examined microscopically
[b] Number of animals with lesion
[c] Average severity grade of lesions in affected animals: 1=minimal, 2=mild, 3=moderate, 4=marked

glomerulosa to a lesser degree) occurred in 500 ppm female mice that died early, and the incidence of this lesion was significantly increased in this group (Table 17). Chronic inflammation was noted at the corticomedullary junction in the adrenal cortex of two 500 ppm females that survived to the end of the study.

Exposure Concentration Selection Rationale: Based on mortality at the highest exposure concentration, changes in organ weights, and the incidences of various nonneoplastic lesions in the 3-month study, 1-bromopropane exposure concentrations selected for the 2-year inhalation study in mice were 62.5, 125, and 250 ppm.

2-YEAR STUDY

Survival

Estimates of 2-year survival probabilities for male and female mice are shown in Table 18 and in the Kaplan-Meier survival curves (Figure 7). Survival of exposed groups of mice was similar to that of the chamber control groups.

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Body Weights and Clinical Findings

Mean body weights of exposed groups of male and female mice were similar to those of the chamber controls throughout the study (Tables 19 and 20; Figure 8). No clinical findings related to exposure to 1-bromopropane were observed.

TABLE 18
Survival of Mice in the 2-Year Inhalation Study of 1-Bromopropane

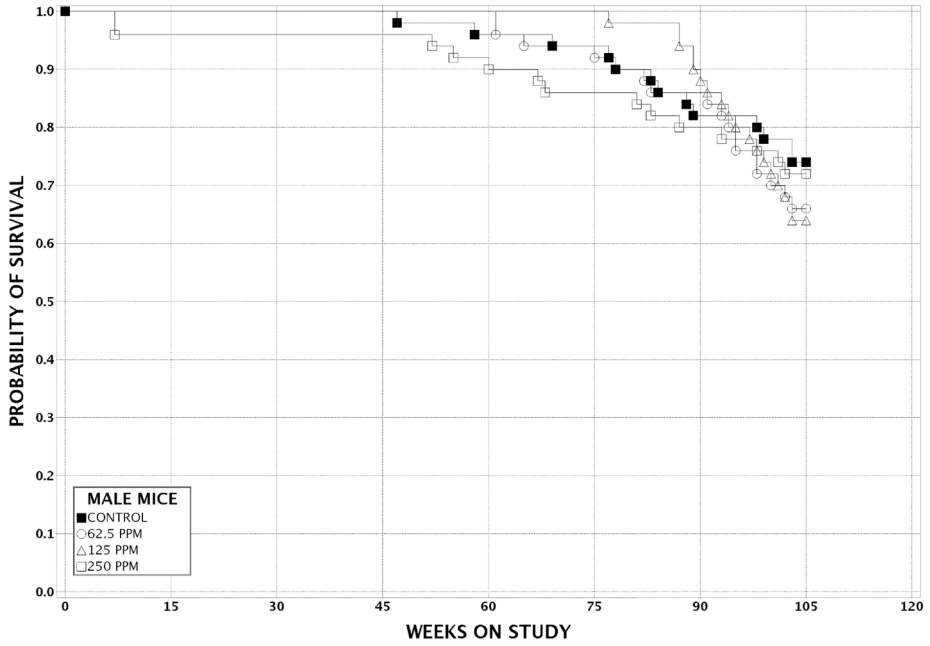
	Chamber Control	62.5 ppm	125 ppm	250 ppm
Male				
Animals initially in study	50	50	50	50
Moribund	6	13	9	9
Natural deaths	7	4	9	5
Animals surviving to study termination	37	33	32	36
Percent probability of survival at end of study[a]	74	66	64	72
Mean survival (days)[b]	689	686	703	658
Survival analysis[c]	P=0.934	P=0.526	P=0.486	P=0.888
Female				
Animals initially in study	50	50	50	50
Moribund	10	7	8	8
Natural deaths	4	3	5	0
Animals surviving to study termination	36[d]	40	37	42[d]
Percent probability of survival at end of study	72	80	74	82
Mean survival (days)	698	713	699	714
Survival analysis	P=0.363N	P=0.413N	P=0.967N	P=0.298N

[a] Kaplan-Meier determinations

[b] Mean of all deaths (uncensored, censored, and terminal sacrifice)

[c] The result of the life table trend test (Tarone, 1975) is in the chamber control column, and the results of the life table pairwise comparisons (Cox, 1972) with the chamber controls are in the exposed group columns. A negative trend or lower mortality in an exposure group is indicated by **N**.

[d] Includes one animal that died during the last week of the study



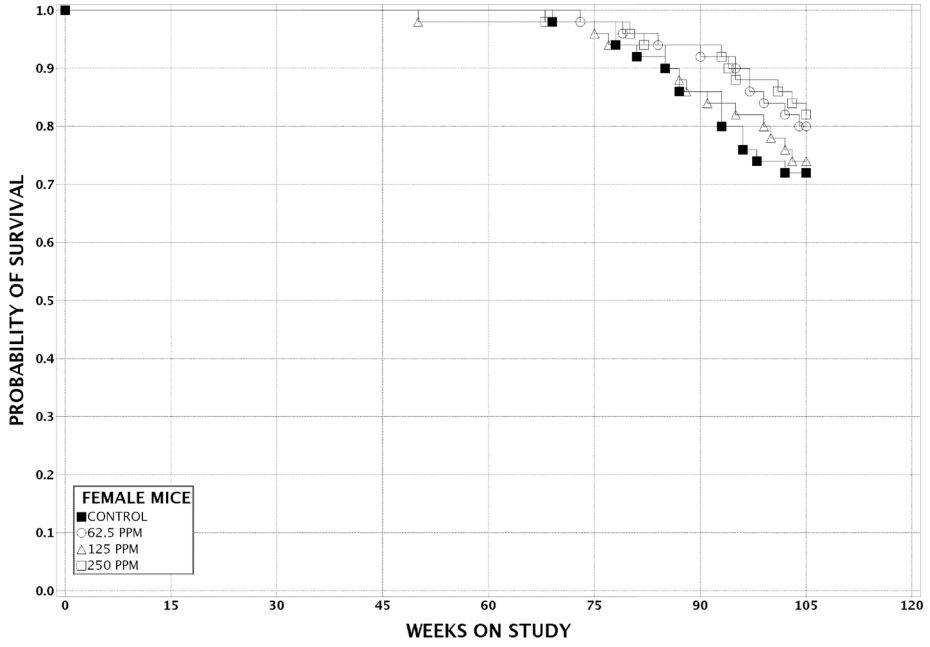


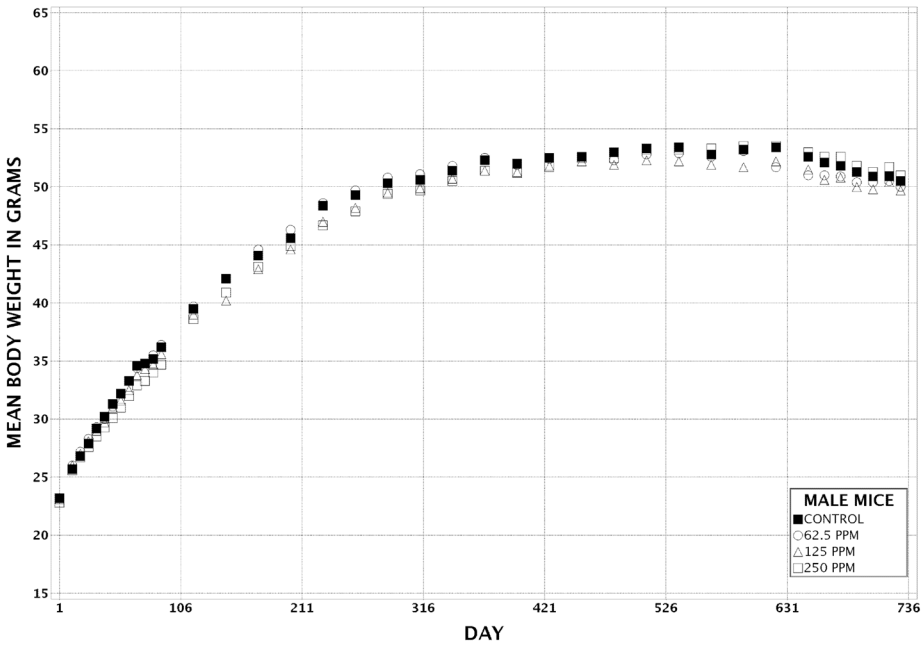
FIGURE 7
Kaplan-Meier Survival Curves for Mice
Exposed to 1-Bromopropane by Inhalation for 2 Years

TABLE 19
Mean Body Weights and Survival of Male Mice in the 2-Year Inhalation Study of 1-Bromopropane

Days on Study	Chamber Control		62.5 ppm			125 ppm			250 ppm		
	Av. Wt. (g)	No. of Survivors	Av. Wt. (g)	Wt. (% of controls)	No. of Survivors	Av. Wt. (g)	Wt. (% of controls)	No. of Survivors	Av. Wt. (g)	Wt. (% of controls)	No. of Survivors
1	23.2	50	23.2	100	50	23.1	100	50	22.8	98	50
12	25.7	50	26.0	101	50	26.0	101	50	25.6	99	50
19	26.8	50	27.2	101	50	27.0	101	50	26.7	100	50
26	27.9	50	28.3	101	50	28.1	101	50	27.6	99	50
33	29.2	50	29.3	100	50	29.0	100	50	28.5	98	50
40	30.2	50	30.0	99	50	29.7	99	50	29.3	97	50
47	31.3	50	31.1	99	50	31.0	99	50	30.1	96	48
54	32.2	50	32.2	100	50	31.6	98	50	31.0	96	48
61	33.3	50	33.3	100	50	32.5	98	50	32.0	96	48
68	34.6	50	34.4	99	50	33.8	98	50	32.9	95	48
75	34.8	50	34.8	100	50	34.3	99	50	33.3	96	48
82	35.2	50	35.5	101	50	34.7	98	50	34.0	96	48
89	36.2	50	36.4	100	50	35.6	98	50	34.7	96	48
117	39.5	50	39.7	100	50	39.0	99	50	38.6	98	48
145	42.1	50	42.1	100	50	40.2	96	50	40.9	97	48
173	44.1	50	44.6	101	50	42.9	97	50	43.1	98	48
201	45.6	50	46.3	102	50	44.6	98	50	44.9	98	48
229	48.4	50	48.6	101	50	47.0	97	50	46.7	97	48
257	49.3	50	49.7	101	50	48.2	98	50	47.9	97	48
285	50.3	50	50.8	101	50	49.5	98	50	49.4	98	48
313	50.6	50	51.1	101	50	49.9	99	50	49.7	98	48
341	51.4	49	51.8	101	50	50.7	99	50	50.5	98	48
369	52.3	49	52.5	101	50	51.4	98	50	51.4	98	47
397	52.0	49	51.9	100	50	51.3	99	50	51.2	99	46
425	52.5	48	52.4	100	48	51.8	99	50	51.7	99	45
453	52.6	48	52.6	100	47	52.2	99	50	52.5	100	45
481	53.0	47	52.3	99	47	51.9	98	50	52.5	99	43
509	53.3	47	52.8	99	47	52.3	98	50	53.2	100	43
537	53.4	47	52.9	99	46	52.2	98	49	53.2	100	43
565	52.8	45	52.6	100	45	51.9	98	49	53.3	101	42
593	53.2	43	53.1	100	43	51.7	97	49	53.5	101	41
621	53.4	41	51.7	97	43	52.2	98	45	53.5	100	40
649	52.6	41	51.0	97	41	51.5	98	42	53.0	101	39
663	52.1	41	51.0	98	38	50.6	97	41	52.6	101	39
677	51.8	41	50.9	98	38	50.8	98	39	52.6	102	39
691	51.3	39	50.4	98	36	50.0	98	37	51.8	101	38
705	50.9	39	50.4	99	35	49.8	98	35	51.3	101	37
719	50.9	37	50.5	99	33	50.4	99	32	51.7	102	36
Mean for weeks											
1-13	30.8		30.9	100		30.5	99		29.9	97	
14-52	46.8		47.2	101		45.8	98		45.7	98	
53-103	52.4		51.8	99		51.4	98		52.4	100	

TABLE 20
Mean Body Weights and Survival of Female Mice in the 2-Year Inhalation Study of 1-Bromopropane

Days on Study	Chamber Control		62.5 ppm			125 ppm			250 ppm		
	Av. Wt. (g)	No. of Survivors	Av. Wt. (g)	Wt. (% of controls)	No. of Survivors	Av. Wt. (g)	Wt. (% of controls)	No. of Survivors	Av. Wt. (g)	Wt. (% of controls)	No. of Survivors
1	19.4	50	19.4	100	50	19.2	99	50	19.1	98	50
12	21.2	50	21.1	100	50	21.3	101	50	21.1	100	50
19	22.3	50	22.3	100	50	22.1	99	50	22.1	99	50
26	23.4	50	23.2	99	50	23.2	99	50	23.3	100	50
33	24.3	50	24.2	99	50	24.2	100	50	24.3	100	50
40	24.8	50	24.7	99	50	25.0	101	50	25.0	101	50
47	26.2	50	25.7	98	50	26.2	100	50	25.8	99	50
54	26.9	50	26.5	99	50	26.4	98	50	26.5	99	50
61	27.3	50	27.0	99	50	27.4	100	50	26.8	98	50
68	27.5	50	27.0	98	50	27.6	100	50	27.2	99	50
75	28.0	50	27.7	99	50	28.2	100	50	27.6	98	50
82	28.7	50	28.4	99	50	28.8	100	50	28.4	99	50
89	29.5	50	28.4	96	50	29.3	100	50	28.5	97	50
117	32.1	50	31.6	99	50	32.6	102	50	31.0	97	50
145	34.6	50	34.3	99	50	34.8	101	50	33.0	95	50
173	37.4	50	37.0	99	50	37.9	101	50	34.8	93	50
201	38.8	50	39.0	100	50	39.9	103	50	36.4	94	50
229	42.1	50	42.1	100	50	43.2	103	50	39.1	93	50
257	44.4	50	44.5	100	50	45.2	102	50	41.0	92	50
285	46.8	50	46.9	100	50	47.5	101	50	43.8	94	50
313	48.2	50	48.1	100	50	48.7	101	50	45.4	94	50
341	49.3	50	49.8	101	50	50.4	102	50	46.6	95	50
369	52.8	50	52.1	99	50	53.2	101	49	49.8	94	50
397	53.6	50	53.6	100	50	54.0	101	49	50.8	95	50
425	55.8	50	55.4	99	50	55.2	99	49	52.7	94	50
453	57.2	50	57.3	100	50	57.8	101	49	55.0	96	50
481	58.6	49	58.5	100	50	58.8	100	49	55.6	95	49
509	60.2	49	59.9	100	49	59.6	99	49	56.9	95	49
537	60.9	49	60.8	100	49	60.3	99	47	58.2	96	49
565	60.4	46	61.3	102	48	60.7	101	47	59.2	98	48
593	61.2	45	62.1	101	47	61.3	100	45	60.1	98	47
621	59.3	43	61.5	104	47	61.0	103	43	60.0	101	47
649	58.2	40	61.6	106	46	59.9	103	42	59.7	103	46
663	56.6	40	60.6	107	45	59.8	106	41	58.8	104	44
677	56.2	38	60.4	107	44	59.5	106	41	58.8	105	44
691	55.8	37	59.5	107	42	58.6	105	40	57.2	103	44
705	55.3	37	59.4	108	42	57.9	105	39	56.5	102	43
719	55.9	36	60.1	108	41	58.0	104	37	55.7	100	42
Mean for weeks											
1-13	25.3		25.0	99		25.3	100		25.1	99	
14-52	41.5		41.5	100		42.2	102		39.0	94	
53-103	57.4		59.0	103		58.5	102		56.6	99	



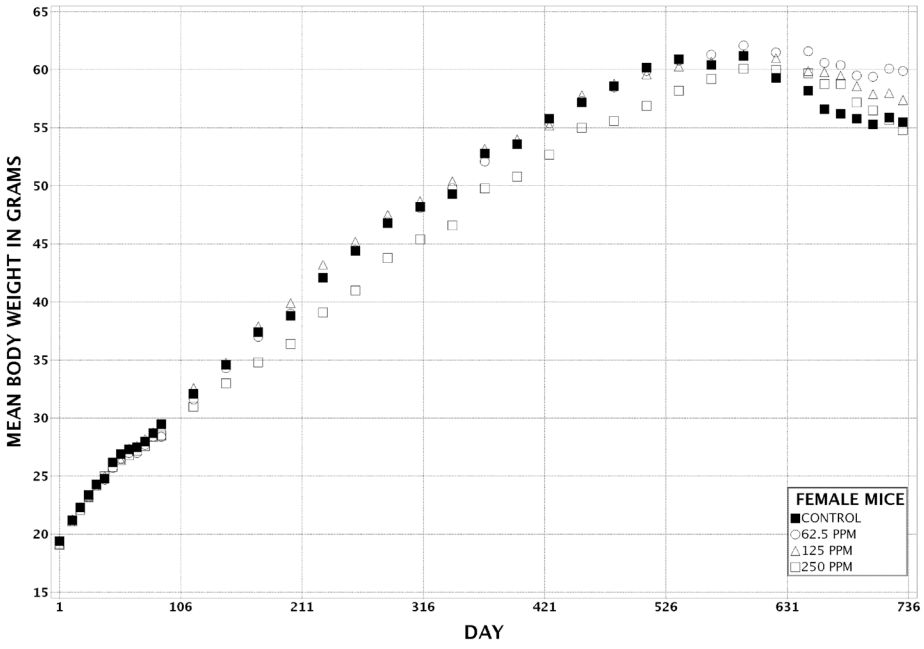


FIGURE 8
Growth Curves for Mice Exposed to 1-Bromopropane by Inhalation for 2 Years

Pathology and Statistical Analyses

This section describes the statistically significant or biologically noteworthy changes in the incidences of neoplasms and/or nonneoplastic lesions of the lung, nose, larynx, and trachea. Summaries of the incidences of neoplasms and nonneoplastic lesions, statistical analyses of primary neoplasms that occurred with an incidence of at least 5% in at least one animal group, and historical incidences for the neoplasms mentioned in this section are presented in Appendix C for male mice and Appendix D for female mice.

Lung: In the females, there were treatment-related increased incidences of alveolar/bronchiolar adenoma, alveolar/bronchiolar carcinoma, and alveolar/bronchiolar adenoma or carcinoma (combined). The incidences of alveolar/bronchiolar adenoma or carcinoma (combined) were significantly increased in all exposed groups (Tables 21, D1, and D2). The incidence of alveolar/bronchiolar adenoma in 250 ppm females and the incidences of alveolar/bronchiolar carcinoma in 62.5 and 125 ppm females were significantly increased. The incidences of alveolar/bronchiolar adenoma in the 250 ppm group and of alveolar/bronchiolar carcinoma and alveolar/bronchiolar adenoma or carcinoma (combined) in all exposed female groups exceeded the respective historical control ranges for inhalation studies (Tables 21 and D3).

Alveolar/bronchiolar carcinomas varied from moderately differentiated, circumscribed lesions to anaplastic, poorly circumscribed, infiltrative lesions. They were variable in size and shape and their growth patterns were papillary or solid with regions of squamous metaplasia, spindle cell differentiation, and necrosis. Alveolar/bronchiolar adenomas were usually smaller, well-differentiated, circumscribed lesions that often compressed the adjacent parenchyma and most were papillary.

Cytoplasmic vacuolization of bronchiolar epithelium occurred in all exposed groups of male and female mice, and the incidences in all exposed groups of male mice were significantly increased (Tables 21, C3, and D4). This lesion did not occur in chamber control mice. Histologically, this lesion was characterized by large, solitary, clear vacuoles, expanding the cytoplasm of bronchiolar epithelial cells (Plates 8 through 10). Affected epithelial cells often lacked cilia. This lesion is similar to that described in the 3-month inhalation study of 1-bromopropane in mice.

Cytoplasmic vacuolization of bronchiolar epithelium often accompanied bronchiolar regeneration and was accompanied by bronchiole necrosis in one 250 ppm male and one 125 ppm female (Tables 21, C3, and D4). Bronchiolar regeneration occurred in most exposed male and female mice, but not in chamber controls (except for a single chamber control male with minimal regeneration). The incidences were similar among exposed groups, although slight exposure concentration-related increases in severities were noted. Histologically, bronchiolar regeneration was characterized by disorganized, elongated, and flattened epithelial cells lining terminal bronchioles (Plates 8 through 10). Regenerative epithelial cells often had increased basophilia, lacked cilia, had variable amounts of cytoplasm, and had increased nuclear to cytoplasmic ratios. Nuclei of regenerative cells tended to be apical rather than basilar, and often varied in size and shape. In the affected bronchioles there was evidence of cell loss. Many of the affected bronchioles contained sloughed epithelial cells and occasional necrotic epithelial cells characterized by small, pyknotic nuclei and homogenous, hypereosinophilic cytoplasm.

Nose: There were exposure concentration-related increased incidences of cytoplasmic vacuolization of respiratory epithelium in male and female mice; the incidences in all exposed groups of males and in 125 and 250 ppm females were significantly increased (Tables 21, C3, and D4). This lesion did not occur in chamber control mice. Cytoplasmic vacuolization involved the respiratory epithelium on the lateral surfaces of nasoturbinates in Levels I and II (Plate 11). Histologically, this lesion was characterized by large, solitary, clear vacuoles expanding the cytoplasm of respiratory epithelial cells. Oftentimes, affected epithelial cells lacked cilia. Occasionally, nuclei of affected epithelial cells were pyknotic. This lesion is similar to that described in the 3-month inhalation study of 1-bromopropane in mice.

There were treatment-related increased incidences of respiratory epithelial hyperplasia in the dorsal meatus(es) in Level I in mice of both sexes (Tables 21, C3, and D4); the incidences in all exposed female groups and in 62.5 and 250 ppm males were significantly increased. Although present in chamber controls, the incidence and severity of respiratory epithelial hyperplasia increased with increasing exposure concentration, and male mice were more severely affected than female mice. Histological criteria for respiratory epithelial

TABLE 21
Incidences of Neoplasms and Nonneoplastic Lesions of the Respiratory System in Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Male				
Lung[a]	50	50	49	49
Bronchiole, Vacuolization Cytoplasmic[b]	0	18** (1.7) [c]	19** (1.7)	17** (1.8)
Bronchiole, Regeneration	1 (1.0)	44** (1.0)	38** (1.3)	47** (1.6)
Bronchiole, Necrosis	0	0	0	1 (3.0)
Nose	50	50	50	50
Respiratory Epithelium, Vacuolization Cytoplasmic	0	12** (1.8)	19** (1.9)	20** (2.2)
Respiratory Epithelium, Hyperplasia	16 (1.3)	29** (2.0)	23 (2.0)	26* (2.7)
Olfactory Epithelium, Metaplasia, Respiratory	0	7** (1.6)	6* (1.3)	3 (1.3)
Olfactory Epithelium, Atrophy	2 (2.0)	4 (2.3)	7 (2.3)	4 (1.8)
Larynx	48	50	48	50
Vacuolization Cytoplasmic	0	5* (1.4)	10** (1.1)	11** (1.6)
Trachea	49	50	47	50
Vacuolization Cytoplasmic	0	15** (1.5)	24** (1.8)	24** (2.3)
Female				
Lung	50	50	50	50
Bronchiole, Vacuolization Cytoplasmic	0	3 (1.7)	4 (1.5)	3 (1.3)
Bronchiole, Regeneration	0	45** (1.2)	43** (1.3)	49** (1.6)
Bronchiole Necrosis	0	0	1 (3.0)	0
Alveolar/bronchiolar Adenoma, Multiple	0	0	0	2
Alveolar/bronchiolar Adenoma (includes multiple)[d]				
Overall rate[e]	1/50 (2%)	6/50 (12%)	4/50 (8%)	10/50 (20%)
Adjusted rate[f]	2.2%	12.8%	8.9%	20.8%
Terminal rate[g]	1/36 (3%)	6/40 (15%)	4/37 (11%)	7/41 (17%)
First incidence (days)	731 (T)	731 (T)	731 (T)	649
Poly-3 test[h]	P=0.007	P=0.064	P=0.181	P=0.006
Alveolar/bronchiolar Carcinoma, Multiple	0	2	1	1
Alveolar/bronchiolar Carcinoma (includes multiple)[i]				
Overall rate	0/50 (0%)	7/50 (14%)	5/50 (10%)	4/50 (8%)
Adjusted rate	0.0%	14.9%	11.1%	8.5%
Terminal rate	0/36 (0%)	6/40 (15%)	5/37 (14%)	4/41 (10%)
First incidence (days)	—[j]	709	731 (T)	731 (T)
Poly-3 test	P=0.277	P=0.009	P=0.031	P=0.068
Alveolar/bronchiolar Adenoma or Carcinoma[k]				
Overall rate	1/50 (2%)	9/50 (18%)	8/50 (16%)	14/50 (28%)
Adjusted rate	2.2%	19.2%	17.8%	29.2%
Terminal rate	1/36 (3%)	8/40 (20%)	8/37 (22%)	11/41 (27%)
First incidence (days)	731 (T)	709	731 (T)	649
Poly-3 test	P<0.001	P=0.010	P=0.016	P<0.001

TABLE 21
Incidences of Neoplasms and Nonneoplastic Lesions of the Respiratory System in Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Female (continued)				
Nose	50	50	50	50
Respiratory Epithelium, Vacuolization Cytoplasmic	0	3 (1.3)	5* (1.6)	8** (1.6)
Respiratory Epithelium, Hyperplasia	11 (1.1)	25** (1.4)	28** (1.5)	27** (1.7)
Olfactory Epithelium, Metaplasia, Respiratory	0	4 (1.0)	5* (1.0)	14** (1.1)
Olfactory Epithelium, Atrophy	0	0	0	6* (1.3)
Larynx	50	50	50	50
Vacuolization Cytoplasmic	0	3 (1.3)	2 (2.0)	2 (2.5)
Trachea	50	49	50	50
Vacuolization Cytoplasmic	0	8** (1.5)	7** (1.9)	4 (2.0)

* Significantly different ($P \leq 0.05$) from the chamber control group by the Poly-3 test

** $P \leq 0.01$

(T) Terminal sacrifice

[a] Number of animals with tissue examined microscopically

[b] Number of animals with lesion

[c] Average severity grade of lesions in affected animals: 1=minimal, 2=mild, 3=moderate, 4=marked

[d] Historical incidence for 2-year inhalation studies with chamber controls (mean ± standard deviation): 18/350 (5.1% ± 3.8%), range 2%-12%; all routes: 73/1,496 (4.9% ± 3.4%), range 0%-12%

[e] Number of animals with neoplasm per number of animals with lung examined microscopically

[f] Poly-3 estimated neoplasm incidence after adjustment for intercurrent mortality

[g] Observed incidence at terminal kill

[h] Beneath the chamber control incidence is the P value associated with the trend test. Beneath the exposed group incidence is the P value corresponding to pairwise comparisons between the chamber controls and that exposed group. The Poly-3 test accounts for differential mortality in animals that do not reach terminal sacrifice.

[i] Historical incidence for inhalation studies: 9/350 (2.6% ± 2.8%), range 0%-6%; all routes: 59/1,496 (3.9% ± 3.4%), range 0%-12%

[j] Not applicable; no neoplasms in animal group

[k] Historical incidence for inhalation studies: 27/350 (7.7% ± 3.6%), range 2%-12%; all routes: 128/1,496 (8.6% ± 3.7%), range 2%-18%

hyperplasia included thickened epithelium due to increased cellularity, submucosal epithelial invaginations with intraepithelial crypt-like structures (pseudoglands), and/or finger-like projections of epithelium that extended into the nasal cavity. Lesions were graded minimal, mild, moderate, or marked (i.e., grades 1, 2, 3, or 4) depending upon the degree of involvement of one or both meatus(es). *Grade 0* was normal. *Grade 1 (minimal*) respiratory epithelial hyperplasia was diagnosed when the respiratory epithelium displayed focal or regional mucosal thickening with submucosal epithelial invaginations in one or both dorsal meatus(es). *Grade 2 (mild)* respiratory epithelial hyperplasia was diagnosed when at least two thirds of one or both dorsal meatus(es) displayed respiratory epithelium with submucosal epithelial invaginations and/or epithelial papillary projections that together involved < 25% into the dorsal meatus(es). *Grade 3 (moderate)* respiratory epithelial hyperplasia was diagnosed when two thirds to all of both dorsal meatus(es) displayed respiratory epithelium with submucosal epithelial invaginations forming intraepithelial crypt-like structures (pseudoglands) and finger-like papillary projections of epithelium that together involved 25% to 50% into the dorsal meatus(es). *Grade 4 (marked)* respiratory epithelial hyperplasia was diagnosed when approximately all of both dorsal meatuses had hyperplastic respiratory epithelium with tortuous, submucosal invaginations

forming intraepithelial crypt-like structures (pseudoglands) and finger-like papillary projections of epithelium that together involved 50% to 75% into the dorsal meatuses.

There were treatment-related increased incidences of respiratory metaplasia of olfactory epithelium in male mice and exposure concentration-related increases in female mice; incidences of this lesion were significantly increased in 62.5 and 125 ppm males and 125 and 250 ppm females (Tables 21, C3, and D4). This lesion was not observed in chamber control mice. Located at Levels II and/or III of the nasal sections, the lesion was characterized as segmental replacement of olfactory epithelium by ciliated, pseudostratified epithelium resembling respiratory epithelium of the anterior nasal cavity; this lesion was often associated with submucosal inflammation and/or glandular hyperplasia. The severity of the lesions was graded minimal to mild based on the degree of involvement of the dorsal meatuses and/or turbinates.

The incidence of olfactory epithelial atrophy was significantly increased in 250 ppm females (Tables 21 and D4). The incidences of this lesion were slightly increased in all exposed groups of males, but the increases were not statistically significant (Tables 21 and C3). This lesion consisted of segmental thinning of the olfactory epithelium at Level II and/or III of the nasal cavity due to loss of neuronal cells with primarily sustentacular and basal cells remaining. It was often associated with submucosal inflammation and/or glandular hyperplasia, and usually was present within the dorsal meatuses.

Larynx and Trachea: There were increased incidences of cytoplasmic vacuolization of respiratory epithelium in the larynx and the trachea of exposed males and females; the incidences were significantly increased in both tissues in all exposed groups of males and in the trachea of 62.5 and 125 ppm females (Tables 21, C3, and D4). This lesion did not occur in chamber control mice. Histologically, this lesion was characterized by large, solitary, clear vacuoles expanding the cytoplasm of respiratory epithelial cells (Plate 12). Affected epithelial cells often lacked cilia. Occasionally, nuclei of affected epithelial cells were pyknotic. This lesion is similar to that described in the 3-month inhalation study of 1-bromopropane in mice.

Other Findings: Hepatocellular carcinomas were not observed in 250 ppm females (Table D2); the incidence was significantly less than in the concurrent chamber controls and lower than the historical control range for inhalation studies [39/350 (11% ± 5%), range 6%-20%]. The incidences of hepatocellular adenomas or carcinomas (combined) in 62.5 and 250 ppm females were significantly less than that in the chamber controls. The incidence of skin sarcoma in 250 ppm females was also significantly less than that in the chamber controls.

GENETIC TOXICOLOGY

1-Bromopropane (doses up to 10,000 µg/plate) was not mutagenic in either of two independent bacterial mutagenicity assays, with or without exogenous metabolic activation (Table E1). Bacterial strains tested included *Salmonella typhimurium* strains TA97, TA98, TA100, and TA1535, as well as *Escherichia coli* strain WP2 *uvrA*/pKM101. In addition to the negative results in the two bacterial tests, no increases in the frequencies of micronucleated normochromatic erythrocytes were seen in peripheral blood of male or female B6C3F1 mice exposed for 3 months to 62.5 to 500 ppm 1-bromopropane via inhalation (Table E2). The percentage of reticulocytes (polychromatic erythrocytes) in the peripheral blood of male and female mice was unaltered by 1-bromopropane exposure, suggesting a lack of chemical-associated bone marrow toxicity.

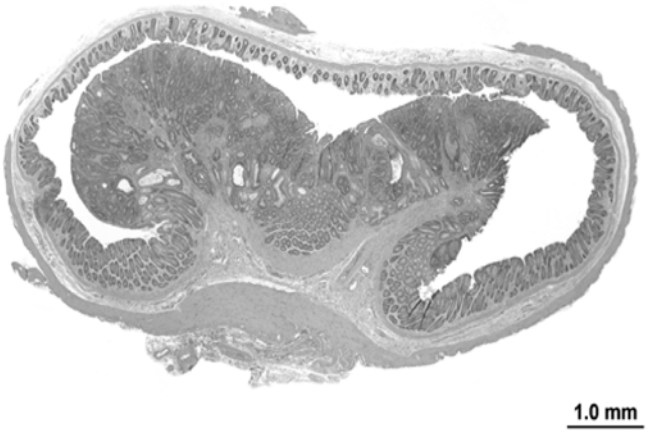


PLATE 1
Polypoid adenoma in the large intestine (rectum) of a female F344/N rat exposed to 500 ppm 1-bromopropane by inhalation for 2 years. The adenoma occupies the majority of the intestinal lumen. H&E

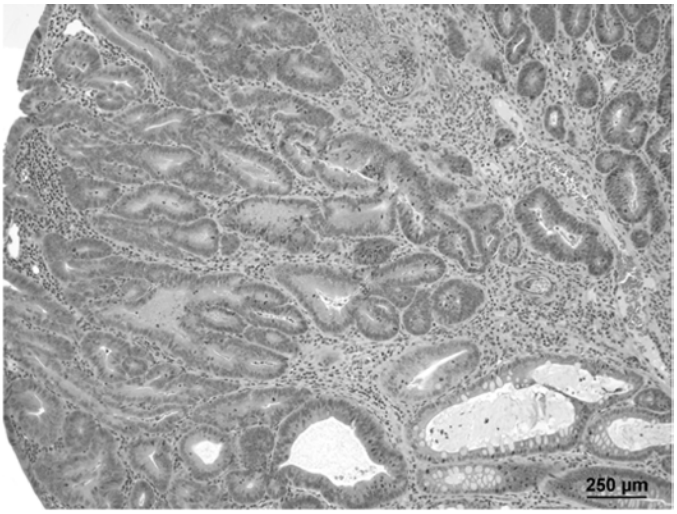


PLATE 2
Higher magnification of Plate 1. Note the multiple layers of basophilic neoplastic epithelial cells lining the glands. H&E

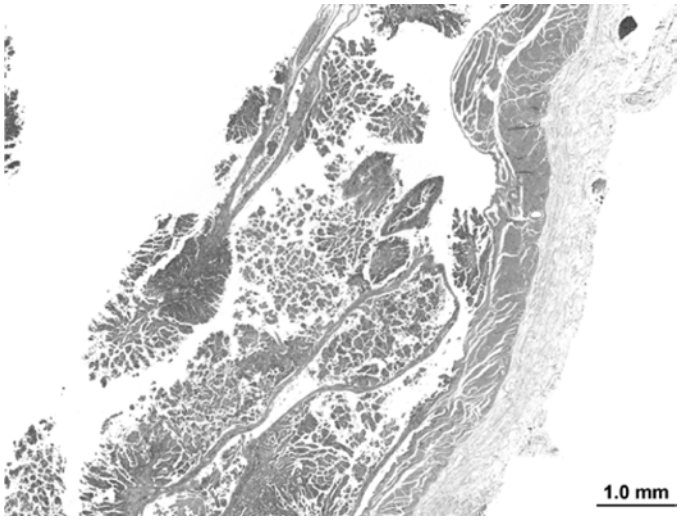


PLATE 3
Malignant mesothelioma on the parietal surface of the peritoneal cavity of a male F344/N rat exposed to 500 ppm 1-bromopropane by inhalation for 2 years. Note the complex papillary structures. H&E

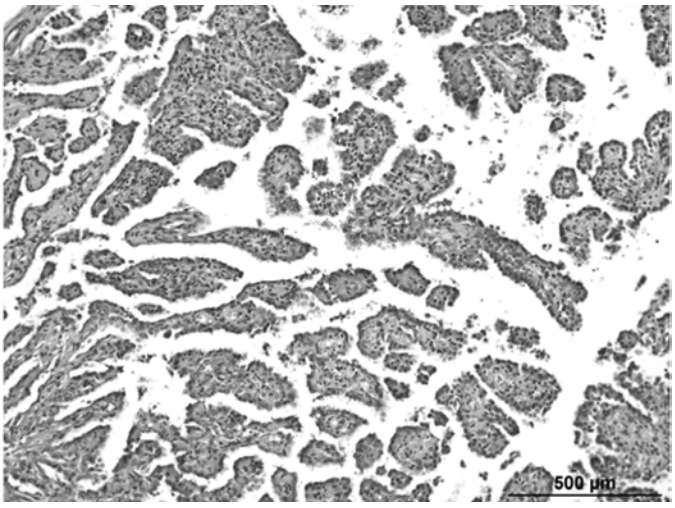


PLATE 4
Higher magnification of Plate 3. Note the cuboidal to flattened neoplastic mesothelial cells lining the papillary structures and the prominent stroma containing clusters of neoplastic mesothelial cells. H&E

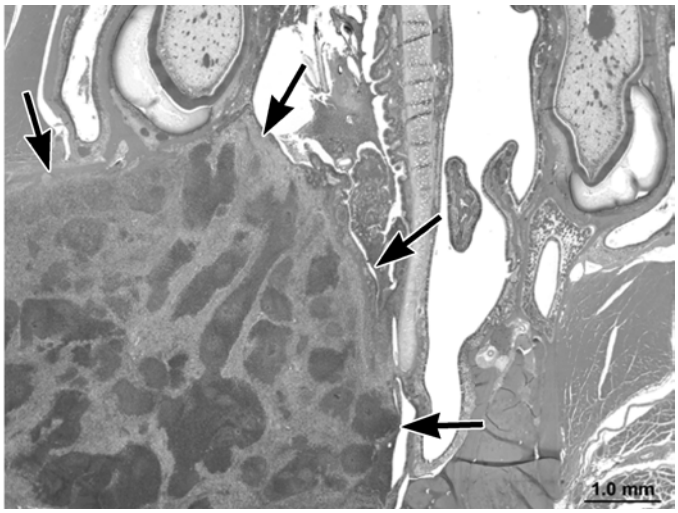


PLATE 5
Suppurative chronic inflammation (arrows) in Level II of the nasal cavity of a male F344/N rat exposed to 500 ppm 1-bromopropane by inhalation for 2 years. H&E

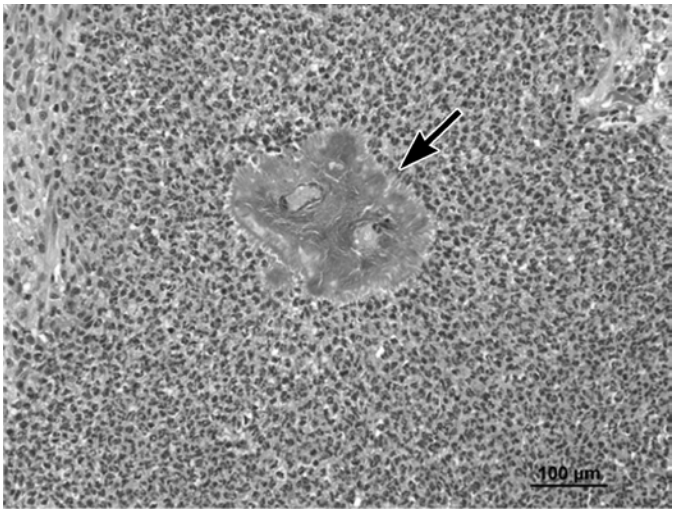


PLATE 6
Higher magnification of Plate 5. The suppurative chronic inflammation is composed of abundant eosinophilic necrotic cellular debris, fibrin, and neutrophils, and contains Splendore-Hoeppli material (arrow). These lesions were often bordered by a dense, fibrous capsule. H&E

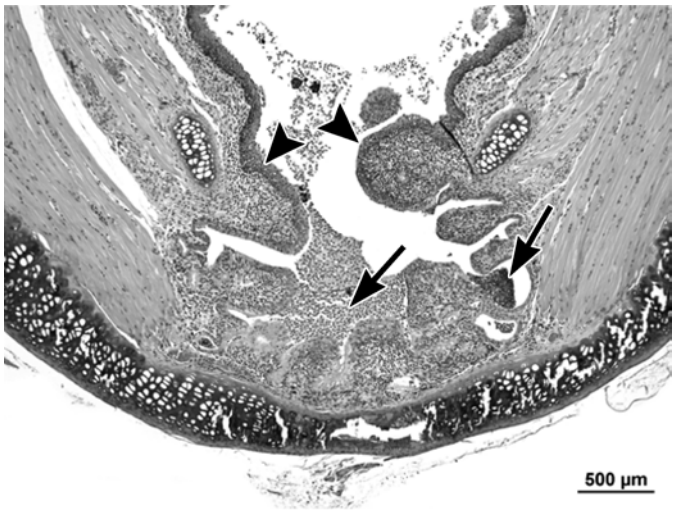


PLATE 7
Larynx of a male F344/N rat exposed to 500 ppm 1-bromopropane by inhalation for 2 years. Note the squamous metaplasia of the lining epithelium (arrowheads) associated with chronic active inflammation within the submucosa and suppurative chronic inflammation within the lumen of the glands (arrows). H&E

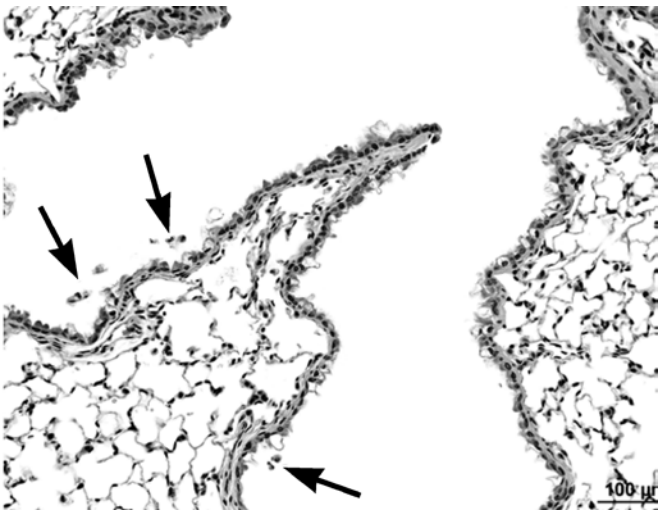


PLATE 8
Bronchiolar epithelial regeneration and vacuolization in the lung of a male B6C3F1 mouse exposed to 250 ppm 1-bromopropane by inhalation for 2 years. There are sloughed epithelial cells in the lumen of the bronchiole (arrows) indicative of epithelial cell loss. H&E

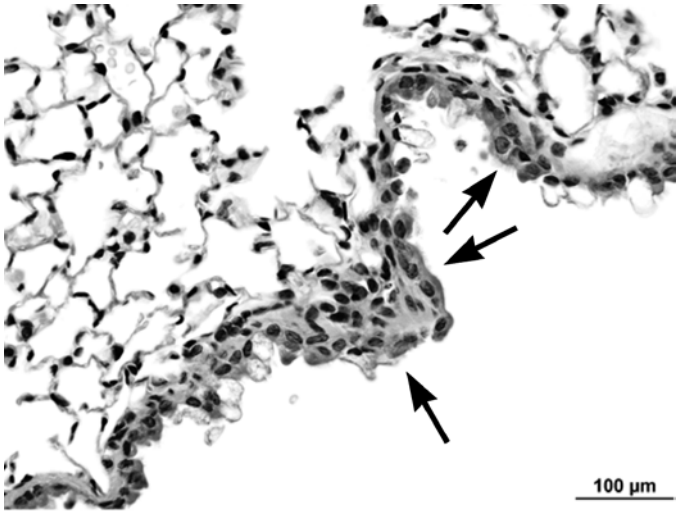


PLATE 9
Higher magnification of Plate 8. The regenerated epithelium has prominent, polyhedral cells and is disorganized with loss of cilia (arrows). There is also vacuolization of the bronchiolar epithelium. H&E

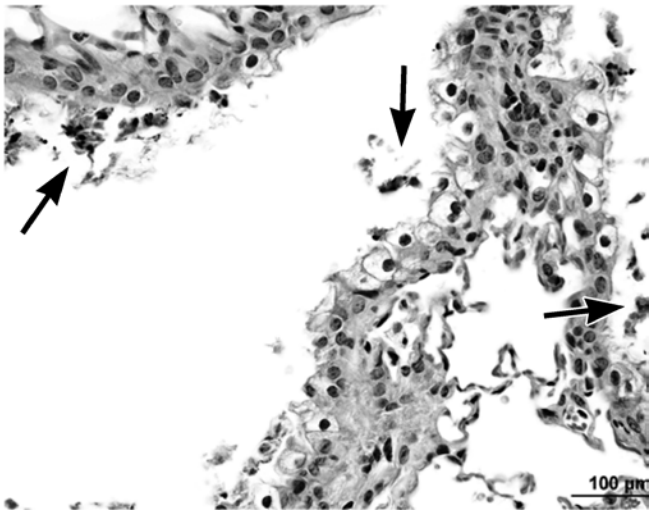


PLATE 10
Bronchiolar epithelial necrosis and cytoplasmic vacuolization in the lung of a male B6C3F1 mouse exposed to 250 ppm 1-bromopropane by inhalation for 2 years. Note the abundant necrotic cells sloughing into the bronchiolar lumen (arrows). H&E

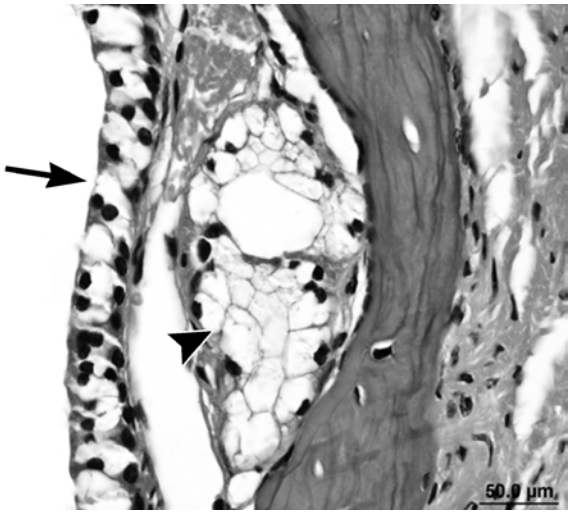


PLATE 11
Cytoplasmic vacuolization of the respiratory epithelium lining the nasoturbinates (arrows) in Level I of the nasal cavity of a male B6C3F1 mouse exposed to 250 ppm 1-bromopropane by inhalation for 2 years. H&E

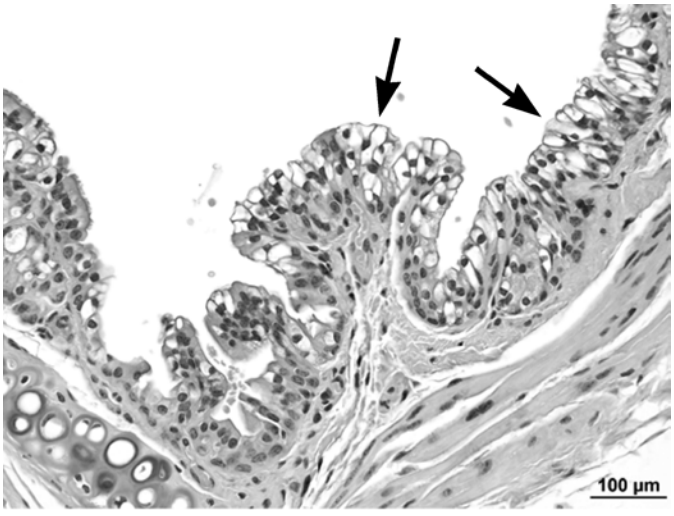


PLATE 12
Vacuolization of the respiratory epithelium (arrows) in the trachea of a male B6C3F1 mouse exposed to 250 ppm 1-bromopropane by inhalation for 2 years. H&E

DISCUSSION AND CONCLUSIONS

Occupational exposure to 1-bromopropane is expected to increase significantly because of its increasing use as a presumably less toxic and less flammable replacement for several high-production volume chemicals used in emissive applications such as vapor and immersion degreasing operations and critical cleaning of electronics and metals. 1-Bromopropane has also been proposed as a replacement for ozone depleting refrigerants (*Fed. Regist.*, 2000). 1-Bromopropane was nominated for study based on the potential for widespread exposure to this chemical and a lack of chronic toxicity and carcinogenicity data. Because previous short-term studies demonstrated that 1-bromopropane is a neurotoxicant and a reproductive toxicant in laboratory animals (Yu *et al*., 1998; Ichihara *et al*., 2000a), the main focus of the current studies was to evaluate the chronic toxicity and carcinogenicity of 1-bromopropane.

Exposure to 1-bromopropane concentrations of 500 ppm or greater in the 2-week and 3-month studies resulted in early deaths of mice. The liver was identified as a primary target site in these mice, and severe centrilobular necrosis was likely the cause of death. In the 2-week study, moderate to marked centrilobular necrosis was present in all males and most females exposed to 500 ppm or greater. In the 3-month study, extensive hepatic centrilobular necrosis was present only in 500 ppm mice that died early. Centrilobular chronic inflammation was present in livers of surviving 500 ppm mice, and no hepatocellular lesions were observed in mice exposed to lower concentrations except in one 250 ppm male. The centrilobular location of the lesions in the region of the liver that contains the highest levels of P450 activity suggests that the hepatotoxicity may be due to a reactive metabolite of 1-bromopropane. In mice, 1-bromopropane is oxidatively metabolized in the liver by cytochrome P450 2E1 (CYP2E1) to reactive metabolites that may be responsible for the toxicity of 1-bromopropane (Garner *et al*., 2006, 2007; Liu *et al*., 2009). 2-Oxo-1-bromopropane, a highly reactive oxidative metabolite of 1-bromopropane (Robinson *et al*., 1989), has been shown to alkylate proteins and inactivate enzymes (Beeley and Neurath, 1968; Cohen *et al*., 1982; Mitchell *et al*., 1998). Detoxification of 1-bromopropane metabolites occurs primarily via glutathione-*S*-transferase (GST)–mediated conjugation with glutathione (Garner *et al*., 2006; Liu *et al*., 2009). Glutathione is important in protecting tissues from alkylating metabolites. Marked reduction or depletion of liver glutathione results in enhanced toxicity (Ketterer *et al*., 1983). In the current studies, mice exposed to concentrations greater than 250 ppm may have produced levels of metabolites that exceeded the amount of glutathione available for conjugation. Oxidative metabolites of 1-bromopropane may also deplete glutathione by inhibiting enzymes required for glutathione synthesis. Alternatively, 1-bromopropane metabolites may inhibit hepatocellular GST thereby preventing glutathione conjugation. These mechanisms are supported by recent studies comparing the susceptibility of three strains of male inbred mice (BALB/cA, C57BL/6, DBA/2J) to 1-bromopropane-mediated hepatotoxicity (Liu *et al*., 2009). BALB/cA mice were the most susceptible to 1-bromopropane hepatotoxicity and also had the highest CYP2E1 activity, the lowest glutathione levels, and the lowest GST activity.

Rats were less sensitive than mice to 1-bromopropane toxicity in the 2-week and 3-month studies. In the 2-week rat study, there were no treatment-related deaths and significantly lower body weights occurred only in the 1,000 and 2,000 ppm groups. Liver weights were increased in most groups of exposed rats; however, there was no histopathologic evidence of toxicity. In the 3-month study, a mild cytoplasmic vacuolization was present in the liver of all 500 and 1,000 ppm rats, and in half of the 250 ppm male rats. Other indications of mild hepatotoxicity in exposed rats were increased liver weights and increases in serum sorbitol dehydrogenase levels. Qualitatively, rats and mice produce the same oxidative metabolites of 1-bromopropane (Garner *et al*., 2007); however, mice were reported to have a greater capacity to oxidatively metabolize 1-bromopropane than rats (Garner *et al*., 2006). This species difference in metabolic capacity may partially explain why extensive hepatotoxicity and early deaths occurred in exposed mice and not in rats.

The differential capacity of mice and rats to metabolize 1-bromopropane to reactive metabolites, and the possible relationship of this capacity to the observed hepatotoxicity in 1-bromopropane-exposed mice, may have a bearing on the design of the bacterial mutagenicity assays conducted with 1-bromopropane (Appendix E). These *in vitro* assays used rat liver microsome preparations (S9) to provide exogenous metabolic activation capability to the assay. Given the reduced levels of CYP2E1 in rat liver, this standard experimental design may not have been optimal for assessing the mutagenicity of 1-bromopropane. However, the mutagenic activity reported by Barber *et al*. (1981) in *Salmonella typhimurium* strains TA100 and TA1535 was seen both with and without rat liver S9, indicating that 1-bromopropane is a direct-acting mutagen, when testing is conducted within a closed system to control for the volatility of 1-bromopropane. Because the NTP bacterial mutagenicity studies were not conducted within a closed system, the negative results in the assays may have resulted from inadequate exposure to the test article. However, given the strength of the responses reported by Barber *et al*. (1981), it is curious that no indication of a mutagenic response was observed in the NTP studies, even allowing for a reduction in exposure due to volatility. Thus, the effects of volatility and reduced CYP2E1 levels in standard rat liver S9 preparations on the mutagenicity of 1-bromopropane remain unresolved.

1-Bromopropane also caused lesions in the respiratory tract of mice, although these lesions were not considered as severe as those in the liver. In the 2-week study, respiratory tract lesions were most severe in mice that died early after exposure to 500 ppm or greater. Necrosis and vacuolization of the nasal respiratory and olfactory epithelium and prominent necrosis of the bronchiolar epithelium were present only in early death mice. In survivors exposed to 500 ppm or greater, lesions consisted primarily of regeneration of the olfactory epithelium and the bronchiolar epithelium. In the 3-month study, mild necrosis of the nasal respiratory and olfactory epithelium, mild necrosis of the epithelium lining the trachea, and minimal necrosis of the epithelium lining the larynx were observed primarily in 500 ppm mice that died early. Minimal regeneration of the bronchiolar epithelium was observed in the majority of 250 and 500 ppm mice that survived to the end of the study. Although the upper respiratory tract lesions in exposed mice may be due to a direct irritant effect, metabolism of 1-bromopropane to a toxic metabolite by the nasal epithelium cannot be ruled out.

In the 2-week and 3-month studies, rats were less susceptible than mice to 1-bromopropane-related lesions in the nasal cavity. In the 2-week study, foci of minimal necrosis of the respiratory epithelium and minimal to mild suppurative inflammation were present in a few 1,000 and 2,000 ppm male rats. There were no exposure-related nasal lesions in rats exposed for 3 months. Although the mechanism of this species difference is unclear, it may be associated with differences in metabolic capability in the nasal epithelium.

The potential immunotoxicity of 1-bromopropane was investigated because of earlier reports of immunosuppression following oral exposure to 1-bromopropane (Lee *et al*., 2007a), and the structurally related chemicals 2-bromopropane (Jeong *et al*., 2002), and 1,3-dibromopropane (Lee *et al*., 2007b). 1-Bromopropane caused some evidence of immunosuppression in F344/N rats and B6C3F1 mice after inhalation exposure for 4 and 10 weeks (Anderson *et al*., 2010). Significant decreases in the IgM response to sheep red blood cells (SRBCs) were observed in both species after exposure to 1-bromopropane (Anderson *et al*., 2010). In addition, significant decreases in the total number of spleen cells and T-cells were observed in mice after 4 weeks of exposure to 1-bromopropane concentrations ranging from 125 to 1,000 ppm. Lee *et al*. (2007a,b) suggested that the immunotoxicity of 1-bromopropane and 1,3-dibromopropane may result from depletion of glutathione. Immunosuppression caused by these two chemicals was correlated with chemical-induced hepatotoxicity and decreases in liver and spleen glutathione. Partial depletion of glutathione has been shown to affect immune function (effector phase of cytotoxic T-cell responses and IL-2 dependent functions) (Dröge *et al*., 1994). Although glutathione levels were not measured in the current studies, it is possible that the hepatotoxicity and immunotoxicity observed in mice and rats after inhalation of 1-bromopropane may be due to reduced levels of glutathione.

In the 2-year study of 1-bromopropane in mice, there was no reduction in survival and no effect on body weights of males or females. Chronic inhalation of 1-bromopropane resulted in neoplasms in female mice but not in male mice. There was clear evidence that 1-bromopropane exposure caused increased incidences of alveolar/bronchiolar adenoma or carcinoma (combined) in female mice. Because alveolar/bronchiolar adenomas are known to progress to carcinomas (Boorman and Eustis, 1990), the NTP combines the incidences of these neoplasms. The incidences of alveolar/bronchiolar adenoma or carcinoma (combined) in female mice increased in an exposure concentration-related manner, were significantly greater than that in the chamber controls at all exposure concentrations, and exceeded the range in historical controls for inhalation

studies. The incidence of alveolar/bronchiolar adenoma in female mice exposed to 250 ppm was significantly increased compared to that in the chamber controls and exceeded the historical control ranges for inhalation studies and for all routes. The incidences of alveolar/bronchiolar carcinoma in 62.5 and 125 ppm female mice were significantly increased compared to the chamber controls; although not statistically increased, alveolar/bronchiolar carcinoma occurred in 8% of the 250 ppm females. The incidences in all exposed groups of female mice exceeded the historical control range for inhalation studies and the incidence in the 62.5 ppm females exceeded the historical control range from all routes. In chamber control female mice alveolar/bronchiolar carcinoma was not observed. Chronic exposure to the structurally related 1,2-dibromoethane also caused alveolar/bronchiolar adenomas and carcinomas in male and female mice and female rats (NTP, 1982a).

In the 2-year study in rats, survival of 500 ppm males was reduced; however, there were no chemical-related effects on body weights. Exposure to 1-bromopropane for 2 years resulted in neoplasms in rats at various sites. The treatment-related increased incidences of adenoma of the large intestine (colon and rectum) in female rats were considered clear evidence of carcinogenic activity. Increased incidences of adenoma were exposure concentration related, and the incidence in the 500 ppm group was significantly greater than that in the chamber controls. Adenomas were found in the large intestine of two 250 ppm male rats (4%) and in one 500 ppm male rat (2%). Although the increased incidences in males were not statistically significant, the presence of these neoplasms in females and the low historical occurrence of these neoplasms suggest that they were treatment related. Adenomas of the large intestine are rare in control male and female rats, occurring at a rate of less than 0.2%. Although no large intestine carcinomas were observed in male or female rats in the current study, adenomas of the large intestine can progress to carcinomas (Deschner, 1983; Chang, 1984; Nigro, 1985). Oral treatment of rats with two brominated methanes (bromodichloromethane and tribromomethane) also resulted in significantly increased incidences of adenocarcinoma of the large intestine in both male and female rats (NTP, 1987, 1989). Thus, the occurrence of adenomas of the large intestine were considered some evidence of carcinogenic activity in male rats.

Chronic exposure to 1-bromopropane caused an increase in the incidences of neoplasms of the skin in male rats, and skin neoplasms in female rats may also have been related to exposure. In males, the combined incidences of all neoplasms of epithelial origin increased in a significant exposure concentration-dependent manner. The incidences were significantly increased in all exposed groups of males relative to chamber controls and exceeded the range in historical controls from inhalation studies. The most numerous neoplasms were identified as keratoacanthoma, a benign neoplasm that morphologically resembles squamous cell carcinoma. In males, the incidences of keratoacanthoma increased with a positive trend; the incidences in the 250 and 500 ppm groups were significantly increased compared to chamber controls and exceeded the historical control range for inhalation studies. The increased incidence of keratoacanthoma is a concern because there is evidence that it can progress to squamous cell carcinoma, a highly malignant tumor (Elwell *et al.*, 1990). Squamous cell carcinoma is a rare neoplasm occurring in only 0.6% of historical controls from all studies. Although not statistically significant, the incidence of squamous cell carcinoma in 500 ppm male rats was greater than that in the chamber controls and exceeded the historical control ranges for inhalation studies and for all routes. In females, the incidence of squamous cell papilloma, keratoacanthoma, basal cell adenoma, or basal cell carcinoma (combined) in the 500 ppm group exceeded the historical control range for inhalation studies. Keratoacanthomas occurred in females, but unlike in the males, the incidences were not increased compared to the concurrent chamber controls or historical controls. No squamous cell carcinomas were seen in female rats.

The evidence that 1-bromopropane exposure was associated with an increased incidence of pancreatic islet adenoma in male rats was equivocal. Although the increased incidences of pancreatic islet adenoma were statistically significant at all exposure concentrations, the incidence in the chamber controls (0/50) was less than the mean in historical controls from inhalation studies (6%), and the incidences in exposed groups did not exceed the historical control ranges of inhalation studies or all routes. The incidences of pancreatic islet carcinoma in exposed male rats were not significantly different from that in the chamber controls and were not considered treatment related.

The evidence for malignant mesothelioma in all organs was equivocal in male rats. The incidences of malignant mesothelioma occurred with a positive trend, but only the incidence in the 500 ppm group was significantly increased. The overall strength of this evidence was considered equivocal because of the common occurrence of malignant mesothelioma in male rats in 2-year NTP

studies. The historical control range for malignant mesothelioma in male rats for inhalation studies and by all routes is 0% to 6%. Although the incidence in the 500 ppm group was significantly increased, it was barely outside the historical control range.

An unusual nonneoplastic finding in rats exposed to 1-bromopropane for 2 years was the presence of Splendore-Hoeppli reaction material associated with suppurative inflammation. Lesions with Splendore-Hoeppli material were present primarily in the nose and skin of exposed male and female rats, although other sites were affected. The exact composition and mechanism of formation of Splendore-Hoeppli material is unknown, but it is thought to represent deposition of antigen-antibody complexes and debris from host inflammatory cells (Hussein, 2008). Alternatively, it may represent glycoproteins, lipid, and calcium derived from host leukocytes (Bhagavan *et al*., 1982). Histologically, Splendore-Hoeppli material appears as strongly eosinophilic, amorphous material surrounding or adjacent to the causative agent, typically fungi, helminthes or bacteria (Hussein, 2008). The Splendore-Hoeppli bodies appear microscopically as distinct radiating, star-like, or club-shaped configurations. In the United States, Splendore-Hoeppli bodies are often seen in association with botryomycotic infections caused by *Proteus*, *Escherichia coli* or *Pseudomonas aeruginosa* (Wenig *et al*., 1996). In this study, cultures from four of five 1-bromopropane-exposed rats with this lesion were positive for *P. aeruginosa*. It is not clear why lesions with Splendore-Hoeppli bodies were only present in rats, since both rats and mice were immunosuppressed after 1-bromopropane exposure. Species differences in the presence of opportunistic bacteria, or differences in innate resistance to infection, may have played a role in this difference.

The presence of lesions with Splendore-Hoeppli bodies in rats was considered to be treatment related. The incidence of lesions with Splendore-Hoeppli bodies increased with 1-bromopropane exposure concentration, and was considerably higher in males (34%) and females (28%) exposed to 500 ppm. Lesions with Splendore-Hoeppli bodies were not present in chamber control rats. Although the mechanism is not clear, immunosuppression in 1-bromopropane-exposed rats may have contributed to the development of Splendore-Hoeppli bodies. The development of botryomycosis and formation of Splendore-Hoeppli bodies in humans may be facilitated by immunosuppression as it has been associated with diabetes, immunoglobulin deficiency, cutaneous anergy, corticosteroid treatment, and AIDS (Schlossberg *et al.*, 1980; Brunken *et al*., 1983; Patterson *et al*., 1987).

Because adequate data were available demonstrating the developmental and reproductive toxicity of 1-bromopropane, only sperm motility and vaginal cytology were evaluated in the current studies. Based upon the effects on sperm count and motility and estrous cyclicity seen in the 3-month studies, we conclude that 1-bromopropane has the potential to produce adverse effects on fertility and reproductive performance in rodent studies involving similar exposures. The NTP Center for the Evaluation of Risks to Human Reproduction (CERHR) conducted an extensive evaluation on the potential effects of 1-bromopropane on human reproduction and development (NTP, 2003). The CERHR concluded that based upon the available data, inhaled 1-bromopropane causes prenatal developmental toxicity and reproductive toxicity in male and female rats; however, available human data were insufficient to draw conclusions on the potential for reproductive or developmental toxicity. The overall NTP conclusion was that "there is serious concern for reproductive and developmental effects of 1-bromopropane at the upper end of the human occupational exposure range (18 to 381 ppm)." "Serious concern" is the highest level of NTP conclusion regarding the possibilities that human development and reproduction might be adversely affected.

CONCLUSIONS

Under the conditions of these 2-year inhalation studies, there was *some evidence of carcinogenic activity** of 1-bromopropane in male F344/N rats based on the occurrence of rare adenomas of the large intestine and increased incidences of epithelial neoplasms of the skin (keratoacanthoma, squamous cell carcinoma, and basal cell neoplasms). Increased incidences of malignant mesothelioma and pancreatic islet adenoma and carcinoma (combined) may also have been related to 1-bromopropane exposure. There was *clear evidence of carcinogenic activity* of 1-bromopropane in female F344/N rats based on increased incidences of adenoma of the large intestine. Increased incidences of skin neoplasms may also have been related to 1-bromopropane exposure. There was *no evidence of carcinogenic activity* of 1-bromopropane in male B6C3F1 mice exposed to concentrations of 62.5, 125, or 250 ppm 1-bromopropane. There was *clear evidence of carcinogenic activity* of 1-bromopropane in female B6C3F1 mice based on increased incidences of alveolar/bronchiolar neoplasms.

Exposure to 1-bromopropane resulted in increased incidences of nonneoplastic lesions in the nose of rats and mice, the larynx of rats and male mice, and the trachea and lung of female rats and male and female mice. Suppurative inflammatory lesions with Splendore-Hoeppli material were present primarily in the nose and skin of male and female rats exposed to 1-bromopropane.

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* Explanation of Levels of Evidence of Carcinogenic Activity is on page 13. A summary of the Technical Reports Review Subcommittee comments and the public discussion on this Technical Report appears on page 15.

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APPENDIX A
SUMMARY OF LESIONS IN MALE RATS IN THE 2-YEAR INHALATION STUDY OF 1-BROMOPROPANE

TABLE A1
Summary of the Incidence of Neoplasms in Male Rats in the 2-Year Inhalation Study of 1-Bromopropane[a]

	Chamber Control	125 ppm	250 ppm	500 ppm
Disposition Summary				
Animals initially in study	50	50	50	50
Early deaths				
Moribund	24	20	28	35
Natural deaths	3	4	4	2
Survivors				
Terminal sacrifice	23	26	18	13
Animals examined microscopically	50	50	50	50
Alimentary System				
Esophagus	(50)	(50)	(50)	(50)
Intestine large, cecum	(47)	(47)	(47)	(48)
Intestine large, colon	(48)	(48)	(48)	(48)
Adenoma				1 (2%)
Intestine large, rectum	(48)	(48)	(48)	(49)
Adenoma			2 (4%)	
Carcinoid tumor malignant		1 (2%)		
Polyp adenomatous		1 (2%)		
Intestine small, duodenum	(48)	(48)	(48)	(48)
Intestine small, ileum	(47)	(47)	(46)	(48)
Intestine small, jejunum	(47)	(47)	(46)	(48)
Adenoma			1 (2%)	
Liver	(50)	(50)	(50)	(50)
Hepatocellular carcinoma	2 (4%)	1 (2%)		
Mesentery	(13)	(13)	(8)	(19)
Sarcoma			1 (13%)	
Oral mucosa	(3)	(0)	(2)	(4)
Squamous cell carcinoma	1 (33%)		1 (50%)	2 (50%)
Pancreas	(50)	(50)	(50)	(50)
Salivary glands	(50)	(50)	(50)	(50)
Stomach, forestomach	(50)	(50)	(50)	(50)
Stomach, glandular	(50)	(50)	(50)	(50)
Tongue	(0)	(0)	(1)	(1)
Cardiovascular System				
Heart	(50)	(50)	(50)	(50)
Endocrine System				
Adrenal cortex	(50)	(50)	(50)	(50)
Adenoma	1 (2%)		1 (2%)	
Adrenal medulla	(50)	(50)	(50)	(50)
Pheochromocytoma benign	8 (16%)	8 (16%)	5 (10%)	7 (14%)
Pheochromocytoma complex	1 (2%)			1 (2%)
Pheochromocytoma malignant		1 (2%)	1 (2%)	1 (2%)
Bilateral, pheochromocytoma benign	2 (4%)	1 (2%)	2 (4%)	1 (2%)
Islets, pancreatic	(50)	(50)	(50)	(50)
Adenoma		5 (10%)	4 (8%)	5 (10%)
Carcinoma	3 (6%)	7 (14%)	5 (10%)	3 (6%)
Parathyroid gland	(46)	(48)	(47)	(46)
Pituitary gland	(50)	(50)	(50)	(49)
Pars distalis, adenoma	37 (74%)	36 (72%)	33 (66%)	31 (63%)

TABLE A1
Summary of the Incidence of Neoplasms in Male Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Endocrine System (continued)				
Thyroid gland	(50)	(50)	(50)	(49)
C-cell, adenoma	5 (10%)	4 (8%)	3 (6%)	3 (6%)
C-cell, carcinoma	4 (8%)	3 (6%)	2 (4%)	
Follicular cell, adenoma				2 (4%)
Follicular cell, carcinoma		2 (4%)	2 (4%)	1 (2%)
General Body System				
Peritoneum	(0)	(0)	(0)	(2)
Genital System				
Coagulating gland	(3)	(0)	(1)	(2)
Epididymis	(50)	(50)	(50)	(50)
Penis	(0)	(3)	(1)	(1)
Preputial gland	(50)	(50)	(49)	(50)
Adenoma	4 (8%)	1 (2%)	1 (2%)	1 (2%)
Carcinoma	2 (4%)	2 (4%)	3 (6%)	
Prostate	(50)	(50)	(50)	(50)
Adenoma		1 (2%)		
Seminal vesicle	(50)	(49)	(49)	(50)
Testes	(50)	(50)	(50)	(50)
Bilateral, interstitial cell, adenoma	22 (44%)	20 (40%)	16 (32%)	17 (34%)
Interstitial cell, adenoma	12 (24%)	18 (36%)	16 (32%)	12 (24%)
Hematopoietic System				
Bone marrow	(50)	(50)	(50)	(50)
Lymph node	(5)	(5)	(5)	(4)
Deep cervical, carcinoma, metastatic, thyroid gland			1 (20%)	
Lymph node, bronchial	(3)	(6)	(5)	(8)
Lymph node, mandibular	(3)	(1)	(0)	(6)
Lymph node, mediastinal	(30)	(25)	(23)	(32)
Carcinoma, metastatic, thyroid gland	1 (3%)			
Lymph node, mesenteric	(50)	(50)	(50)	(50)
Spleen	(50)	(50)	(49)	(50)
Thymus	(43)	(41)	(47)	(46)
Integumentary System				
Mammary gland	(40)	(36)	(39)	(36)
Fibroadenoma	2 (5%)	2 (6%)	1 (3%)	1 (3%)
Skin	(50)	(50)	(50)	(50)
Basal cell adenoma		1 (2%)	2 (4%)	1 (2%)
Basal cell carcinoma		2 (4%)	1 (2%)	2 (4%)
Fibrous histiocytoma			1 (2%)	
Hemangiosarcoma			1 (2%)	
Keratoacanthoma		3 (6%)	4 (8%)	5 (10%)
Keratoacanthoma, multiple			2 (4%)	1 (2%)
Squamous cell carcinoma	1 (2%)	1 (2%)		2 (4%)
Pinna, neural crest tumor	1 (2%)			
Sebaceous gland, adenoma			2 (4%)	1 (2%)
Sebaceous gland, carcinoma		1 (2%)		
Subcutaneous tissue, fibroma	3 (6%)	5 (10%)	1 (2%)	
Subcutaneous tissue, lipoma	2 (4%)		1 (2%)	1 (2%)

TABLE A1
Summary of the Incidence of Neoplasms in Male Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Musculoskeletal System				
Bone	(50)	(50)	(50)	(50)
Chordoma	1 (2%)			
Osteosarcoma	1 (2%)			
Skeletal muscle	(1)	(1)	(3)	(3)
Chordoma, metastatic, bone	1 (100%)			
Nervous System				
Brain	(50)	(50)	(50)	(50)
Oligodendroglioma malignant		1 (2%)		
Osteosarcoma	1 (2%)			
Spinal cord	(1)	(1)	(2)	(1)
Respiratory System				
Larynx	(50)	(50)	(50)	(50)
Carcinoma, metastatic, thyroid gland			1 (2%)	
Lung	(50)	(50)	(50)	(50)
Alveolar/bronchiolar adenoma	2 (4%)	3 (6%)	1 (2%)	5 (10%)
Alveolar/bronchiolar adenoma, multiple		1 (2%)		
Alveolar/bronchiolar carcinoma	1 (2%)	1 (2%)	2 (4%)	1 (2%)
Carcinoma, metastatic, preputial gland	1 (2%)			
Carcinoma, metastatic, thyroid gland			1 (2%)	
Chordoma, metastatic, bone	1 (2%)			
Nose	(50)	(48)	(48)	(50)
Trachea	(50)	(50)	(50)	(50)
Carcinoma, metastatic, thyroid gland			1 (2%)	
Special Senses System				
Eye	(49)	(49)	(50)	(50)
Melanoma malignant		1 (2%)		
Harderian gland	(50)	(50)	(50)	(50)
Lacrimal gland	(1)	(0)	(0)	(0)
Zymbal's gland	(0)	(1)	(2)	(2)
Adenoma				1 (50%)
Carcinoma		1 (100%)	2 (100%)	1 (50%)
Urinary System				
Kidney	(50)	(50)	(50)	(49)
Liposarcoma				1 (2%)
Stromal nephroma				1 (2%)
Cortex, renal tubule, adenoma	1 (2%)			
Pelvis, papilloma	1 (2%)			
Urinary bladder	(50)	(50)	(50)	(50)
Transitional epithelium, carcinoma		1 (2%)		
Systemic Lesions				
Multiple organs[b]	(50)	(50)	(50)	(50)
Histiocytic sarcoma			1 (2%)	2 (4%)
Leukemia mononuclear	29 (58%)	22 (44%)	16 (32%)	24 (48%)
Lymphoma malignant				1 (2%)
Mesothelioma malignant		2 (4%)	2 (4%)	4 (8%)

TABLE A1
Summary of the Incidence of Neoplasms in Male Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Neoplasm Summary				
Total animals with primary neoplasms[c]	50	50	48	49
Total primary neoplasms	150	160	139	143
Total animals with benign neoplasms	48	49	47	48
Total benign neoplasms	102	110	98	96
Total animals with malignant neoplasms	38	33	32	32
Total malignant neoplasms	47	50	41	47
Total animals with metastatic neoplasms	3		1	
Total metastatic neoplasms	4		4	
Total animals with uncertain neoplasms-benign or malignant	1			
Total uncertain neoplasms	1			

[a] Number of animals examined microscopically at the site and the number of animals with neoplasm
[b] Number of animals with any tissue examined microscopically
[c] Primary neoplasms: all neoplasms except metastatic neoplasms

TABLE A2
Statistical Analysis of Primary Neoplasms in Male Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Adrenal Medulla: Benign Pheochromocytoma				
Overall rate[a]	10/50 (20%)	9/50 (18%)	7/50 (14%)	8/50 (16%)
Adjusted rate[b]	23.9%	21.7%	18.1%	21.6%
Terminal rate[c]	8/23 (35%)	7/26 (27%)	3/18 (17%)	3/13 (23%)
First incidence (days)	660	585	618	453
Poly-3 test[d]	P=0.432N	P=0.506N	P=0.354N	P=0.509N
Adrenal Medulla: Benign, Malignant, or Complex Pheochromocytoma				
Overall rate	11/50 (22%)	10/50 (20%)	8/50 (16%)	10/50 (20%)
Adjusted rate	25.8%	24.1%	20.6%	26.7%
Terminal rate	8/23 (35%)	8/26 (31%)	4/18 (22%)	4/13 (31%)
First incidence (days)	429	585	618	453
Poly-3 test	P=0.525	P=0.526N	P=0.385N	P=0.566
Lung: Alveolar/bronchiolar Adenoma				
Overall rate	2/50 (4%)	4/50 (8%)	1/50 (2%)	5/50 (10%)
Adjusted rate	4.8%	9.8%	2.7%	13.7%
Terminal rate	2/23 (9%)	3/26 (12%)	1/18 (6%)	3/13 (23%)
First incidence (days)	729 (T)	648	729 (T)	575
Poly-3 test	P=0.162	P=0.331	P=0.533N	P=0.165
Lung: Alveolar/bronchiolar Adenoma or Carcinoma				
Overall rate	3/50 (6%)	5/50 (10%)	3/50 (6%)	6/50 (12%)
Adjusted rate	7.2%	12.2%	8.0%	16.4%
Terminal rate	3/23 (13%)	4/26 (15%)	2/18 (11%)	3/13 (23%)
First incidence (days)	729 (T)	648	725	575
Poly-3 test	P=0.169	P=0.349	P=0.620	P=0.181
Pancreatic Islets: Adenoma				
Overall rate	0/50 (0%)	5/50 (10%)	4/50 (8%)	5/50 (10%)
Adjusted rate	0.0%	12.2%	10.4%	13.9%
Terminal rate	0/23 (0%)	4/26 (15%)	1/18 (6%)	4/13 (31%)
First incidence (days)	—[e]	666	608	697
Poly-3 test	P=0.043	P=0.029	P=0.050	P=0.019
Pancreatic Islets: Carcinoma				
Overall rate	3/50 (6%)	7/50 (14%)	5/50 (10%)	3/50 (6%)
Adjusted rate	7.2%	17.0%	13.0%	8.3%
Terminal rate	2/23 (9%)	3/26 (12%)	3/18 (17%)	2/13 (15%)
First incidence (days)	686	687	578	687
Poly-3 test	P=0.516N	P=0.149	P=0.312	P=0.594
Pancreatic Islets: Adenoma or Carcinoma				
Overall rate	3/50 (6%)	10/50 (20%)	9/50 (18%)	8/50 (16%)
Adjusted rate	7.2%	24.2%	23.1%	22.2%
Terminal rate	2/23 (9%)	5/26 (19%)	4/18 (22%)	6/13 (46%)
First incidence (days)	686	666	578	687
Poly-3 test	P=0.093	P=0.031	P=0.043	P=0.057
Pituitary Gland (Pars Distalis): Adenoma				
Overall rate	37/50 (74%)	36/50 (72%)	33/50 (66%)	31/49 (63%)
Adjusted rate	79.8%	76.0%	73.4%	71.3%
Terminal rate	19/23 (83%)	20/26 (77%)	12/18 (67%)	9/13 (69%)
First incidence (days)	534	505	514	394
Poly-3 test	P=0.189N	P=0.421N	P=0.307N	P=0.226N

TABLE A2
Statistical Analysis of Primary Neoplasms in Male Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Preputial Gland: Adenoma				
Overall rate	4/50 (8%)	1/50 (2%)	1/49 (2%)	1/50 (2%)
Adjusted rate	9.6%	2.5%	2.7%	2.8%
Terminal rate	3/23 (13%)	0/26 (0%)	0/18 (0%)	0/13 (0%)
First incidence (days)	669	711	688	642
Poly-3 test	P=0.152N	P=0.184N	P=0.212N	P=0.224N
Preputial Gland: Carcinoma				
Overall rate	2/50 (4%)	2/50 (4%)	3/49 (6%)	0/50 (0%)
Adjusted rate	4.8%	4.9%	7.9%	0.0%
Terminal rate	1/23 (4%)	1/26 (4%)	1/18 (6%)	0/13 (0%)
First incidence (days)	660	660	575	—
Poly-3 test	P=0.257N	P=0.688	P=0.458	P=0.272N
Preputial Gland: Adenoma or Carcinoma				
Overall rate	6/50 (12%)	3/50 (6%)	4/49 (8%)	1/50 (2%)
Adjusted rate	14.3%	7.3%	10.5%	2.8%
Terminal rate	4/23 (17%)	1/26 (4%)	1/18 (6%)	0/13 (0%)
First incidence (days)	660	660	575	642
Poly-3 test	P=0.080N	P=0.251N	P=0.428N	P=0.082N
Skin: Keratoacanthoma				
Overall rate	0/50 (0%)	3/50 (6%)	6/50 (12%)	6/50 (12%)
Adjusted rate	0.0%	7.4%	15.4%	16.2%
Terminal rate	0/23 (0%)	3/26 (12%)	2/18 (11%)	2/13 (15%)
First incidence (days)	—	729 (T)	488	562
Poly-3 test	P=0.008	P=0.115	P=0.012	P=0.010
Skin: Basal Cell Adenoma or Basal Cell Carcinoma				
Overall rate	0/50 (0%)	3/50 (6%)	3/50 (6%)	3/50 (6%)
Adjusted rate	0.0%	7.3%	7.8%	8.3%
Terminal rate	0/23 (0%)	2/26 (8%)	0/18 (0%)	2/13 (15%)
First incidence (days)	—	585	618	687
Poly-3 test	P=0.111	P=0.117	P=0.105	P=0.094
Skin: Keratoacanthoma or Squamous Cell Carcinoma				
Overall rate	1/50 (2%)	4/50 (8%)	6/50 (12%)	8/50 (16%)
Adjusted rate	2.4%	9.8%	15.4%	21.4%
Terminal rate	0/23 (0%)	3/26 (12%)	2/18 (11%)	3/13 (23%)
First incidence (days)	669	711	488	562
Poly-3 test	P=0.006	P=0.171	P=0.044	P=0.009
Skin: Basal Cell Carcinoma or Squamous Cell Carcinoma				
Overall rate	1/50 (2%)	3/50 (6%)	1/50 (2%)	4/50 (8%)
Adjusted rate	2.4%	7.4%	2.6%	11.0%
Terminal rate	0/23 (0%)	2/26 (8%)	0/18 (0%)	2/13 (15%)
First incidence (days)	669	711	697	659
Poly-3 test	P=0.124	P=0.297	P=0.739	P=0.138
Skin: Keratoacanthoma, Basal Cell Adenoma, Basal Cell Carcinoma, or Squamous Cell Carcinoma				
Overall rate	1/50 (2%)	7/50 (14%)	9/50 (18%)	10/50 (20%)
Adjusted rate	2.4%	17.0%	22.6%	26.7%
Terminal rate	0/23 (0%)	5/26 (19%)	2/18 (11%)	4/13 (31%)
First incidence (days)	669	585	488	562
Poly-3 test	P=0.003	P=0.028	P=0.006	P=0.002

TABLE A2
Statistical Analysis of Primary Neoplasms in Male Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Skin (Subcutaneous Tissue): Fibroma				
Overall rate	3/50 (6%)	5/50 (10%)	1/50 (2%)	0/50 (0%)
Adjusted rate	7.1%	12.0%	2.6%	0.0%
Terminal rate	1/23 (4%)	2/26 (8%)	0/18 (0%)	0/13 (0%)
First incidence (days)	646	560	690	—
Poly-3 test	P=0.053N	P=0.349	P=0.342N	P=0.149N
Skin: Fibroma or Fibrous Histiocytoma				
Overall rate	3/50 (6%)	5/50 (10%)	2/50 (4%)	0/50 (0%)
Adjusted rate	7.1%	12.0%	5.2%	0.0%
Terminal rate	1/23 (4%)	2/26 (8%)	0/18 (0%)	0/13 (0%)
First incidence (days)	646	560	614	—
Poly-3 test	P=0.075N	P=0.349	P=0.542N	P=0.149N
Testes: Adenoma				
Overall rate	34/50 (68%)	38/50 (76%)	32/50 (64%)	29/50 (58%)
Adjusted rate	73.9%	82.7%	75.9%	68.7%
Terminal rate	17/23 (74%)	22/26 (85%)	18/18 (100%)	8/13 (62%)
First incidence (days)	501	528	520	500
Poly-3 test	P=0.198N	P=0.204	P=0.516	P=0.371N
Thyroid Gland (C-Cell): Adenoma				
Overall rate	5/50 (10%)	4/50 (8%)	3/50 (6%)	3/49 (6%)
Adjusted rate	11.9%	9.7%	7.7%	8.5%
Terminal rate	2/23 (9%)	3/26 (12%)	1/18 (6%)	2/13 (15%)
First incidence (days)	660	505	488	710
Poly-3 test	P=0.356N	P=0.510N	P=0.397N	P=0.451N
Thyroid Gland (C-Cell): Carcinoma				
Overall rate	4/50 (8%)	3/50 (6%)	2/50 (4%)	0/49 (0%)
Adjusted rate	9.7%	7.3%	5.2%	0.0%
Terminal rate	4/23 (17%)	2/26 (8%)	1/18 (6%)	0/13 (0%)
First incidence (days)	729 (T)	687	554	—
Poly-3 test	P=0.051N	P=0.508N	P=0.373N	P=0.082N
Thyroid Gland (C-Cell): Adenoma or Carcinoma				
Overall rate	9/50 (18%)	7/50 (14%)	5/50 (10%)	3/49 (6%)
Adjusted rate	21.4%	16.9%	12.6%	8.5%
Terminal rate	6/23 (26%)	5/26 (19%)	2/18 (11%)	2/13 (15%)
First incidence (days)	660	505	488	710
Poly-3 test	P=0.066N	P=0.400N	P=0.223N	P=0.102N
Thyroid Gland (Follicular Cell): Adenoma or Carcinoma				
Overall rate	0/50 (0%)	2/50 (4%)	2/50 (4%)	3/49 (6%)
Adjusted rate	0.0%	4.9%	5.2%	8.4%
Terminal rate	0/23 (0%)	2/26 (8%)	1/18 (6%)	2/13 (15%)
First incidence (days)	—	729 (T)	590	687
Poly-3 test	P=0.075	P=0.233	P=0.219	P=0.093
All Organs: Malignant Mesothelioma				
Overall rate	0/50 (0%)	2/50 (4%)	2/50 (4%)	4/50 (8%)
Adjusted rate	0.0%	4.9%	5.2%	10.8%
Terminal rate	0/23 (0%)	2/26 (8%)	0/18 (0%)	1/13 (8%)
First incidence (days)	—	729 (T)	536	394
Poly-3 test	P=0.031	P=0.233	P=0.220	P=0.046

TABLE A2
Statistical Analysis of Primary Neoplasms in Male Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
All Organs: Mononuclear Cell Leukemia				
Overall rate	29/50 (58%)	22/50 (44%)	16/50 (32%)	24/50 (48%)
Adjusted rate	62.7%	49.8%	40.1%	59.9%
Terminal rate	13/23 (57%)	10/26 (39%)	8/18 (44%)	9/13 (69%)
First incidence (days)	501	540	578	527
Poly-3 test	P=0.429N	P=0.147N	P=0.024N	P=0.483N
All Organs: Benign Neoplasms				
Overall rate	48/50 (96%)	49/50 (98%)	47/50 (94%)	48/50 (96%)
Adjusted rate	97.6%	99.8%	97.8%	97.9%
Terminal rate	23/23 (100%)	26/26 (100%)	18/18 (100%)	13/13 (100%)
First incidence (days)	429	505	488	394
Poly-3 test	P=0.611N	P=0.444	P=0.794	P=0.760
All Organs: Malignant Neoplasms				
Overall rate	38/50 (76%)	33/50 (66%)	32/50 (64%)	32/50 (64%)
Adjusted rate	79.4%	72.5%	73.5%	74.5%
Terminal rate	18/23 (78%)	19/26 (73%)	15/18 (83%)	11/13 (85%)
First incidence (days)	429	316	488	394
Poly-3 test	P=0.366N	P=0.287N	P=0.328N	P=0.373N
All Organs: Benign or Malignant Neoplasms				
Overall rate	50/50 (100%)	50/50 (100%)	48/50 (96%)	49/50 (98%)
Adjusted rate	100.0%	100.0%	98.8%	99.4%
Terminal rate	23/23 (100%)	26/26 (100%)	18/18 (100%)	13/13 (100%)
First incidence (days)	429	316	488	394
Poly-3 test	P=0.592N	—[f]	P=0.826N	P=0.992N

(T) Terminal sacrifice

[a] Number of neoplasm-bearing animals/number of animals examined. Denominator is number of animals examined microscopically for adrenal medulla, lung, pancreatic islets, pituitary gland, preputial gland, testes, and thyroid gland; for other tissues, denominator is number of animals necropsied.

[b] Poly-3 estimated neoplasm incidence after adjustment for intercurrent mortality

[c] Observed incidence at terminal kill

[d] Beneath the chamber control incidence is the P value associated with the trend test. Beneath the exposed group incidence are the P values corresponding to pairwise comparisons between the chamber controls and that exposed group. The Poly-3 test accounts for the differential mortality in animals that do not reach terminal sacrifice. A negative trend or a lower incidence in an exposure group is indicated by **N**.

[e] Not applicable; no neoplasms in animal group

[f] Value of statistic cannot be computed.

TABLE A3a
Historical Incidence of Large Intestine (Colon or Rectum) Neoplasms in Control Male F344/N Rats[a]

Study (Study Start)	Incidence in Controls		
	Adenoma	Carcinoma	Adenoma or Carcinoma
Historical Incidence: Inhalation Studies			
1-Bromopropane (July 2003)	0/50	0/50	0/50
Cumene (June 2001)	0/50	0/50	0/50
Diethylamine (August 2003)	0/50	0/50	0/50
Methyl isobutyl ketone (May 2000)	0/50	0/50	0/50
α-Methylstyrene (August 2001)	0/50	0/50	0/50
Propargyl alcohol (October 2001)	0/49	0/49	0/49
Tetralin (June 2003)	0/50	0/50	0/50
Total (%)	0/349	0/349	0/349
Overall Historical Incidence: All Routes			
Total (%)	2/1,398 (0.1%)	1/1,398 (0.1%)	3/1,398 (0.2%)
Mean ± standard deviation	0.1% ± 0.5%	0.1% ± 0.4%	0.2% ± 0.6%
Range	0%-2%	0%-2%	0%-2%

[a] Data as of April 29, 2009

TABLE A3b
Historical Incidence of Skin Neoplasms in Control Male F344/N Rats[a]

Study (Study Start)	Incidence in Controls		
	Keratoacanthoma	**Basal Cell Adenoma**	**Basal Cell Carcinoma**
Historical Incidence: Inhalation Studies			
1-Bromopropane (July 2003)	0/50	0/50	0/50
Cumene (June 2001)	4/50	1/50	0/50
Diethylamine (August 2003)	0/50	0/50	1/50
Methyl isobutyl ketone (May 2000)	0/50	0/50	0/50
α-Methylstyrene (August 2001)	1/50	1/50	0/50
Propargyl alcohol (October 2001)	4/49	1/49	0/49
Tetralin (June 2003)	1/50	1/50	3/50
Total (%)	10/349 (2.9%)	4/349 (1.2%)	4/349 (1.2%)
Mean ± standard deviation	2.9% ± 3.7%	1.2% ± 1.1%	1.1% ± 2.3%
Range	0%-8%	0%-2%	0%-6%
Overall Historical Incidence: All Routes			
Total (%)	66/1,398 (4.7%)	15/1,398 (1.1%)	11/1,398 (0.8%)
Mean ± standard deviation	4.7% ± 4.2%	1.1% ± 1.4%	0.8% ± 1.5%
Range	0%-16%	0%-4%	0%-6%

	Squamous Cell Carcinoma	**Keratoacanthoma or Squamous Cell Carcinoma**	**Keratoacanthoma, Basal Cell Adenoma, Basal Cell Carcinoma, or Squamous Cell Carcinoma**
Historical Incidence: Inhalation Studies			
1-Bromopropane (July 2003)	1/50	1/50	1/50
Cumene (June 2001)	0/50	4/50	5/50
Diethylamine (August 2003)	0/50	0/50	1/50
Methyl isobutyl ketone (May 2000)	0/50	0/50	0/50
α-Methylstyrene (August 2001)	0/50	1/50	2/50
Propargyl alcohol (October 2001)	0/49	4/49	5/49
Tetralin (June 2003)	0/50	1/50	5/50
Total (%)	1/349 (0.3%)	11/349 (3.2%)	19/349 (5.4%)
Mean ± standard deviation	0.3% ± 0.8%	3.2% ± 3.5%	5.5% ± 4.5%
Range	0%-2%	0%-8%	0%-10%
Overall Historical Incidence: All Routes			
Total (%)	8/1,398 (0.6%)	74/1,398 (5.3%)	97/1,398 (6.9%)
Mean ± standard deviation	0.6% ± 0.9%	5.3% ± 4.1%	6.9% ± 4.9%
Range	0%-2%	0%-16%	0%-20%

[a] Data as of April 29, 2009

TABLE A3c
Historical Incidence of Malignant Mesothelioma in Control Male F344/N Rats[a]

Study (Study Start)	Incidence in Controls
Historical Incidence: Inhalation Studies	
1-Bromopropane (July 2003)	0/50
Cumene (June 2001)	3/50
Diethylamine (August 2003)	0/50
Methyl isobutyl ketone (May 2000)	1/50
α-Methylstyrene (August 2001)	0/50
Propargyl alcohol (October 2001)	1/49
Tetralin (June 2003)	0/50
Total (%)	5/349 (1.4%)
Mean ± standard deviation	1.4% ± 2.2%
Range	0%-6%
Overall Historical Incidence: All Routes	
Total (%)	35/1,398 (2.5%)
Mean ± standard deviation	2.5% ± 2.3%
Range	0%-6%

[a] Data as of April 29, 2009

TABLE A3d
Historical Incidence of Pancreatic Islet Neoplasms in Control Male F344/N Rats[a]

Study (Study Start)	Incidence in Controls		
	Adenoma	Carcinoma	Adenoma or Carcinoma
Historical Incidence: Inhalation Studies			
1-Bromopropane (July 2003)	0/50	3/50	3/50
Cumene (June 2001)	1/50	2/50	3/50
Diethylamine (August 2003)	6/50	1/50	7/50
Methyl isobutyl ketone (May 2000)	3/50	4/50	7/50
α-Methylstyrene (August 2001)	3/50	1/50	4/50
Propargyl alcohol (October 2001)	4/49	5/49	9/49
Tetralin (June 2003)	3/50	1/50	4/50
Total (%)	20/349 (5.7%)	17/349 (4.9%)	37/349 (10.6%)
Mean ± standard deviation	5.7% ± 3.9%	4.9.% ± 3.3%	10.6% ± 4.8%
Range	0%-12%	2%-10%	6%-18%
Overall Historical Incidence: All Routes			
Total (%)	90/1,394 (6.5%)	29/1,394 (2.1%)	119/1,394 (8.5%)
Mean ± standard deviation	6.5% ± 3.6%	2.1% ± 2.6%	8.6% ± 4.0%
Range	0%-14%	0%-10%	0%-18%

[a] Data as of April 29, 2009

TABLE A4
Summary of the Incidence of Nonneoplastic Lesions in Male Rats in the 2-Year Inhalation Study of 1-Bromopropane[a]

	Chamber Control	125 ppm	250 ppm	500 ppm
Disposition Summary				
Animals initially in study	50	50	50	50
Early deaths				
Moribund	24	20	28	35
Natural deaths	3	4	4	2
Survivors				
Terminal sacrifice	23	26	18	13
Animals examined microscopically	50	50	50	50
Alimentary System				
Esophagus	(50)	(50)	(50)	(50)
Intestine large, cecum	(47)	(47)	(47)	(48)
Edema	1 (2%)			
Inflammation, chronic active			1 (2%)	
Necrosis	1 (2%)			
Artery, inflammation	1 (2%)			
Intestine large, colon	(48)	(48)	(48)	(48)
Artery, inflammation	1 (2%)			
Intestine large, rectum	(48)	(48)	(48)	(49)
Necrosis			1 (2%)	1 (2%)
Intestine small, duodenum	(48)	(48)	(48)	(48)
Fibrosis	1 (2%)			
Intestine small, ileum	(47)	(47)	(46)	(48)
Inflammation, chronic active			1 (2%)	
Necrosis	1 (2%)		1 (2%)	
Intestine small, jejunum	(47)	(47)	(46)	(48)
Liver	(50)	(50)	(50)	(50)
Angiectasis		2 (4%)	1 (2%)	
Basophilic focus	9 (18%)	9 (18%)	8 (16%)	8 (16%)
Clear cell focus	16 (32%)	25 (50%)	17 (34%)	15 (30%)
Degeneration, cystic	2 (4%)	2 (4%)	1 (2%)	4 (8%)
Eosinophilic focus		1 (2%)	1 (2%)	1 (2%)
Hemorrhage				1 (2%)
Hepatodiaphragmatic nodule	4 (8%)	1 (2%)		2 (4%)
Mixed cell focus	2 (4%)	1 (2%)	3 (6%)	
Necrosis	2 (4%)	1 (2%)	1 (2%)	1 (2%)
Thrombosis	1 (2%)		1 (2%)	
Vacuolization cytoplasmic	31 (62%)	38 (76%)	32 (64%)	37 (74%)
Serosa, fibrosis	1 (2%)			
Mesentery	(13)	(13)	(8)	(19)
Necrosis	13 (100%)	13 (100%)	7 (88%)	17 (89%)
Oral mucosa	(3)	(0)	(2)	(4)
Foreign body	1 (33%)		1 (50%)	2 (50%)
Hyperplasia, squamous	1 (33%)		1 (50%)	
Inflammation, chronic active	1 (33%)			2 (50%)
Pancreas	(50)	(50)	(50)	(50)
Basophilic focus	1 (2%)			1 (2%)
Cyst				1 (2%)
Fibrosis				1 (2%)
Necrosis				1 (2%)
Acinus, atrophy	23 (46%)	13 (26%)	20 (40%)	23 (46%)
Artery, inflammation	1 (2%)			1 (2%)
Salivary glands	(50)	(50)	(50)	(50)
Hyperplasia	1 (2%)			
Inflammation, chronic active		1 (2%)	3 (6%)	2 (4%)

[a] Number of animals examined microscopically at the site and the number of animals with lesion

TABLE A4
Summary of the Incidence of Nonneoplastic Lesions in Male Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Alimentary System (continued)				
Stomach, forestomach	(50)	(50)	(50)	(50)
Erosion	1 (2%)			
Hyperplasia, squamous	1 (2%)	1 (2%)	1 (2%)	1 (2%)
Ulcer	7 (14%)	3 (6%)	5 (10%)	4 (8%)
Stomach, glandular	(50)	(50)	(50)	(50)
Erosion	1 (2%)		1 (2%)	
Ulcer	1 (2%)	2 (4%)	1 (2%)	
Artery, inflammation	1 (2%)			
Epithelium, hyperplasia				1 (2%)
Tongue	(0)	(0)	(1)	(1)
Hyperplasia, squamous			1 (100%)	1 (100%)
Cardiovascular System				
Heart	(50)	(50)	(50)	(50)
Cardiomyopathy	26 (52%)	23 (46%)	23 (46%)	15 (30%)
Atrium, thrombosis	4 (8%)			2 (4%)
Ventricle, thrombosis				1 (2%)
Endocrine System				
Adrenal cortex	(50)	(50)	(50)	(50)
Hyperplasia	27 (54%)	21 (42%)	22 (44%)	20 (40%)
Vacuolization cytoplasmic	9 (18%)	16 (32%)	9 (18%)	12 (24%)
Adrenal medulla	(50)	(50)	(50)	(50)
Hemorrhage	1 (2%)			
Hyperplasia	23 (46%)	18 (36%)	23 (46%)	19 (38%)
Islets, pancreatic	(50)	(50)	(50)	(50)
Hyperplasia	10 (20%)	7 (14%)	6 (12%)	9 (18%)
Parathyroid gland	(46)	(48)	(47)	(46)
Hyperplasia	2 (4%)			
Pituitary gland	(50)	(50)	(50)	(49)
Cyst	1 (2%)	2 (4%)	1 (2%)	
Hemorrhage	4 (8%)	2 (4%)	5 (10%)	
Pars distalis, hyperplasia	8 (16%)	5 (10%)	11 (22%)	7 (14%)
Thyroid gland	(50)	(50)	(50)	(49)
Ultimobranchial cyst				1 (2%)
C-cell, hyperplasia	32 (64%)	31 (62%)	29 (58%)	34 (69%)
Follicular cell, hyperplasia		1 (2%)		
General Body System				
Peritoneum	(0)	(0)	(0)	(2)
Inflammation, suppurative, chronic				1 (50%)
Genital System				
Coagulating gland	(3)	(0)	(1)	(2)
Hyperplasia	1 (33%)			
Inflammation, suppurative	2 (67%)			
Epididymis	(50)	(50)	(50)	(50)
Penis	(0)	(3)	(1)	(1)
Concretion			1 (100%)	
Hyperplasia, squamous		1 (33%)		

TABLE A4
Summary of the Incidence of Nonneoplastic Lesions in Male Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Genital System (continued)				
Preputial gland	(50)	(50)	(49)	(50)
Ectasia			1 (2%)	1 (2%)
Hyperplasia	3 (6%)			1 (2%)
Inflammation, suppurative, chronic		1 (2%)		
Inflammation, chronic active	30 (60%)	33 (66%)	31 (63%)	37 (74%)
Prostate	(50)	(50)	(50)	(50)
Hyperplasia	12 (24%)	13 (26%)	10 (20%)	17 (34%)
Inflammation, suppurative, chronic				1 (2%)
Inflammation, suppurative	38 (76%)	42 (84%)	40 (80%)	36 (72%)
Seminal vesicle	(50)	(49)	(49)	(50)
Congestion			1 (2%)	
Dilatation				1 (2%)
Hyperplasia				1 (2%)
Inflammation, suppurative	2 (4%)	1 (2%)		2 (4%)
Testes	(50)	(50)	(50)	(50)
Mineralization		1 (2%)	1 (2%)	
Germinal epithelium, atrophy	12 (24%)	8 (16%)	7 (14%)	6 (12%)
Interstitial cell, hyperplasia	1 (2%)		2 (4%)	1 (2%)
Hematopoietic System				
Bone marrow	(50)	(50)	(50)	(50)
Thrombosis	1 (2%)			
Lymph node	(5)	(5)	(5)	(4)
Deep cervical, angiectasis			1 (20%)	
Pancreatic, angiectasis		2 (40%)		
Pancreatic, hemorrhage				1 (25%)
Lymph node, bronchial	(3)	(6)	(5)	(8)
Angiectasis	1 (33%)	2 (33%)	2 (40%)	
Fibrosis		1 (17%)		
Hemorrhage	1 (33%)	1 (17%)	1 (20%)	1 (13%)
Hyperplasia, lymphoid				4 (50%)
Lymph node, mandibular	(3)	(1)	(0)	(6)
Angiectasis				1 (17%)
Lymph node, mediastinal	(30)	(25)	(23)	(32)
Angiectasis			1 (4%)	3 (9%)
Hyperplasia, lymphoid		1 (4%)	1 (4%)	2 (6%)
Lymph node, mesenteric	(50)	(50)	(50)	(50)
Angiectasis		1 (2%)	1 (2%)	2 (4%)
Hyperplasia, lymphoid				1 (2%)
Infiltration cellular, histiocyte			1 (2%)	
Artery, necrosis		1 (2%)		
Spleen	(50)	(50)	(49)	(50)
Accessory spleen	1 (2%)		1 (2%)	
Fibrosis	1 (2%)	2 (4%)	1 (2%)	
Hematopoietic cell proliferation	10 (20%)	17 (34%)	13 (27%)	6 (12%)
Hemorrhage, chronic	4 (8%)		1 (2%)	3 (6%)
Hyperplasia, lymphoid		1 (2%)		
Infarct, chronic		1 (2%)	2 (4%)	3 (6%)
Infiltration cellular, mononuclear cell			1 (2%)	
Thymus	(43)	(41)	(47)	(46)
Cyst			1 (2%)	

TABLE A4
Summary of the Incidence of Nonneoplastic Lesions in Male Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Integumentary System				
Mammary gland	(40)	(36)	(39)	(36)
Galactocele	1 (3%)	1 (3%)		
Hyperplasia		1 (3%)		
Epithelium, hyperplasia	1 (3%)	1 (3%)		1 (3%)
Skin	(50)	(50)	(50)	(50)
Cyst epithelial inclusion		1 (2%)	1 (2%)	
Foreign body		1 (2%)		
Hyperkeratosis	3 (6%)		2 (4%)	
Hyperplasia, squamous				1 (2%)
Inflammation, suppurative, chronic		1 (2%)	2 (4%)	10 (20%)
Inflammation, chronic active	1 (2%)	3 (6%)	3 (6%)	4 (8%)
Thrombosis		1 (2%)		
Musculoskeletal System				
Bone	(50)	(50)	(50)	(50)
Fibrosis				1 (2%)
Hyperostosis				1 (2%)
Inflammation, suppurative, chronic				2 (4%)
Skeletal muscle	(1)	(1)	(3)	(3)
Inflammation, suppurative, chronic				1 (33%)
Nervous System				
Brain	(50)	(50)	(50)	(50)
Hemorrhage	1 (2%)	1 (2%)	2 (4%)	2 (4%)
Spinal cord	(1)	(1)	(2)	(1)
Respiratory System				
Larynx	(50)	(50)	(50)	(50)
Foreign body	6 (12%)	1 (2%)	5 (10%)	1 (2%)
Inflammation, suppurative, chronic				1 (2%)
Inflammation, chronic active	21 (42%)	28 (56%)	31 (62%)	26 (52%)
Metaplasia, squamous	4 (8%)	6 (12%)	8 (16%)	5 (10%)
Respiratory epithelium, hyperplasia	1 (2%)		1 (2%)	1 (2%)
Lung	(50)	(50)	(50)	(50)
Foreign body			1 (2%)	
Hemorrhage	2 (4%)	5 (10%)	1 (2%)	1 (2%)
Inflammation, suppurative, chronic			1 (2%)	3 (6%)
Inflammation, chronic active	5 (10%)			3 (6%)
Metaplasia, osseous		1 (2%)		
Alveolar epithelium, hyperplasia	15 (30%)	11 (22%)	13 (26%)	11 (22%)
Alveolar epithelium, metaplasia, squamous			1 (2%)	
Alveolus, infiltration cellular, histiocyte	16 (32%)	14 (28%)	13 (26%)	11 (22%)
Artery, inflammation				1 (2%)

TABLE A4
Summary of the Incidence of Nonneoplastic Lesions in Male Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Respiratory System (continued)				
Nose	(50)	(48)	(48)	(50)
Degeneration, hyaline		1 (2%)		
Foreign body	11 (22%)	10 (21%)	15 (31%)	11 (22%)
Inflammation, suppurative, chronic		1 (2%)	2 (4%)	7 (14%)
Inflammation, chronic active	29 (58%)	33 (69%)	34 (71%)	35 (70%)
Epithelium, accumulation, hyaline droplet	44 (88%)	39 (81%)	36 (75%)	44 (88%)
Glands, hyperplasia	5 (10%)	14 (29%)	14 (29%)	15 (30%)
Olfactory epithelium, atrophy			1 (2%)	
Olfactory epithelium, metaplasia, respiratory	7 (14%)	10 (21%)	12 (25%)	12 (24%)
Respiratory epithelium, hyperplasia	14 (28%)	15 (31%)	20 (42%)	17 (34%)
Respiratory epithelium, metaplasia, squamous	1 (2%)	1 (2%)		2 (4%)
Trachea	(50)	(50)	(50)	(50)
Inflammation, chronic active	1 (2%)	1 (2%)	1 (2%)	4 (8%)
Epithelium, hyperplasia	1 (2%)			1 (2%)
Special Senses System				
Eye	(49)	(49)	(50)	(50)
Inflammation, chronic active			1 (2%)	2 (4%)
Lens, cataract	2 (4%)		3 (6%)	2 (4%)
Retina, degeneration		1 (2%)		
Harderian gland	(50)	(50)	(50)	(50)
Inflammation, suppurative, chronic				2 (4%)
Lacrimal gland	(1)	(0)	(0)	(0)
Inflammation, chronic active	1 (100%)			
Zymbal's gland	(0)	(1)	(2)	(2)
Urinary System				
Kidney	(50)	(50)	(50)	(49)
Cyst			1 (2%)	1 (2%)
Inflammation, suppurative	1 (2%)			
Nephropathy, chronic	44 (88%)	45 (90%)	39 (78%)	44 (90%)
Cortex, infarct	1 (2%)			
Cortex, renal tubule, casts granular			2 (4%)	
Pelvis, inflammation, chronic active				2 (4%)
Urinary bladder	(50)	(50)	(50)	(50)
Hemorrhage			1 (2%)	
Inflammation, chronic active	1 (2%)			1 (2%)
Transitional epithelium, hyperplasia				1 (2%)

APPENDIX B
SUMMARY OF LESIONS IN FEMALE RATS IN THE 2-YEAR INHALATION STUDY OF 1-BROMOPROPANE

TABLE B1
Summary of the Incidence of Neoplasms in Female Rats in the 2-Year Inhalation Study of 1-Bromopropane[a]

	Chamber Control	125 ppm	250 ppm	500 ppm
Disposition Summary				
Animals initially in study	50	50	50	50
Early deaths				
Accidental deaths				2
Moribund	13	17	17	23
Natural deaths	3		3	1
Survivors				
Terminal sacrifice	34	33	30	24
Animals examined microscopically	50	50	50	50
Alimentary System				
Intestine large, colon	(47)	(50)	(48)	(49)
Adenoma		1 (2%)	1 (2%)	1 (2%)
Carcinoid tumor malignant			1 (2%)	
Intestine large, rectum	(47)	(50)	(48)	(49)
Adenoma			1 (2%)	4 (8%)
Intestine small, ileum	(46)	(50)	(47)	(48)
Carcinoma		1 (2%)		
Intestine small, jejunum	(47)	(50)	(47)	(48)
Liver	(50)	(50)	(50)	(50)
Hepatocellular carcinoma	1 (2%)			
Mesentery	(13)	(26)	(17)	(23)
Sarcoma			1 (6%)	
Sarcoma, metastatic, stomach, forestomach				1 (4%)
Oral mucosa	(2)	(0)	(1)	(1)
Pancreas	(50)	(50)	(50)	(50)
Sarcoma, metastatic, mesentery			1 (2%)	
Salivary glands	(50)	(50)	(50)	(50)
Stomach, forestomach	(50)	(50)	(50)	(50)
Sarcoma				1 (2%)
Stomach, glandular	(50)	(50)	(50)	(50)
Sarcoma				1 (2%)
Tongue	(0)	(1)	(0)	(1)
Tooth	(0)	(0)	(1)	(0)
Cardiovascular System				
Heart	(50)	(50)	(50)	(50)
Alveolar/bronchiolar carcinoma, metastatic, lung			1 (2%)	
Endocrine System				
Adrenal cortex	(50)	(50)	(50)	(50)
Adenoma	3 (6%)	3 (6%)		
Adrenal medulla	(50)	(50)	(50)	(50)
Pheochromocytoma benign	2 (4%)	4 (8%)	1 (2%)	
Pheochromocytoma complex	1 (2%)			
Islets, pancreatic	(50)	(50)	(50)	(50)
Adenoma		2 (4%)	1 (2%)	
Carcinoma	1 (2%)	1 (2%)	1 (2%)	
Pituitary gland	(50)	(50)	(50)	(50)
Pars distalis, adenoma	35 (70%)	33 (66%)	28 (56%)	30 (60%)
Pars distalis, carcinoma		1 (2%)	1 (2%)	

TABLE B1
Summary of the Incidence of Neoplasms in Female Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Endocrine System (continued)				
Thyroid gland	(50)	(50)	(50)	(50)
C-cell, adenoma	3 (6%)	3 (6%)	3 (6%)	4 (8%)
C-cell, carcinoma	1 (2%)	2 (4%)	2 (4%)	2 (4%)
Follicular cell, adenoma		1 (2%)	1 (2%)	
Follicular cell, carcinoma				1 (2%)
General Body System				
None				
Genital System				
Clitoral gland	(49)	(50)	(49)	(49)
Adenoma	3 (6%)	9 (18%)	3 (6%)	3 (6%)
Carcinoma	1 (2%)	3 (6%)	2 (4%)	1 (2%)
Ovary	(50)	(50)	(50)	(50)
Cystadenoma			1 (2%)	
Granulosa cell tumor malignant			1 (2%)	
Granulosa-theca tumor malignant	2 (4%)			
Uterus	(50)	(50)	(50)	(50)
Adenoma				1 (2%)
Polyp stromal	8 (16%)	8 (16%)	8 (16%)	7 (14%)
Sarcoma stromal	2 (4%)		1 (2%)	1 (2%)
Vagina	(3)	(0)	(0)	(0)
Sarcoma	1 (33%)			
Hematopoietic System				
Bone marrow	(50)	(50)	(50)	(50)
Lymph node	(1)	(3)	(2)	(2)
Lymph node, bronchial	(1)	(4)	(5)	(7)
Squamous cell carcinoma, metastatic, lung		1 (25%)		
Lymph node, mediastinal	(34)	(30)	(31)	(28)
Carcinoma, metastatic, thyroid gland			1 (3%)	
Lymph node, mesenteric	(49)	(49)	(50)	(50)
Spleen	(50)	(50)	(50)	(50)
Thymus	(46)	(45)	(45)	(43)
Thymoma benign	1 (2%)			
Integumentary System				
Mammary gland	(50)	(50)	(50)	(50)
Carcinoma	4 (8%)	3 (6%)	3 (6%)	1 (2%)
Fibroadenoma	19 (38%)	14 (28%)	15 (30%)	15 (30%)
Fibroadenoma, multiple	6 (12%)	6 (12%)	6 (12%)	1 (2%)
Skin	(50)	(50)	(50)	(50)
Basal cell adenoma		1 (2%)		1 (2%)
Basal cell carcinoma				1 (2%)
Keratoacanthoma	1 (2%)		1 (2%)	1 (2%)
Squamous cell papilloma				1 (2%)
Pinna, neural crest tumor	1 (2%)	1 (2%)		
Sebaceous gland, adenoma		1 (2%)		
Subcutaneous tissue, fibroma	2 (4%)			
Subcutaneous tissue, lipoma			1 (2%)	

TABLE B1
Summary of the Incidence of Neoplasms in Female Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Musculoskeletal System				
Bone	(50)	(50)	(50)	(50)
Skeletal muscle	(0)	(0)	(1)	(4)
Alveolar/bronchiolar carcinoma, metastatic, lung			1 (100%)	
Nervous System				
Brain	(50)	(50)	(50)	(50)
Carcinoma, metastatic, pituitary gland		1 (2%)	1 (2%)	
Glioma malignant	1 (2%)			
Spinal cord	(0)	(1)	(0)	(3)
Respiratory System				
Larynx	(50)	(50)	(50)	(50)
Lung	(50)	(50)	(50)	(50)
Alveolar/bronchiolar adenoma	1 (2%)	4 (8%)		2 (4%)
Alveolar/bronchiolar adenoma, multiple			1 (2%)	
Alveolar/bronchiolar carcinoma	1 (2%)	1 (2%)	2 (4%)	
Squamous cell carcinoma		1 (2%)		
Nose	(50)	(50)	(49)	(50)
Fibroma				1 (2%)
Pleura	(0)	(0)	(1)	(0)
Alveolar/bronchiolar carcinoma, metastatic, lung			1 (100%)	
Trachea	(50)	(50)	(50)	(50)
Special Senses System				
Ear	(0)	(0)	(1)	(1)
Eye	(50)	(50)	(50)	(50)
Harderian gland	(50)	(50)	(50)	(50)
Zymbal's gland	(1)	(1)	(2)	(0)
Carcinoma	1 (100%)	1 (100%)	2 (100%)	
Urinary System				
Kidney	(49)	(50)	(50)	(50)
Sarcoma, metastatic, stomach, forestomach				1 (2%)
Renal tubule, carcinoma	1 (2%)			
Systemic Neoplasms				
Multiple organs[b]	(50)	(50)	(50)	(50)
Histiocytic sarcoma		1 (2%)		
Leukemia mononuclear	16 (32%)	13 (26%)	17 (34%)	15 (30%)
Lymphoma malignant		1 (2%)		

TABLE B1
Summary of the Incidence of Neoplasms in Female Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Neoplasm Summary				
Total animals with primary neoplasms[c]	49	48	46	43
Total primary neoplasms	119	120	106	96
Total animals with benign neoplasms	45	46	36	41
Total benign neoplasms	84	90	72	72
Total animals with malignant neoplasms	26	23	28	20
Total malignant neoplasms	34	29	34	24
Total animals with metastatic neoplasms		2	4	1
Total metastatic neoplasms		2	6	2
Total animals with uncertain neoplasms-benign or malignant	1	1		
Total uncertain neoplasms	1	1		

[a] Number of animals examined microscopically at the site and the number of animals with neoplasm
[b] Number of animals with any tissue examined microscopically
[c] Primary neoplasms: all neoplasms except metastatic neoplasms

TABLE B2
Statistical Analysis of Primary Neoplasms in Female Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Adrenal Cortex: Adenoma				
Overall rate[a]	3/50 (6%)	3/50 (6%)	0/50 (0%)	0/50 (0%)
Adjusted rate[b]	6.6%	6.9%	0.0%	0.0%
Terminal rate[c]	2/34 (6%)	3/33 (9%)	0/30 (0%)	0/24 (0%)
First incidence (days)	701	730 (T)	—[e]	—
Poly-3 test[d]	P=0.042N	P=0.645	P=0.131N	P=0.156N
Adrenal Medulla: Benign Pheochromocytoma				
Overall rate	2/50 (4%)	4/50 (8%)	1/50 (2%)	0/50 (0%)
Adjusted rate	4.4%	9.1%	2.3%	0.0%
Terminal rate	2/34 (6%)	3/33 (9%)	0/30 (0%)	0/24 (0%)
First incidence (days)	730 (T)	687	607	—
Poly-3 test	P=0.118N	P=0.323	P=0.519N	P=0.281N
Adrenal Medulla: Benign or Complex Pheochromocytoma				
Overall rate	3/50 (6%)	4/50 (8%)	1/50 (2%)	0/50 (0%)
Adjusted rate	6.6%	9.1%	2.3%	0.0%
Terminal rate	2/34 (6%)	3/33 (9%)	0/30 (0%)	0/24 (0%)
First incidence (days)	677	687	607	—
Poly-3 test	P=0.062N	P=0.481	P=0.327N	P=0.156N
Clitoral Gland: Adenoma				
Overall rate	3/49 (6%)	9/50 (18%)	3/49 (6%)	3/49 (6%)
Adjusted rate	6.7%	20.5%	7.2%	8.0%
Terminal rate	2/33 (6%)	8/33 (24%)	3/30 (10%)	1/24 (4%)
First incidence (days)	677	719	730 (T)	627
Poly-3 test	P=0.390N	P=0.054	P=0.631	P=0.583
Clitoral Gland: Carcinoma				
Overall rate	1/49 (2%)	3/50 (6%)	2/49 (4%)	1/49 (2%)
Adjusted rate	2.3%	6.8%	4.8%	2.7%
Terminal rate	1/33 (3%)	2/33 (6%)	2/30 (7%)	0/24 (0%)
First incidence (days)	730 (T)	632	730 (T)	724
Poly-3 test	P=0.556N	P=0.303	P=0.478	P=0.720
Clitoral Gland: Adenoma or Carcinoma				
Overall rate	4/49 (8%)	12/50 (24%)	4/49 (8%)	4/49 (8%)
Adjusted rate	9.0%	27.2%	9.6%	10.6%
Terminal rate	3/33 (9%)	10/33 (30%)	4/30 (13%)	1/24 (4%)
First incidence (days)	677	632	730 (T)	627
Poly-3 test	P=0.353N	P=0.023	P=0.606	P=0.549
Large Intestine (Colon and Rectum): Adenoma				
Overall rate	0/50 (0%)	1/50 (2%)	2/50 (4%)	5/50 (10%)
Adjusted rate	0.0%	2.3%	4.7%	13.3%
Terminal rate	0/34 (0%)	1/33 (3%)	1/30 (3%)	4/24 (17%)
First incidence (days)	—	730 (T)	607	719
Poly-3 test	P=0.004	P=0.493	P=0.225	P=0.018
Lung: Alveolar/bronchiolar Adenoma				
Overall rate	1/50 (2%)	4/50 (8%)	1/50 (2%)	2/50 (4%)
Adjusted rate	2.2%	9.1%	2.4%	5.3%
Terminal rate	1/34 (3%)	3/33 (9%)	1/30 (3%)	0/24 (0%)
First incidence (days)	730 (T)	635	730 (T)	627
Poly-3 test	P=0.509	P=0.170	P=0.746	P=0.440

TABLE B2
Statistical Analysis of Primary Neoplasms in Female Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Lung: Alveolar/bronchiolar Adenoma or Carcinoma				
Overall rate	2/50 (4%)	5/50 (10%)	3/50 (6%)	2/50 (4%)
Adjusted rate	4.4%	11.3%	6.9%	5.3%
Terminal rate	1/34 (3%)	3/33 (9%)	1/30 (3%)	0/24 (0%)
First incidence (days)	694	635	417	627
Poly-3 test	P=0.521N	P=0.205	P=0.482	P=0.628
Mammary Gland: Fibroadenoma				
Overall rate	25/50 (50%)	20/50 (40%)	21/50 (42%)	16/50 (32%)
Adjusted rate	53.8%	42.8%	48.4%	41.3%
Terminal rate	18/34 (53%)	12/33 (36%)	16/30 (53%)	11/24 (46%)
First incidence (days)	638	528	642	501
Poly-3 test	P=0.200N	P=0.194N	P=0.383N	P=0.172N
Mammary Gland: Carcinoma				
Overall rate	4/50 (8%)	3/50 (6%)	3/50 (6%)	1/50 (2%)
Adjusted rate	8.7%	6.8%	7.0%	2.7%
Terminal rate	0/34 (0%)	2/33 (6%)	2/30 (7%)	1/24 (4%)
First incidence (days)	612	575	684	730 (T)
Poly-3 test	P=0.196N	P=0.524N	P=0.544N	P=0.248N
Mammary Gland: Fibroadenoma or Carcinoma				
Overall rate	27/50 (54%)	20/50 (40%)	23/50 (46%)	16/50 (32%)
Adjusted rate	57.1%	42.8%	53.1%	41.3%
Terminal rate	18/34 (53%)	12/33 (36%)	18/30 (60%)	11/24 (46%)
First incidence (days)	612	528	642	501
Poly-3 test	P=0.149N	P=0.115N	P=0.428N	P=0.102N
Pancreatic Islets: Adenoma or Carcinoma				
Overall rate	1/50 (2%)	3/50 (6%)	2/50 (4%)	0/50 (0%)
Adjusted rate	2.2%	6.9%	4.7%	0.0%
Terminal rate	1/34 (3%)	3/33 (9%)	2/30 (7%)	0/24 (0%)
First incidence (days)	730 (T)	730 (T)	730 (T)	—
Poly-3 test	P=0.315N	P=0.293	P=0.477	P=0.537N
Pituitary Gland (Pars Distalis): Adenoma				
Overall rate	35/50 (70%)	33/50 (66%)	28/50 (56%)	30/50 (60%)
Adjusted rate	73.9%	68.2%	62.0%	71.3%
Terminal rate	25/34 (74%)	21/33 (64%)	20/30 (67%)	17/24 (71%)
First incidence (days)	633	433	534	450
Poly-3 test	P=0.418N	P=0.347N	P=0.149N	P=0.485N
Pituitary Gland (Pars Distalis): Adenoma or Carcinoma				
Overall rate	35/50 (70%)	34/50 (68%)	29/50 (58%)	30/50 (60%)
Adjusted rate	73.9%	70.3%	64.1%	71.3%
Terminal rate	25/34 (74%)	22/33 (67%)	20/30 (67%)	17/24 (71%)
First incidence (days)	633	433	534	450
Poly-3 test	P=0.404N	P=0.433N	P=0.206N	P=0.485N
Skin: Squamous Cell Papilloma, Keratoacanthoma, Basal Cell Adenoma, or Basal Cell Carcinoma				
Overall rate	1/50 (2%)	1/50 (2%)	1/50 (2%)	4/50 (8%)
Adjusted rate	2.2%	2.3%	2.4%	10.6%
Terminal rate	1/34 (3%)	1/33 (3%)	1/30 (3%)	4/24 (17%)
First incidence (days)	730 (T)	730 (T)	730 (T)	730 (T)
Poly-3 test	P=0.050	P=0.753	P=0.746	P=0.128

TABLE B2
Statistical Analysis of Primary Neoplasms in Female Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Thyroid Gland (C-Cell): Adenoma				
Overall rate	3/50 (6%)	3/50 (6%)	3/50 (6%)	4/50 (8%)
Adjusted rate	6.6%	6.8%	7.0%	10.6%
Terminal rate	3/34 (9%)	2/33 (6%)	2/30 (7%)	4/24 (17%)
First incidence (days)	730 (T)	659	694	730 (T)
Poly-3 test	P=0.310	P=0.649	P=0.634	P=0.400
Thyroid Gland (C-Cell): Adenoma or Carcinoma				
Overall rate	4/50 (8%)	5/50 (10%)	5/50 (10%)	6/50 (12%)
Adjusted rate	8.8%	11.4%	11.7%	15.8%
Terminal rate	4/34 (12%)	4/33 (12%)	4/30 (13%)	5/24 (21%)
First incidence (days)	730 (T)	659	694	626
Poly-3 test	P=0.212	P=0.482	P=0.462	P=0.264
Uterus: Stromal Polyp				
Overall rate	8/50 (16%)	8/50 (16%)	8/50 (16%)	7/50 (14%)
Adjusted rate	17.3%	18.0%	18.5%	17.9%
Terminal rate	6/34 (18%)	6/33 (18%)	5/30 (17%)	3/24 (13%)
First incidence (days)	438	577	607	590
Poly-3 test	P=0.519	P=0.573	P=0.551	P=0.584
Uterus: Stromal Polyp or Stromal Sarcoma				
Overall rate	10/50 (20%)	8/50 (16%)	8/50 (16%)	8/50 (16%)
Adjusted rate	21.3%	18.0%	18.5%	20.3%
Terminal rate	7/34 (21%)	6/33 (18%)	5/30 (17%)	3/24 (13%)
First incidence (days)	438	577	607	590
Poly-3 test	P=0.516N	P=0.449N	P=0.472N	P=0.558N
All Organs: Mononuclear Cell Leukemia				
Overall rate	16/50 (32%)	13/50 (26%)	17/50 (34%)	15/50 (30%)
Adjusted rate	34.3%	29.2%	37.6%	38.7%
Terminal rate	9/34 (27%)	10/33 (30%)	8/30 (27%)	10/24 (42%)
First incidence (days)	633	590	541	611
Poly-3 test	P=0.294	P=0.385N	P=0.453	P=0.423
All Organs: Benign Neoplasms				
Overall rate	45/50 (90%)	46/50 (92%)	36/50 (72%)	41/50 (82%)
Adjusted rate	93.3%	92.2%	79.3%	92.9%
Terminal rate	33/34 (97%)	30/33 (91%)	26/30 (87%)	23/24 (96%)
First incidence (days)	438	433	534	450
Poly-3 test	P=0.399N	P=0.577N	P=0.028N	P=0.654N
All Organs: Malignant Neoplasms				
Overall rate	26/50 (52%)	23/50 (46%)	28/50 (56%)	20/50 (40%)
Adjusted rate	53.3%	50.0%	58.9%	50.4%
Terminal rate	13/34 (38%)	15/33 (46%)	15/30 (50%)	12/24 (50%)
First incidence (days)	493	575	417	578
Poly-3 test	P=0.519N	P=0.452N	P=0.363	P=0.475N

TABLE B2
Statistical Analysis of Primary Neoplasms in Female Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
All Organs: Benign or Malignant Neoplasms				
Overall rate	49/50 (98%)	48/50 (96%)	46/50 (92%)	43/50 (86%)
Adjusted rate	98.0%	96.0%	92.7%	96.6%
Terminal rate	33/34 (97%)	31/33 (94%)	27/30 (90%)	24/24 (100%)
First incidence (days)	438	433	417	450
Poly-3 test	P=0.415N	P=0.500N	P=0.211N	P=0.613N

(T) Terminal sacrifice

[a] Number of neoplasm-bearing animals/number of animals examined. Denominator is number of animals examined microscopically for adrenal gland, clitoral gland, lung, pancreatic islets, pituitary gland, and thyroid gland; for other tissues, denominator is number of animals necropsied.

[b] Poly-3 estimated neoplasm incidence after adjustment for intercurrent mortality

[c] Observed incidence at terminal kill

[d] Beneath the chamber control incidence is the P value associated with the trend test. Beneath the exposed group incidence are the P values corresponding to pairwise comparisons between the chamber controls and that exposed group. The Poly-3 test accounts for the differential mortality in animals that do not reach terminal sacrifice. A negative trend or a lower incidence in an exposure group is indicated by **N**.

[e] Not applicable; no neoplasms in animal group

TABLE B3a
Historical Incidence of Large Intestine (Colon or Rectum) Neoplasms in Control Female F344/N Rats[a]

Study (Study Start)	Incidence in Controls		
	Adenoma	Carcinoma	Adenoma or Carcinoma
Historical Incidence: Inhalation Studies			
1-Bromopropane (July 2003)	0/50	0/50	0/50
Cumene (June 2001)	0/50	0/50	0/50
Diethylamine (August 2003)	0/50	0/50	0/50
Methyl isobutyl ketone (May 2000)	0/50	0/50	0/50
α-Methylstyrene (August 2001)	0/50	0/50	0/50
Propargyl alcohol (October 2001)	0/50	0/50	0/50
Tetralin (June 2003)	0/50	0/50	0/50
Total (%)	0/350	0/350	0/350
Overall Historical Incidence: All Routes			
Total (%)	3/1,350 (0.2%)	1/1,350 (0.1%)	4/1,350 (0.3%)
Mean ± standard deviation	0.2% ± 0.6%	0.1% ± 0.4%	0.3% ± 0.7%
Range	0%-2%	0%-2%	0%-2%

[a] Data as of April 29, 2009

TABLE B3b
Historical Incidence of Skin Neoplasms in Control Female F344/N Rats[a]

Study (Study Start)	Incidence in Controls		
	Squamous Cell Papilloma	Keratoacanthoma	Basal Cell Adenoma
Historical Incidence: Inhalation Studies			
1-Bromopropane (July 2003)	0/50	1/50	0/50
Cumene (June 2001)	0/50	0/50	0/50
Diethylamine (August 2003)	0/50	1/50	0/50
Methyl isobutyl ketone (May 2000)	0/50	0/50	0/50
α-Methylstyrene (August 2001)	0/50	0/50	0/50
Propargyl alcohol (October 2001)	0/50	0/50	0/50
Tetralin (June 2003)	0/50	0/50	0/50
Total (%)	0/350	2/350 (0.6%)	0/350
Mean ± standard deviation		0.6.% ± 1.0%	
Range		0%-2%	
Overall Historical Incidence: All Routes			
Total (%)	5/1,350 (0.4%)	8/1,350 (0.6%)	3/1,350 (0.2%)
Mean ± standard deviation	0.4% ± 0.8%	0.6% ± 1.2%	0.2% ± 0.6%
Range	0%-2%	0%-4%	0%-2%

	Basal Cell Carcinoma	Squamous Cell Papilloma, Keratoacanthoma, Basal Cell Adenoma, or Basal Cell Carcinoma
Historical Incidence: Inhalation Studies		
1-Bromopropane (July 2003)	0/50	1/50
Cumene (June 2001)	0/50	0/50
Diethylamine (August 2003)	0/50	1/50
Methyl isobutyl ketone (May 2000)	0/50	0/50
α-Methylstyrene (August 2001)	0/50	0/50
Propargyl alcohol (October 2001)	0/50	0/50
Tetralin (June 2003)	0/50	0/50
Total (%)	0/350	2/350 (0.6%)
Mean ± standard deviation		0.6.% ± 1.0%
Range		0%-2%
Overall Historical Incidence: All Routes		
Total (%)	1/1,350 (0.1%)	16/1,350 (1.2%)
Mean ± standard deviation	0.1% ± 0.4%	1.2% ± 1.8%
Range	0%-2%	0%-6%

[a] Data as of April 29, 2009

TABLE B4
Summary of the Incidence of Nonneoplastic Lesions in Female Rats in the 2-Year Inhalation Study of 1-Bromopropane[a]

	Chamber Control	125 ppm	250 ppm	500 ppm
Disposition Summary				
Animals initially in study	50	50	50	50
Early deaths				
Accidental deaths				2
Moribund	13	17	17	23
Natural deaths	3		3	1
Survivors				
Terminal sacrifice	34	33	30	24
Animals examined microscopically	50	50	50	50
Alimentary System				
Intestine large, colon	(47)	(50)	(48)	(49)
Intestine large, rectum	(47)	(50)	(48)	(49)
Inflammation, suppurative				1 (2%)
Necrosis				1 (2%)
Intestine small, ileum	(46)	(50)	(47)	(48)
Intestine small, jejunum	(47)	(50)	(47)	(48)
Liver	(50)	(50)	(50)	(50)
Angiectasis	3 (6%)	2 (4%)	2 (4%)	1 (2%)
Basophilic focus	40 (80%)	43 (86%)	42 (84%)	39 (78%)
Clear cell focus	30 (60%)	34 (68%)	35 (70%)	35 (70%)
Cyst	1 (2%)			
Degeneration, cystic				1 (2%)
Eosinophilic focus	1 (2%)		1 (2%)	
Hepatodiaphragmatic nodule	4 (8%)	9 (18%)	5 (10%)	5 (10%)
Mixed cell focus	2 (4%)	2 (4%)	3 (6%)	3 (6%)
Necrosis	1 (2%)		1 (2%)	
Vacuolization cytoplasmic	40 (80%)	41 (82%)	39 (78%)	47 (94%)
Bile duct, cyst				1 (2%)
Serosa, fibrosis	1 (2%)			
Mesentery	(13)	(26)	(17)	(23)
Necrosis	13 (100%)	26 (100%)	16 (94%)	22 (96%)
Oral mucosa	(2)	(0)	(1)	(1)
Foreign body			1 (100%)	1 (100%)
Hyperplasia, squamous	2 (100%)		1 (100%)	1 (100%)
Inflammation, chronic active				1 (100%)
Pancreas	(50)	(50)	(50)	(50)
Acinus, atrophy	9 (18%)	12 (24%)	7 (14%)	7 (14%)
Salivary glands	(50)	(50)	(50)	(50)
Hyperplasia			1 (2%)	
Inflammation, chronic active				1 (2%)
Necrosis			1 (2%)	
Stomach, forestomach	(50)	(50)	(50)	(50)
Hyperplasia, basal cell	1 (2%)			1 (2%)
Hyperplasia, squamous	4 (8%)		2 (4%)	2 (4%)
Ulcer	2 (4%)	3 (6%)	3 (6%)	6 (12%)
Stomach, glandular	(50)	(50)	(50)	(50)
Erosion		1 (2%)		
Ulcer	3 (6%)	1 (2%)	1 (2%)	1 (2%)
Tongue	(0)	(1)	(0)	(1)
Hyperplasia, squamous		1 (100%)		
Tooth	(0)	(0)	(1)	(0)
Inflammation, chronic active			1 (100%)	

[a] Number of animals examined microscopically at the site and the number of animals with lesion

TABLE B4
Summary of the Incidence of Nonneoplastic Lesions in Female Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Cardiovascular System				
Heart	(50)	(50)	(50)	(50)
Cardiomyopathy	35 (70%)	32 (64%)	34 (68%)	21 (42%)
Atrium, thrombosis	1 (2%)			
Endocrine System				
Adrenal cortex	(50)	(50)	(50)	(50)
Degeneration, cystic	1 (2%)		1 (2%)	
Hyperplasia	27 (54%)	30 (60%)	24 (48%)	26 (52%)
Necrosis				1 (2%)
Vacuolization cytoplasmic	12 (24%)	16 (32%)	16 (32%)	11 (22%)
Adrenal medulla	(50)	(50)	(50)	(50)
Hyperplasia	4 (8%)	9 (18%)	7 (14%)	6 (12%)
Metaplasia, osseous	1 (2%)			
Islets, pancreatic	(50)	(50)	(50)	(50)
Hyperplasia		3 (6%)	4 (8%)	4 (8%)
Metaplasia, hepatocyte			2 (4%)	1 (2%)
Pituitary gland	(50)	(50)	(50)	(50)
Cyst	2 (4%)	1 (2%)	2 (4%)	2 (4%)
Hemorrhage	2 (4%)	4 (8%)	5 (10%)	3 (6%)
Pars distalis, hyperplasia	7 (14%)	11 (22%)	7 (14%)	7 (14%)
Thyroid gland	(50)	(50)	(50)	(50)
Ultimobranchial cyst	1 (2%)	1 (2%)		
C-cell, hyperplasia	39 (78%)	36 (72%)	37 (74%)	37 (74%)
Follicular cell, hyperplasia		1 (2%)	2 (4%)	
General Body System				
None				
Genital System				
Clitoral gland	(49)	(50)	(49)	(49)
Hyperplasia	3 (6%)	5 (10%)	9 (18%)	4 (8%)
Inflammation, chronic active	22 (45%)	26 (52%)	26 (53%)	18 (37%)
Ovary	(50)	(50)	(50)	(50)
Cyst	3 (6%)	1 (2%)	7 (14%)	3 (6%)
Interstitial cell, hyperplasia			1 (2%)	
Uterus	(50)	(50)	(50)	(50)
Congestion			1 (2%)	
Cyst				1 (2%)
Decidual reaction				2 (4%)
Hemorrhage		2 (4%)		1 (2%)
Hydrometra		1 (2%)		
Inflammation, chronic active				1 (2%)
Endometrium, hyperplasia			1 (2%)	
Vagina	(3)	(0)	(0)	(0)
Muscularis, hypertrophy	2 (67%)			

TABLE B4
Summary of the Incidence of Nonneoplastic Lesions in Female Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Hematopoietic System				
Bone marrow	(50)	(50)	(50)	(50)
Hyperplasia, histiocytic	1 (2%)	1 (2%)	1 (2%)	1 (2%)
Lymph node	(1)	(3)	(2)	(2)
Inflammation, chronic active	1 (100%)			
Deep cervical, ectasia		1 (33%)		
Deep cervical, hyperplasia, lymphoid		1 (33%)		
Pancreatic, hemorrhage				1 (50%)
Lymph node, bronchial	(1)	(4)	(5)	(7)
Angiectasis	1 (100%)		2 (40%)	1 (14%)
Hemorrhage			1 (20%)	1 (14%)
Lymph node, mediastinal	(34)	(30)	(31)	(28)
Angiectasis			2 (6%)	2 (7%)
Hemorrhage				2 (7%)
Lymph node, mesenteric	(49)	(49)	(50)	(50)
Hemorrhage			1 (2%)	
Hyperplasia, lymphoid	1 (2%)			
Infiltration cellular, histiocyte	1 (2%)			1 (2%)
Spleen	(50)	(50)	(50)	(50)
Hematopoietic cell proliferation	33 (66%)	32 (64%)	29 (58%)	21 (42%)
Hemorrhage, chronic	1 (2%)		1 (2%)	2 (4%)
Infarct, chronic	1 (2%)	1 (2%)	1 (2%)	1 (2%)
Inflammation, granulomatous		2 (4%)		
Lymphoid follicle, atrophy		1 (2%)		
Thymus	(46)	(45)	(45)	(43)
Cyst	1 (2%)			
Integumentary System				
Mammary gland	(50)	(50)	(50)	(50)
Galactocele	1 (2%)		3 (6%)	1 (2%)
Hyperplasia	1 (2%)			
Inflammation, chronic active			1 (2%)	1 (2%)
Skin	(50)	(50)	(50)	(50)
Hyperkeratosis	1 (2%)	2 (4%)	2 (4%)	
Hyperplasia, squamous		1 (2%)		
Inflammation, suppurative, chronic		1 (2%)		1 (2%)
Inflammation, chronic active	1 (2%)	2 (4%)	1 (2%)	
Ulcer		1 (2%)	1 (2%)	
Musculoskeletal System				
Bone	(50)	(50)	(50)	(50)
Fracture				1 (2%)
Inflammation, suppurative, chronic				1 (2%)
Skeletal muscle	(0)	(0)	(1)	(4)
Nervous System				
Brain	(50)	(50)	(50)	(50)
Angiectasis		1 (2%)		
Hemorrhage			1 (2%)	
Spinal cord	(0)	(1)	(0)	(3)
Cyst epithelial inclusion				1 (33%)

TABLE B4
Summary of the Incidence of Nonneoplastic Lesions in Female Rats in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	125 ppm	250 ppm	500 ppm
Respiratory System				
Larynx	(50)	(50)	(50)	(50)
Foreign body	8 (16%)	6 (12%)	3 (6%)	4 (8%)
Inflammation, suppurative, chronic				3 (6%)
Inflammation, chronic active	18 (36%)	25 (50%)	30 (60%)	32 (64%)
Metaplasia, squamous	3 (6%)	2 (4%)	6 (12%)	21 (42%)
Necrosis				1 (2%)
Respiratory epithelium, hyperplasia		1 (2%)		
Lung	(50)	(50)	(50)	(50)
Hemorrhage	4 (8%)	6 (12%)	4 (8%)	4 (8%)
Inflammation, suppurative, chronic				4 (8%)
Inflammation, chronic active	6 (12%)	10 (20%)	12 (24%)	2 (4%)
Alveolar epithelium, hyperplasia	11 (22%)	10 (20%)	16 (32%)	8 (16%)
Alveolus, infiltration cellular, histiocyte	30 (60%)	30 (60%)	29 (58%)	26 (52%)
Nose	(50)	(50)	(49)	(50)
Foreign body	6 (12%)	6 (12%)	6 (12%)	9 (18%)
Inflammation, suppurative, chronic		1 (2%)	3 (6%)	7 (14%)
Inflammation, chronic active	24 (48%)	37 (74%)	37 (76%)	36 (72%)
Epithelium, accumulation, hyaline droplet	48 (96%)	48 (96%)	48 (98%)	47 (94%)
Glands, hyperplasia	6 (12%)	23 (46%)	28 (57%)	30 (60%)
Olfactory epithelium, hyperplasia	1 (2%)			
Olfactory epithelium, metaplasia, respiratory	3 (6%)	4 (8%)	6 (12%)	9 (18%)
Respiratory epithelium, hyperplasia	5 (10%)	13 (26%)	9 (18%)	18 (36%)
Respiratory epithelium, metaplasia, squamous	1 (2%)	1 (2%)	5 (10%)	1 (2%)
Pleura	(0)	(0)	(1)	(0)
Trachea	(50)	(50)	(50)	(50)
Inflammation, chronic active		4 (8%)	1 (2%)	6 (12%)
Epithelium, hyperplasia				4 (8%)
Epithelium, metaplasia, squamous				1 (2%)
Epithelium, necrosis				1 (2%)
Special Senses System				
Ear	(0)	(0)	(1)	(1)
Inflammation, suppurative, chronic				1 (100%)
Eye	(50)	(50)	(50)	(50)
Cornea, inflammation, chronic active			1 (2%)	
Lens, cataract	4 (8%)	3 (6%)	5 (10%)	2 (4%)
Harderian gland	(50)	(50)	(50)	(50)
Inflammation, suppurative, chronic				1 (2%)
Inflammation, chronic active			1 (2%)	1 (2%)
Zymbal's gland	(1)	(1)	(2)	(0)
Urinary System				
Kidney	(49)	(50)	(50)	(50)
Cyst	1 (2%)	1 (2%)		
Nephropathy, chronic	35 (71%)	35 (70%)	31 (62%)	29 (58%)
Cortex, infarct			1 (2%)	
Cortex, renal tubule, accumulation, hyaline droplet			1 (2%)	
Pelvis, inflammation, suppurative			1 (2%)	
Pelvis, inflammation, chronic active			1 (2%)	

APPENDIX C
SUMMARY OF LESIONS IN MALE MICE IN THE 2-YEAR INHALATION STUDY OF 1-BROMOPROPANE

TABLE C1
Summary of the Incidence of Neoplasms in Male Mice in the 2-Year Inhalation Study of 1-Bromopropane[a]

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Disposition Summary				
Animals initially in study	50	50	50	50
Early deaths				
Moribund	6	13	9	9
Natural deaths	7	4	9	5
Survivors				
Terminal sacrifice	37	33	32	36
Animals examined microscopically	50	50	50	50
Alimentary System				
Gallbladder	(40)	(40)	(34)	(38)
Intestine large, rectum	(45)	(48)	(43)	(48)
Intestine small, duodenum	(44)	(48)	(43)	(46)
Polyp adenomatous	1 (2%)	1 (2%)		
Intestine small, jejunum	(44)	(48)	(43)	(46)
Carcinoma	4 (9%)	1 (2%)	1 (2%)	
Liver	(50)	(50)	(50)	(50)
Hemangiosarcoma	1 (2%)	1 (2%)	1 (2%)	1 (2%)
Hepatoblastoma	1 (2%)	1 (2%)	1 (2%)	
Hepatocellular adenoma	16 (32%)	14 (28%)	19 (38%)	15 (30%)
Hepatocellular adenoma, multiple	14 (28%)	18 (36%)	14 (28%)	8 (16%)
Hepatocellular carcinoma	9 (18%)	8 (16%)	15 (30%)	7 (14%)
Hepatocellular carcinoma, multiple	6 (12%)	7 (14%)	4 (8%)	4 (8%)
Hepatocholangiocarcinoma	1 (2%)	1 (2%)	1 (2%)	1 (2%)
Hepatocholangiocarcinoma, multiple				1 (2%)
Capsule, carcinoma, metastatic, pancreas			1 (2%)	
Mesentery	(9)	(11)	(6)	(7)
Carcinoma, metastatic, pancreas	1 (11%)		1 (17%)	
Hepatocholangiocarcinoma, metastatic, liver				1 (14%)
Pancreas	(48)	(50)	(48)	(50)
Carcinoma	1 (2%)			
Hepatocholangiocarcinoma, metastatic, liver				1 (2%)
Acinar cell, carcinoma			1 (2%)	
Salivary glands	(49)	(50)	(49)	(50)
Stomach, forestomach	(47)	(50)	(48)	(50)
Squamous cell papilloma				2 (4%)
Stomach, glandular	(47)	(49)	(47)	(49)
Tooth	(8)	(8)	(9)	(11)
Cardiovascular System				
Blood vessel	(3)	(1)	(0)	(0)
Heart	(50)	(50)	(50)	(50)
Alveolar/bronchiolar carcinoma, metastatic, lung			1 (2%)	
Carcinoma, metastatic, pancreas	1 (2%)			
Hemangiosarcoma		1 (2%)		
Hepatocellular carcinoma, metastatic, liver		1 (2%)		
Hepatocholangiocarcinoma, metastatic, liver	1 (2%)		1 (2%)	2 (4%)

TABLE C1
Summary of the Incidence of Neoplasms in Male Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Endocrine System				
Adrenal cortex	(49)	(50)	(49)	(50)
Adenoma	1 (2%)		1 (2%)	
Carcinoma, metastatic, pancreas			1 (2%)	
Hepatocholangiocarcinoma, metastatic, liver				1 (2%)
Subcapsular, adenoma	3 (6%)	3 (6%)	1 (2%)	
Adrenal medulla	(50)	(49)	(49)	(50)
Pheochromocytoma benign	1 (2%)		1 (2%)	
Pheochromocytoma malignant				1 (2%)
Islets, pancreatic	(48)	(50)	(48)	(50)
Adenoma		1 (2%)		1 (2%)
Pituitary gland	(46)	(48)	(46)	(48)
Pars distalis, adenoma	1 (2%)	1 (2%)		
Thyroid gland	(49)	(50)	(48)	(50)
General Body System				
Peritoneum	(0)	(0)	(1)	(0)
Carcinoma, metastatic, pancreas			1 (100%)	
Genital System				
Coagulating gland	(1)	(0)	(0)	(0)
Epididymis	(50)	(50)	(50)	(50)
Preputial gland	(49)	(49)	(50)	(50)
Bilateral, squamous cell carcinoma		1 (2%)		
Prostate	(49)	(50)	(48)	(50)
Seminal vesicle	(48)	(50)	(49)	(50)
Adenoma	1 (2%)			
Hepatocholangiocarcinoma, metastatic, liver		1 (2%)		
Testes	(50)	(50)	(50)	(50)
Interstitial cell, adenoma				1 (2%)
Hematopoietic System				
Bone marrow	(48)	(48)	(48)	(50)
Hemangiosarcoma		1 (2%)		1 (2%)
Mast cell tumor benign			1 (2%)	
Lymph node	(1)	(1)	(3)	(0)
Alveolar/bronchiolar carcinoma, metastatic, lung			1 (33%)	
Lumbar, carcinoma, metastatic, uncertain primary site			1 (33%)	
Popliteal, carcinoma, metastatic, uncertain primary site			1 (33%)	
Lymph node, bronchial	(23)	(30)	(32)	(31)
Carcinoma, metastatic, pancreas			1 (3%)	
Hepatocellular carcinoma, metastatic, liver		1 (3%)	1 (3%)	
Hepatocholangiocarcinoma, metastatic, liver		1 (3%)	1 (3%)	2 (6%)
Lymph node, mandibular	(27)	(28)	(21)	(26)

TABLE C1
Summary of the Incidence of Neoplasms in Male Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Hematopoietic System (continued)				
Lymph node, mediastinal	(35)	(32)	(35)	(34)
Carcinoma, metastatic, pancreas	1 (3%)		1 (3%)	
Hepatocellular carcinoma, metastatic, liver		1 (3%)		
Hepatocholangiocarcinoma, metastatic, liver	1 (3%)	1 (3%)	1 (3%)	2 (6%)
Lymph node, mesenteric	(48)	(46)	(44)	(48)
Carcinoma, metastatic, pancreas	1 (2%)		1 (2%)	
Hepatocellular carcinoma, metastatic, liver		1 (2%)		
Hepatocholangiocarcinoma, metastatic, liver				1 (2%)
Spleen	(48)	(50)	(49)	(50)
Hemangiosarcoma	2 (4%)	1 (2%)		
Mast cell tumor malignant, metastatic, uncertain primary site		1 (2%)		
Thymus	(38)	(35)	(37)	(32)
Hepatocholangiocarcinoma, metastatic, liver	1 (3%)	1 (3%)		
Integumentary System				
Skin	(50)	(50)	(50)	(50)
Hepatocholangiocarcinoma, metastatic, liver			1 (2%)	
Subcutaneous tissue, fibrous histiocytoma				1 (2%)
Subcutaneous tissue, hemangiosarcoma		1 (2%)	1 (2%)	
Subcutaneous tissue, sarcoma			1 (2%)	
Musculoskeletal System				
Bone	(50)	(49)	(50)	(50)
Hemangiosarcoma				1 (2%)
Osteosarcoma	1 (2%)			
Skeletal muscle	(0)	(2)	(1)	(2)
Alveolar/bronchiolar carcinoma, metastatic, lung			1 (100%)	
Hemangiosarcoma				1 (50%)
Hepatocholangiocarcinoma, metastatic, liver		1 (50%)		1 (50%)
Nervous System				
Brain	(50)	(50)	(49)	(50)

TABLE C1
Summary of the Incidence of Neoplasms in Male Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Respiratory System				
Larynx	(48)	(50)	(48)	(50)
Lung	(50)	(50)	(49)	(49)
Alveolar/bronchiolar adenoma	6 (12%)	4 (8%)	4 (8%)	4 (8%)
Alveolar/bronchiolar adenoma, multiple		1 (2%)		1 (2%)
Alveolar/bronchiolar carcinoma	8 (16%)	7 (14%)	8 (16%)	9 (18%)
Alveolar/bronchiolar carcinoma, multiple			2 (4%)	1 (2%)
Carcinoma, metastatic, harderian gland			1 (2%)	
Carcinoma, metastatic, pancreas	1 (2%)		1 (2%)	
Hemangiosarcoma, metastatic, liver			1 (2%)	
Hemangiosarcoma, metastatic, spleen	1 (2%)			
Hepatocellular carcinoma, metastatic, liver	7 (14%)	10 (20%)	12 (24%)	4 (8%)
Hepatocholangiocarcinoma, metastatic, liver	1 (2%)	1 (2%)	1 (2%)	2 (4%)
Osteosarcoma, metastatic, uncertain primary site		1 (2%)		
Bronchiole, adenoma		1 (2%)		
Nose	(50)	(50)	(50)	(50)
Carcinoma, metastatic, harderian gland			1 (2%)	
Hepatocholangiocarcinoma, metastatic, liver			1 (2%)	
Pleura	(0)	(0)	(1)	(1)
Alveolar/bronchiolar carcinoma, metastatic, lung			1 (100%)	
Hepatocholangiocarcinoma, metastatic, liver				1 (100%)
Trachea	(49)	(50)	(47)	(50)
Special Senses System				
Eye	(45)	(47)	(44)	(47)
Harderian gland	(46)	(49)	(49)	(48)
Adenoma	6 (13%)	3 (6%)	2 (4%)	3 (6%)
Carcinoma	3 (7%)	1 (2%)	7 (14%)	5 (10%)
Hepatocholangiocarcinoma, metastatic, liver				1 (2%)
Bilateral, adenoma	1 (2%)			
Urinary System				
Kidney	(49)	(50)	(50)	(49)
Alveolar/bronchiolar carcinoma, metastatic, lung			1 (2%)	
Hepatocellular carcinoma, metastatic, liver		1 (2%)	1 (2%)	
Hepatocholangiocarcinoma, metastatic, liver	1 (2%)	1 (2%)	1 (2%)	2 (4%)
Urethra	(0)	(0)	(0)	(1)
Urinary bladder	(47)	(50)	(47)	(48)
Systemic Lesions				
Multiple organs[b]	(50)	(50)	(50)	(50)
Histiocytic sarcoma		2 (4%)	1 (2%)	
Lymphoma malignant	2 (4%)		3 (6%)	

TABLE C1
Summary of the Incidence of Neoplasms in Male Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Neoplasm Summary				
Total animals with primary neoplasms[c]	47	47	46	36
Total primary neoplasms	90	81	90	69
Total animals with benign neoplasms	36	35	35	29
Total benign neoplasms	51	47	43	35
Total animals with malignant neoplasms	30	29	34	24
Total malignant neoplasms	39	34	47	34
Total animals with metastatic neoplasms	9	12	17	6
Total metastatic neoplasms	18	24	39	21
Total animals with malignant neoplasms of uncertain primary site		2	1	

[a] Number of animals examined microscopically at the site and the number of animals with neoplasm
[b] Number of animals with any tissue examined microscopically
[c] Primary neoplasms: all neoplasms except metastatic neoplasms

TABLE C2
Statistical Analysis of Primary Neoplasms in Male Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Adrenal Cortex: Adenoma				
Overall rate[a]	4/49 (8%)	3/50 (6%)	2/49 (4%)	0/50 (0%)
Adjusted rate[b]	9.1%	6.9%	4.5%	0.0%
Terminal rate[c]	3/37 (8%)	2/33 (6%)	2/32 (6%)	0/36 (0%)
First incidence (days)	586	655	729 (T)	—[e]
Poly-3 test[d]	P=0.040N	P=0.502N	P=0.329N	P=0.066N
Harderian Gland: Adenoma				
Overall rate	7/50 (14%)	3/50 (6%)	2/50 (4%)	3/50 (6%)
Adjusted rate	15.9%	6.9%	4.4%	7.2%
Terminal rate	7/37 (19%)	1/33 (3%)	1/32 (3%)	3/36 (8%)
First incidence (days)	729 (T)	684	702	729 (T)
Poly-3 test	P=0.133N	P=0.161N	P=0.072N	P=0.179N
Harderian Gland: Carcinoma				
Overall rate	3/50 (6%)	1/50 (2%)	7/50 (14%)	5/50 (10%)
Adjusted rate	6.8%	2.3%	15.1%	12.0%
Terminal rate	3/37 (8%)	1/33 (3%)	3/32 (9%)	5/36 (14%)
First incidence (days)	729 (T)	729 (T)	604	729 (T)
Poly-3 test	P=0.123	P=0.312N	P=0.178	P=0.327
Harderian Gland: Adenoma or Carcinoma				
Overall rate	10/50 (20%)	4/50 (8%)	8/50 (16%)	8/50 (16%)
Adjusted rate	22.7%	9.2%	17.2%	19.1%
Terminal rate	10/37 (27%)	2/33 (6%)	3/32 (9%)	8/36 (22%)
First incidence (days)	729 (T)	684	604	729 (T)
Poly-3 test	P=0.547	P=0.074N	P=0.350N	P=0.446N
Small Intestine (Jejunum): Carcinoma				
Overall rate	4/50 (8%)	1/50 (2%)	1/50 (2%)	0/50 (0%)
Adjusted rate	9.1%	2.3%	2.2%	0.0%
Terminal rate	4/37 (11%)	0/33 (0%)	0/32 (0%)	0/36 (0%)
First incidence (days)	729 (T)	631	682	—
Poly-3 test	P=0.034N	P=0.181N	P=0.170N	P=0.067N
Liver: Hepatocellular Adenoma				
Overall rate	30/50 (60%)	32/50 (64%)	33/50 (66%)	23/50 (46%)
Adjusted rate	62.5%	67.6%	68.3%	53.3%
Terminal rate	23/37 (62%)	24/33 (73%)	21/32 (66%)	20/36 (56%)
First incidence (days)	328	424	537	417
Poly-3 test	P=0.186N	P=0.377	P=0.347	P=0.248N
Liver: Hepatocellular Carcinoma				
Overall rate	15/50 (30%)	15/50 (30%)	19/50 (38%)	11/50 (22%)
Adjusted rate	32.7%	33.1%	39.1%	25.4%
Terminal rate	9/37 (24%)	8/33 (24%)	8/32 (25%)	7/36 (19%)
First incidence (days)	539	520	537	470
Poly-3 test	P=0.289N	P=0.571	P=0.332	P=0.302N
Liver: Hepatocellular Adenoma or Carcinoma				
Overall rate	38/50 (76%)	38/50 (76%)	41/50 (82%)	28/50 (56%)
Adjusted rate	77.7%	78.1%	82.3%	63.1%
Terminal rate	28/37 (76%)	25/33 (76%)	24/32 (75%)	22/36 (61%)
First incidence (days)	328	424	537	417
Poly-3 test	P=0.066N	P=0.578	P=0.370	P=0.089N

TABLE C2
Statistical Analysis of Primary Neoplasms in Male Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Liver: Hepatocellular Carcinoma or Hepatoblastoma				
Overall rate	16/50 (32%)	16/50 (32%)	19/50 (38%)	11/50 (22%)
Adjusted rate	34.9%	35.3%	39.1%	25.4%
Terminal rate	10/37 (27%)	9/33 (27%)	8/32 (25%)	7/36 (19%)
First incidence (days)	539	520	537	470
Poly-3 test	P=0.208N	P=0.569	P=0.415	P=0.230N
Liver: Hepatocellular Adenoma, Hepatocellular Carcinoma, or Hepatoblastoma				
Overall rate	38/50 (76%)	39/50 (78%)	41/50 (82%)	28/50 (56%)
Adjusted rate	77.7%	80.1%	82.3%	63.1%
Terminal rate	28/37 (76%)	26/33 (79%)	24/32 (75%)	22/36 (61%)
First incidence (days)	328	424	537	417
Poly-3 test	P=0.055N	P=0.479	P=0.370	P=0.089N
Lung: Alveolar/bronchiolar Adenoma				
Overall rate	6/50 (12%)	5/50 (10%)	4/49 (8%)	5/49 (10%)
Adjusted rate	13.3%	11.5%	9.0%	11.9%
Terminal rate	4/37 (11%)	5/33 (15%)	4/32 (13%)	4/36 (11%)
First incidence (days)	544	729 (T)	729 (T)	360
Poly-3 test	P=0.476N	P=0.530N	P=0.380N	P=0.554N
Lung: Alveolar/bronchiolar Carcinoma				
Overall rate	8/50 (16%)	7/50 (14%)	10/49 (20%)	10/49 (20%)
Adjusted rate	17.8%	15.9%	22.0%	24.3%
Terminal rate	6/37 (16%)	6/33 (18%)	5/32 (16%)	9/36 (25%)
First incidence (days)	539	520	617	708
Poly-3 test	P=0.209	P=0.518N	P=0.407	P=0.317
Lung: Alveolar/bronchiolar Adenoma or Carcinoma				
Overall rate	13/50 (26%)	12/50 (24%)	14/49 (29%)	15/49 (31%)
Adjusted rate	28.3%	27.3%	30.8%	35.7%
Terminal rate	9/37 (24%)	11/33 (33%)	9/32 (28%)	13/36 (36%)
First incidence (days)	539	520	617	360
Poly-3 test	P=0.225	P=0.552N	P=0.484	P=0.301
All Organs: Hemangiosarcoma				
Overall rate	2/50 (4%)	4/50 (8%)	2/50 (4%)	2/50 (4%)
Adjusted rate	4.5%	9.2%	4.4%	4.8%
Terminal rate	2/37 (5%)	3/33 (9%)	1/32 (3%)	1/36 (3%)
First incidence (days)	729 (T)	661	718	708
Poly-3 test	P=0.477N	P=0.331	P=0.684N	P=0.675
All Organs: Malignant Lymphoma				
Overall rate	2/50 (4%)	0/50 (0%)	3/50 (6%)	0/50 (0%)
Adjusted rate	4.5%	0.0%	6.6%	0.0%
Terminal rate	1/37 (3%)	0/33 (0%)	3/32 (9%)	0/36 (0%)
First incidence (days)	578	—	729 (T)	—
Poly-3 test	P=0.313N	P=0.244N	P=0.506	P=0.252N
All Organs: Benign Neoplasms				
Overall rate	36/50 (72%)	35/50 (70%)	35/50 (70%)	29/50 (58%)
Adjusted rate	73.2%	73.5%	72.4%	65.9%
Terminal rate	26/37 (70%)	26/33 (79%)	23/32 (72%)	25/36 (69%)
First incidence (days)	328	424	537	360
Poly-3 test	P=0.232N	P=0.582	P=0.556N	P=0.290N

TABLE C2
Statistical Analysis of Primary Neoplasms in Male Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
All Organs: Malignant Neoplasms				
Overall rate	30/50 (60%)	30/50 (60%)	34/50 (68%)	24/50 (48%)
Adjusted rate	62.7%	63.5%	69.0%	54.8%
Terminal rate	20/37 (54%)	18/33 (55%)	19/32 (59%)	19/36 (53%)
First incidence (days)	328	520	537	470
Poly-3 test	P=0.270N	P=0.554	P=0.329	P=0.289N
All Organs: Benign or Malignant Neoplasms				
Overall rate	47/50 (94%)	47/50 (94%)	46/50 (92%)	36/50 (72%)
Adjusted rate	94.0%	94.3%	92.4%	78.6%
Terminal rate	34/37 (92%)	31/33 (94%)	29/32 (91%)	28/36 (78%)
First incidence (days)	328	424	537	360
Poly-3 test	P=0.005N	P=0.640	P=0.529N	P=0.024N

(T) Terminal sacrifice

[a] Number of neoplasm-bearing animals/number of animals examined. Denominator is number of animals examined microscopically for adrenal cortex, liver, and lung; for other tissues, denominator is number of animals necropsied.

[b] Poly-3 estimated neoplasm incidence after adjustment for intercurrent mortality

[c] Observed incidence at terminal kill

[d] Beneath the chamber control incidence is the P value associated with the trend test. Beneath the exposed group incidence are the P values corresponding to pairwise comparisons between the chamber controls and that exposed group. The Poly-3 test accounts for the differential mortality in animals that do not reach terminal sacrifice. A negative trend or a lower incidence in an exposure group is indicated by **N**.

[e] Not applicable; no neoplasms in animal group

TABLE C3
Summary of the Incidence of Nonneoplastic Lesions in Male Mice in the 2-Year Inhalation Study of 1-Bromopropane[a]

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Disposition Summary				
Animals initially in study	50	50	50	50
Early deaths				
Moribund	6	13	9	9
Natural deaths	7	4	9	5
Survivors				
Terminal sacrifice	37	33	32	36
Animals examined microscopically	50	50	50	50
Alimentary System				
Gallbladder	(40)	(40)	(34)	(38)
Degeneration, hyaline	2 (5%)			1 (3%)
Hemorrhage				1 (3%)
Hyperplasia	1 (3%)			
Infiltration cellular, polymorphonuclear			1 (3%)	
Intestine large, rectum	(45)	(48)	(43)	(48)
Inflammation, suppurative				1 (2%)
Ulcer				1 (2%)
Artery, inflammation, chronic active	1 (2%)			
Intestine small, duodenum	(44)	(48)	(43)	(46)
Inflammation, acute		1 (2%)		
Intestine small, jejunum	(44)	(48)	(43)	(46)
Liver	(50)	(50)	(50)	(50)
Angiectasis		3 (6%)		
Basophilic focus	1 (2%)	2 (4%)	1 (2%)	3 (6%)
Clear cell focus	9 (18%)	14 (28%)	16 (32%)	12 (24%)
Eosinophilic focus	20 (40%)	16 (32%)	8 (16%)	9 (18%)
Fatty change, focal	1 (2%)	3 (6%)		5 (10%)
Inflammation, chronic			1 (2%)	1 (2%)
Mineralization				1 (2%)
Mixed cell focus	2 (4%)	7 (14%)	1 (2%)	1 (2%)
Necrosis	5 (10%)	4 (8%)	3 (6%)	9 (18%)
Tension lipidosis	4 (8%)	3 (6%)	6 (12%)	2 (4%)
Vacuolization cytoplasmic				1 (2%)
Bile duct, hyperplasia				1 (2%)
Mesentery	(9)	(11)	(6)	(7)
Artery, inflammation				1 (14%)
Artery, mineralization	1 (11%)			
Fat, necrosis	7 (78%)	11 (100%)	5 (83%)	5 (71%)
Pancreas	(48)	(50)	(48)	(50)
Atrophy	1 (2%)			1 (2%)
Basophilic focus	1 (2%)		1 (2%)	
Cyst	1 (2%)			
Inflammation, chronic active	1 (2%)			
Salivary glands	(49)	(50)	(49)	(50)
Atrophy	1 (2%)		1 (2%)	
Stomach, forestomach	(47)	(50)	(48)	(50)
Hyperplasia, squamous	5 (11%)	7 (14%)	5 (10%)	3 (6%)
Inflammation	2 (4%)	2 (4%)	1 (2%)	
Ulcer	2 (4%)	4 (8%)	3 (6%)	2 (4%)
Artery, inflammation, chronic active	1 (2%)			

[a] Number of animals examined microscopically at the site and the number of animals with lesion

TABLE C3
Summary of the Incidence of Nonneoplastic Lesions in Male Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Alimentary System (continued)				
Stomach, glandular	(47)	(49)	(47)	(49)
Mineralization	2 (4%)			
Artery, inflammation, chronic active	1 (2%)			
Tooth	(8)	(8)	(9)	(11)
Dysplasia	7 (88%)	8 (100%)	9 (100%)	11 (100%)
Malformation	1 (13%)			
Cardiovascular System				
Blood vessel	(3)	(1)	(0)	(0)
Inflammation, chronic active	1 (33%)			
Thrombosis		1 (100%)		
Aorta, mineralization	2 (67%)			
Heart	(50)	(50)	(50)	(50)
Cardiomyopathy	12 (24%)	10 (20%)	14 (28%)	9 (18%)
Hemorrhage				1 (2%)
Inflammation, acute	1 (2%)			
Mineralization	2 (4%)			
Thrombosis	1 (2%)	2 (4%)	1 (2%)	
Artery, inflammation, chronic active	1 (2%)	1 (2%)		
Epicardium, inflammation, chronic		1 (2%)		
Endocrine System				
Adrenal cortex	(49)	(50)	(49)	(50)
Hyperplasia	16 (33%)	18 (36%)	14 (29%)	12 (24%)
Hypertrophy	23 (47%)	19 (38%)	22 (45%)	28 (56%)
Subcapsular, hyperplasia		1 (2%)		
Adrenal medulla	(50)	(49)	(49)	(50)
Hyperplasia	1 (2%)	2 (4%)	1 (2%)	2 (4%)
Islets, pancreatic	(48)	(50)	(48)	(50)
Hyperplasia	1 (2%)			
Pituitary gland	(46)	(48)	(46)	(48)
Cyst			1 (2%)	1 (2%)
Pars distalis, hyperplasia	2 (4%)	2 (4%)	1 (2%)	1 (2%)
Pars intermedia, hyperplasia			1 (2%)	
Thyroid gland	(49)	(50)	(48)	(50)
Follicular cell, hyperplasia	1 (2%)			
General Body System				
Peritoneum	(0)	(0)	(1)	(0)
Genital System				
Coagulating gland	(1)	(0)	(0)	(0)
Inflammation, suppurative	1 (100%)			
Epididymis	(50)	(50)	(50)	(50)
Granuloma sperm	1 (2%)	1 (2%)	1 (2%)	
Necrosis				2 (4%)
Preputial gland	(49)	(49)	(50)	(50)
Inflammation, suppurative			1 (2%)	
Inflammation, granulomatous			1 (2%)	1 (2%)

TABLE C3
Summary of the Incidence of Nonneoplastic Lesions in Male Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Genital System (continued)				
Prostate	(49)	(50)	(48)	(50)
Inflammation, suppurative	2 (4%)		2 (4%)	2 (4%)
Inflammation, chronic active			1 (2%)	1 (2%)
Artery, inflammation, chronic active	1 (2%)			
Seminal vesicle	(48)	(50)	(49)	(50)
Amyloid deposition	1 (2%)			
Inflammation, suppurative	1 (2%)			
Inflammation, chronic active			1 (2%)	
Testes	(50)	(50)	(50)	(50)
Atrophy	1 (2%)	1 (2%)	1 (2%)	3 (6%)
Hematopoietic System				
Bone marrow	(48)	(48)	(48)	(50)
Lymph node	(1)	(1)	(3)	(0)
Lymph node, bronchial	(23)	(30)	(32)	(31)
Infiltration cellular, mixed cell		1 (3%)		
Lymph node, mandibular	(27)	(28)	(21)	(26)
Lymph node, mediastinal	(35)	(32)	(35)	(34)
Infiltration cellular, mixed cell		1 (3%)		
Lymph node, mesenteric	(48)	(46)	(44)	(48)
Angiectasis	1 (2%)			
Spleen	(48)	(50)	(49)	(50)
Angiectasis		1 (2%)		
Atrophy	7 (15%)	8 (16%)	2 (4%)	3 (6%)
Congestion	4 (8%)	4 (8%)	1 (2%)	3 (6%)
Hematopoietic cell proliferation	20 (42%)	19 (38%)	14 (29%)	16 (32%)
Hyperplasia, lymphoid	5 (10%)	2 (4%)	5 (10%)	2 (4%)
Infarct	1 (2%)			
Necrosis	1 (2%)			
Necrosis, lymphoid	1 (2%)		1 (2%)	2 (4%)
Thymus	(38)	(35)	(37)	(32)
Atrophy	13 (34%)	12 (34%)	19 (51%)	12 (38%)
Cyst	5 (13%)	1 (3%)	7 (19%)	4 (13%)
Necrosis, lymphoid	1 (3%)		1 (3%)	3 (9%)
Medulla, hyperplasia, lymphoid	16 (42%)	9 (26%)	14 (38%)	6 (19%)
Integumentary System				
Skin	(50)	(50)	(50)	(50)
Cyst epithelial inclusion				1 (2%)
Inflammation, chronic active	5 (10%)	5 (10%)	3 (6%)	8 (16%)
Ulcer	1 (2%)			
Musculoskeletal System				
Bone	(50)	(49)	(50)	(50)
Hyperostosis		2 (4%)		1 (2%)
Necrosis				1 (2%)
Skeletal muscle	(0)	(2)	(1)	(2)
Nervous System				
Brain	(50)	(50)	(49)	(50)

TABLE C3
Summary of the Incidence of Nonneoplastic Lesions in Male Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Respiratory System				
Larynx	(48)	(50)	(48)	(50)
Vacuolization cytoplasmic		5 (10%)	10 (21%)	11 (22%)
Lung	(50)	(50)	(49)	(49)
Congestion, chronic	1 (2%)		1 (2%)	
Hemorrhage	2 (4%)	2 (4%)		1 (2%)
Inflammation, chronic active			2 (4%)	
Mineralization	1 (2%)			
Pigmentation		1 (2%)		
Thrombosis	1 (2%)			
Alveolar epithelium, hyperplasia	2 (4%)	2 (4%)	1 (2%)	5 (10%)
Alveolus, infiltration cellular, histiocyte		3 (6%)	1 (2%)	1 (2%)
Bronchiole, hyperplasia		3 (6%)		1 (2%)
Bronchiole, necrosis				1 (2%)
Bronchiole, regeneration	1 (2%)	44 (88%)	38 (78%)	47 (96%)
Bronchiole, vacuolization cytoplasmic		18 (36%)	19 (39%)	17 (35%)
Nose	(50)	(50)	(50)	(50)
Foreign body		1 (2%)		
Inflammation, suppurative	2 (4%)	3 (6%)	2 (4%)	3 (6%)
Polyp, inflammatory				3 (6%)
Glands, hyperplasia		1 (2%)		
Olfactory epithelium, atrophy	2 (4%)	4 (8%)	7 (14%)	4 (8%)
Olfactory epithelium, metaplasia, respiratory		7 (14%)	6 (12%)	3 (6%)
Respiratory epithelium, hyperplasia	16 (32%)	29 (58%)	23 (46%)	26 (52%)
Respiratory epithelium, vacuolization cytoplasmic		12 (24%)	19 (38%)	20 (40%)
Pleura	(0)	(0)	(1)	(1)
Trachea	(49)	(50)	(47)	(50)
Vacuolization cytoplasmic		15 (30%)	24 (51%)	24 (48%)
Special Senses System				
Eye	(45)	(47)	(44)	(47)
Cataract	1 (2%)	1 (2%)	1 (2%)	
Degeneration	1 (2%)	1 (2%)		1 (2%)
Cornea, inflammation, acute			1 (2%)	
Cornea, inflammation, chronic active	2 (4%)		1 (2%)	
Retina, atrophy	1 (2%)		1 (2%)	
Harderian gland	(46)	(49)	(49)	(48)
Hyperplasia	3 (7%)	4 (8%)	1 (2%)	4 (8%)
Urinary System				
Kidney	(49)	(50)	(50)	(49)
Amyloid deposition				1 (2%)
Cyst		1 (2%)		
Hydronephrosis	1 (2%)			1 (2%)
Infarct	1 (2%)	2 (4%)	3 (6%)	1 (2%)
Inflammation, suppurative	1 (2%)		2 (4%)	2 (4%)
Metaplasia, osseous	2 (4%)	1 (2%)		
Mineralization	2 (4%)			
Nephropathy	44 (90%)	46 (92%)	47 (94%)	41 (84%)
Papilla, necrosis			1 (2%)	
Renal tubule, hyperplasia	2 (4%)	1 (2%)	2 (4%)	
Renal tubule, necrosis				1 (2%)

TABLE C3
Summary of the Incidence of Nonneoplastic Lesions in Male Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Urinary System (continued)				
Urethra	(0)	(0)	(0)	(1)
Bulbourethral gland, angiectasis				1 (100%)
Urinary bladder	(47)	(50)	(47)	(48)
Inflammation, chronic active	1 (2%)		1 (2%)	1 (2%)
Mineralization	1 (2%)			

APPENDIX D
SUMMARY OF LESIONS IN FEMALE MICE IN THE 2-YEAR INHALATION STUDY OF 1-BROMOPROPANE

TABLE D1
Summary of the Incidence of Neoplasms in Female Mice in the 2-Year Inhalation Study of 1-Bromopropane[a]

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Disposition Summary				
Animals initially in study	50	50	50	50
Early deaths				
Moribund	10	7	8	8
Natural deaths	4	3	5	
Survivors				
Died last week of study	1			1
Terminal sacrifice	35	40	37	41
Animals examined microscopically	50	50	50	50
Alimentary System				
Esophagus	(50)	(50)	(50)	(50)
Squamous cell carcinoma	1 (2%)			
Gallbladder	(43)	(36)	(36)	(44)
Intestine large, cecum	(48)	(49)	(46)	(50)
Leiomyoma		1 (2%)	1 (2%)	
Intestine large, colon	(48)	(49)	(46)	(50)
Leiomyoma			1 (2%)	
Intestine large, rectum	(47)	(49)	(46)	(50)
Intestine small, duodenum	(46)	(49)	(46)	(50)
Hepatocholangiocarcinoma, metastatic, liver				1 (2%)
Intestine small, ileum	(48)	(49)	(46)	(50)
Hemangiosarcoma		1 (2%)		
Intestine small, jejunum	(46)	(49)	(46)	(50)
Hemangiosarcoma		1 (2%)		
Leiomyosarcoma				1 (2%)
Liver	(50)	(50)	(50)	(50)
Hemangiosarcoma				1 (2%)
Hepatoblastoma			1 (2%)	
Hepatocellular adenoma	13 (26%)	8 (16%)	13 (26%)	9 (18%)
Hepatocellular adenoma, multiple	6 (12%)	5 (10%)	2 (4%)	3 (6%)
Hepatocellular carcinoma	5 (10%)	4 (8%)	7 (14%)	
Hepatocholangiocarcinoma				1 (2%)
Mesentery	(11)	(9)	(11)	(13)
Hemangiosarcoma	1 (9%)			
Hepatocholangiocarcinoma, metastatic, liver				1 (8%)
Pancreas	(50)	(49)	(50)	(50)
Hepatocholangiocarcinoma, metastatic, liver				1 (2%)
Salivary glands	(50)	(50)	(50)	(50)
Stomach, forestomach	(50)	(50)	(49)	(50)
Hepatocholangiocarcinoma, metastatic, liver				1 (2%)
Squamous cell papilloma				2 (4%)
Stomach, glandular	(50)	(49)	(48)	(50)
Hepatocholangiocarcinoma, metastatic, liver				1 (2%)
Leiomyosarcoma, metastatic, intestine small, jejunum				1 (2%)
Tongue	(0)	(0)	(1)	(0)
Squamous cell carcinoma			1 (100%)	
Tooth	(0)	(1)	(0)	(0)

TABLE D1
Summary of the Incidence of Neoplasms in Female Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Cardiovascular System				
Heart	(50)	(50)	(50)	(50)
Hemangiosarcoma	1 (2%)			
Endocrine System				
Adrenal cortex	(50)	(50)	(50)	(50)
Hepatocholangiocarcinoma, metastatic, liver				1 (2%)
Adrenal medulla	(50)	(50)	(49)	(50)
Pheochromocytoma benign	3 (6%)		2 (4%)	
Pheochromocytoma malignant	1 (2%)			
Islets, pancreatic	(50)	(49)	(49)	(49)
Carcinoma		1 (2%)		
Pituitary gland	(46)	(49)	(47)	(48)
Pars distalis, adenoma	5 (11%)	7 (14%)	5 (11%)	9 (19%)
Pars distalis, carcinoma	1 (2%)			
Pars intermedia, adenoma			4 (9%)	
Thyroid gland	(49)	(50)	(46)	(50)
General Body System				
Peritoneum	(0)	(0)	(0)	(1)
Leiomyosarcoma, metastatic, intestine small, jejunum				1 (100%)
Genital System				
Ovary	(50)	(50)	(50)	(49)
Cystadenoma	3 (6%)	1 (2%)	1 (2%)	2 (4%)
Granulosa cell tumor benign			1 (2%)	
Granulosa cell tumor malignant		1 (2%)		
Luteoma	1 (2%)		1 (2%)	
Uterus	(50)	(50)	(50)	(50)
Carcinoma		1 (2%)		
Choriocarcinoma, metastatic, uncertain primary site		1 (2%)		
Leiomyosarcoma				1 (2%)
Polyp stromal		2 (4%)	2 (4%)	3 (6%)
Hematopoietic System				
Bone marrow	(50)	(50)	(49)	(50)
Lymph node	(6)	(5)	(3)	(8)
Lymph node, bronchial	(37)	(32)	(39)	(34)
Hepatocholangiocarcinoma, metastatic, liver				1 (3%)
Lymph node, mandibular	(45)	(30)	(30)	(37)
Lymph node, mediastinal	(45)	(39)	(41)	(37)
Hemangiosarcoma			1 (2%)	
Hepatocholangiocarcinoma, metastatic, liver				1 (3%)
Lymph node, mesenteric	(48)	(47)	(47)	(49)
Spleen	(50)	(49)	(50)	(50)
Hemangiosarcoma	1 (2%)	1 (2%)	1 (2%)	
Thymus	(49)	(45)	(45)	(45)
Hepatocholangiocarcinoma, metastatic, liver				1 (2%)
Thymoma benign			1 (2%)	

TABLE D1
Summary of the Incidence of Neoplasms in Female Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Integumentary System				
Mammary gland	(50)	(50)	(50)	(50)
Adenosquamous carcinoma			1 (2%)	2 (4%)
Carcinoma	2 (4%)			1 (2%)
Skin	(50)	(50)	(50)	(50)
Sebaceous gland, carcinoma			1 (2%)	
Subcutaneous tissue, fibrosarcoma			1 (2%)	
Subcutaneous tissue, fibrous histiocytoma	1 (2%)	2 (4%)		
Subcutaneous tissue, hemangioma		1 (2%)		
Subcutaneous tissue, hemangiosarcoma	1 (2%)			
Subcutaneous tissue, sarcoma	7 (14%)	2 (4%)	3 (6%)	1 (2%)
Musculoskeletal System				
Bone	(50)	(50)	(50)	(50)
Hemangiosarcoma	1 (2%)			
Skeletal muscle	(0)	(0)	(0)	(1)
Hepatocholangiocarcinoma, metastatic, liver				1 (100%)
Nervous System				
Brain	(50)	(50)	(50)	(50)
Carcinoma, metastatic, pituitary gland	1 (2%)			
Meningioma benign			1 (2%)	
Peripheral nerve	(1)	(1)	(1)	(1)
Spinal cord	(1)	(1)	(1)	(1)
Respiratory System				
Larynx	(50)	(50)	(50)	(50)
Lung	(50)	(50)	(50)	(50)
Alveolar/bronchiolar adenoma	1 (2%)	6 (12%)	4 (8%)	8 (16%)
Alveolar/bronchiolar adenoma, multiple				2 (4%)
Alveolar/bronchiolar carcinoma		5 (10%)	4 (8%)	3 (6%)
Alveolar/bronchiolar carcinoma, multiple		2 (4%)	1 (2%)	1 (2%)
Choriocarcinoma, metastatic, uterus		1 (2%)		
Hepatoblastoma, metastatic, liver			1 (2%)	
Hepatocellular carcinoma, metastatic, liver		1 (2%)		
Hepatocholangiocarcinoma, metastatic, liver				1 (2%)
Osteosarcoma, metastatic, uncertain primary site			1 (2%)	
Sarcoma, metastatic, skin	1 (2%)			
Bronchiole, adenoma		1 (2%)		1 (2%)
Nose	(50)	(50)	(50)	(50)
Pleura	(0)	(0)	(1)	(0)
Trachea	(50)	(49)	(50)	(50)
Special Senses System				
Eye	(47)	(49)	(45)	(50)
Harderian gland	(49)	(49)	(48)	(50)
Adenoma	3 (6%)	2 (4%)	4 (8%)	2 (4%)
Carcinoma	2 (4%)	1 (2%)		1 (2%)
Zymbal's gland	(0)	(1)	(0)	(0)
Carcinoma		1 (100%)		

TABLE D1
Summary of the Incidence of Neoplasms in Female Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Urinary System				
Kidney	(50)	(49)	(49)	(50)
Hepatoblastoma, metastatic, liver			1 (2%)	
Hepatocholangiocarcinoma, metastatic, liver				1 (2%)
Urinary bladder	(50)	(49)	(48)	(50)
Hemangiosarcoma	1 (2%)			
Systemic Lesions				
Multiple organs[b]	(50)	(50)	(50)	(50)
Histiocytic sarcoma	1 (2%)		2 (4%)	2 (4%)
Lymphoma malignant	17 (34%)	9 (18%)	15 (30%)	11 (22%)
Neoplasm Summary				
Total animals with primary neoplasms[c]	42	37	43	34
Total primary neoplasms	79	66	82	67
Total animals with benign neoplasms	22	24	30	28
Total benign neoplasms	35	34	43	41
Total animals with malignant neoplasms	33	27	32	22
Total malignant neoplasms	44	32	39	26
Total animals with metastatic neoplasms	2	2	2	2
Total metastatic neoplasms	2	3	3	14
Total animals with malignant neoplasms of uncertain primary site		1	1	

[a] Number of animals examined microscopically at the site and the number of animals with neoplasm
[b] Number of animals with any tissue examined microscopically
[c] Primary neoplasms: all neoplasms except metastatic neoplasms

TABLE D2
Statistical Analysis of Primary Neoplasms in Female Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Adrenal Medulla: Benign Pheochromocytoma				
Overall rate[a]	3/50 (6%)	0/50 (0%)	2/49 (4%)	0/50 (0%)
Adjusted rate[b]	6.7%	0.0%	4.5%	0.0%
Terminal rate[c]	2/36 (6%)	0/40 (0%)	1/37 (3%)	0/41 (0%)
First incidence (days)	708	—[e]	631	—
Poly-3 test[d]	P=0.123N	P=0.110N	P=0.503N	P=0.109N
Adrenal Medulla: Benign or Malignant Pheochromocytoma				
Overall rate	4/50 (8%)	0/50 (0%)	2/49 (4%)	0/50 (0%)
Adjusted rate	9.0%	0.0%	4.5%	0.0%
Terminal rate	3/36 (8%)	0/40 (0%)	1/37 (3%)	0/41 (0%)
First incidence (days)	708	—	631	—
Poly-3 test	P=0.060N	P=0.054N	P=0.340N	P=0.054N
Harderian Gland: Adenoma				
Overall rate	3/50 (6%)	2/50 (4%)	4/50 (8%)	2/50 (4%)
Adjusted rate	6.7%	4.3%	8.9%	4.2%
Terminal rate	3/36 (8%)	2/40 (5%)	4/37 (11%)	1/41 (2%)
First incidence (days)	731 (T)	731 (T)	731 (T)	663
Poly-3 test	P=0.456N	P=0.477N	P=0.505	P=0.472N
Harderian Gland: Adenoma or Carcinoma				
Overall rate	5/50 (10%)	2/50 (4%)	4/50 (8%)	3/50 (6%)
Adjusted rate	11.2%	4.3%	8.9%	6.3%
Terminal rate	5/36 (14%)	2/40 (5%)	4/37 (11%)	2/41 (5%)
First incidence (days)	731 (T)	731 (T)	731 (T)	663
Poly-3 test	P=0.372N	P=0.196N	P=0.494N	P=0.323N
Liver: Hepatocellular Adenoma				
Overall rate	19/50 (38%)	13/50 (26%)	15/50 (30%)	12/50 (24%)
Adjusted rate	40.8%	27.6%	32.3%	25.4%
Terminal rate	14/36 (39%)	12/40 (30%)	11/37 (30%)	11/41 (27%)
First incidence (days)	481	678	589	703
Poly-3 test	P=0.108N	P=0.127N	P=0.263N	P=0.083N
Liver: Hepatocellular Carcinoma				
Overall rate	5/50 (10%)	4/50 (8%)	7/50 (14%)	0/50 (0%)
Adjusted rate	11.1%	8.5%	15.4%	0.0%
Terminal rate	3/36 (8%)	3/40 (8%)	5/37 (14%)	0/41 (0%)
First incidence (days)	669	678	589	—
Poly-3 test	P=0.047N	P=0.471N	P=0.388	P=0.027N
Liver: Hepatocellular Adenoma or Carcinoma				
Overall rate	24/50 (48%)	15/50 (30%)	20/50 (40%)	12/50 (24%)
Adjusted rate	51.1%	31.8%	43.0%	25.4%
Terminal rate	17/36 (47%)	14/40 (35%)	15/37 (41%)	11/41 (27%)
First incidence (days)	481	678	589	703
Poly-3 test	P=0.017N	P=0.043N	P=0.282N	P=0.007N
Lung: Alveolar/bronchiolar Adenoma				
Overall rate	1/50 (2%)	6/50 (12%)	4/50 (8%)	10/50 (20%)
Adjusted rate	2.2%	12.8%	8.9%	20.8%
Terminal rate	1/36 (3%)	6/40 (15%)	4/37 (11%)	7/41 (17%)
First incidence (days)	731 (T)	731 (T)	731 (T)	649
Poly-3 test	P=0.007	P=0.064	P=0.181	P=0.006

TABLE D2
Statistical Analysis of Primary Neoplasms in Female Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Lung: Alveolar/bronchiolar Carcinoma				
Overall rate	0/50 (0%)	7/50 (14%)	5/50 (10%)	4/50 (8%)
Adjusted rate	0.0%	14.9%	11.1%	8.5%
Terminal rate	0/36 (0%)	6/40 (15%)	5/37 (14%)	4/41 (10%)
First incidence (days)	—	709	731 (T)	731 (T)
Poly-3 test	P=0.277	P=0.009	P=0.031	P=0.068
Lung: Alveolar/bronchiolar Adenoma or Carcinoma				
Overall rate	1/50 (2%)	9/50 (18%)	8/50 (16%)	14/50 (28%)
Adjusted rate	2.2%	19.2%	17.8%	29.2%
Terminal rate	1/36 (3%)	8/40 (20%)	8/37 (22%)	11/41 (27%)
First incidence (days)	731 (T)	709	731 (T)	649
Poly-3 test	P<0.001	P=0.010	P=0.016	P<0.001
Ovary: Cystadenoma				
Overall rate	3/50 (6%)	1/50 (2%)	1/50 (2%)	2/49 (4%)
Adjusted rate	6.7%	2.1%	2.2%	4.3%
Terminal rate	3/36 (8%)	1/40 (3%)	1/37 (3%)	1/40 (3%)
First incidence (days)	731 (T)	731 (T)	731 (T)	730
Poly-3 test	P=0.480N	P=0.287N	P=0.302N	P=0.484N
Pituitary Gland (Pars Distalis): Adenoma				
Overall rate	5/46 (11%)	7/49 (14%)	5/47 (11%)	9/48 (19%)
Adjusted rate	12.2%	15.2%	11.8%	19.1%
Terminal rate	5/34 (15%)	6/39 (15%)	5/35 (14%)	6/41 (15%)
First incidence (days)	731 (T)	723	731 (T)	470
Poly-3 test	P=0.237	P=0.461	P=0.608N	P=0.278
Pituitary Gland (Pars Distalis): Adenoma or Carcinoma				
Overall rate	6/46 (13%)	7/49 (14%)	5/47 (11%)	9/48 (19%)
Adjusted rate	14.6%	15.2%	11.8%	19.1%
Terminal rate	5/34 (15%)	6/39 (15%)	5/35 (14%)	6/41 (15%)
First incidence (days)	708	723	731 (T)	470
Poly-3 test	P=0.322	P=0.586	P=0.477N	P=0.391
Pituitary Gland (Pars Intermedia): Adenoma				
Overall rate	0/46 (0%)	0/49 (0%)	4/47 (9%)	0/48 (0%)
Adjusted rate	0.0%	0.0%	9.4%	0.0%
Terminal rate	0/34 (0%)	0/39 (0%)	4/35 (11%)	0/41 (0%)
First incidence (days)	—	—	731 (T)	—
Poly-3 test	P=0.551	—[f]	P=0.064	—
Skin (Subcutaneous Tissue): Sarcoma				
Overall rate	7/50 (14%)	2/50 (4%)	3/50 (6%)	1/50 (2%)
Adjusted rate	15.0%	4.2%	6.6%	2.1%
Terminal rate	1/36 (3%)	0/40 (0%)	2/37 (5%)	1/41 (2%)
First incidence (days)	562	505	610	731 (T)
Poly-3 test	P=0.029N	P=0.074N	P=0.169N	P=0.029N
Skin (Subcutaneous Tissue): Fibrous Histiocytoma, Fibrosarcoma, or Sarcoma				
Overall rate	8/50 (16%)	4/50 (8%)	4/50 (8%)	1/50 (2%)
Adjusted rate	17.1%	8.3%	8.7%	2.1%
Terminal rate	2/36 (6%)	1/40 (3%)	2/37 (5%)	1/41 (2%)
First incidence (days)	562	505	537	731 (T)
Poly-3 test	P=0.014N	P=0.164N	P=0.184N	P=0.015N

TABLE D2
Statistical Analysis of Primary Neoplasms in Female Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Uterus: Stromal Polyp				
Overall rate	0/50 (0%)	2/50 (4%)	2/50 (4%)	3/50 (6%)
Adjusted rate	0.0%	4.2%	4.4%	6.4%
Terminal rate	0/36 (0%)	1/40 (3%)	1/37 (3%)	2/41 (5%)
First incidence (days)	—	547	589	719
Poly-3 test	P=0.121	P=0.252	P=0.241	P=0.129
All Organs: Hemangiosarcoma				
Overall rate	4/50 (8%)	2/50 (4%)	2/50 (4%)	1/50 (2%)
Adjusted rate	9.0%	4.3%	4.4%	2.1%
Terminal rate	4/36 (11%)	1/40 (3%)	1/37 (3%)	1/41 (2%)
First incidence (days)	731 (T)	687	603	731 (T)
Poly-3 test	P=0.131N	P=0.312N	P=0.328N	P=0.162N
All Organs: Hemangioma or Hemangiosarcoma				
Overall rate	4/50 (8%)	3/50 (6%)	2/50 (4%)	1/50 (2%)
Adjusted rate	9.0%	6.4%	4.4%	2.1%
Terminal rate	4/36 (11%)	2/40 (5%)	1/37 (3%)	1/41 (2%)
First incidence (days)	731 (T)	687	603	731 (T)
Poly-3 test	P=0.107N	P=0.470N	P=0.328N	P=0.162N
All Organs: Malignant Lymphoma				
Overall rate	17/50 (34%)	9/50 (18%)	15/50 (30%)	11/50 (22%)
Adjusted rate	37.2%	19.0%	33.0%	23.2%
Terminal rate	13/36 (36%)	8/40 (20%)	13/37 (35%)	8/41 (20%)
First incidence (days)	591	583	631	649
Poly-3 test	P=0.198N	P=0.040N	P=0.421N	P=0.104N
All Organs: Benign Neoplasms				
Overall rate	22/50 (44%)	24/50 (48%)	30/50 (60%)	28/50 (56%)
Adjusted rate	47.2%	50.3%	64.0%	57.3%
Terminal rate	17/36 (47%)	21/40 (53%)	25/37 (68%)	21/41 (51%)
First incidence (days)	481	547	589	470
Poly-3 test	P=0.150	P=0.463	P=0.072	P=0.217
All Organs: Malignant Neoplasms				
Overall rate	33/50 (66%)	28/50 (56%)	33/50 (66%)	22/50 (44%)
Adjusted rate	68.4%	57.0%	67.9%	45.2%
Terminal rate	22/36 (61%)	20/40 (50%)	23/37 (62%)	15/41 (37%)
First incidence (days)	540	505	345	568
Poly-3 test	P=0.021N	P=0.168N	P=0.566N	P=0.016N

TABLE D2
Statistical Analysis of Primary Neoplasms in Female Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
All Organs: Benign or Malignant Neoplasms				
Overall rate	42/50 (84%)	37/50 (74%)	44/50 (88%)	34/50 (68%)
Adjusted rate	85.0%	74.4%	89.6%	68.8%
Terminal rate	29/36 (81%)	28/40 (70%)	33/37 (89%)	26/41 (63%)
First incidence (days)	481	505	345	470
Poly-3 test	P=0.064N	P=0.142N	P=0.349	P=0.044N

(T) Terminal sacrifice

[a] Number of neoplasm-bearing animals/number of animals examined. Denominator is number of animals examined microscopically for adrenal medulla, liver, lung, ovary, and pituitary gland; for other tissues, denominator is number of animals necropsied.

[b] Poly-3 estimated neoplasm incidence after adjustment for intercurrent mortality

[c] Observed incidence at terminal kill

[d] Beneath the chamber control incidence is the P value associated with the trend test. Beneath the exposed group incidence are the P values corresponding to pairwise comparisons between the chamber controls and that exposed group. The Poly-3 test accounts for the differential mortality in animals that do not reach terminal sacrifice. A negative trend or a lower incidence in an exposure group is indicated by **N**.

[e] Not applicable; no neoplasms in animal group

[f] Value of statistic cannot be computed.

TABLE D3
Historical Incidence of Alveolar/bronchiolar Neoplasms in Control Female B6C3F1 Mice[a]

Study (Study Start)	Incidence in Controls		
	Adenoma	Carcinoma	Adenoma or Carcinoma
Historical Incidence: Inhalation Studies			
1-Bromopropane (July 2003)	1/50	0/50	1/50
Cumene (June 2001)	1/50	3/50	4/50
Diethylamine (August 2003)	2/50	3/50	5/50
Methyl isobutyl ketone (June 2000)	4/50	0/50	4/50
α-Methylstyrene (July 2001)	1/50	1/50	2/50
Propargyl alcohol (September 2001)	3/50	2/50	5/50
Tetralin (June 2003)	6/50	0/50	6/50
Total (%)	18/350 (5.1%)	9/350 (2.6%)	27/350 (7.7%)
Mean ± standard deviation	5.1% ± 3.8%	2.6% ± 2.8%	7.7% ± 3.6%
Range	2%-12%	0%-6%	2%-12%
Overall Historical Incidence: All Routes			
Total (%)	73/1,496 (4.9%)	59/1,496 (3.9%)	128/1,496 (8.6%)
Mean ± standard deviation	4.9% ± 3.4%	3.9% ± 3.4%	8.6% ± 3.7%
Range	0%-12%	0%-12%	2%-18%

[a] Data as of April 29, 2009

TABLE D4
Summary of the Incidence of Nonneoplastic Lesions in Female Mice in the 2-Year Inhalation Study of 1-Bromopropane[a]

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Disposition Summary				
Animals initially in study	50	50	50	50
Early deaths				
Moribund	10	7	8	8
Natural deaths	4	3	5	
Survivors				
Died last week of study	1			1
Terminal sacrifice	35	40	37	41
Animals examined microscopically	50	50	50	50
Alimentary System				
Esophagus	(50)	(50)	(50)	(50)
Gallbladder	(43)	(36)	(36)	(44)
Inflammation, chronic active		1 (3%)		
Intestine large, cecum	(48)	(49)	(46)	(50)
Hemorrhage	1 (2%)			
Inflammation, suppurative		1 (2%)		
Intestine large, colon	(48)	(49)	(46)	(50)
Necrosis	1 (2%)			
Intestine large, rectum	(47)	(49)	(46)	(50)
Artery, inflammation, chronic active				1 (2%)
Intestine small, duodenum	(46)	(49)	(46)	(50)
Intestine small, ileum	(48)	(49)	(46)	(50)
Epithelium, hyperplasia			1 (2%)	
Intestine small, jejunum	(46)	(49)	(46)	(50)
Epithelium, hyperplasia			1 (2%)	
Liver	(50)	(50)	(50)	(50)
Angiectasis		1 (2%)	1 (2%)	2 (4%)
Basophilic focus	2 (4%)	3 (6%)	2 (4%)	3 (6%)
Clear cell focus	2 (4%)		1 (2%)	2 (4%)
Eosinophilic focus	6 (12%)	7 (14%)	7 (14%)	5 (10%)
Fatty change	1 (2%)	1 (2%)		2 (4%)
Fatty change, focal	2 (4%)			
Hematopoietic cell proliferation			1 (2%)	
Infarct		1 (2%)		
Inflammation, acute	1 (2%)			
Inflammation, chronic		1 (2%)	1 (2%)	
Malformation, lobular			1 (2%)	
Mineralization			1 (2%)	
Mixed cell focus	2 (4%)	3 (6%)	3 (6%)	
Necrosis	1 (2%)	1 (2%)	2 (4%)	1 (2%)
Tension lipidosis	3 (6%)	10 (20%)	4 (8%)	1 (2%)
Thrombosis			1 (2%)	
Vacuolization cytoplasmic		1 (2%)		
Bile duct, hyperplasia		1 (2%)		
Hepatocyte, mitotic alteration	1 (2%)			
Mesentery	(11)	(9)	(11)	(13)
Artery, inflammation	1 (9%)			
Fat, hemorrhage		1 (11%)	1 (9%)	
Fat, necrosis	9 (82%)	9 (100%)	9 (82%)	12 (92%)
Pancreas	(50)	(49)	(50)	(50)
Atrophy	7 (14%)	4 (8%)	5 (10%)	2 (4%)
Basophilic focus	1 (2%)	2 (4%)		
Inflammation, chronic active		1 (2%)		1 (2%)
Salivary glands	(50)	(50)	(50)	(50)

[a] Number of animals examined microscopically at the site and the number of animals with lesion

TABLE D4
Summary of the Incidence of Nonneoplastic Lesions in Female Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Alimentary System (continued)				
Stomach, forestomach	(50)	(50)	(49)	(50)
Hyperplasia, squamous	3 (6%)	2 (4%)	2 (4%)	1 (2%)
Inflammation	1 (2%)			
Ulcer	2 (4%)	2 (4%)		2 (4%)
Artery, inflammation, chronic active				1 (2%)
Stomach, glandular	(50)	(49)	(48)	(50)
Infiltration cellular, mixed cell		1 (2%)		
Infiltration cellular, mononuclear cell	1 (2%)			
Mineralization			1 (2%)	1 (2%)
Necrosis		1 (2%)	1 (2%)	
Artery, inflammation, chronic active				1 (2%)
Tongue	(0)	(0)	(1)	(0)
Tooth	(0)	(1)	(0)	(0)
Dysplasia		1 (100%)		
Cardiovascular System				
Heart	(50)	(50)	(50)	(50)
Cardiomyopathy	6 (12%)	12 (24%)	8 (16%)	8 (16%)
Inflammation, suppurative			1 (2%)	
Mineralization	3 (6%)		2 (4%)	
Necrosis		1 (2%)		
Thrombosis			2 (4%)	
Artery, inflammation, chronic active	1 (2%)	1 (2%)		
Endocrine System				
Adrenal cortex	(50)	(50)	(50)	(50)
Hyperplasia	3 (6%)	10 (20%)	4 (8%)	6 (12%)
Hypertrophy	4 (8%)	3 (6%)	1 (2%)	9 (18%)
Inflammation, suppurative				1 (2%)
Subcapsular, hyperplasia		1 (2%)	1 (2%)	
Adrenal medulla	(50)	(50)	(49)	(50)
Hyperplasia		5 (10%)	1 (2%)	4 (8%)
Islets, pancreatic	(50)	(49)	(49)	(49)
Hyperplasia		2 (4%)	1 (2%)	
Pituitary gland	(46)	(49)	(47)	(48)
Pars distalis, angiectasis	1 (2%)	1 (2%)	2 (4%)	
Pars distalis, hyperplasia	15 (33%)	14 (29%)	11 (23%)	15 (31%)
Pars intermedia, hyperplasia		1 (2%)	2 (4%)	
Pars intermedia, hypertrophy	1 (2%)	1 (2%)		
Thyroid gland	(49)	(50)	(46)	(50)
Follicular cell, hyperplasia	1 (2%)	1 (2%)	1 (2%)	1 (2%)
General Body System				
Peritoneum	(0)	(0)	(0)	(1)

TABLE D4
Summary of the Incidence of Nonneoplastic Lesions in Female Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Genital System				
Ovary	(50)	(50)	(50)	(49)
Angiectasis	1 (2%)	2 (4%)		2 (4%)
Congestion, chronic		1 (2%)		
Cyst	12 (24%)	8 (16%)	12 (24%)	2 (4%)
Hyperplasia, tubular				1 (2%)
Thrombosis	2 (4%)	2 (4%)		1 (2%)
Uterus	(50)	(50)	(50)	(50)
Angiectasis	2 (4%)	3 (6%)	6 (12%)	3 (6%)
Fibrosis			1 (2%)	
Inflammation, suppurative			1 (2%)	
Inflammation, acute	1 (2%)			
Thrombosis	1 (2%)	1 (2%)	2 (4%)	1 (2%)
Endometrium, hyperplasia, cystic	39 (78%)	43 (86%)	43 (86%)	41 (82%)
Hematopoietic System				
Bone marrow	(50)	(50)	(49)	(50)
Angiectasis	2 (4%)			
Atrophy				1 (2%)
Lymph node	(6)	(5)	(3)	(8)
Iliac, angiectasis	2 (33%)	1 (20%)		
Iliac, hyperplasia, lymphoid	1 (17%)			
Lumbar, angiectasis		1 (20%)	1 (33%)	1 (13%)
Renal, angiectasis				1 (13%)
Renal, hyperplasia, lymphoid	1 (17%)	1 (20%)		
Lymph node, bronchial	(37)	(32)	(39)	(34)
Lymph node, mandibular	(45)	(30)	(30)	(37)
Hyperplasia, lymphoid		1 (3%)		
Lymph node, mediastinal	(45)	(39)	(41)	(37)
Hyperplasia, lymphoid	1 (2%)			
Lymph node, mesenteric	(48)	(47)	(47)	(49)
Angiectasis	2 (4%)	2 (4%)		1 (2%)
Hyperplasia, lymphoid			1 (2%)	
Spleen	(50)	(49)	(50)	(50)
Atrophy	4 (8%)	1 (2%)	2 (4%)	5 (10%)
Congestion			1 (2%)	2 (4%)
Hematopoietic cell proliferation	13 (26%)	10 (20%)	13 (26%)	16 (32%)
Hemorrhage				1 (2%)
Hyperplasia, histiocytic	2 (4%)			
Hyperplasia, lymphoid	7 (14%)	7 (14%)	5 (10%)	3 (6%)
Hyperplasia, plasma cell				1 (2%)
Necrosis, lymphoid		1 (2%)	1 (2%)	
Pigmentation, hemosiderin	1 (2%)	2 (4%)		
Thymus	(49)	(45)	(45)	(45)
Atrophy	14 (29%)	13 (29%)	17 (38%)	16 (36%)
Cyst	5 (10%)	7 (16%)	6 (13%)	9 (20%)
Necrosis, lymphoid		2 (4%)	4 (9%)	
Medulla, hyperplasia, lymphoid	25 (51%)	29 (64%)	30 (67%)	26 (58%)
Integumentary System				
Mammary gland	(50)	(50)	(50)	(50)
Hyperplasia	1 (2%)	1 (2%)	1 (2%)	
Skin	(50)	(50)	(50)	(50)
Cyst	1 (2%)			
Inflammation, chronic active		4 (8%)	2 (4%)	1 (2%)

TABLE D4
Summary of the Incidence of Nonneoplastic Lesions in Female Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Musculoskeletal System				
Bone	(50)	(50)	(50)	(50)
Cyst		1 (2%)		
Skeletal muscle	(0)	(0)	(0)	(1)
Nervous System				
Brain	(50)	(50)	(50)	(50)
Necrosis		1 (2%)	1 (2%)	
Artery, inflammation	1 (2%)			
Peripheral nerve	(1)	(1)	(1)	(1)
Degeneration		1 (100%)		
Spinal cord	(1)	(1)	(1)	(1)
Inflammation, chronic active		1 (100%)		
Respiratory System				
Larynx	(50)	(50)	(50)	(50)
Vacuolization cytoplasmic		3 (6%)	2 (4%)	2 (4%)
Artery, inflammation, chronic active				1 (2%)
Lung	(50)	(50)	(50)	(50)
Inflammation, chronic active		1 (2%)		
Alveolar epithelium, hyperplasia	1 (2%)	1 (2%)		3 (6%)
Alveolus, infiltration cellular, histiocyte			4 (8%)	1 (2%)
Bronchiole, hyperplasia	1 (2%)			1 (2%)
Bronchiole, necrosis			1 (2%)	
Bronchiole, regeneration		45 (90%)	43 (86%)	49 (98%)
Bronchiole, vacuolization cytoplasmic		3 (6%)	4 (8%)	3 (6%)
Nose	(50)	(50)	(50)	(50)
Inflammation, suppurative				1 (2%)
Olfactory epithelium, atrophy				6 (12%)
Olfactory epithelium, metaplasia, respiratory		4 (8%)	5 (10%)	14 (28%)
Respiratory epithelium, hyperplasia	11 (22%)	25 (50%)	28 (56%)	27 (54%)
Respiratory epithelium, vacuolization cytoplasmic		3 (6%)	5 (10%)	8 (16%)
Pleura	(0)	(0)	(1)	(0)
Trachea	(50)	(49)	(50)	(50)
Vacuolization cytoplasmic		8 (16%)	7 (14%)	4 (8%)
Special Senses System				
Eye	(47)	(49)	(45)	(50)
Cataract	3 (6%)	1 (2%)		2 (4%)
Cornea, hyperplasia, squamous	1 (2%)			
Cornea, inflammation, chronic active	1 (2%)		2 (4%)	1 (2%)
Cornea, mineralization	1 (2%)		2 (4%)	1 (2%)
Harderian gland	(49)	(49)	(48)	(50)
Hyperplasia	3 (6%)	1 (2%)	2 (4%)	6 (12%)
Inflammation, chronic active				1 (2%)
Zymbal's gland	(0)	(1)	(0)	(0)

TABLE D4
Summary of the Incidence of Nonneoplastic Lesions in Female Mice in the 2-Year Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm
Urinary System				
Kidney	(50)	(49)	(49)	(50)
Amyloid deposition				1 (2%)
Cyst			1 (2%)	
Inflammation, suppurative			1 (2%)	
Inflammation, chronic active		1 (2%)		
Metaplasia, osseous	3 (6%)	4 (8%)		2 (4%)
Nephropathy	33 (66%)	37 (76%)	40 (82%)	39 (78%)
Renal tubule, hyperplasia			1 (2%)	
Transitional epithelium, hyperplasia		1 (2%)		
Urinary bladder	(50)	(49)	(48)	(50)
Inflammation, chronic active			1 (2%)	
Artery, inflammation, chronic active				2 (4%)

APPENDIX E
GENETIC TOXICOLOGY

GENETIC TOXICOLOGY

BACTERIAL MUTAGENICITY TEST PROTOCOL

Testing at BioReliance Corporation was performed as reported by Zeiger *et al.* (1992). 1-Bromopropane was sent to the laboratory as a coded aliquot. In the tests conducted at BioReliance Corporation, it was incubated with the *Salmonella typhimurium* tester strains TA97, TA98, TA100, and TA1535 either in buffer or S9 mix (metabolic activation enzymes and cofactors from Aroclor 1254-induced male Sprague-Dawley rat or Syrian hamster liver) for 20 minutes at 37° C. Top agar supplemented with L-histidine and d-biotin was added, and the contents of the tubes were mixed and poured onto the surfaces of minimal glucose agar plates. Histidine-independent mutant colonies arising on these plates were counted following incubation for 2 days at 37° C. The slightly modified protocol used at SITEK Research Laboratories tested the same lot (1581313004) of 1-bromopropane used in the 2-year studies, used only 10% rat liver S9 for exogenous metabolic activation, and employed *Escherichia coli* strain WP2 *uvrA*/pKM101 as a bacterial tester strain in addition to *S. typhimurium* strains TA98 and TA100. Incubation of bacterial strains with 1-bromopropane and subsequent plating were carried out as described above for the traditional protocol.

Each trial consisted of triplicate plates of concurrent positive and negative controls and five doses of 1-bromopropane. The high dose was limited by toxicity in some trials and by the limit dose of 10,000 µg/plate in those trials where only slight toxicity was observed. All trials were repeated, and those that were conducted with S9 activation enzymes were repeated using the same or higher concentrations of S9.

In this assay, a positive response is defined as a reproducible, dose-related increase in histidine-independent (revertant) colonies in any one strain/activation combination. An equivocal response is defined as an increase in revertants that is not dose related, is not reproducible, or is not of sufficient magnitude to support a determination of mutagenicity. A negative response is obtained when no increase in revertant colonies is observed following chemical treatment. There is no minimum percentage or fold increase required for a chemical to be judged positive or weakly positive, although positive calls are typically reserved for increases in mutant colonies that are at least twofold over background.

MOUSE PERIPHERAL BLOOD MICRONUCLEUS TEST PROTOCOL

A detailed discussion of this assay is presented by MacGregor *et al.* (1990). At the end of the 3-month toxicity study, peripheral blood samples were obtained from male and female mice. Smears were immediately prepared and fixed in absolute methanol. The methanol-fixed slides were stained with acridine orange and coded. Slides were scanned to determine the frequency of micronuclei in 2,000 normochromatic erythrocytes (NCEs; immature erythrocytes) per animal. In addition, the percentage of polychromatic erythrocytes (PCEs or reticulocytes) in a population of 1,000 erythrocytes was determined as a measure of bone marrow toxicity.

The results were tabulated as the mean of the pooled results from all animals within an exposure group plus or minus the standard error of the mean. The frequency of micronucleated cells among NCEs was analyzed by a statistical software package that tested for increasing trend over exposure groups with a one-tailed Cochran-Armitage trend test, followed by pairwise comparisons between each exposed group and the chamber control group. In the presence of excess binomial variation, as detected by a binomial dispersion test, the binomial variance of the Cochran-Armitage test was adjusted upward in proportion to the excess variation. In the micronucleus test, an individual trial is considered positive if the trend test P value is less than or equal to 0.025 or if the P value for any single exposed group is less than or equal to 0.025 divided by the number of exposed groups. A final call of positive for micronucleus induction is preferably based on reproducibly positive trials (as noted above). Results of the 3-month study were accepted without repeat tests, because additional test data could not be obtained. Ultimately, the final call is determined by the scientific staff after considering the results of statistical analyses, the reproducibility of any effects observed, and the magnitudes of those effects.

EVALUATION PROTOCOL

These are the basic guidelines for arriving at an overall assay result for assays performed by the National Toxicology Program. Statistical as well as biological factors are considered. For an individual assay, the statistical procedures for data analysis have been described in the preceding protocols. There have been instances, however, in which multiple aliquots of a chemical were tested in the same assay, and different results were obtained among aliquots and/or among laboratories. Results from more than one aliquot or from more than one laboratory are not simply combined into an overall result. Rather, all the data are critically evaluated, particularly with regard to pertinent protocol variations, in determining the weight of evidence for an overall conclusion of chemical activity in an assay. In addition to multiple aliquots, the *in vitro* assays have another variable that must be considered in arriving at an overall test result. *In vitro* assays are conducted with and without exogenous metabolic activation. Results obtained in the absence of activation are not combined with results obtained in the presence of activation; each testing condition is evaluated separately. The summary table in the Abstract of this Technical Report presents a result that represents a scientific judgment of the overall evidence for activity of the chemical in an assay.

RESULTS

1-Bromopropane (doses up to 10,000 µg/plate) was not mutagenic in either of two independent bacterial mutagenicity assays, with or without exogenous metabolic activation (Table E1). Bacterial strains tested included *S. typhimurium* strains TA97, TA98, TA100, and TA1535, as well as *E. coli* strain WP2 *uvrA*/pKM101. In addition to the negative results in the two bacterial tests, no increases in the frequencies of micronucleated NCEs were seen in peripheral blood of male or female B6C3F1 mice exposed for 3 months to 62.5 to 500 ppm 1-bromopropane via inhalation (Table E2). The percentage of reticulocytes (PCEs) in the peripheral blood of male and female mice was unaltered by 1-bromopropane exposure, suggesting a lack of chemical-associated bone marrow toxicity.

TABLE E1
Mutagenicity of 1-Bromopropane in Bacterial Tester Strains[a]

Strain	Dose (µg/Plate)	Revertants/Plate[b] −S9 Trial 1	−S9 Trial 2	+hamster S9 10%	+hamster S9 30%	+rat S9 10%	+rat S9 30%
Study performed at BioReliance Corporation							
TA100	0	150 ± 4	121 ± 16	104 ± 2	103 ± 9	124 ± 2	140 ± 8
	33		115 ± 6	118 ± 6		121 ± 7	
	100	144 ± 10	124 ± 4	109 ± 12	104 ± 8	123 ± 2	133 ± 5
	333	139 ± 7	127 ± 7	92 ± 8	108 ± 6	128 ± 10	117 ± 12
	1,000	149 ± 7	128 ± 6	111 ± 7	98 ± 6	144 ± 5	103 ± 7
	3,333	117 ± 2[c]	107 ± 11[c]	98 ± 19[c]	118 ± 6[c]	107 ± 7[c]	123 ± 7[c]
	10,000	Toxic			Toxic		Toxic
Trial summary		Negative	Negative	Negative	Negative	Negative	Negative
Positive control[d]		728 ± 27	279 ± 14	661 ± 22	615 ± 14	523 ± 32	636 ± 27
TA1535	0	16 ± 2	10 ± 2	10 ± 2	12 ± 2	12 ± 2	14 ± 3
	33		9 ± 1	9 ± 2		9 ± 2	
	100	19 ± 2	11 ± 0	12 ± 2	18 ± 2	8 ± 1[e]	14 ± 2
	333	14 ± 3	10 ± 1	11 ± 2	15 ± 4	8 ± 1	18 ± 3
	1,000	14 ± 1	10 ± 1	12 ± 1	17 ± 1	13 ± 2	15 ± 0
	3,333	6 ± 3[f]	9 ± 1[c]	8 ± 1[c]	10 ± 1[c]	8 ± 2[c]	8 ± 0[c]
	10,000	2 ± 1[c]			6 ± 1[c]		4 ± 1[c]
Trial summary		Negative	Negative	Negative	Negative	Negative	Negative
Positive control		226 ± 10	199 ± 19	60 ± 6	137 ± 9	127 ± 13	80 ± 8
TA97	0	134 ± 27	105 ± 12	156 ± 6	125 ± 9	161 ± 7	168 ± 13
	33		130 ± 12	163 ± 13		156 ± 9	
	100	108 ± 9	121 ± 3	114 ± 17	141 ± 11	141 ± 9	170 ± 9
	333	98 ± 5	132 ± 6	124 ± 4	133 ± 12	149 ± 14	168 ± 5
	1,000	117 ± 4	125 ± 6	143 ± 7	166 ± 3[e]	147 ± 8	180 ± 13
	3,333	86 ± 3[c]	117 ± 7[c]	113 ± 9[c]	91 ± 16[c]	122 ± 10[c]	121 ± 13[c]
	10,000	40 ± 22[f]			53 ± 3[c]		70 ± 15[c]
Trial summary		Negative	Negative	Negative	Negative	Negative	Negative
Positive control		354 ± 17	211 ± 4	988 ± 30	1,063 ± 138	1,084 ± 32	418 ± 16
TA98	0	23 ± 3	14 ± 1	16 ± 1	29 ± 1	25 ± 2	31 ± 4
	33		18 ± 2	21 ± 3		24 ± 3	
	100	23 ± 2	15 ± 2	24 ± 3	23 ± 2	22 ± 2	27 ± 3
	333	17 ± 2	16 ± 1	14 ± 1	17 ± 2	21 ± 5	23 ± 2
	1,000	19 ± 2	15 ± 1	20 ± 2	22 ± 1	20 ± 2	21 ± 1
	3,333	19 ± 1	17 ± 2[c]	12 ± 3[c]	19 ± 2[c]	15 ± 1[c]	15 ± 2[c]
	10,000	Toxic			11 ± 2[c]		13 ± 0[c]
Trial summary		Negative	Negative	Negative	Negative	Negative	Negative
Positive control		340 ± 17	166 ± 3	875 ± 24	578 ± 17	302 ± 22	824 ± 28

TABLE E1
Mutagenicity of 1-Bromopropane in Bacterial Tester Strains

Strain	Dose (µg/Plate)	Revertants/Plate −S9 Trial 1	−S9 Trial 2	+10% rat S9 Trial 1	+10% rat S9 Trial 2
Study Performed at SITEK Research Laboratories (lot 1581313004 used in the 2-year studies)					
TA100	0	62 ± 2	67 ± 4	76 ± 3	65 ± 2
	500	57 ± 6	57 ± 3		
	1,000		62 ± 4		
	1,500	64 ± 3	53 ± 4		
	2,500	50 ± 2	35 ± 1	84 ± 2	67 ± 3
	3,500	38 ± 6	16 ± 1[c]	75 ± 3	63 ± 6
	5,000	11 ± 2[c]		66 ± 3	66 ± 4
	7,500			62 ± 5	60 ± 4
	10,000			59 ± 7	53 ± 7
Trial summary		Negative	Negative	Negative	Negative
Positive control		488 ± 27	635 ± 35	878 ± 19	982 ± 37
TA98	0	18 ± 1	22 ± 3	29 ± 3	24 ± 2
	500	16 ± 0	16 ± 1		
	1,000		19 ± 2		
	1,500	17 ± 3	18 ± 1		
	2,500	18 ± 2	14 ± 1	24 ± 0	23 ± 1
	3,500	8 ± 2[c]	5 ± 1[c]	21 ± 4	21 ± 0
	5,000	2 ± 1[c]		26 ± 4	26 ± 1
	7,500			23 ± 3	20 ± 1
	10,000			6 ± 1[c]	8 ± 2[c]
Trial summary		Negative	Negative	Negative	Negative
Positive control		693 ± 26	693 ± 26	1,163 ± 77	742 ± 37
***Escherichia coli* WP2 *uvrA*/pKM101**					
	0	177 ± 3	163 ± 7	185 ± 8	190 ± 23
	500	197 ± 25	190 ± 15		
	1,500	211 ± 20	194 ± 6		
	2,500	153 ± 7	139 ± 12	226 ± 17	214 ± 14
	3,500	148 ± 10	124 ± 3	203 ± 14	201 ± 8
	5,000	88 ± 3[c]	76 ± 17[c]	192 ± 6	195 ± 10
	7,500			171 ± 6	164 ± 25
	10,000			149 ± 7	142 ± 4
Trial summary		Negative	Negative	Negative	Negative
Positive control		1,883 ± 20	1,955 ± 18	993 ± 79	1,164 ± 25

[a] The detailed protocol for the BioReliance Corporation study is presented by Zeiger *et al.* (1992); the study performed at SITEK Research Laboratories used a slight modification of those procedures. 0 µg/plate was the solvent control.

[b] Revertants are presented as mean ± standard error from three plates.

[c] Slight toxicity

[d] The positive controls in the absence of metabolic activation were sodium azide (TA100 and TA1535), 9-aminoacridine (TA97), 4-nitro-*o*-phenylenediamine (TA98), and methyl methanesulfonate (*E. coli*). The positive control for metabolic activation with all strains was 2-aminoanthracene.

[e] Contamination

[f] Slight toxicity and precipitate

TABLE E2
Frequency of Micronuclei in Peripheral Blood Erythrocytes of Mice Following Treatment with 1-Bromopropane by Inhalation for 3 Months[a]

Compound	Exposure Concentration (ppm)	Number of Mice with Erythrocytes Scored	Micronucleated NCEs/ 1,000 NCEs[b]	P value[c]	PCEs (%)[b]
Male					
Air[d]	0	5	2.00 ± 0.61		2.52 ± 0.30
1-Bromopropane	62.5	5	3.10 ± 0.81	0.0615	2.16 ± 0.31
	125	5	2.70 ± 0.64	0.1533	2.96 ± 0.11
	250	5	1.30 ± 0.41	0.8887	2.80 ± 0.18
	500	5	2.30 ± 0.46	0.3235	2.80 ± 0.44
			P=0.757[e]		
Female					
Air	0	5	1.80 ± 0.25		2.24 ± 0.52
1-Bromopropane	62.5	5	1.70 ± 0.25	0.5672	2.94 ± 0.23
	125	5	1.60 ± 0.19	0.6343	2.74 ± 0.52
	250	5	1.40 ± 0.33	0.7604	2.42 ± 0.27
	500	5	1.80 ± 0.20	0.5000	2.48 ± 0.29
			P=0.500		

[a] Study was performed at ILS, Inc. The detailed protocol is presented by MacGregor *et al.* (1990). PCE=polychromatic erythrocyte; NCE=normochromatic erythrocyte.

[b] Mean ± standard error

[c] Pairwise comparison with the chamber control group; significant at P≤0.006

[d] Chamber control

[e] Significance of micronucleated NCEs/1,000 NCEs tested by the one-tailed trend test; significant at P≤0.025

APPENDIX F
CLINICAL PATHOLOGY RESULTS

TABLE F1
Hematology and Clinical Chemistry Data for Rats in the 3-Month Inhalation Study of 1-Bromopropane[a]

	Chamber Control	62.5 ppm	125 ppm	250 ppm	500 ppm	1,000 ppm
Male						
Hematology						
n						
Day 3	10	10	10	9	10	10
Day 23	10	10	10	10	10	10
Week 14	10	10	10	10	10	10
Hematocrit (%)						
Day 3	45.3 ± 0.5	44.2 ± 0.4	44.7 ± 0.4	44.9 ± 0.5	45.0 ± 0.6	47.0 ± 0.6
Day 23	47.2 ± 0.5	47.2 ± 0.4	47.9 ± 0.4	47.4 ± 0.4	47.2 ± 0.5	47.9 ± 0.5
Week 14	48.2 ± 0.2	48.4 ± 0.4	48.0 ± 0.3	48.5 ± 0.3	47.9 ± 0.3	49.1 ± 0.5
Packed cell volume (mL/dL)						
Day 3	44.3 ± 0.6	43.8 ± 0.4	43.8 ± 0.4	44.3 ± 0.4	44.5 ± 0.5	46.5 ± 0.5**
Day 23	46.0 ± 0.6	46.0 ± 0.5	46.8 ± 0.4	46.4 ± 0.5	46.1 ± 0.4	47.1 ± 0.4
Week 14	47.8 ± 0.3	47.8 ± 0.3	47.7 ± 0.4	47.8 ± 0.3	47.6 ± 0.4	48.4 ± 0.5
Hemoglobin (g/dL)						
Day 3	13.5 ± 0.2	13.5 ± 0.1	13.6 ± 0.2	13.8 ± 0.2	13.7 ± 0.1	14.3 ± 0.2**
Day 23	14.8 ± 0.2	14.7 ± 0.2	14.9 ± 0.1	14.8 ± 0.2	14.7 ± 0.1	14.9 ± 0.1
Week 14	15.4 ± 0.1	15.5 ± 0.1	15.5 ± 0.1	15.6 ± 0.1	15.5 ± 0.1	15.8 ± 0.2
Erythrocytes (10^6/µL)						
Day 3	7.12 ± 0.10	7.07 ± 0.08	7.14 ± 0.09	7.30 ± 0.08	7.34 ± 0.09	7.71 ± 0.11**
Day 23	7.80 ± 0.10	7.74 ± 0.09	7.84 ± 0.10	7.78 ± 0.07	7.67 ± 0.09	7.84 ± 0.05
Week 14	9.07 ± 0.06	9.02 ± 0.05	9.01 ± 0.05	9.06 ± 0.04	8.98 ± 0.08	9.15 ± 0.09
Reticulocytes (10^3/µL)						
Day 3	360.8 ± 28.8	356.4 ± 32.6	381.7 ± 25.1	350.3 ± 30.4	362.4 ± 33.7	346.3 ± 17.0
Day 23	209.2 ± 19.6	187.6 ± 16.7	233.4 ± 19.1	240.4 ± 15.2	222.6 ± 10.8	222.0 ± 11.8
Week 14	106.1 ± 14.6	131.2 ± 12.4	137.4 ± 12.4	127.6 ± 15.7	97.8 ± 12.6	119.3 ± 10.7
Nucleated erythrocytes/100 leukocytes						
Day 3	0.70 ± 0.15	1.10 ± 0.28	0.90 ± 0.35	0.78 ± 0.28	0.20 ± 0.13	0.10 ± 0.10*
Day 23	0.20 ± 0.13	0.00 ± 0.00	0.10 ± 0.10	0.00 ± 0.00	0.30 ± 0.15	0.00 ± 0.00
Week 14	0.00 ± 0.00	0.30 ± 0.15	0.10 ± 0.10	0.30 ± 0.21	0.50 ± 0.22	0.30 ± 0.21
Mean cell volume (fL)						
Day 3	62.2 ± 0.2	61.9 ± 0.4	61.4 ± 0.4	60.7 ± 0.3**	60.6 ± 0.4**	60.3 ± 0.4**
Day 23	59.0 ± 0.3	59.4 ± 0.3	59.8 ± 0.4	59.6 ± 0.2	60.2 ± 0.4*	60.0 ± 0.3
Week 14	52.7 ± 0.2	53.0 ± 0.2	52.9 ± 0.2	52.8 ± 0.2	53.0 ± 0.3	52.9 ± 0.2
Mean cell hemoglobin (pg)						
Day 3	18.9 ± 0.1	19.0 ± 0.1	19.0 ± 0.1	18.9 ± 0.1	18.7 ± 0.1	18.6 ± 0.1*
Day 23	18.9 ± 0.1	19.0 ± 0.1	19.0 ± 0.1	19.0 ± 0.1	19.2 ± 0.1	19.0 ± 0.1
Week 14	17.1 ± 0.0	17.2 ± 0.1	17.1 ± 0.1	17.2 ± 0.1	17.3 ± 0.1	17.2 ± 0.1
Mean cell hemoglobin concentration (g/dL)						
Day 3	30.4 ± 0.1	30.7 ± 0.2	30.8 ± 0.2	31.1 ± 0.2*	30.8 ± 0.2	30.8 ± 0.2
Day 23	32.1 ± 0.1	32.0 ± 0.1	31.8 ± 0.1	31.8 ± 0.2	31.9 ± 0.1	31.7 ± 0.1
Week 14	32.4 ± 0.1	32.5 ± 0.1	32.4 ± 0.1	32.6 ± 0.2	32.6 ± 0.1	32.5 ± 0.2
Platelets (10^3/µL)						
Day 3	905.9 ± 33.7	875.4 ± 16.8	888.2 ± 16.3	799.0 ± 30.6**	759.8 ± 29.9**	820.9 ± 33.4**
Day 23	754.5 ± 18.1	798.8 ± 14.0	820.2 ± 16.4*	802.4 ± 20.1*	843.3 ± 14.8**	845.8 ± 15.6**
Week 14	678.6 ± 10.6	674.2 ± 8.3	653.8 ± 14.4	661.8 ± 13.9	647.8 ± 16.1	651.7 ± 6.4
Leukocytes (10^3/µL)						
Day 3	10.39 ± 0.50	9.77 ± 0.71	10.77 ± 0.47	10.95 ± 0.56	8.97 ± 0.53	7.79 ± 0.43**
Day 23	7.90 ± 0.53	8.40 ± 0.40	8.13 ± 0.57	7.73 ± 0.32	8.24 ± 0.44	7.68 ± 0.44
Week 14	7.25 ± 0.17	6.78 ± 0.22	7.48 ± 0.41	8.09 ± 0.37	8.07 ± 0.34	8.56 ± 0.33*
Segmented neutrophils (10^3/µL)						
Day 3	1.22 ± 0.06	1.19 ± 0.06	1.34 ± 0.06	1.34 ± 0.09	1.10 ± 0.09	0.92 ± 0.05**
Day 23	1.16 ± 0.03	1.25 ± 0.04	1.15 ± 0.06	1.18 ± 0.04	1.07 ± 0.05	0.93 ± 0.03**
Week 14	1.19 ± 0.09	1.18 ± 0.09	1.34 ± 0.11	1.32 ± 0.07	1.40 ± 0.08	1.37 ± 0.11

TABLE F1
Hematology and Clinical Chemistry Data for Rats in the 3-Month Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm	500 ppm	1,000 ppm
Male (continued)						
Hematology (continued)						
n						
Day 3	10	10	10	9	10	10
Day 23	10	10	10	10	10	10
Week 14	10	10	10	10	10	10
Bands (10^3/µL)						
Day 3	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
Day 23	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
Week 14	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
Lymphocytes (10^3/µL)						
Day 3	8.93 ± 0.46	8.39 ± 0.66	9.19 ± 0.43	9.38 ± 0.49	7.65 ± 0.45	6.72 ± 0.39**
Day 23	6.62 ± 0.50	7.01 ± 0.36	6.86 ± 0.51	6.42 ± 0.29	7.03 ± 0.39	6.63 ± 0.43
Week 14	5.79 ± 0.17	5.35 ± 0.16	5.73 ± 0.35	6.49 ± 0.34	6.33 ± 0.34	6.91 ± 0.27*
Monocytes (10^3/µL)						
Day 3	0.14 ± 0.02	0.09 ± 0.01	0.14 ± 0.02	0.09 ± 0.02	0.14 ± 0.03	0.08 ± 0.02*
Day 23	0.04 ± 0.01	0.06 ± 0.01	0.04 ± 0.01	0.05 ± 0.01	0.05 ± 0.01	0.05 ± 0.01
Week 14	0.17 ± 0.05	0.16 ± 0.04	0.27 ± 0.08	0.18 ± 0.05	0.22 ± 0.07	0.21 ± 0.03
Basophils (10^3/µL)						
Day 3	0.007 ± 0.002	0.004 ± 0.002	0.006 ± 0.002	0.007 ± 0.002	0.003 ± 0.002	0.005 ± 0.002
Day 23	0.007 ± 0.002	0.005 ± 0.002	0.008 ± 0.002	0.006 ± 0.002	0.006 ± 0.002	0.007 ± 0.002
Week 14	0.002 ± 0.001	0.002 ± 0.002	0.010 ± 0.004	0.004 ± 0.002	0.003 ± 0.002	0.009 ± 0.009
Eosinophils (10^3/µL)						
Day 3	0.09 ± 0.01	0.10 ± 0.01	0.10 ± 0.01	0.13 ± 0.02	0.09 ± 0.01	0.06 ± 0.01
Day 23	0.08 ± 0.01	0.08 ± 0.01	0.08 ± 0.01	0.07 ± 0.01	0.08 ± 0.01	0.06 ± 0.01
Week 14	0.09 ± 0.01	0.10 ± 0.01	0.13 ± 0.01	0.09 ± 0.03	0.12 ± 0.02	0.07 ± 0.02
Clinical Chemistry						
n	10	10	10	10	10	10
Urea nitrogen (mg/dL)						
Day 3	8.1 ± 0.4	9.0 ± 0.2*	9.0 ± 0.4*	10.3 ± 0.4**	11.0 ± 0.5**	16.7 ± 0.6**
Day 23	10.7 ± 0.3	11.5 ± 0.6	11.5 ± 0.3	12.3 ± 0.3**	14.6 ± 0.5**	16.5 ± 0.4**
Week 14	16.9 ± 0.3	15.8 ± 0.5	15.6 ± 0.5*	16.3 ± 0.5	17.0 ± 0.3	14.5 ± 0.5**
Creatinine (mg/dL)						
Day 3	0.24 ± 0.02	0.26 ± 0.02	0.26 ± 0.02	0.26 ± 0.02	0.26 ± 0.02	0.30 ± 0.00*
Day 23	0.31 ± 0.01	0.30 ± 0.00	0.33 ± 0.02	0.36 ± 0.02*	0.37 ± 0.02**	0.38 ± 0.01**
Week 14	0.61 ± 0.03	0.63 ± 0.02	0.62 ± 0.02	0.62 ± 0.01	0.63 ± 0.02	0.67 ± 0.02
Total protein (g/dL)						
Day 3	5.9 ± 0.1	5.8 ± 0.0	5.7 ± 0.1*	5.6 ± 0.1**	5.6 ± 0.1**	5.7 ± 0.1*
Day 23	6.3 ± 0.0	6.2 ± 0.0	6.3 ± 0.0	6.3 ± 0.1	6.2 ± 0.1	6.2 ± 0.1
Week 14	7.3 ± 0.1	7.2 ± 0.1	7.1 ± 0.1	7.3 ± 0.0	7.2 ± 0.1	7.2 ± 0.1
Albumin (g/dL)						
Day 3	4.3 ± 0.0	4.2 ± 0.0	4.1 ± 0.0**	4.1 ± 0.0**	4.1 ± 0.0**	4.2 ± 0.0**
Day 23	4.5 ± 0.0	4.4 ± 0.0	4.4 ± 0.0	4.4 ± 0.0	4.3 ± 0.0**	4.3 ± 0.0**
Week 14	4.9 ± 0.0	4.8 ± 0.0	4.7 ± 0.1	4.8 ± 0.0	4.7 ± 0.0	4.8 ± 0.0
Globulin (g/dL)						
Day 3	1.6 ± 0.1	1.6 ± 0.0	1.6 ± 0.0	1.5 ± 0.0	1.5 ± 0.0	1.6 ± 0.0
Day 23	1.8 ± 0.0	1.8 ± 0.0	1.9 ± 0.0	1.8 ± 0.0	1.9 ± 0.0	1.8 ± 0.0
Week 14	2.4 ± 0.1	2.5 ± 0.0	2.4 ± 0.0	2.5 ± 0.0	2.5 ± 0.0	2.5 ± 0.0
Albumin/globulin ratio						
Day 3	2.7 ± 0.1	2.7 ± 0.0	2.7 ± 0.1	2.7 ± 0.0	2.7 ± 0.1	2.7 ± 0.0
Day 23	2.5 ± 0.0	2.4 ± 0.0	2.4 ± 0.0	2.4 ± 0.1	2.3 ± 0.0**	2.4 ± 0.0**
Week 14	2.0 ± 0.0	2.0 ± 0.0	1.9 ± 0.1	1.9 ± 0.0*	1.9 ± 0.0*	1.9 ± 0.0*

TABLE F1
Hematology and Clinical Chemistry Data for Rats in the 3-Month Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm	500 ppm	1,000 ppm
Male (continued)						
Clinical Chemistry (continued)						
n	10	10	10	10	10	10
Alanine aminotransferase (IU/L)						
Day 3	56 ± 2	49 ± 1**	40 ± 1**	32 ± 1**	50 ± 7**	31 ± 5**
Day 23	47 ± 1	44 ± 1	38 ± 1**	30 ± 1**	25 ± 1**	22 ± 1**
Week 14	141 ± 16	115 ± 9	121 ± 8	124 ± 9	120 ± 7	71 ± 5**
Alkaline phosphatase (IU/L)						
Day 3	689 ± 13	691 ± 15	679 ± 7	686 ± 12	706 ± 22	645 ± 18
Day 23	438 ± 11	467 ± 13	476 ± 9	470 ± 12	429 ± 9	453 ± 9
Week 14	258 ± 6	231 ± 3**	238 ± 6*	236 ± 4*	230 ± 5**	232 ± 5**
Creatine kinase (IU/L)						
Day 3	400 ± 43[b]	385 ± 35	417 ± 37	397 ± 44	400 ± 22[b]	311 ± 25
Day 23	417 ± 63	350 ± 41	380 ± 48	712 ± 190	363 ± 49	358 ± 39
Week 14	355 ± 113	191 ± 26	189 ± 40[b]	224 ± 46	223 ± 38	137 ± 18
Sorbitol dehydrogenase (IU/L)						
Day 23	17 ± 1	17 ± 0	17 ± 1	16 ± 1	18 ± 1	17 ± 0
Week 14	27 ± 2	24 ± 1	27 ± 1	30 ± 1	35 ± 1**	34 ± 1**
Bile salts (µmol/L)						
Day 3	19.5 ± 1.0	16.6 ± 0.5	16.4 ± 0.5	16.4 ± 0.9	25.3 ± 2.6	21.6 ± 2.2
Day 23	15.7 ± 0.4	15.0 ± 0.6	15.3 ± 0.3	15.2 ± 0.6	13.5 ± 0.4**	14.6 ± 0.5
Week 14	17.7 ± 0.9	15.7 ± 0.5	18.1 ± 1.2	16.4 ± 0.6	18.7 ± 0.6	17.5 ± 1.1
Female						
Hematology						
n	10	10	10	10	10	10
Hematocrit (%)						
Day 3	48.4 ± 0.5	47.3 ± 0.4	47.7 ± 0.5	47.7 ± 0.5	47.8 ± 0.5	47.4 ± 0.7
Day 23	47.6 ± 0.5	48.1 ± 0.4	47.7 ± 0.5	48.2 ± 0.6	48.7 ± 0.4	50.5 ± 0.4**
Week 14	48.2 ± 0.3	47.8 ± 0.4	47.6 ± 0.5	48.8 ± 0.3	47.3 ± 0.4	48.4 ± 0.4
Packed cell volume (mL/dL)						
Day 3	48.2 ± 0.4	47.2 ± 0.4	46.9 ± 0.5	47.5 ± 0.6	47.7 ± 0.6	47.8 ± 0.6
Day 23	47.9 ± 0.5	47.9 ± 0.4	47.5 ± 0.3	48.4 ± 0.5	48.5 ± 0.4	50.1 ± 0.4**
Week 14	48.2 ± 0.4	47.6 ± 0.4	47.5 ± 0.5	48.5 ± 0.3	47.3 ± 0.3	47.7 ± 0.4
Hemoglobin (g/dL)						
Day 3	15.0 ± 0.2	14.5 ± 0.1	14.5 ± 0.2	14.5 ± 0.2	14.7 ± 0.2	14.7 ± 0.2
Day 23	15.4 ± 0.2	15.3 ± 0.1	15.4 ± 0.1	15.6 ± 0.2	15.8 ± 0.1*	16.2 ± 0.2**
Week 14	15.7 ± 0.1	15.6 ± 0.1	15.5 ± 0.2	15.8 ± 0.1	15.6 ± 0.1	15.6 ± 0.1
Erythrocytes (10^6/µL)						
Day 3	7.97 ± 0.11	7.69 ± 0.07	7.69 ± 0.12	7.75 ± 0.11	7.87 ± 0.14	7.95 ± 0.11
Day 23	8.16 ± 0.12	8.04 ± 0.06	8.03 ± 0.06	8.08 ± 0.11	8.23 ± 0.10	8.48 ± 0.09
Week 14	8.39 ± 0.07	8.36 ± 0.06	8.34 ± 0.09	8.49 ± 0.05	8.35 ± 0.04	8.45 ± 0.07
Reticulocytes (10^3/µL)						
Day 3	293.9 ± 16.6	295.4 ± 22.4	316.4 ± 10.9	274.5 ± 14.3	294.0 ± 14.6	290.2 ± 18.3
Day 23	156.7 ± 9.9	158.4 ± 11.8	148.5 ± 10.9	160.7 ± 8.2	162.3 ± 8.7	168.4 ± 7.6
Week 14	110.2 ± 11.5	114.7 ± 15.5	112.4 ± 18.5	113.0 ± 8.2	104.9 ± 17.8	115.9 ± 16.8
Nucleated erythrocytes/100 leukocytes						
Day 3	0.20 ± 0.20	0.20 ± 0.13	0.20 ± 0.13	0.60 ± 0.22	0.60 ± 0.31	0.20 ± 0.13
Day 23	0.20 ± 0.13	0.00 ± 0.00	0.20 ± 0.13	0.10 ± 0.10	0.00 ± 0.00	0.10 ± 0.10
Week 14	0.70 ± 0.21	0.40 ± 0.22	0.30 ± 0.15	0.50 ± 0.27	0.80 ± 0.39	0.70 ± 0.40

TABLE F1
Hematology and Clinical Chemistry Data for Rats in the 3-Month Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm	500 ppm	1,000 ppm
Female (continued)						
Hematology (continued)						
n	10	10	10	10	10	10
Mean cell volume (fL)						
Day 3	60.5 ± 0.5	61.4 ± 0.2	61.0 ± 0.4	61.3 ± 0.2	60.6 ± 0.4	60.2 ± 0.2
Day 23	58.8 ± 0.4	59.5 ± 0.3	59.2 ± 0.6	59.9 ± 0.4	59.0 ± 0.4	59.1 ± 0.4
Week 14	57.4 ± 0.2	57.0 ± 0.2	56.9 ± 0.1	57.2 ± 0.1	56.6 ± 0.1**	56.5 ± 0.2**
Mean cell hemoglobin (pg)						
Day 3	18.8 ± 0.1	18.9 ± 0.1	18.8 ± 0.1	18.8 ± 0.1	18.7 ± 0.1	18.6 ± 0.1
Day 23	18.8 ± 0.1	19.1 ± 0.1	19.1 ± 0.1	19.3 ± 0.1*	19.2 ± 0.2	19.1 ± 0.1
Week 14	18.7 ± 0.1	18.7 ± 0.1	18.6 ± 0.1	18.6 ± 0.1	18.7 ± 0.1	18.4 ± 0.1*
Mean cell hemoglobin concentration (g/dL)						
Day 3	31.1 ± 0.1	30.8 ± 0.1	30.8 ± 0.2	30.6 ± 0.1	30.8 ± 0.2	30.8 ± 0.1
Day 23	32.1 ± 0.1	32.0 ± 0.1	32.4 ± 0.2	32.2 ± 0.1	32.5 ± 0.2	32.2 ± 0.1
Week 14	32.6 ± 0.1	32.8 ± 0.1	32.6 ± 0.1	32.6 ± 0.1	33.0 ± 0.1	32.6 ± 0.1
Platelets (10^3/µL)						
Day 3	812.0 ± 25.7	824.1 ± 21.5	796.1 ± 19.3	805.1 ± 35.1	740.4 ± 30.7	778.1 ± 29.5
Day 23	773.3 ± 17.5	791.6 ± 13.7	806.4 ± 10.6	757.4 ± 19.3	791.4 ± 15.1	775.0 ± 21.8
Week 14	681.6 ± 12.3	647.1 ± 10.7	656.6 ± 12.0	660.7 ± 15.7	658.2 ± 13.6	647.5 ± 11.6
Leukocytes (10^3/µL)						
Day 3	12.82 ± 0.51	11.64 ± 0.33	11.75 ± 0.57	11.64 ± 0.51	11.46 ± 0.53	8.41 ± 0.45**
Day 23	7.39 ± 0.54	7.48 ± 0.27	9.13 ± 0.39*	8.90 ± 0.36	9.27 ± 0.41*	7.98 ± 0.36
Week 14	6.34 ± 0.34	6.54 ± 0.26	6.51 ± 0.31	6.55 ± 0.31	6.44 ± 0.32	7.64 ± 0.43
Segmented neutrophils (10^3/µL)						
Day 3	1.41 ± 0.07	1.43 ± 0.07	1.29 ± 0.06	1.40 ± 0.11	1.21 ± 0.07*	1.10 ± 0.14**
Day 23	1.10 ± 0.11	1.09 ± 0.08	1.33 ± 0.19	1.23 ± 0.15	1.28 ± 0.14	1.12 ± 0.12
Week 14	1.03 ± 0.10	0.90 ± 0.05	1.03 ± 0.06	1.10 ± 0.09	0.89 ± 0.05	1.33 ± 0.10
Bands (10^3/µL)						
Day 3	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
Day 23	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
Week 14	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
Lymphocytes (10^3/µL)						
Day 3	11.11 ± 0.50	9.97 ± 0.35	10.20 ± 0.54	9.98 ± 0.41	10.02 ± 0.48	7.10 ± 0.40**
Day 23	6.13 ± 0.49	6.22 ± 0.20	7.59 ± 0.31*	7.46 ± 0.32	7.78 ± 0.35*	6.64 ± 0.30
Week 14	5.04 ± 0.28	5.49 ± 0.24	5.25 ± 0.28	5.19 ± 0.27	5.29 ± 0.31	5.95 ± 0.38
Monocytes (10^3/µL)						
Day 3	0.12 ± 0.01	0.09 ± 0.02	0.12 ± 0.02	0.11 ± 0.02	0.10 ± 0.01	0.08 ± 0.01
Day 23	0.05 ± 0.01	0.07 ± 0.01	0.07 ± 0.02	0.07 ± 0.01	0.06 ± 0.01	0.10 ± 0.02
Week 14	0.17 ± 0.06	0.06 ± 0.02	0.13 ± 0.04	0.16 ± 0.05	0.16 ± 0.04	0.29 ± 0.07
Basophils (10^3/µL)						
Day 3	0.018 ± 0.002	0.013 ± 0.003	0.015 ± 0.004	0.015 ± 0.003	0.013 ± 0.003	0.009 ± 0.002
Day 23	0.008 ± 0.007	0.005 ± 0.002	0.006 ± 0.002	0.006 ± 0.002	0.009 ± 0.002	0.006 ± 0.002
Week 14	0.004 ± 0.002	0.002 ± 0.001	0.015 ± 0.008	0.005 ± 0.002	0.007 ± 0.002	0.007 ± 0.003
Eosinophils (10^3/µL)						
Day 3	0.17 ± 0.02	0.14 ± 0.01	0.12 ± 0.01	0.13 ± 0.02	0.12 ± 0.01	0.12 ± 0.01
Day 23	0.10 ± 0.01	0.10 ± 0.01	0.13 ± 0.01	0.13 ± 0.01	0.15 ± 0.02*	0.10 ± 0.01
Week 14	0.09 ± 0.01	0.10 ± 0.01	0.08 ± 0.01	0.10 ± 0.01	0.10 ± 0.02	0.07 ± 0.01

TABLE F1
Hematology and Clinical Chemistry Data for Rats in the 3-Month Inhalation Study of 1-Bromopropane

	Chamber Control	62.5 ppm	125 ppm	250 ppm	500 ppm	1,000 ppm
Female (continued)						
Clinical Chemistry						
n	10	10	10	10	10	10
Urea nitrogen (mg/dL)						
Day 3	10.3 ± 0.5	10.4 ± 0.5	8.9 ± 0.5	10.2 ± 0.4	12.4 ± 0.4*	14.5 ± 0.4**
Day 23	11.6 ± 0.4	11.4 ± 0.3	11.5 ± 0.3	11.3 ± 0.2	12.8 ± 0.3**	16.6 ± 0.3**
Week 14	16.2 ± 0.4	17.0 ± 0.7	15.5 ± 0.4	16.4 ± 0.5	16.9 ± 0.4	14.8 ± 0.5
Creatinine (mg/dL)						
Day 3	0.27 ± 0.02	0.25 ± 0.02	0.27 ± 0.02	0.27 ± 0.02	0.28 ± 0.01	0.31 ± 0.01
Day 23	0.24 ± 0.02	0.27 ± 0.02	0.28 ± 0.01	0.28 ± 0.01	0.30 ± 0.02**	0.37 ± 0.02**
Week 14	0.70 ± 0.02	0.69 ± 0.01	0.68 ± 0.01	0.68 ± 0.01	0.63 ± 0.02	0.71 ± 0.02
Total protein (g/dL)						
Day 3	6.1 ± 0.1	5.8 ± 0.1**	5.9 ± 0.1*	5.8 ± 0.1**	5.7 ± 0.0**	5.7 ± 0.1**
Day 23	6.2 ± 0.0	6.1 ± 0.0	6.3 ± 0.1	6.2 ± 0.1	6.2 ± 0.1	6.1 ± 0.1
Week 14	7.4 ± 0.1	7.4 ± 0.1	7.3 ± 0.1	7.3 ± 0.1	7.4 ± 0.1	7.4 ± 0.1
Albumin (g/dL)						
Day 3	4.6 ± 0.0	4.4 ± 0.0**	4.4 ± 0.0**	4.4 ± 0.1**	4.3 ± 0.0**	4.3 ± 0.0**
Day 23	4.6 ± 0.0	4.5 ± 0.0	4.6 ± 0.1	4.5 ± 0.0	4.5 ± 0.0	4.5 ± 0.0
Week 14	5.3 ± 0.0	5.3 ± 0.0	5.2 ± 0.1	5.2 ± 0.1	5.3 ± 0.0	5.2 ± 0.1
Globulin (g/dL)						
Day 3	1.5 ± 0.0	1.4 ± 0.0	1.5 ± 0.0	1.4 ± 0.0	1.4 ± 0.0	1.4 ± 0.0
Day 23	1.7 ± 0.0	1.6 ± 0.0	1.8 ± 0.0	1.7 ± 0.0	1.7 ± 0.0	1.6 ± 0.0
Week 14	2.1 ± 0.0	2.1 ± 0.0	2.1 ± 0.1	2.1 ± 0.1	2.1 ± 0.1	2.2 ± 0.0
Albumin/globulin ratio						
Day 3	3.2 ± 0.1	3.1 ± 0.1	3.0 ± 0.0	3.1 ± 0.1	3.1 ± 0.1	3.2 ± 0.1
Day 23	2.8 ± 0.1	2.9 ± 0.1	2.6 ± 0.0	2.7 ± 0.0	2.7 ± 0.0	2.9 ± 0.1
Week 14	2.6 ± 0.0	2.5 ± 0.0	2.5 ± 0.0	2.6 ± 0.0	2.5 ± 0.1**	2.4 ± 0.0**
Alanine aminotransferase (IU/L)						
Day 3	51 ± 2	41 ± 1**	33 ± 1**	26 ± 1**	25 ± 2**	19 ± 1**
Day 23	44 ± 1	34 ± 1**	29 ± 1**	22 ± 1**	17 ± 1**	20 ± 5**
Week 14	71 ± 8	61 ± 5	57 ± 5	53 ± 3*	41 ± 4**	37 ± 7**
Alkaline phosphatase (IU/L)						
Day 3	598 ± 15	589 ± 10	579 ± 17	563 ± 7	529 ± 13**	515 ± 11**
Day 23	326 ± 9	342 ± 8	325 ± 9	326 ± 7	314 ± 9	337 ± 9
Week 14	215 ± 7	191 ± 5**	182 ± 5**	201 ± 6*	179 ± 5**	187 ± 8**
Creatine kinase (IU/L)						
Day 3	373 ± 33[b]	329 ± 48	326 ± 25[b]	338 ± 26	303 ± 29	217 ± 10**
Day 23	285 ± 25[b]	331 ± 25	334 ± 25	399 ± 57	311 ± 17	296 ± 18
Week 14	179 ± 20	182 ± 12[b]	226 ± 37	168 ± 23	192 ± 34	157 ± 22
Sorbitol dehydrogenase (IU/L)						
Day 23	16 ± 1	17 ± 0	15 ± 1	14 ± 2	16 ± 1	67 ± 27*
Week 14	19 ± 1	18 ± 1	18 ± 1	18 ± 1	18 ± 1	87 ± 33
Bile salts (µmol/L)						
Day 3	17.3 ± 0.7	17.8 ± 1.3	16.1 ± 0.9	14.0 ± 0.8**	14.7 ± 0.7**	15.3 ± 0.9*
Day 23	16.9 ± 1.0	13.6 ± 0.5	14.3 ± 0.9	13.1 ± 0.5*	13.3 ± 0.5*	19.2 ± 2.9
Week 14	16.2 ± 0.9	17.7 ± 1.2	15.9 ± 2.1	13.7 ± 0.5	15.7 ± 1.2	30.7 ± 6.3

* Significantly different ($P \leq 0.05$) from the chamber control group by Dunn's or Shirley's test

** $P \leq 0.01$

[a] Mean ± standard error. Statistical tests were performed on unrounded data.

[b] n=9

TABLE F2
Hematology Data for Mice in the 3-Month Inhalation Study of 1-Bromopropane[a]

	Chamber Control	62.5 ppm	125 ppm	250 ppm	500 ppm
Male					
n	10	10	10	9	6
Hematocrit (%)	47.8 ± 0.4	48.4 ± 0.3	49.3 ± 0.4*	48.1 ± 0.2	47.9 ± 0.5
Packed cell volume (mL/dL)	47.9 ± 0.5	49.2 ± 0.3	50.1 ± 0.4**	49.1 ± 0.2	48.9 ± 0.7
Hemoglobin (g/dL)	15.2 ± 0.2	15.5 ± 0.1	15.9 ± 0.1**	15.4 ± 0.1	15.1 ± 0.2
Erythrocytes (10^6/µL)	9.79 ± 0.09	9.91 ± 0.04	9.98 ± 0.09	9.62 ± 0.05	9.46 ± 0.15
Reticulocytes (10^3/µL)	143.8 ± 8.9	158.6 ± 12.0	145.6 ± 12.6	130.2 ± 11.7	153.5 ± 26.9
Nucleated erythrocytes /100 leukocytes	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
Howell-Jolly bodies (%) erythrocytes	0.1 ± 0.0	0.2 ± 0.0	0.2 ± 0.0	0.2 ± 0.1	0.1 ± 0.0
Mean cell volume (fL)	48.9 ± 0.2	49.6 ± 0.2*	50.2 ± 0.3**	51.1 ± 0.2**	51.7 ± 0.2**
Mean cell hemoglobin (pg)	15.5 ± 0.1	15.6 ± 0.1	15.9 ± 0.1**	16.0 ± 0.1**	15.9 ± 0.1**
Mean cell hemoglobin concentration (g/dL)	31.7 ± 0.1	31.6 ± 0.1	31.6 ± 0.1	31.4 ± 0.1*	30.9 ± 0.1**
Platelets (10^3/µL)	891.7 ± 27.6	894.0 ± 17.4	874.9 ± 14.7	870.0 ± 15.5	880.3 ± 43.4
Leukocytes (10^3/µL)	2.36 ± 0.28	2.50 ± 0.25	2.42 ± 0.22	2.72 ± 0.17	3.86 ± 0.41**
Segmented neutrophils (10^3/µL)	0.29 ± 0.05	0.31 ± 0.03	0.29 ± 0.04	0.31 ± 0.03	0.43 ± 0.03*
Bands (10^3/µL)	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
Lymphocytes (10^3/µL)	1.94 ± 0.23	2.09 ± 0.21	2.02 ± 0.20	2.28 ± 0.16	3.31 ± 0.36**
Monocytes (10^3/µL)	0.06 ± 0.01	0.03 ± 0.01	0.04 ± 0.02	0.07 ± 0.02	0.05 ± 0.02
Basophils (10^3/µL)	0.013 ± 0.002	0.009 ± 0.003	0.012 ± 0.003	0.009 ± 0.002	0.012 ± 0.003
Eosinophils (10^3/µL)	0.06 ± 0.01	0.06 ± 0.02	0.05 ± 0.01	0.05 ± 0.01	0.07 ± 0.02
Female					
n	10	10	10	10	5
Hematocrit (%)	50.1 ± 0.3	50.1 ± 0.5	49.4 ± 0.3	48.5 ± 0.4**	48.6 ± 0.5*
Packed cell volume (mL/dL)	50.9 ± 0.3	50.7 ± 0.4	50.0 ± 0.2	49.1 ± 0.5**	49.1 ± 0.5*
Hemoglobin (g/dL)	16.2 ± 0.1	16.2 ± 0.1	15.9 ± 0.1	15.6 ± 0.1**	15.6 ± 0.2*
Erythrocytes (10^6/µL)	10.18 ± 0.08	10.13 ± 0.07	9.92 ± 0.04*	9.60 ± 0.08**	9.53 ± 0.11**
Reticulocytes (10^3/µL)	153.9 ± 10.6	148.8 ± 10.9	134.9 ± 17.3	123.9 ± 8.0	112.2 ± 10.1
Nucleated erythrocytes /100 leukocytes	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
Howell-Jolly bodies (%) erythrocytes	0.0 ± 0.0	0.1 ± 0.0	0.1 ± 0.0	0.1 ± 0.0	0.0 ± 0.0
Mean cell volume (fL)	50.0 ± 0.2	50.1 ± 0.1	50.4 ± 0.2	51.1 ± 0.2**	51.6 ± 0.2**
Mean cell hemoglobin (pg)	15.9 ± 0.1	16.0 ± 0.0	16.1 ± 0.1	16.3 ± 0.1**	16.3 ± 0.1**
Mean cell hemoglobin concentration (g/dL)	31.8 ± 0.1	32.0 ± 0.1	31.9 ± 0.1	31.8 ± 0.1	31.7 ± 0.2
Platelets (10^3/µL)	846.7 ± 18.9	821.5 ± 30.0	835.0 ± 17.5	819.0 ± 16.9	732.4 ± 50.0
Leukocytes (10^3/µL)	2.73 ± 0.24	2.81 ± 0.23	2.33 ± 0.16	2.78 ± 0.33	2.57 ± 0.25
Segmented neutrophils (10^3/µL)	0.32 ± 0.05	0.29 ± 0.03	0.26 ± 0.02	0.31 ± 0.03	0.26 ± 0.03
Bands (10^3/µL)	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
Lymphocytes (10^3/µL)	2.35 ± 0.19	2.45 ± 0.21	1.97 ± 0.14	2.36 ± 0.32	2.25 ± 0.22
Monocytes (10^3/µL)	0.04 ± 0.01	0.03 ± 0.01	0.05 ± 0.02	0.08 ± 0.02	0.04 ± 0.02
Basophils (10^3/µL)	0.007 ± 0.002	0.010 ± 0.002	0.009 ± 0.003	0.012 ± 0.002	0.008 ± 0.004
Eosinophils (10^3/µL)	0.02 ± 0.01	0.03 ± 0.01	0.04 ± 0.01	0.03 ± 0.00	0.01 ± 0.00

* Significantly different ($P \leq 0.05$) from the chamber control group by Dunn's or Shirley's test

** $P \leq 0.01$

[a] Mean ± standard error. Statistical tests were performed on unrounded data.

APPENDIX G
ORGAN WEIGHTS
AND ORGAN-WEIGHT-TO-BODY-WEIGHT RATIOS

TABLE G1
Organ Weights and Organ-Weight-to-Body-Weight Ratios for Rats in the 2-Week Inhalation Study of 1-Bromopropane[a]

	Chamber Control	125 ppm	250 ppm	500 ppm	1,000 ppm	2,000 ppm
Male						
n	5	5	5	4	5	5
Necropsy body wt	169 ± 4	167 ± 4	169 ± 5	166 ± 6	164 ± 5	123 ± 4**
Heart						
Absolute	0.62 ± 0.01	0.61 ± 0.01	0.63 ± 0.02	0.63 ± 0.02	0.62 ± 0.02	0.56 ± 0.02
Relative	3.686 ± 0.026	3.648 ± 0.116	3.711 ± 0.032	3.779 ± 0.075	3.802 ± 0.046	4.550 ± 0.103**
R. Kidney						
Absolute	0.65 ± 0.02	0.70 ± 0.02	0.70 ± 0.02	0.72 ± 0.03	0.79 ± 0.03**	0.63 ± 0.02
Relative	3.843 ± 0.096	4.191 ± 0.076*	4.162 ± 0.074*	4.350 ± 0.095**	4.813 ± 0.091**	5.144 ± 0.090**
Liver						
Absolute	7.57 ± 0.14	7.26 ± 0.17	7.61 ± 0.22	7.98 ± 0.22	8.93 ± 0.19**	6.57 ± 0.24**
Relative	44.895 ± 0.729	43.588 ± 0.572	45.158 ± 0.735	48.215 ± 0.754**	54.535 ± 0.909**	53.600 ± 0.936**
Lung						
Absolute	1.37 ± 0.15	1.36 ± 0.13	1.26 ± 0.11	1.16 ± 0.11	1.10 ± 0.05	0.90 ± 0.05**
Relative	8.122 ± 0.878	8.241 ± 0.991	7.449 ± 0.556	7.001 ± 0.617	6.674 ± 0.131	7.365 ± 0.314
R. Testis						
Absolute	1.000 ± 0.027	1.022 ± 0.019	1.018 ± 0.033	1.016 ± 0.046	1.022 ± 0.037	0.893 ± 0.042
Relative	5.924 ± 0.075	6.142 ± 0.087	6.036 ± 0.100	6.127 ± 0.102	6.229 ± 0.142	7.265 ± 0.119**
Thymus						
Absolute	0.461 ± 0.025	0.457 ± 0.016	0.477 ± 0.028	0.523 ± 0.017	0.452 ± 0.016	0.286 ± 0.011**
Relative	2.742 ± 0.189	2.743 ± 0.087	2.826 ± 0.142	3.177 ± 0.200	2.761 ± 0.104	2.342 ± 0.103
Female						
n	5	5	5	5	5	5
Necropsy body wt	120 ± 2	124 ± 3	120 ± 4	124 ± 5	119 ± 2	107 ± 2*
Heart						
Absolute	0.48 ± 0.01	0.48 ± 0.01	0.48 ± 0.01	0.54 ± 0.01**	0.48 ± 0.01	0.51 ± 0.01
Relative	3.984 ± 0.068	3.877 ± 0.143	3.982 ± 0.081	4.325 ± 0.069	4.058 ± 0.036	4.759 ± 0.103**
R. Kidney						
Absolute	0.48 ± 0.01	0.58 ± 0.01**	0.54 ± 0.02**	0.57 ± 0.03**	0.60 ± 0.02**	0.61 ± 0.02**
Relative	3.999 ± 0.074	4.647 ± 0.116**	4.521 ± 0.036**	4.615 ± 0.067**	5.049 ± 0.107**	5.718 ± 0.161**
Liver						
Absolute	4.75 ± 0.12	5.24 ± 0.09	5.22 ± 0.28	6.01 ± 0.26**	6.19 ± 0.19**	5.82 ± 0.13**
Relative	39.548 ± 0.581	42.259 ± 0.673*	43.253 ± 0.977**	48.342 ± 0.856**	52.105 ± 1.280**	54.698 ± 0.832**
Lung						
Absolute	0.83 ± 0.03	1.07 ± 0.09*	0.91 ± 0.04	1.22 ± 0.11	0.86 ± 0.04	0.79 ± 0.02
Relative	6.915 ± 0.201	8.609 ± 0.579	7.586 ± 0.377	9.874 ± 1.027**	7.245 ± 0.304	7.395 ± 0.258
Thymus						
Absolute	0.360 ± 0.007	0.400 ± 0.012	0.365 ± 0.022	0.424 ± 0.031	0.374 ± 0.023	0.300 ± 0.013
Relative	3.000 ± 0.023	3.220 ± 0.095	3.025 ± 0.086	3.417 ± 0.203	3.138 ± 0.155	2.817 ± 0.116

* Significantly different (P≤0.05) from the chamber control group by Williams' or Dunnett's test

** P≤0.01

[a] Organ weights (absolute weights) and body weights are given in grams; organ-weight-to-body-weight ratios (relative weights) are given as mg organ weight/g body weight (mean ± standard error).

TABLE G2
Organ Weights and Organ-Weight-to-Body-Weight Ratios for Rats in the 3-Month Inhalation Study of 1-Bromopropane[a]

	Chamber Control	62.5 ppm	125 ppm	250 ppm	500 ppm	1,000 ppm
n	10	10	10	10	10	10
Male						
Necropsy body wt	345 ± 7	346 ± 5	348 ± 6	350 ± 7	344 ± 10	305 ± 6**
Heart						
Absolute	0.93 ± 0.02	0.93 ± 0.02	0.95 ± 0.01	0.93 ± 0.01	0.92 ± 0.02	0.86 ± 0.02*
Relative	2.690 ± 0.023	2.698 ± 0.027	2.727 ± 0.038	2.662 ± 0.037	2.675 ± 0.059	2.819 ± 0.034
R. Kidney						
Absolute	1.01 ± 0.02	1.02 ± 0.02	1.06 ± 0.02	1.08 ± 0.03	1.08 ± 0.07	1.02 ± 0.03
Relative	2.929 ± 0.024	2.936 ± 0.041	3.035 ± 0.019	3.102 ± 0.110	3.161 ± 0.209	3.347 ± 0.050**
Liver						
Absolute	11.21 ± 0.28	11.21 ± 0.30	11.71 ± 0.33	12.45 ± 0.31*	13.34 ± 0.43**	13.23 ± 0.36**
Relative	32.435 ± 0.387	32.318 ± 0.481	33.674 ± 0.638	35.543 ± 0.696**	38.800 ± 0.435**	43.386 ± 0.496**
Lung						
Absolute	1.73 ± 0.06	2.01 ± 0.18	1.82 ± 0.08	1.80 ± 0.04	1.69 ± 0.05	1.55 ± 0.05
Relative	5.007 ± 0.184	5.797 ± 0.465	5.257 ± 0.248	5.152 ± 0.165	4.950 ± 0.202	5.101 ± 0.123
Spleen						
Absolute	0.678 ± 0.015	0.682 ± 0.012	0.677 ± 0.015	0.689 ± 0.014	0.691 ± 0.021	0.625 ± 0.015
Relative	1.963 ± 0.024	1.970 ± 0.017	1.947 ± 0.027	1.968 ± 0.031	2.011 ± 0.030	2.053 ± 0.033
R. Testis						
Absolute	1.430 ± 0.027	1.434 ± 0.022	1.436 ± 0.034	1.405 ± 0.022	1.320 ± 0.078	1.336 ± 0.017
Relative	4.142 ± 0.050	4.147 ± 0.077	4.132 ± 0.079	4.015 ± 0.044	3.809 ± 0.170	4.396 ± 0.069
Thymus						
Absolute	0.346 ± 0.012	0.321 ± 0.010	0.327 ± 0.012	0.338 ± 0.015	0.355 ± 0.016	0.323 ± 0.015
Relative	1.002 ± 0.033	0.927 ± 0.028	0.941 ± 0.028	0.965 ± 0.042	1.029 ± 0.021	1.064 ± 0.050
Female						
Necropsy body wt	202 ± 3	203 ± 5	207 ± 4	209 ± 2	205 ± 3	190 ± 4
Heart						
Absolute	0.59 ± 0.01	0.60 ± 0.01	0.62 ± 0.01	0.63 ± 0.01	0.63 ± 0.01	0.61 ± 0.01
Relative	2.942 ± 0.064	2.978 ± 0.053	3.009 ± 0.059	3.001 ± 0.028	3.090 ± 0.058	3.207 ± 0.051**
R. Kidney						
Absolute	0.65 ± 0.01	0.66 ± 0.02	0.67 ± 0.01	0.68 ± 0.01	0.68 ± 0.01	0.71 ± 0.02**
Relative	3.209 ± 0.037	3.237 ± 0.043	3.252 ± 0.081	3.263 ± 0.045	3.298 ± 0.045	3.719 ± 0.058**
Liver						
Absolute	6.02 ± 0.09	6.38 ± 0.20	6.62 ± 0.16*	6.60 ± 0.18*	7.29 ± 0.17**	7.98 ± 0.18**
Relative	29.910 ± 0.219	31.398 ± 0.538	32.001 ± 0.573*	31.497 ± 0.754*	35.490 ± 0.666**	41.895 ± 0.398**
Lung						
Absolute	1.16 ± 0.03	1.19 ± 0.03	1.30 ± 0.06	1.24 ± 0.05	1.25 ± 0.05	1.08 ± 0.03
Relative	5.752 ± 0.126	5.904 ± 0.199	6.283 ± 0.276	5.945 ± 0.232	6.089 ± 0.252	5.658 ± 0.172
Spleen						
Absolute	0.420 ± 0.009	0.422 ± 0.010	0.446 ± 0.006*	0.461 ± 0.005*	0.451 ± 0.009*	0.439 ± 0.010*
Relative	2.085 ± 0.030	2.084 ± 0.053	2.162 ± 0.048	2.204 ± 0.030	2.200 ± 0.052	2.311 ± 0.057**
Thymus						
Absolute	0.261 ± 0.009	0.271 ± 0.008	0.290 ± 0.012	0.301 ± 0.011*	0.302 ± 0.010*	0.271 ± 0.014
Relative	1.296 ± 0.042	1.336 ± 0.039	1.397 ± 0.039	1.437 ± 0.042	1.471 ± 0.050*	1.425 ± 0.073*

* Significantly different ($P \leq 0.05$) from the chamber control group by Williams' or Dunnett's test

** ($P \leq 0.01$)

[a] Organ weights (absolute weights) and body weights are given in grams; organ-weight-to-body-weight ratios (relative weights) are given as mg organ weight/g body weight (mean ± standard error).

TABLE G3
Organ Weights and Organ-Weight-to-Body-Weight Ratios for Mice in the 2-Week Inhalation Study of 1-Bromopropane[a]

	Chamber Control	125 ppm	250 ppm	500 ppm	1,000 ppm	2,000 ppm
Male						
n	5	5	5	1	4	0[b]
Necropsy body wt	28.0 ± 0.4	28.9 ± 0.5	27.0 ± 0.4	25.8	26.6 ± 0.5	
Heart						
Absolute	0.14 ± 0.01	0.14 ± 0.01	0.12 ± 0.00	0.12	0.11 ± 0.01**	
Relative	4.995 ± 0.216	4.906 ± 0.149	4.594 ± 0.075	4.651	4.243 ± 0.232*	
R. Kidney						
Absolute	0.24 ± 0.01	0.25 ± 0.01	0.24 ± 0.00	0.24	0.24 ± 0.01	
Relative	8.641 ± 0.099	8.638 ± 0.301	8.743 ± 0.127	9.302	8.941 ± 0.161	
Liver						
Absolute	1.46 ± 0.04	1.50 ± 0.05	1.39 ± 0.02	1.44	1.65 ± 0.05*	
Relative	52.109 ± 0.653	51.841 ± 0.992	51.568 ± 0.464	55.814	61.936 ± 1.839**	
Lung						
Absolute	0.18 ± 0.01	0.19 ± 0.01	0.19 ± 0.00	0.19	0.18 ± 0.01	
Relative	6.562 ± 0.238	6.489 ± 0.291	7.116 ± 0.111	7.364	6.679 ± 0.125	
R. Testis						
Absolute	0.107 ± 0.002	0.102 ± 0.005	0.107 ± 0.002	0.109	0.103 ± 0.003	
Relative	3.807 ± 0.029	3.516 ± 0.162	3.955 ± 0.127	4.225	3.863 ± 0.149	
Thymus						
Absolute	0.059 ± 0.002	0.057 ± 0.004	0.060 ± 0.003	0.049	0.050 ± 0.006	
Relative	2.107 ± 0.080	1.960 ± 0.150	2.222 ± 0.109	1.899	1.876 ± 0.191	
Female						
n	5	5	5	5	4	3
Necropsy body wt	22.9 ± 0.2	22.2 ± 0.5	22.8 ± 0.2	23.4 ± 0.6	22.8 ± 0.5	22.7 ± 0.3
Heart						
Absolute	0.12 ± 0.00	0.12 ± 0.00	0.12 ± 0.01	0.12 ± 0.01	0.11 ± 0.01	0.12 ± 0.00
Relative	5.146 ± 0.129	5.410 ± 0.082	5.258 ± 0.256	4.990 ± 0.314	4.923 ± 0.161	5.152 ± 0.205
R. Kidney						
Absolute	0.16 ± 0.00	0.17 ± 0.00	0.18 ± 0.00*	0.17 ± 0.01*	0.19 ± 0.01**	0.19 ± 0.00**
Relative	6.977 ± 0.135	7.762 ± 0.174	7.795 ± 0.169	7.377 ± 0.315	8.355 ± 0.425**	8.235 ± 0.034**
Liver						
Absolute	1.19 ± 0.03	1.13 ± 0.03	1.23 ± 0.02	1.35 ± 0.03*	1.52 ± 0.07**	1.69 ± 0.10**
Relative	51.985 ± 0.718	51.014 ± 0.533	53.680 ± 0.826	57.791 ± 1.092**	66.739 ± 2.104**	74.471 ± 3.176**
Lung						
Absolute	0.17 ± 0.00	0.18 ± 0.01	0.19 ± 0.01	0.19 ± 0.01	0.19 ± 0.01	0.20 ± 0.01
Relative	7.506 ± 0.163	8.287 ± 0.259	8.404 ± 0.220	8.147 ± 0.370	8.348 ± 0.330	8.677 ± 0.274*
Thymus						
Absolute	0.074 ± 0.004	0.078 ± 0.004	0.077 ± 0.002	0.062 ± 0.006	0.058 ± 0.005*	0.048 ± 0.005**
Relative	3.208 ± 0.169	3.527 ± 0.111	3.389 ± 0.080	2.689 ± 0.319	2.552 ± 0.262*	2.114 ± 0.199**

* Significantly different ($P \leq 0.05$) from the chamber control group by Williams' or Dunnett's test

** $P \leq 0.01$

[a] Organ weights (absolute weights) and body weights are given in grams; organ-weight-to-body-weight ratios (relative weights) are given as mg organ weight/g body weight (mean ± standard error).

[b] No data were available for the 2,000 ppm males due to 100% mortality.

TABLE G4
Organ Weights and Organ-Weight-to-Body-Weight Ratios for Mice in the 3-Month Inhalation Study of 1-Bromopropane[a]

	Chamber Control	62.5 ppm	125 ppm	250 ppm	500 ppm
Male					
n	10	10	10	9	6
Necropsy body wt	39.6 ± 0.8	39.0 ± 0.9	39.6 ± 0.6	37.9 ± 0.7	37.4 ± 1.4
Heart					
Absolute	0.16 ± 0.00	0.16 ± 0.00	0.16 ± 0.00	0.15 ± 0.00	0.16 ± 0.00
Relative	4.051 ± 0.054	4.014 ± 0.050	4.023 ± 0.049	4.080 ± 0.058	4.210 ± 0.148
R. Kidney					
Absolute	0.32 ± 0.01	0.31 ± 0.01	0.30 ± 0.00	0.29 ± 0.01**	0.28 ± 0.01**
Relative	8.133 ± 0.216	7.947 ± 0.123	7.669 ± 0.117	7.687 ± 0.208	7.458 ± 0.258*
Liver					
Absolute	1.68 ± 0.05	1.66 ± 0.05	1.71 ± 0.03	1.71 ± 0.03	1.80 ± 0.08
Relative	42.485 ± 0.503	42.584 ± 0.641	43.288 ± 0.379	45.129 ± 0.393**	48.038 ± 1.126**
Lung					
Absolute	0.21 ± 0.01	0.21 ± 0.01	0.23 ± 0.01	0.21 ± 0.00	0.21 ± 0.01
Relative	5.236 ± 0.147	5.477 ± 0.168	5.734 ± 0.259	5.505 ± 0.167	5.742 ± 0.302
Spleen					
Absolute	0.067 ± 0.002	0.067 ± 0.002	0.069 ± 0.002	0.067 ± 0.002	0.063 ± 0.002
Relative	1.696 ± 0.051	1.724 ± 0.050	1.743 ± 0.032	1.763 ± 0.051	1.699 ± 0.069
R. Testis					
Absolute	0.116 ± 0.003	0.120 ± 0.002	0.108 ± 0.009	0.114 ± 0.002	0.115 ± 0.001
Relative	2.933 ± 0.053	3.099 ± 0.051	2.747 ± 0.226	3.014 ± 0.050	3.081 ± 0.098
Thymus					
Absolute	0.049 ± 0.005	0.045 ± 0.004	0.047 ± 0.004	0.049 ± 0.003	0.056 ± 0.006
Relative	1.236 ± 0.107	1.154 ± 0.084	1.192 ± 0.087	1.277 ± 0.060	1.481 ± 0.104
Female					
n	10	10	10	10	5
Necropsy body wt	30.9 ± 0.7	33.0 ± 1.3	31.9 ± 0.8	30.2 ± 0.9	31.3 ± 1.4
Heart					
Absolute	0.14 ± 0.00	0.14 ± 0.00	0.14 ± 0.00	0.14 ± 0.01	0.14 ± 0.01
Relative	4.378 ± 0.087	4.332 ± 0.112	4.404 ± 0.078	4.651 ± 0.110	4.365 ± 0.210
R. Kidney					
Absolute	0.20 ± 0.00	0.20 ± 0.00	0.21 ± 0.01	0.20 ± 0.01	0.22 ± 0.01**
Relative	6.330 ± 0.155	6.199 ± 0.179	6.552 ± 0.184	6.780 ± 0.164	7.187 ± 0.265**
Liver					
Absolute	1.42 ± 0.03	1.49 ± 0.06	1.51 ± 0.03	1.49 ± 0.04	1.65 ± 0.11*
Relative	46.102 ± 0.767	45.104 ± 0.424	47.586 ± 0.880	49.377 ± 0.640**	52.666 ± 1.904**
Lung					
Absolute	0.20 ± 0.01	0.22 ± 0.01	0.22 ± 0.01	0.21 ± 0.01	0.26 ± 0.02**
Relative	6.518 ± 0.276	6.642 ± 0.298	7.018 ± 0.222	7.028 ± 0.190	8.497 ± 0.929**
Spleen					
Absolute	0.093 ± 0.002	0.098 ± 0.003	0.096 ± 0.004	0.092 ± 0.004	0.082 ± 0.007
Relative	3.019 ± 0.073	2.980 ± 0.072	3.006 ± 0.079	3.060 ± 0.114	2.643 ± 0.257
Thymus					
Absolute	0.058 ± 0.005	0.058 ± 0.003	0.054 ± 0.002	0.054 ± 0.002	0.055 ± 0.004
Relative	1.861 ± 0.145	1.749 ± 0.081	1.692 ± 0.073	1.797 ± 0.062	1.767 ± 0.138

* Significantly different ($P \leq 0.05$) from the chamber control group by Williams' or Dunnett's test

** $P \leq 0.01$

[a] Organ weights (absolute weights) and body weights are given in grams; organ-weight-to-body-weight ratios (relative weights) are given as mg organ weight/g body weight (mean ± standard error).

APPENDIX H
REPRODUCTIVE TISSUE EVALUATIONS AND ESTROUS CYCLE CHARACTERIZATION

TABLE H1
Summary of Reproductive Tissue Evaluations for Male Rats in the 3-Month Inhalation Study of 1-Bromopropane[a]

	Chamber Control	250 ppm	500 ppm	1,000 ppm
n	10	10	10	10
Weights (g)				
Necropsy body wt	345 ± 7	350 ± 7	344 ± 10	305 ± 6**
L. Cauda epididymis	0.1865 ± 0.0039	0.1929 ± 0.0038	0.1740 ± 0.0106	0.1603 ± 0.0067*
L. Epididymis	0.4862 ± 0.0081	0.4994 ± 0.0042	0.4622 ± 0.0192	0.4423 ± 0.0083*
L. Testis	1.5128 ± 0.0313	1.4833 ± 0.0252	1.3867 ± 0.0839	1.4104 ± 0.0269
Spermatid measurement				
Spermatid heads (10^6/g testis)	123.6 ± 2.3	129.9 ± 6.1	112.9 ± 12.3	130.5 ± 3.8
Spermatid heads (10^6/testis)	171.8 ± 4.6	172.0 ± 9.2	150.8 ± 16.9	169.5 ± 6.0
Epididymal spermatozoal measurements				
Motility (%)	90.99 ± 0.83	84.87 ± 0.94**	81.84 ± 0.80**[b]	65.82 ± 3.17**
Sperm (10^6/g cauda epididymis)	699 ± 26	638 ± 34	576 ± 63	523 ± 30**
Sperm (10^6/cauda epididymis)	130.27 ± 5.19	122.90 ± 6.63	105.27 ± 12.01	82.39 ± 2.51**

* Significantly different (P≤0.05) from the chamber control group by Williams' test (cauda epididymis and epididymis weights)

** Significantly different (P≤0.01) from the chamber control group by Williams' test (body weights) or Shirley's test (epididymal spermatozoal measurements)

[a] Data are presented as mean ± standard error. Differences from the chamber control group are not significant by Dunnett's test (testis weights) or Dunn's test (spermatid measurements).

[b] n=9

TABLE H2
Estrous Cycle Characterization for Female Rats in the 3-Month Inhalation Study of 1-Bromopropane[a]

	Chamber Control	250 ppm	500 ppm	1,000 ppm
Number weighed at necropsy	10	10	10	10
Necropsy body wt (g)	202 ± 3	209 ± 2	205 ± 3	190 ± 4
Proportion of regular cycling females[b]	9/10	10/10	10/10	10/10
Estrous cycle length (days)	4.9 ± 0.11[c]	5.0 ± 0.05	4.9 ± 0.07	5.0 ± 0.05
Estrous stages (% of cycle)[d]				
Diestrus	55.8	43.3	40.0	41.7
Proestrus	17.5	5.0	4.2	6.7
Estrus	20.8	32.5	36.7	33.3
Metestrus	5.0	19.2	19.2	18.3
Uncertain diagnoses	0.8	0.0	0.0	0.0

[a] Necropsy body weights and estrous cycle length data are presented as mean ± standard error. Differences from the chamber control group are not significant by Dunnett's test (body weight) or Dunn's test (estrous cycle length).

[b] Number of females with a regular cycle/number of females cycling

[c] Estrous cycle was longer than 12 days or unclear in 1 of 10 animals.

[d] Evidence shows that females exposed to 250, 500, or 1,000 ppm differ significantly (Wilk's Criterion, P≤0.05) from the chamber control females in the relative length of time spent in the estrous stages. The tests for equality of transition probability matrices among exposed groups and between the chamber control group and each exposed group indicated that exposed females spent significantly more time in extended estrus (P<0.001) and significantly less time in extended diestrus (P<0.005) than the chamber control group.

TABLE H3
Summary of Reproductive Tissue Evaluations for Male Mice in the 3-Month Inhalation Study of 1-Bromopropane[a]

	Chamber Control	125 ppm	250 ppm	500 ppm
n	10	10	9	6
Weights (g)				
Necropsy body wt	39.6 ± 0.8	39.6 ± 0.6	37.9 ± 0.7	37.4 ± 1.4
L. Cauda epididymis	0.0164 ± 0.0008	0.0170 ± 0.0009	0.0179 ± 0.0005	0.0192 ± 0.0007
L. Epididymis	0.0538 ± 0.0010	0.0492 ± 0.0016*	0.0532 ± 0.0014	0.0535 ± 0.0012
L. Testis	0.1137 ± 0.0020	0.1063 ± 0.0085	0.1104 ± 0.0028	0.1103 ± 0.0010
Spermatid measurement				
Spermatid heads (10^6/g testis)	188.7 ± 8.1	164.9 ± 19.7	180.5 ± 3.7	199.3 ± 8.0
Spermatid heads (10^6/testis)	19.83 ± 0.89	17.57 ± 2.09	18.17 ± 0.48	20.11 ± 0.59
Epididymal spermatozoal measurements				
Motility (%)	91.57 ± 0.73	89.70 ± 0.66[b]	87.78 ± 1.10*	88.13 ± 0.56**
Sperm (10^6/g cauda epididymis)	1,237 ± 60	1,204 ± 66[b]	1,062 ± 70	890 ± 68**
Sperm (10^6/cauda epididymis)	19.93 ± 0.47	19.66 ± 0.69[b]	18.92 ± 0.97	16.93 ± 1.15

* Significantly different (P≤0.05) from the chamber control group by Dunnett's test (epididymis weights) or Shirley's test (sperm motility)

** Significantly different (P≤0.01) from the chamber control group by Shirley's test

[a] Data are presented as mean ± standard error. Differences from the chamber control group are not significant by Dunnett's test (body, cauda epididymis, and testis weights) or Dunn's test (spermatid measurements and sperm/cauda epididymis).

[b] n=9

TABLE H4
Estrous Cycle Characterization for Female Mice in the 3-Month Inhalation Study of 1-Bromopropane[a]

	Chamber Control	125 ppm	250 ppm	500 ppm
Number weighed at necropsy	10	10	10	5
Necropsy body wt (g)	30.9 ± 0.7	31.9 ± 0.8	30.2 ± 0.9	31.3 ± 1.4
Proportion of regular cycling females[b]	9/10	8/10	6/10	5/5
Estrous cycle length (days)	4.2 ± 0.11	4.6 ± 0.50	4.2 ± 0.08	4.6 ± 0.10*
Estrous stages (% of cycle)[c]				
Diestrus	27.5	27.5	25.8	41.7
Proestrus	0.0	0.0	0.0	0.0
Estrus	49.2	48.3	52.5	41.7
Metestrus	23.3	24.2	21.7	16.7

* Significantly different (P≤0.05) from the chamber control group by Dunn's test

[a] Necropsy body weights and estrous cycle length data are presented as mean ± standard error. Differences from the chamber control group are not significant by Dunnett's test (body weight).

[b] Number of females with a regular cycle/number of females cycling

[c] Evidence shows that females exposed to 500 ppm differ significantly (Wilk's Criterion, P≤0.05) from the chamber control females in the relative length of time spent in the estrous stages. The tests for equality of transition probability matrices among exposed groups and between the chamber control group and each exposed group indicated that 500 ppm females spent significantly more time in extended diestrus (P=0.035) and 250 ppm females spent significantly more time in extended estrus (P<0.001) than the chamber control group.

APPENDIX I
CHEMICAL CHARACTERIZATION AND GENERATION OF CHAMBER CONCENTRATIONS

CHEMICAL CHARACTERIZATION AND GENERATION OF CHAMBER CONCENTRATIONS

PROCUREMENT AND CHARACTERIZATION OF 1-BROMOPROPANE

1-Bromopropane was obtained in 55-gallon metal drums from Diaz Chemical Corporation (Holley, NY) in one lot (106-015) and from Albemarle PCC (Thann, France) in one lot (1581313004). Lot 106-015 was used in the 2-week and 3-month studies, and lot 1581313004 was used during the 2-year studies. Identity and purity analyses were conducted by the study laboratory at Battelle Toxicology Northwest (Richland, WA), and by Chemir/Polytech Laboratories, Inc. (Maryland Heights, MO). Reports on analyses performed in support of the 1-bromopropane studies are on file at the National Institute of Environmental Health Sciences.

Both lots of the chemical, a colorless to pale yellow liquid with a strong, characteristic odor, were identified as 1-bromopropane by Chemir/Polytech Laboratories, Inc., by infrared (IR) and ^{1}H-nuclear magnetic resonance (NMR) spectroscopy. All spectra were consistent with the literature reference spectra (*Aldrich*, 1993, 1997) of 1-bromopropane. Representative IR and ^{1}H-NMR spectra are presented in Figures I1 and I2, respectively.

The purity of each lot was determined by the study laboratory using gas chromatography (GC) by system A (Table I1). In addition, Chemir/Polytech Laboratories, Inc., determined the moisture content of each lot by Karl Fischer titration and the purity of each lot by elemental and hydrogen bromide (HBr) analyses. The analyses of the bulk chemical for HBr were performed by volumetric extraction of the samples with deionized water subsequent to an analysis of the extractables for free bromide ion using ion chromatography (IC). IC was performed on a Dionex DX100 ion chromatograph (Dionex Corporation, Sunnyvale, CA) equipped with a conductivity detector and IonPac (Dionex) column.

For lot 106-015, Karl Fischer titration indicated a water content of 39 ppm. Elemental analyses for carbon, hydrogen, nitrogen, and sulfur were in agreement with the theoretical values for 1-bromopropane; residual HBr was determined to be 1.3 ppm. GC indicated one major peak and three impurities with areas exceeding 0.1% of the total peak area; these peaks matched the retention times for prepared standards of 1-propanol (0.14%), 2-bromopropane (0.11%), and di-*n*-propyl ether (0.74%). Measured purity of the bulk chemical was consistent throughout the sampled metal drum, and the overall purity of lot 106-015 was determined to be approximately 99%.

For lot 1581313004, Karl Fischer titration indicated a water content of 119 ppm. Elemental analyses for carbon, hydrogen, nitrogen, and sulfur were in agreement with the theoretical values for 1-bromopropane; residual HBr was determined to be 61.6 ppm. GC detected one major peak and no impurities with areas greater than 0.1% of the total peak area. Three impurities identified with prepared standards were 1-propanol (0.03%), 2-bromopropane (0.02%), and di-*n*-propyl ether (0.02%). Measured purity of the bulk chemical was consistent throughout the sampled metal drums, and the overall purity of lot 1581313004 was determined to be 99.9% or greater.

To ensure stability, the bulk chemical was stored at controlled room temperature in the original shipping containers (55-gallon metal drums). Periodic reanalyses of the bulk chemical were performed during the 2-week, 3-month, and 2-year studies by the study laboratory using GC by system A, and no degradation of the bulk chemical was detected.

VAPOR GENERATION AND EXPOSURE SYSTEM

A diagram of the 1-bromopropane vapor generation and delivery system used in the studies is shown in Figure I3. 1-Bromopropane was pumped through a preheater (for the 2-week and 3-month studies) and into a heated glass column filled with glass beads that increased the surface area for vaporization. Heated nitrogen entered the column from below and assisted in vaporizing the chemical while conveying it into a short distribution manifold. Concentration in the manifold was determined by the chemical pump rate and nitrogen flow rate. The pressure in the distribution manifold was kept fixed to ensure constant flow through the manifold and into all chambers as the flow of vapor to each chamber was adjusted.

Metering valves at the manifold controlled flow to each chamber through individual Teflon® delivery lines that carried the vapor from the manifold to three-way exposure valves at the chamber inlets. The exposure valves diverted vapor delivery to exposure chamber exhaust until the generation system was stable and exposures were ready to proceed. To initiate exposure, the chamber exposure valves were rotated to allow the 1-bromopropane vapor to flow to each exposure chamber inlet duct where it was further diluted with filtered, conditioned air to achieve the desired exposure concentration.

The study laboratory designed the inhalation exposure chamber (Harford Systems Division of Lab Products, Inc., Aberdeen, MD) so that uniform vapor concentrations could be maintained throughout the chamber with the catch pans in place. The total active mixing volume of each chamber was 1.7 m^3. A small particle detector (Model 3022A, TSI Incorporated, St. Paul, MN) was used with and without animals in the exposure chambers to ensure that 1-bromopropane vapor, and not aerosol, was produced. No particle counts above the minimum resolvable level (approximately 200 particles/cm^3) were detected.

VAPOR CONCENTRATION MONITORING

Summaries of the chamber vapor concentrations are given in Tables I2 through I4. Chamber and room concentrations of 1-bromopropane were monitored by an on-line gas chromatograph (system B, Table I1). Samples were drawn from each exposure chamber approximately every 20 (2-week and 3-month studies) or 30 (2-year studies) minutes during each 6-hour exposure period using Hastelloy®-C stream-select and gas-sampling valves (VALCO Instruments Company, Houston, TX) in a separate, heated valve oven. The sample lines composing each sample loop were made from Teflon® tubing and were connected to the exposure chamber relative humidity sampling lines at a location close to the gas chromatograph. A vacuum regulator maintained a constant vacuum in the sample loop to compensate for variations in sample line pressure. An in-line flow meter between the vacuum regulator and the gas chromatograph allowed digital measurement of sample flow.

The on-line gas chromatograph was checked throughout the day for instrument drift against an on-line standard vapor of 1-bromopropane in nitrogen supplied by a standard generator (Kin-Tek; Precision Calibration Systems, La Marque, TX). The on-line gas chromatograph was recalibrated as required to meet acceptance criteria. Calibration was performed by a comparison of chamber concentration data to data from grab samples that were collected with activated coconut charcoal gas sampling tubes (ORBO™-32; Supelco, Bellefonte, PA), extracted with methylene chloride containing 1-bromobutane as an internal standard, and analyzed using an off-line gas chromatograph (system C). Known volumes of chamber atmosphere were sampled at a constant flow rate ensured by a calibrated critical orifice. The off-line gas chromatograph was calibrated with gravimetrically prepared standards of 1-bromopropane containing 1-bromobutane as an internal standard in methylene chloride.

CHAMBER ATMOSPHERE CHARACTERIZATION

Buildup and decay rates for chamber vapor concentrations were determined with and without animals present in the chambers. At a chamber airflow rate of 15 air changes per hour, the theoretical value for the time to achieve 90% of the target concentration after the beginning of vapor generation (T_{90}) and the time for the chamber concentration to decay to 10% of the target concentration after vapor generation was terminated (T_{10}) was approximately 12.5 minutes. For rats and mice in the 2-week studies, T_{90} and T_{10} values ranged from 9 to 10 minutes with animals present. For rats and mice in the 3-month studies, T_{90} values averaged 9 minutes without animals present and ranged from 9 to 10 minutes with animals present; T_{10} values ranged from 8 to 9 minutes without animals present and from 9 to 11 minutes with animals present. For rats and mice in the 2-year studies, T_{90} values ranged from 9 to 10 minutes without animals present and from 9 to 11 minutes with animals present; T_{10} values ranged from 9 to 10 minutes without animals present and from 10 to 11 minutes with animals present. A T_{90} value of 12 minutes was selected for the 2-week studies, and a T_{90} value of 10 minutes was selected for the 3-month and 2-year studies.

The uniformity of vapor concentration in the inhalation exposure chambers without animals present was evaluated before the 3-month and 2-year studies began; in addition, concentration uniformity with animals present in the chambers was measured once during the 2-week studies, once during the 3-month studies, and quarterly during the 2-year studies. The vapor concentration was measured using the on-line gas chromatograph (system B, Table I1) with the stream-selection valve fixed in one position to allow continuous monitoring from a single input line.

During the 2-week studies and prior to the 3-month and 2-year studies, concentrations were measured at 12 chamber positions, one in front and one in back for each of the six possible animal cage unit positions per chamber. During the 3-month and 2-year studies, concentrations were measured at the regular monitoring port and from sample ports at levels where animals were present. Chamber concentration uniformity was maintained throughout the studies.

The persistence of 1-bromopropane in the chambers after vapor delivery ended was determined by monitoring the vapor concentration in the 2,000 ppm chambers in the 2-week studies, the 500 ppm (rats and mice) and 1,000 ppm (rats) chambers in the 3-month studies, and the 250 ppm (mice) and 500 ppm (rats) chambers in the 2-year studies, with (all studies) and without (3-month and 2-year studies) animals present in the chambers. In the 2-week studies, the concentration decreased to 1% of the target concentration within 19 minutes with animals present. In the 3-month studies, the concentration decreased to 1% of the target concentration within 20 minutes without animals present and within 25 (rat and mouse chambers) and 21 (rat chamber) minutes with animals present. In the 2-year studies, the concentration decreased to 1% of the target concentration within 19 (rats) and 20 (mice) minutes without animals present and within 25 (rats) and 22 (mice) minutes with animals present.

Samples of the test atmosphere from the distribution lines and low and high exposure concentration chambers were collected prior to the 3-month and 2-year studies and also at the beginning and end of one generation day during the 2-week, 3-month, and 2-year studies; the atmosphere samples were collected with adsorbent gas sampling tubes containing activated charcoal (ORBO™-32), followed by a tube containing silica gel (ORBO™-52; Supelco, Inc.) and extracted with methylene chloride. Additional samples were collected from the generator reservoir, and all of the samples were analyzed using GC by system A to measure the stability and purity of 1-bromopropane in the generation and delivery system. To assess whether impurities or degradation products co-eluted with 1-bromopropane or the solvent, a second GC analysis of the samples was performed using a polar column capable of resolving compounds with similar boiling points and polarities (system D). Fourier transform IR was used to determine if HBr was present in the atmosphere samples; spectra were generated using a MIDAC I-1101 spectrometer with a 9.5 m path-length gas cell (MIDAC Corporation, Irvine, CA) and were compared to those of prepared HBr standards.

No evidence of degradation of 1-bromopropane was noted in any part of the exposure system. Three impurity peaks with areas greater than 0.1% of the total peak area were consistently detected in atmosphere and generator reservoir samples collected prior to the 3-month studies and during the 2-week and 3-month studies. These peaks matched the retention times for prepared standards of 1-propanol, 2-bromopropane, and di-*n*-propyl ether and had area percent values similar to those measured during the initial bulk purity analyses of lot 106-015. No impurity peaks were detected in atmosphere or generator reservoir samples collected prior to or during the 2-year studies. Using the polar column, no additional impurities were detected in any of the atmosphere or generator reservoir samples collected prior to or during any of the studies. HBr levels in the exposure atmosphere were determined to be less than 0.07% during the 2-week studies and less than 1% prior to and during the 3-month and 2-year studies.

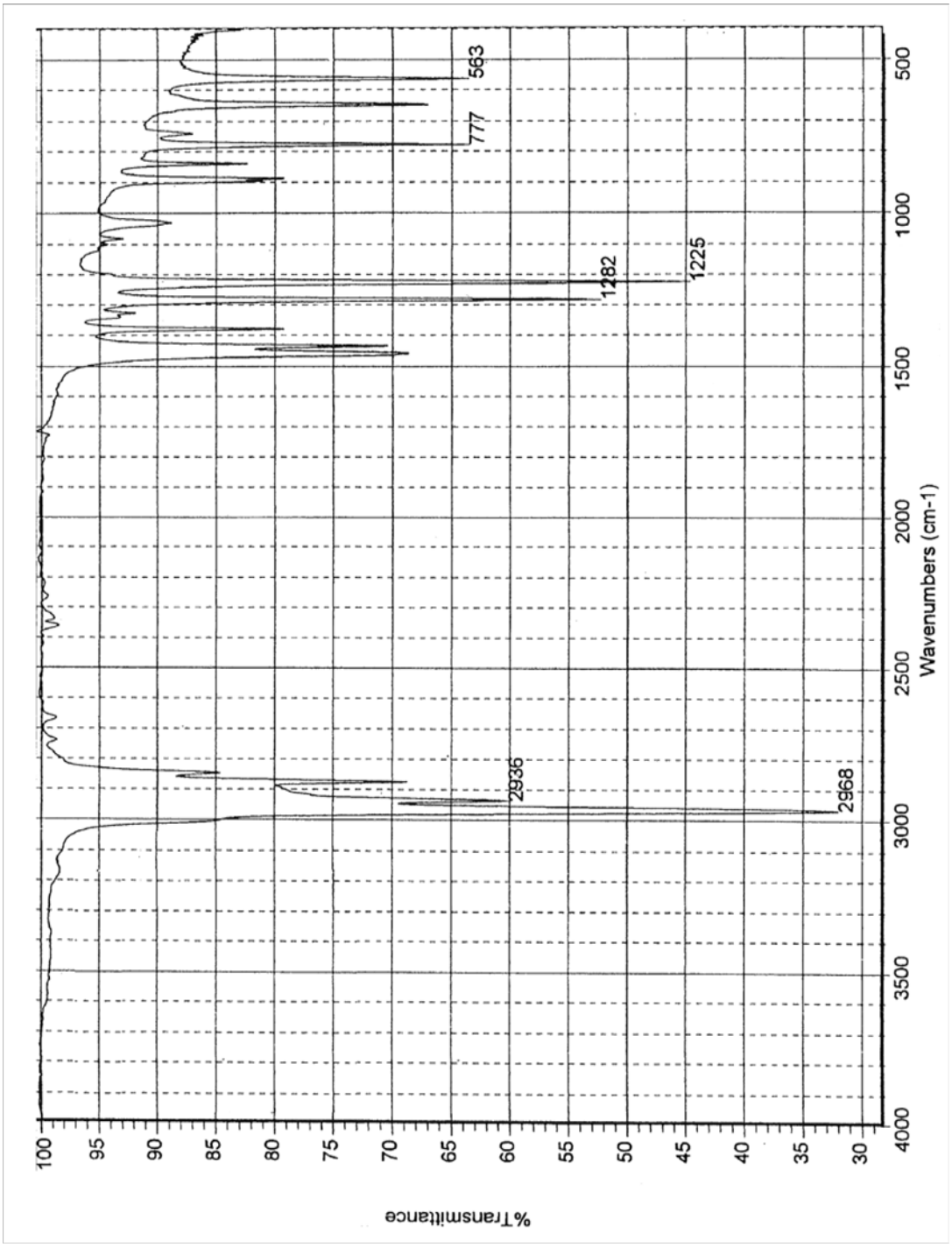

FIGURE I1
Infrared Absorption Spectrum of 1-Bromopropane

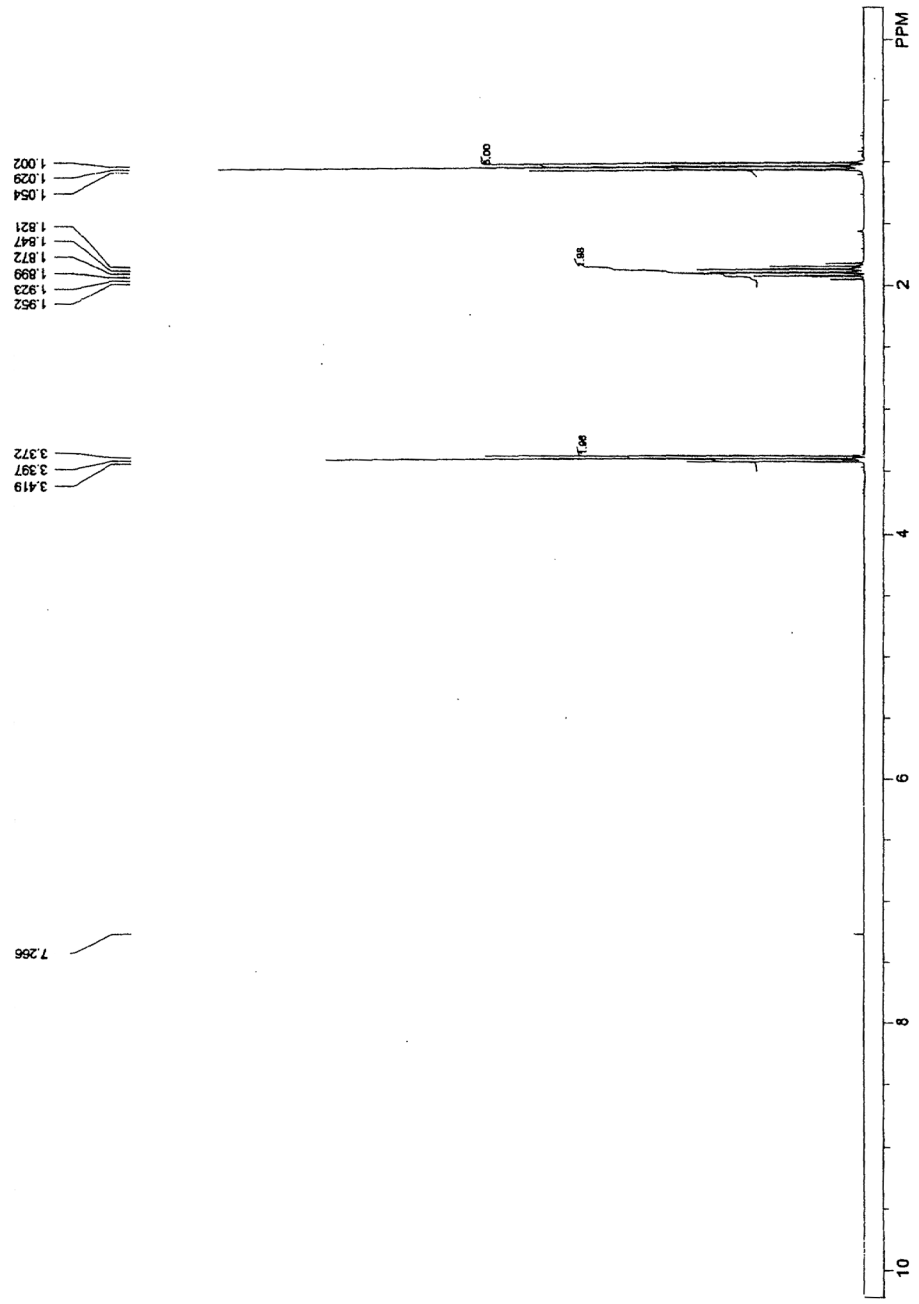

FIGURE I2
^{1}H-Nuclear Magnetic Resonance Spectrum of 1-Bromopropane

TABLE I1
Gas Chromatography Systems Used in the Inhalation Studies of 1-Bromopropane[a]

Detection System	Column	Carrier Gas	Oven Temperature Program
System A			
Flame ionization	DB-5, 30 m × 0.53 mm, 5.0-µm film thickness (J&W Scientific, Folsom, CA) or Rtx-5, 30 m × 0.53 mm, 5.0-µm film thickness (Restek, Bellefonte, PA)	Helium at 4 psi head pressure	35° C for 3 minutes, then 3° C/minute to 65° C, then 7° C/minute to 260° C, held for 1 minute
System B			
Flame ionization	DB-5, 15 m × 0.53 mm, 1.5-µm film thickness (J&W Scientific)	Nitrogen at 17 mL/minute	Isothermal at 35° C
System C			
Flame ionization	DB-5, 30 m × 0.53 mm, 5.0-µm film thickness (J&W Scientific) or Rtx-5, 30 m × 0.53 mm, 5.0-µm film thickness (Restek)	Helium at 6 psi head pressure	40° C for 1 minute, then 6° C/minute to 95° C, then 15° C/minute to 150° C, held for 2 minutes
System D			
Flame ionization	DB-Wax, 30 m × 0.53 mm, 1.0-µm film thickness (J&W Scientific)	Helium	40° C for 1 minute, then 6° C/minute to 95° C, then 15° C/minute to 150° C, held for 2 minutes

[a] The gas chromatographs were manufactured by Hewlett-Packard (Palo Alto, CA).

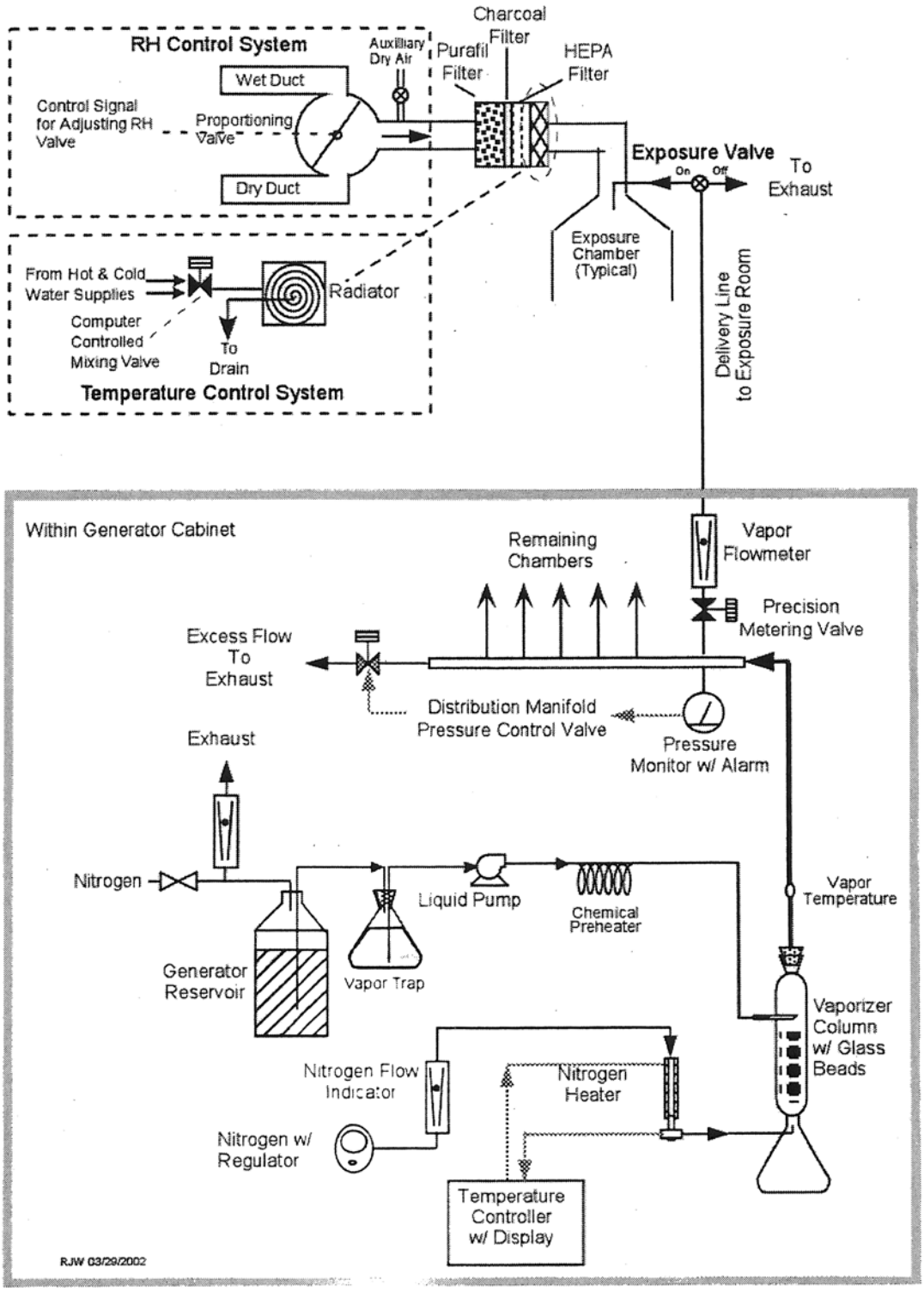

FIGURE I3
Schematic of the Vapor Generation and Delivery System in the Inhalation Studies of 1-Bromopropane

TABLE I2
Summary of Chamber Concentrations in the 2-Week Inhalation Studies of 1-Bromopropane

	Target Concentration (ppm)	Total Number of Readings	Average Concentration[a] (ppm)
Rat Chambers			
	125	225	127 ± 2
	250	226	252 ± 3
	500	227	499 ± 4
	1,000	226	1,015 ± 12
	2,000	228	2,028 ± 17
Mouse Chambers			
	125	245	127 ± 2
	250	246	251 ± 3
	500	247	499 ± 3
	1,000	246	1,013 ± 13
	2,000	248	2,027 ± 17

[a] Mean ± standard deviation

TABLE I3
Summary of Chamber Concentrations in the 3-Month Inhalation Studies of 1-Bromopropane

	Target Concentration (ppm)	Total Number of Readings	Average Concentration[a] (ppm)
Rat Chambers			
	62.5	1,222	62.2 ± 1.2
	125	1,237	124 ± 2
	250	1,247	247 ± 4
	500	1,241	497 ± 8
	1,000	1,243	1,005 ± 15
Mouse Chambers			
	62.5	1,264	62.1 ± 1.2
	125	1,277	124 ± 2
	250	1,287	247 ± 4
	500	1,281	497 ± 8

[a] Mean ± standard deviation

TABLE I4
Summary of Chamber Concentrations in the 2-Year Inhalation Studies of 1-Bromopropane

	Target Concentration (ppm)	Total Number of Readings	Average Concentration[a] (ppm)
Rat Chambers			
	125	8,018	125 ± 3
	250	8,011	250 ± 6
	500	8,042	500 ± 11
Mouse Chambers			
	62.5	8,494	62.5 ± 1.6
	125	8,009	125 ± 3
	250	8,038	250 ± 5

[a] Mean ± standard deviation

APPENDIX J
INGREDIENTS, NUTRIENT COMPOSITION, AND CONTAMINANT LEVELS IN NTP-2000 RAT AND MOUSE RATION

TABLE J1
Ingredients of NTP-2000 Rat and Mouse Ration

Ingredients	Percent by Weight
Ground hard winter wheat	22.26
Ground #2 yellow shelled corn	22.18
Wheat middlings	15.0
Oat hulls	8.5
Alfalfa meal (dehydrated, 17% protein)	7.5
Purified cellulose	5.5
Soybean meal (49% protein)	5.0
Fish meal (60% protein)	4.0
Corn oil (without preservatives)	3.0
Soy oil (without preservatives)	3.0
Dried brewer's yeast	1.0
Calcium carbonate (USP)	0.9
Vitamin premix[a]	0.5
Mineral premix[b]	0.5
Calcium phosphate, dibasic (USP)	0.4
Sodium chloride	0.3
Choline chloride (70% choline)	0.26
Methionine	0.2

[a] Wheat middlings as carrier
[b] Calcium carbonate as carrier

TABLE J2
Vitamins and Minerals in NTP-2000 Rat and Mouse Ration[a]

	Amount	Source
Vitamins		
A	4,000 IU	Stabilized vitamin A palmitate or acetate
D	1,000 IU	D-activated animal sterol
K	1.0 mg	Menadione sodium bisulfite complex
α-Tocopheryl acetate	100 IU	
Niacin	23 mg	
Folic acid	1.1 mg	
d-Pantothenic acid	10 mg	*d*-Calcium pantothenate
Riboflavin	3.3 mg	
Thiamine	4 mg	Thiamine mononitrate
B_{12}	52 µg	
Pyridoxine	6.3 mg	Pyridoxine hydrochloride
Biotin	0.2 mg	*d*-Biotin
Minerals		
Magnesium	514 mg	Magnesium oxide
Iron	35 mg	Iron sulfate
Zinc	12 mg	Zinc oxide
Manganese	10 mg	Manganese oxide
Copper	2.0 mg	Copper sulfate
Iodine	0.2 mg	Calcium iodate
Chromium	0.2 mg	Chromium acetate

[a] Per kg of finished product

TABLE J3
Nutrient Composition of NTP-2000 Rat and Mouse Ration

Nutrient	Mean ± Standard Deviation	Range	Number of Samples
Protein (% by weight)	14.8 ± 0.65	13.7 – 16.1	24
Crude fat (% by weight)	8.1 ± 0.36	7.4 – 9.0	24
Crude fiber (% by weight)	9.2 ± 0.45	8.2 – 9.9	24
Ash (% by weight)	5.0 ± 0.25	4.4 – 5.4	24
Amino Acids (% of total diet)			
Arginine	0.770 ± 0.070	0.670 – 0.970	18
Cystine	0.225 ± 0.023	0.150 – 0.250	18
Glycine	0.706 ± 0.043	0.620 – 0.800	18
Histidine	0.362 ± 0.082	0.310 – 0.680	18
Isoleucine	0.524 ± 0.046	0.430 – 0.660	18
Leucine	1.087 ± 0.066	0.960 – 1.240	18
Lysine	0.712 ± 0.118	0.310 – 0.840	18
Methionine	0.407 ± 0.051	0.260 – 0.490	18
Phenylalanine	0.626 ± 0.043	0.540 – 0.720	18
Threonine	0.500 ± 0.046	0.430 – 0.610	18
Tryptophan	0.142 ± 0.024	0.110 – 0.200	18
Tyrosine	0.388 ± 0.058	0.280 – 0.540	18
Valine	0.667 ± 0.045	0.550 – 0.730	18
Essential Fatty Acids (% of total diet)			
Linoleic	3.92 ± 0.243	3.49 – 4.54	18
Linolenic	0.30 ± 0.035	0.21 – 0.35	18
Vitamins			
Vitamin A (IU/kg)	4,870 ± 121	3,230 – 8,900	24
Vitamin D (IU/kg)	1,000[a]		
α-Tocopherol (ppm)	84.2 ± 16.60	52.0 – 110.0	15
Thiamine (ppm)[b]	9.0 ± 3.71	6.4 – 25.2	24
Riboflavin (ppm)	6.8 ± 2.11	4.20 – 11.20	15
Niacin (ppm)	79.0 ± 10.50	66.4 – 98.2	15
Pantothenic acid (ppm)	23.9 ± 3.73	17.4 – 29.8	15
Pyridoxine (ppm)[b]	9.21 ± 2.20	6.4 – 13.7	15
Folic acid (ppm)	1.75 ± 0.54	1.20 – 3.27	15
Biotin (ppm)	0.332 ± 0.12	0.225 – 0.704	15
Vitamin B_{12} (ppb)	60.5 ± 46.5	18.3 – 174.0	15
Choline (ppm)[b]	3,064 ± 270	2,700 – 3,790	15
Minerals			
Calcium (%)	0.964 ± 0.043	0.884 – 1.030	24
Phosphorus (%)	0.578 ± 0.026	0.535 – 0.623	24
Potassium (%)	0.665 ± 0.023	0.626 – 0.694	15
Chloride (%)	0.376 ± 0.041	0.300 – 0.474	15
Sodium (%)	0.191 ± 0.017	0.160 – 0.222	15
Magnesium (%)	0.201 ± 0.009	0.185 – 0.217	15
Sulfur (%)	0.170 ± 0.029	0.116 – 0.209	15
Iron (ppm)	182 ± 46.7	135 – 311	15
Manganese (ppm)	54.1 ± 7.89	42.1 – 73.1	15
Zinc (ppm)	55.0 ± 9.55	43.3 – 78.5	15
Copper (ppm)	6.65 ± 1.790	3.21 – 10.50	15
Iodine (ppm)	0.512 ± 0.221	0.233 – 0.972	15
Chromium (ppm)	0.604 ± 0.253	0.330 – 1.380	14
Cobalt (ppm)	0.25 ± 0.074	0.20 – 0.47	14

[a] From formulation
[b] As hydrochloride (thiamine and pyridoxine) or chloride (choline)

TABLE J4
Contaminant Levels in NTP-2000 Rat and Mouse Ration[a]

	Mean ± Standard Deviation[b]	Range	Number of Samples
Contaminants			
Arsenic (ppm)	0.25 ± 0.120	0.14 – 0.50	24
Cadmium (ppm)	0.07 ± 0.022	0.04 – 0.10	24
Lead (ppm)	0.09 ± 0.023	0.05 – 0.13	24
Mercury (ppm)	<0.02		24
Selenium (ppm)	0.21 ± 0.058	0.16 – 0.45	24
Aflatoxins (ppb)	<5.00		24
Nitrate nitrogen (ppm)[c]	13.1 ± 4.17	7.89 – 24.4	24
Nitrite nitrogen (ppm)[c]	<0.61		24
BHA (ppm)[d]	<1.0		24
BHT (ppm)[d]	<1.0		24
Aerobic plate count (CFU/g)	10 ± 0.0	10	24
Coliform (MPN/g)	3.0 ± 0.0	3.0	24
Escherichia coli (MPN/g)	<10		24
Salmonella (MPN/g)	Negative		24
Total nitrosoamines (ppb)[e]	4.4 ± 1.73	2.3 – 8.5	24
N-Nitrosodimethylamine (ppb)[e]	2.4 ± 1.29	1.1 – 5.6	24
N-Nitrosopyrrolidine (ppb)[e]	1.9 ± 0.80	0.9 – 4.1	24
Pesticides (ppm)			
α-BHC	<0.01		24
β-BHC	<0.02		24
γ-BHC	<0.01		24
δ-BHC	<0.01		24
Heptachlor	<0.01		24
Aldrin	<0.01		24
Heptachlor epoxide	<0.01		24
DDE	<0.01		24
DDD	<0.01		24
DDT	<0.01		24
HCB	<0.01		24
Mirex	<0.01		24
Methoxychlor	<0.05		24
Dieldrin	<0.01		24
Endrin	<0.01		24
Telodrin	<0.01		24
Chlordane	<0.05		24
Toxaphene	<0.10		24
Estimated PCBs	<0.20		24
Ronnel	<0.01		24
Ethion	<0.02		24
Trithion	<0.05		24
Diazinon	<0.10		24
Methyl chlorpyrifos	0.122 ± 0.139	0.020 – 0.416	24
Methyl parathion	<0.02		24
Ethyl parathion	<0.02		24
Malathion	0.165 ± 0.178	0.020 – 0.589	24
Endosulfan I	<0.01		24
Endosulfan II	<0.01		24
Endosulfan sulfate	<0.03		24

[a] All samples were irradiated. CFU=colony-forming units; MPN=most probable number; BHC=hexachlorocyclohexane or benzene hexachloride

[b] For values less than the limit of detection, the detection limit is given as the mean.

[c] Sources of contamination: alfalfa, grains, and fish meal

[d] Sources of contamination: soy oil and fish meal

[e] All values were corrected for percent recovery.

APPENDIX K
SENTINEL ANIMAL PROGRAM

SENTINEL ANIMAL PROGRAM

METHODS

Rodents used in the Carcinogenesis Program of the National Toxicology Program are produced in optimally clean facilities to eliminate potential pathogens that may affect study results. The Sentinel Animal Program is part of the periodic monitoring of animal health that occurs during the toxicologic evaluation of chemical compounds. Under this program, the disease state of the rodents is monitored via serology on sera from extra (sentinel) animals in the study rooms. These animals and the study animals are subject to identical environmental conditions. The sentinel animals come from the same production source and weanling groups as the animals used for the studies of chemical compounds.

Serum samples were collected from five male and five female chamber control rats and mice at the end of the 2-week and 3-month studies, five male and five female sentinel rats and mice 1 week after the start of the 3-month and 2-year studies, five male and five female sentinel rats and mice at 6 and 12 months in the 2-year studies, five male and four female sentinel rats at 18 months in the 2-year studies, five male and five female mice at 18 months in the 2-year studies, and five males and five females from the 500 ppm rats and 250 ppm mice at the end of the 2-year studies. Blood from each animal was collected and allowed to clot, and the serum was separated; fecal samples were collected from five male and five female mice at 18 months in the 2-year study for *Helicobacter* species by polymerase chain reaction testing. The samples were processed appropriately and sent to MA Bioservices/ BioReliance (Rockville, MD) for determination of antibody titers. The laboratory methods and agents for which testing was performed are tabulated below; the times at which samples were collected during the studies are also listed.

Method and Test	Time of Collection
RATS	
2-Week Study	
ELISA	
H-1 (Toolan's H-1 virus)	Study termination
KRV (Kilham rat virus)	Study termination
Mycoplasma pulmonis	Study termination
PVM (pneumonia virus of mice)	Study termination
RCV/SDA (rat coronavirus/sialodacryoadenitis virus)	Study termination
Sendai	Study termination
3-Month Study	
ELISA	
H-1	1 week
KRV	1 week
Mycoplasma arthritidis	Study termination
M. pulmonis	1 week, study termination
PVM	1 week, study termination
RCV/SDA	1 week, study termination
Sendai	1 week, study termination
Immunofluorescence Assay	
Parvovirus	Study termination

Method and Test	Time of Collection
RATS (continued)	
2-Year Study	
ELISA	
H-1	1 week
KRV	1 week
M. pulmonis	1 week, study termination
M. arthritidis	Study termination
PVM	1 week, 6, 12, and 18 months, study termination
RCV/SDA	1 week, 6, 12, and 18 months, study termination
Sendai	1 week, 6, 12, and 18 months, study termination
Immunofluorescence Assay	
M. arthritidis	Study termination
PVM	18 months
RCV/SDA	12 and 18 months
Sendai	18 months
Parvovirus	6, 12, and 18 months, study termination
MICE	
2-Week Study	
ELISA	
GDVII (mouse encephalomyelitis virus)	Study termination
MVM (minute virus of mice)	Study termination
MHV (mouse hepatitis virus)	Study termination
M. pulmonis	Study termination
PVM	Study termination
Sendai	Study termination
3-Month Study	
ELISA	
Ectromelia virus	Study termination
EDIM (epizootic diarrhea of infant mice)	Study termination
GDVII	1 week, study termination
LCM (lymphocytic choriomeningitis virus)	Study termination
MVM	1 week
Mouse adenoma virus-FL	Study termination
MHV	1 week, study termination
M. arthritidis	Study termination
M. pulmonis	1 week, study termination
PVM	1 week, study termination
Reovirus 3	Study termination
Sendai	1 week, study termination
Immunofluorescence Assay	
MCMV (mouse cytomegalovirus)	Study termination
Parvovirus	Study termination

Method and Test	Time of Collection
MICE (continued)	
2-Year Study	
ELISA	
Ectromelia virus	6, 12, and 18 months, study termination
EDIM	6, 12, and 18 months, study termination
GDVII	1 week, 6, 12, and 18 months, study termination
LCM	6, 12, and 18 months, study termination
MVM	1 week, 12 and 18 months, study termination
Mouse adenoma virus	6, 12, and 18 months, study termination
MHV	1 week, 6, 12, and 18 months, study termination
M. arthritidis	Study termination
M. pulmonis	1 week, study termination
PVM	1 week, 6, 12, and 18 months, study termination
Reovirus 3	6, 12, and 18 months, study termination
Sendai	1 week, 6, 12, and 18 months, study termination
Immunofluorescence Assay	
Ectromelia virus	12 months, study termination
EDIM	12 months
GDVII	12 months
Mouse adenoma virus-FL	18 months
MCMV	Study termination
MHV	Study termination
M. pulmonis	Study termination
Reovirus 3	12 months
Parvovirus	6 and 18 months, study termination
Polymerase Chain Reaction	
Helicobacter species	18 months

RESULTS

All test results were negative.