Miller, Diane M. (CDC/NIOSH/EID)

From: Paul Dugard [pdugard@mindspring.com]
Sent: Monday, December 14, 2009 7:03 PM
To: NIOSH Docket Office (CDC)
Subject: 1-Bromopropane: Request for Information
Attachments: nPB NTP carc draft abstract 2009-11-19.pdf; nPB Response to NIOSH 2009-12.doc; XBNA 2009-11-20 nPB NTP Studies.htm

Dear Sir:

Please find attached the response to the request for information on1-bromopropane (nPB) published in the Federal Register of September 16, 2009. The items attached are the review of the toxicity and occupational exposure limits for nPB prepared by the Halogenated Solvents Industry Alliance (HSIA), an abstract of the draft report of the carcinogenicity and other studies conducted on behalf of the NTP and the report of the findings of the Technical Reports Review Subcommittee of the NTP. The full draft report of the NTP studies is available via the NTP website. An assessment of the occupational exposure limit by Dr. G. Rusch could not be submitted in electronic format and will be submitted in hard copy, or in a scanned version.

Thank you for the opportunity to contribute information.

Sincerely,

Paul H. Dugard, PhD
Director of Scientific Programs

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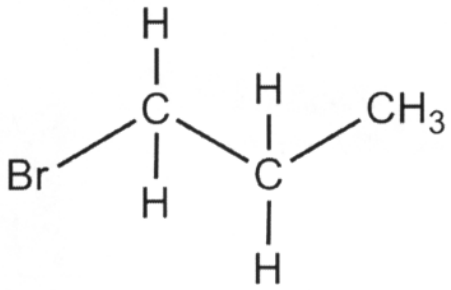
ABSTRACT



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. Results of sperm count and vaginal cytology evaluations showed exposure concentration-related decreases in sperm motility and counts in male rats, reaching 28% and 37%, respectively, in the 1,000 ppm group. 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. Male mice in the 500 ppm group had decreased sperm counts that 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) in 500 ppm females was significantly greater than that 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) in all exposed groups of males were significantly greater than that 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 in 500 ppm males was significantly greater than that 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. 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 *S. typhimurium* strains TA97, TA98, TA100, and TA1535, as well as *E. 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 neoplasms of the skin. Increased incidences of malignant mesothelioma and pancreatic islet adenoma 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 neoplasms of the skin 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, the trachea of female rats and male and female mice, and the lung of 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 14.

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) 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)	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
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.

HALOGENATED SOLVENTS INDUSTRY ALLIANCE, INC.

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December 14, 2009

NIOSH Docket Office
Robert A. Taft Laboratories, MS-C34
4676 Columbia Parkway
Cincinnati, OH 45226

Re: NIOSH-057

Dear Sirs:

By notice published on September 16, 2009, the National Institute for Occupational Safety & Health (NIOSH) announced that it intends to evaluate the scientific data on 1-bromopropane (n-propyl bromide, nPB, or 1-BP) and to establish a Recommended Exposure Level (REL) for nPB. 74 Fed. Reg. 47593. Specifically, NIOSH requested information on the following: (1) published and unpublished reports and findings from in vitro and in vivo toxicity studies with 1-BP, (2) information on possible health effects observed in workers exposed to 1-BP, (3) information on workplaces and products in which 1-BP can be found, (4) description of work tasks and scenarios with a potential for exposure to 1-BP, (5) workplace exposure data, and (6) information on control measures (*e.g.*, engineering controls, work practices, personal protective equipment) that are being used in workplaces where potential exposures to 1-BP occur. The Halogenated Solvents Industry Alliance, Inc. (HSIA) supports this activity by NIOSH, and provides these comments on the items requested.

1. Reproductive Toxicity

In a proposed rule to list nPB as acceptable for certain applications and unacceptable for others under its Significant New Alternatives Policy (SNAP) program, the Environmental Protection Agency (EPA) discussed the available reproductive toxicity data in some detail. 72 Fed. Reg. 30168 (May 30, 2007). EPA based its analysis of occupational exposures on effects on sperm motility observed in a two-generation reproductive toxicity study employing a benchmark dose (BMD) calculation.[1] Since that time, additional BMD calculations have been developed based on effects on estrus cycle in the same multigeneration study. HSIA has reviewed the documents provided by EPA detailing the selection of endpoints, the choice of study, and the preferred calculations of BMD and the lower 95% confidence limit on the BMD (BMDL). We agree that the BMDL values selected (169 ppm based on sperm motility, 162 ppm based on effects on estrus cycle) are appropriate and follow accepted methods and principles. These values were then adjusted to take into account the temporal differences between the exposure regime in the rat study (6 hours per day, 7 days per week) and occupational exposures of 8 hours per day, 5 days per week. The resultant human equivalent concentrations (HECs) are 177 ppm and 170 ppm for male and female reproductive toxicity, respectively. The HEC value provides the point of departure (POD) for calculation of an REL.

[1] WIL, An Inhalation Two-Generation Reproductive Toxicity Study of 1-Bromopropane in Rats, sponsored by the Brominated Solvents Consortium (May 24, 2001).

EPA modified its normal procedure to calculate the reference concentration (RfC), which is considered to be the highest concentration having no significant effect when inhaled throughout life by sensitive individuals, to establish a possible range for the REL. Although designed to set the RfC for the general population, the approach does provide a reasonable framework for assessing an REL. This calculation involves applying a sequence of uncertainty factors to the POD based on the extrapolation of animal data to human exposures, the variability among the worker population, and the completeness of the database.

The maximum uncertainty factor applied for the animal to human extrapolation is 10-fold (*i.e.,* man is 10-fold more sensitive than the laboratory animal). This factor of 10 is considered to include a 3-fold safety factor for pharmacodynamic species differences (sensitivity at the site of action) and a 3-fold factor for pharmacokinetic differences (delivery of the active moiety to the site of action). For nPB, EPA adopted a 3-fold uncertainty factor for pharmacodynamic differences between rat and man, but elected to include no uncertainty factor (i.e., a factor of 1) for pharmacokinetic differences.

EPA's judgment that no uncertainty factor is necessary for dose-response extrapolation from rat to man was based on a higher blood/air partition coefficient for nPB in rats than in man (11.7 versus 7.1). EPA concluded that, for a given external concentration, "the delivered dose of nPB into the bloodstream of rats is slightly higher than in humans." This relatively small difference in blood/air partition coefficients is insufficient, however, to overcome the many remaining uncertainties. The partition coefficient itself is only an indication of relative solubility and the target tissues in rat and man may be similar in ability to dissolve nPB such that, if nPB is poorly metabolized/cleared, the level achieved in the target tissue may be the same in the two species at equilibrium.

There are only certain situations in which the blood/air partition coefficient is the controlling factor in determining the level of the active moiety at the site of action. Without information on nPB metabolism in humans, this level cannot be predicted with physiologically-based pharmacokinetic (PBPK) modeling for humans. The factors that support the application of a full 3-fold uncertainty factor for pharmacokinetics are the following:

- the active moiety or moieties for reproductive effects are not known,
- the relationship between blood level and active moiety is unknown,
- the nature and extent of nPB metabolism and general pharmacokinetics in man are not known,
- the difference in blood/air partition coefficients is small, and
- the blood/air partition coefficient is only controlling under certain conditions that can only be defined through PBPK modeling for man.

Taken together, these uncertainties are highly significant and support the application of a safety factor of 3 for pharmacokinetics for extrapolation from rat to man.

Combined with the 3-fold safety factor for pharmacodynamics, HSIA believes that the full 10-fold uncertainty factor should be applied for the interspecies (rat to man) extrapolation. This is particularly important when one considers the greater human sensitivity to reproductive toxins relative to responses in the rat. The rat possesses huge overcapacity in reproductive capabilities whereas man is considered to be much closer to infertility as a species. This fact suggests that humans are fundamentally more susceptible to reprotoxins than the rat. Although this sensitivity is captured, to a limited extent, in the 3-fold uncertainty factor for pharmacodynamics, it may not be sufficient to account for the greater sensitivity of humans.

Regarding human variability, the RfC methodology allows up to a 10-fold uncertainty factor to account for the variability in sensitivity within the general population. In some circumstances, HSIA acknowledges that this factor can be reduced for a working population because of the general good health required for employment. HSIA agrees with EPA's conclusion that there may be sensitive individuals for reproduction in the workforce because poor reproductive health may exist in individuals generally fit enough to work. EPA considers that variability in the worker population may be less than in the general population and that an uncertainty factor of 10-fold is greater than necessary. This is also a reasonable assumption and HSIA supports the use of an intraspecies uncertainty factor of 3-fold for variations in sensitivity in a worker population exposed to nPB.

In its analysis, EPA did not include an uncertainty factor for the relatively incomplete nature of the database for nPB toxicity, despite the fact that it used data from a subchronic (*e.g.*, 90-day) study in its exposure calculations. As discussed below, 2-year bioassays in both rats and mice conducted under the auspices of the National Toxicology Program (NTP) show clear evidence of carcinogenicity. The appropriate response may be to add nPB to the NIOSH Carcinogen List with the consequent recommendation that workplace exposures not exceed the "lowest feasible concentration". This new finding certainly should not lessen the safety/uncertainty factors applied and, in the absence of a formal risk assessment for cancer, a factor of 3 or even 10-fold should be applied

Based on the above analysis, the total uncertainty factor to be applied to the HEC values for male and female reproductive toxicity to yield REL values is at least 30 fold. Thus, the REL based on female or male reproductive toxicity should be 6 ppm or less.

2. Neurotoxicity

Earlier reviews of the neurotoxicity of nPB focused on animal studies in which hind limb weakness and neurophysiological measurements indicated effects on the peripheral nervous system, although neurotoxicity was not reported in two recent 90-day studies. More recently, as NIOSH is

aware, there have been a number of reports of severe neurotoxicity in workers exposed to nPB. In particular, the human effects noted relate to lower limb peripheral nerve effects with adverse sensory responses. These effects are qualitatively similar to those seen in rat experiments -- limb weakness progressing in some cases to paralysis. Central nervous system effects also were reported in a proportion of human cases.

The consistency of severe effects across multiple independent investigations is striking and, although the measurements of atmospheric levels have limitations, the similarity of exposure levels associated with effects is also noteworthy. Many of the case subjects needed hospitalization and the reversibility of the effects is still in question. A publication by Majersik *et al.* provides a complete clinical analysis of the effects and indications of the slow recovery, if any, observed in patient follow-up.[2] More recently, two cases were reported involving workers exposed to nPB (in vapor degreasing and dry cleaning, respectively) who were diagnosed with clinical manifestations of nPB.[3]

While acknowledging that "severe, possibly irreversible, neurological effects may occur at sustained concentrations of 100 ppm or greater," EPA elected to disregard these reports of severe responses in humans because "the data on neurotoxic effects of nPB on workers are limited and are not sufficient to determine acceptable levels of exposure." Based on the more recent information, however, HSIA believes that neurotoxicity in humans is of substantial concern. Indeed, there is one published study showing neurophysiological effects in workers exposed at levels between 0.34 and 50 ppm.[4] This study suggests the nature of the dose-response relationship for neurological effects and provides a basis for setting an REL.

Enclosed is an analysis of alternative RELs for nPB based on the human and animal data on neurotoxicity developed by a prominent toxicologist and industrial hygienist (to be submitted in hard copy). This analysis suggests an REL of 1 to 4.3 ppm based on the human data and an REL of 1 to 6.7 ppm using the animal data. These values are consistent with the evidence from Ichihara *et al.* suggesting that minimal neurophysiological effects were detectable in workers where exposures were predominantly 10 ppm or less. These exposure limits do not involve undue conservatism, moreover, since the cumulative nature of neurological effects is not fully understood.

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[2] Majersik *et al.*, Severe Neurotoxicity Associated with Exposure to the Solvent 1-Bromopropane (n-Propyl Bromide), *Clinical Toxicology* 45: 270-276 (2007).

[3] Perrone *et al.*, Neurologic Illness Associated with Occupational Exposure to the Solvent 1-Bromopropane – New Jersey and Pennsylvania, 2007 – 2008, *Morbidity and Mortality Weekly Report* 57(48): 1300-1302 (2008).

[4] Ichihara *et al.*, Neurological Abnormalities in Workers of a 1-Bromopropane Factory, *Environmental Health Perspectives* 112(13): 1319-1325 (2004).

3. Carcinogenicity

Perhaps the most significant recent information relevant to the health hazard on nPB is the report of 2-year carcinogenesis studies recently conducted and reviewed by NTP. The enclosed draft report, which was unanimously approved by the NTP Technical Reports Review Subcommittee, shows:

- Clear evidence of carcinogenicity in female F344 rats (adenomas in large intestine and equivocal evidence for skin tumors)
- Some evidence of carcinogenicity in male F344 rats (adenomas in large intestine, skin tumors, and equivocal findings for mesotheliomas and pancreatic adenomas)
- Clear evidence of carcinogenicity in female B6C3F1 mice (lung tumors)
- No evidence of carcinogenicity in male B6C3F1 mice

4. Manufacturers' Recommendations

Exposure information provided by current or recent nPB manufacturers (Albemarle, Great Lakes Chemical/ Chemtura, ATOFINA, ICL Industrial Products) indicates that their RELs are between 5 and 25 ppm.[5] The ACGIH TLV® of 10 ppm is another relevant recommendation.

5. Conclusion

The application of standard industrial hygiene and risk assessment practices supports an REL in the lower part of a range between 1 and 10 ppm for nPB using the available data for both reproductive toxicity and neurotoxicity endpoints. The carcinogenicity data suggest addition to the NIOSH Carcinogen List and a recommendation that workplace exposures not exceed the "lowest feasible concentration."

Sincerely,

Paul H. Dugard, PhD
Director of Scientific Programs

Enclosures

[5] These documents also suggest the need for warnings concerning absorption through skin, which may occur very readily from the liquid phase.

Daily Environment Report™

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Toxic Substances
NTP Tests Show Cancerous Effects
In Rodents After Exposure to Pulegone

RESEARCH TRIANGLE PARK, N.C.—Tests conducted by the National Toxicology Program showed clear evidence of cancerous effects in mice due to exposure to pulegone, an organic compound found in the essential oils of peppermint, catnip, and pennyroyal, a scientific review panel said Nov. 19.

NTP's Board of Scientific Counselors' Technical Reports Review Subcommittee agreed with the findings in a draft agency report (TR 563) that said a two-year study using pulegone (CAS No. 89-82-7) delivered through corn oil showed clear evidence of cancerous effects in both male and female mice, based on growths found in their livers.

The review panel also agreed with the conclusions that there was no evidence of cancerous effects of exposure to pulegone in male rats, but said the cancer effects on female rats was understated in the draft report.

Split Panel Agrees to Modify Conclusion

On a 6-4 vote, the scientific panel said NTP should revise its conclusion that there was some evidence of carcinogenicity in female rats because of growths found in their bladders. The majority of scientists on the panel believed the observed effects should be described as clear evidence of cancerous effects.

Pulegone is used as a flavoring, a fragrance agent, and in herbal medicines. The substance was nominated for toxicological and cancer studies by the National Institute for Environmental Health Sciences because of the potential for human exposure and the lack of carcinogenicity data.

Members of the review panel also said the study had a significant mortality rate among the rodents tested, at the higher doses selected. The level of mortality—due to liver toxicity—was central to the debate over whether the cancerous effects on female rats were understated or not.

1-Bromopropane Findings

At the meeting, the Technical Reports Review Subcommittee unanimously agreed with NTP's findings of cancerous effects found in rodents exposed to 1-bromopropane (CAS No. 106-94-5). 1-bromopropane is used as a solvent for adhesive resins, a replacement for ozone-depleting refrigerants, and in cleaning and degreasing applications.

NTP's two-year inhalation study of exposure to 1-bromopropane, described in the draft report (TR 564), found clear evidence of cancerous effects in female rats and female mice, based on growths found in their large intestines and lungs, respectively. The study also found some evidence of carcinogenicity in male rats based on growths on their large intestines and skin, and no evidence of cancerous effects in male mice.

No Effects Found for Certain Herbs, Chemicals

According to the review panel, NTP also was correct in its determination, that under the conditions of their studies, no cancer effects were found in exposure to:

• bis(2-chloroethoxy)methane (CAS No. 111-91-1), a substance used as a solvent and a starting agent in the production of fungicides and polysulfide polymers, that was tested in a two-year study of exposure through rodents' skin (TR 536),

• diethylamine (CAS No. 109-89-7), which is used as a chemical intermediate for corrosion inhibitors, pesticides, and rubber processing chemicals, and was tested in a two-year inhalation study (TR 566),

• ginseng (CAS No. 50647-08-0), an herb that is widely used in herbal remedies, dietary supplements, cosmetics, and as a food additive, that was tested in a two-year drinking water study (TR 567), and

• milk thistle extract (CAS No. 84604-20-6), which is used as an herbal medicine to treat a variety of diseases and as a food additive, that was tested in a two-year feed study (TR 565).

Regarding the diethylamine report, the panelists agreed to add to the study summary page an observation that exposure to the substance resulted in increased lesions to male rats' corneas, given that occupational exposure is of interest.

Members of the review panel debated whether to clarify language in the ginseng report that said "[t]he incidence of mammary gland fibroadenoma was significantly decreased" in female rats exposed to levels of ginseng at 5,000 milligrams per kilogram, the highest dose used in the two-year study. Some panelists believed that additional language clarifying that it was unknown whether the observed decrease in such growths was due to exposure to ginseng should be added to the report.

Concerns about the use of the statement to claim certain unproven health benefits of taking ginseng were voiced, but the panel ultimately approved the conclusions as written.

The conclusions of the report on milk thistle extract also noted decreased incidences of mammary gland growths in female mice and liver growths in male mice, when compared with unexposed—or control—rodents.

John Bucher, associate director of NTP, told panelists that there was debate among agency staff on how to address the findings of reduced incidences of certain growths found during the exposure studies. "These studies are not designed to detect decreases in tumor incidences," he told meeting participants.

The apparent inconsistencies regarding how such decreases are presented in the reports under consideration represent "some disagreement" about the significance of such findings among agency staff, according to Bucher.

By Andrew M. Ballard

Copies of the draft NTP studies of the carcinogenic and toxic effects of exposure to certain chemicals and substances related to the review panel meeting are available at http://ntp.niehs.nih.gov/?objectid=93F86CF4-F1F6-975E-799E4CD8D5A90F6A&#20091119.

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