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Provisional Peer Reviewed Toxicity Values for
1,2-Dibromo-3-Chloropropane
(CASRN 96-12-8)

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

Acronyms and Abbreviations

bw	body weight
cc	cubic centimeters
CD	Caesarean Delivered
CERCLA	Comprehensive Environmental Response, Compensation and Liability Act of 1980
CNS	central nervous system
cu.m	cubic meter
DWEL	Drinking Water Equivalent Level
FEL	frank-effect level
FIFRA	Federal Insecticide, Fungicide, and Rodenticide Act
g	grams
GI	gastrointestinal
HEC	human equivalent concentration
Hgb	hemoglobin
i.m.	intramuscular
i.p.	intraperitoneal
i.v.	intravenous
IRIS	Integrated Risk Information System
IUR	inhalation unit risk
kg	kilogram
L	liter
LEL	lowest-effect level
LOAEL	lowest-observed-adverse-effect level
LOAEL(ADJ)	LOAEL adjusted to continuous exposure duration
LOAEL(HEC)	LOAEL adjusted for dosimetric differences across species to a human
m	meter
MCL	maximum contaminant level
MCLG	maximum contaminant level goal
MF	modifying factor
mg	milligram
mg/kg	milligrams per kilogram
mg/L	milligrams per liter
MRL	minimal risk level

MTD	maximum tolerated dose
MTL	median threshold limit
NAAQS	National Ambient Air Quality Standards
NOAEL	no-observed-adverse-effect level
NOAEL(ADJ)	NOAEL adjusted to continuous exposure duration
NOAEL(HEC)	NOAEL adjusted for dosimetric differences across species to a human
NOEL	no-observed-effect level
OSF	oral slope factor
p-IUR	provisional inhalation unit risk
p-OSF	provisional oral slope factor
p-RfC	provisional inhalation reference concentration
p-RfD	provisional oral reference dose
PBPK	physiologically based pharmacokinetic
ppb	parts per billion
ppm	parts per million
PPRTV	Provisional Peer Reviewed Toxicity Value
RBC	red blood cell(s)
RCRA	Resource Conservation and Recovery Act
RDDR	Regional deposited dose ratio (for the indicated lung region)
REL	relative exposure level
RfC	inhalation reference concentration
RfD	oral reference dose
RGDR	Regional gas dose ratio (for the indicated lung region)
s.c.	subcutaneous
SCE	sister chromatid exchange
SDWA	Safe Drinking Water Act
sq.cm.	square centimeters
TSCA	Toxic Substances Control Act
UF	uncertainty factor
µg	microgram
µmol	micromoles
VOC	volatile organic compound

**PROVISIONAL PEER REVIEWED TOXICITY VALUES FOR
1,2-DIBROMO-3-CHLOROPROPANE (CASRN 96-12-8)**

Background

On December 5, 2003, the U.S. Environmental Protection Agency's (EPA's) Office of Superfund Remediation and Technology Innovation (OSRTI) revised its hierarchy of human health toxicity values for Superfund risk assessments, establishing the following three tiers as the new hierarchy:

1. EPA's Integrated Risk Information System (IRIS).
2. Provisional Peer-Reviewed Toxicity Values (PPRTV) used in EPA's Superfund Program.
3. Other (peer-reviewed) toxicity values, including:
 - ▶ Minimal Risk Levels produced by the Agency for Toxic Substances and Disease Registry (ATSDR),
 - ▶ California Environmental Protection Agency (CalEPA) values, and
 - ▶ EPA Health Effects Assessment Summary Table (HEAST) values.

A PPRTV is defined as a toxicity value derived for use in the Superfund Program when such a value is not available in EPA's Integrated Risk Information System (IRIS). PPRTVs are developed according to a Standard Operating Procedure (SOP) and are derived after a review of the relevant scientific literature using the same methods, sources of data, and Agency guidance for value derivation generally used by the EPA IRIS Program. All provisional toxicity values receive internal review by two EPA scientists and external peer review by three independently selected scientific experts. PPRTVs differ from IRIS values in that PPRTVs do not receive the multi-program consensus review provided for IRIS values. This is because IRIS values are generally intended to be used in all EPA programs, while PPRTVs are developed specifically for the Superfund Program.

Because science and available information evolve, PPRTVs are initially derived with a three-year life-cycle. However, EPA Regions or the EPA Headquarters Superfund Program sometimes request that a frequently used PPRTV be reassessed. Once an IRIS value for a specific chemical becomes available for Agency review, the analogous PPRTV for that same chemical is retired. It should also be noted that some PPRTV manuscripts conclude that a PPRTV cannot be derived based on inadequate data.

Disclaimers

Users of this document should first check to see if any IRIS values exist for the chemical of concern before proceeding to use a PPRTV. If no IRIS value is available, staff in the regional Superfund and RCRA program offices are advised to carefully review the information provided in this document to ensure that the PPRTVs used are appropriate for the types of exposures and circumstances at the Superfund site or RCRA facility in question. PPRTVs are periodically updated; therefore, users should ensure that the values contained in the PPRTV are current at the time of use.

It is important to remember that a provisional value alone tells very little about the adverse effects of a chemical or the quality of evidence on which the value is based. Therefore, users are strongly encouraged to read the entire PPRTV manuscript and understand the strengths and limitations of the derived provisional values. PPRTVs are developed by the EPA Office of Research and Development's National Center for Environmental Assessment, Superfund Health Risk Technical Support Center for OSRTI. Other EPA programs or external parties who may choose of their own initiative to use these PPRTVs are advised that Superfund resources will not generally be used to respond to challenges of PPRTVs used in a context outside of the Superfund Program.

Questions Regarding PPRTVs

Questions regarding the contents of the PPRTVs and their appropriate use (e.g., on chemicals not covered, or whether chemicals have pending IRIS toxicity values) may be directed to the EPA Office of Research and Development's National Center for Environmental Assessment, Superfund Health Risk Technical Support Center (513-569-7300), or OSRTI.

INTRODUCTION

Dibromochloropropane (1,2-dibromo-3-chloropropane; DBCP) is not included in the HEAST non-cancer table (U.S. EPA, 1997). There is no RfD assessment for DBCP on IRIS (U.S. EPA, 2006) or in the Drinking Water Standards and Health Advisories list (U.S. EPA, 2002). A Health Effects Assessment (HEA) and a Drinking Water Criteria Document (DWCD) for DBCP both opted not to perform an oral non-cancer assessment for the chemical in favor of a carcinogenicity assessment (U.S. EPA, 1988a, 1989a). No other relevant documents were found in the CARA list (U.S. EPA, 1991, 1994a). ATSDR (1992) derived an intermediate-duration oral MRL of 0.002 mg/kg-day for DBCP based on a subchronic LOAEL of 1.88 mg/kg-day for effects on spermatogenesis and sperm morphology in rabbits (Foote et al., 1986a, 1986b). A chronic oral MRL was not derived because the critical reproductive endpoints were not

evaluated at doses less than those producing tumors or death in chronic studies. No Environmental Health Criteria Document is available for DBCP (WHO, 2002).

An RfC assessment for DBCP is on IRIS (U.S. EPA, 2006). Based on a subchronic NOAEL of 0.94 mg/m³ (0.1 ppm) for testicular effects in rabbits (Rao et al., 1982), an RfC of 2 x 10⁻⁴ mg/m³ was verified on 8/15/1991. ATSDR (1992) derived an intermediate-duration inhalation MRL of 0.0002 ppm for DBCP based on the same subchronic NOAEL from the Rao et al. (1982) study (0.1 ppm for changes in spermatogenesis and testicular atrophy in rabbits). A chronic inhalation MRL was not derived because the critical reproductive endpoints were not evaluated at concentrations less than those producing tumors or death in chronic studies. ACGIH (2002) does not recommend an occupational exposure limit for DBCP, and no non-cancer-based limits are recommended by NIOSH (2002) or promulgated by OSHA (2002).

Oral and inhalation slope factors and unit risk values for DBCP are listed in the HEAST (U.S. EPA, 1997). The CRAVE Work Group meeting notes indicate that the cancer classification was verified, the oral quantitation was to be included on IRIS, and the inhalation quantitation was under review (U.S. EPA, 1993). However, the cancer assessment for DBCP was never added to IRIS (U.S. EPA, 2006). The oral slope factor of 1.4 (mg/kg-day)⁻¹ was derived based on stomach, kidney, and liver tumors in an unpublished chronic dietary study (Hazelton Laboratories, 1977), in Office of Drinking Water and Carcinogen Assessment Group assessments (U.S. EPA, 1988a, 1988b), and is also presented in the HEA (U.S. EPA, 1989a). The inhalation unit risk of 6.9E-4 (mg/m³)⁻¹ was derived based on nasal tumors in an NTP (1982) chronic inhalation bioassay and sourced to the CRAVE Work Group (U.S. EPA, 1989b). An earlier assessment based on the NTP (1982) study had been derived in the HEA (U.S. EPA, 1989a). The carcinogenicity of DBCP has been tested in NCI (1978) oral and NTP (1982) inhalation bioassays, and evaluated by IARC (1999), who categorized DBCP as possibly carcinogenic to humans (Group B) based on inadequate evidence in humans and sufficient evidence in experimental animals.

Literature searches were conducted from 1986 thru 2002 for studies relevant to the derivation of provisional toxicity values for DBCP. Databases searched included: TOXLINE (including NTIS and BIOSIS updates), MEDLINE, CANCERLIT, TSCATS, RTECS, GENETOX, DART, EMIC, HSDB and CCRIS. An updated literature search was conducted through April 2004 and no relevant information was found.

REVIEW OF PERTINENT DATA

Human Studies

The toxicity of DBCP to the human male reproductive system has been evaluated in epidemiological studies of exposed production workers, pesticide applicators and farmers. A number of these studies found adverse effects on testicular function attributable to DBCP, including altered sperm morphology, clinically significant decreases in spermatogenic activity and sperm counts (oligospermia or azoospermia), secondary increases in serum levels of follicle-stimulation hormone (FSH) and luteinizing hormone (LH), and increases in frequency of spontaneous abortion in wives of exposed workers (ATSDR, 1992; IARC, 1999; Goldsmith, 1997; Potashnik and Porath, 1995; U.S. EPA, 2003). The available evidence has established that DBCP is a human testicular toxicant in some occupational exposure scenarios, but none of the studies provide data that are sufficient for quantitative exposure-response analysis and/or uncomplicated by concurrent exposure to other chemicals.

The risk of cancer among humans exposed to DBCP (among other chemicals) has been evaluated in several occupational cohort mortality studies and general population-based case-control studies. Excesses of lung, liver, biliary tract and/or cervical cancers were observed in the cohort mortality studies (Amoateng-Adjepong et al., 1995; Brown, 1992; IARC, 1987; Olsen et al., 1995; Wesseling et al., 1996), but the findings cannot be clearly attributed to DBCP due to small numbers of cases and/or exposures to other chemicals (IARC, 1999). Case-control studies found no significant associations between gastric cancer or leukemia and DBCP in drinking water (Wong et al., 1989).

Animal Studies

Oral Systemic Toxicity Studies:

Groups of 20 male Sprague-Dawley rats were exposed to drinking water that provided reported average DBCP intakes of 0, 0.4, 3.3, 5.4 or 9.7 mg/kg-day for 64 days (Heindel et al., 1989). Water consumption, body weight, and serum levels of liver-related enzymes (AST, ALT, BUN, SDH and OCT) and reproductive hormones (FSH and LH) were evaluated throughout the study. Endpoints evaluated at the end of the study included selected organ weights (liver, kidneys, prostate, left epididymis, full and fluid-expelled seminal vesicles, testes), histopathology (liver, kidney, testis), sperm count (total sperm per epididymis), and intratesticular testosterone level. The testicular histological examinations included qualitative assessment of spermatogenesis abnormalities in the seminiferous epithelium. Water intake decreased in a dose-related manner during the first 30 days and thereafter remained generally constant in all groups until the end of the study. Water consumption was significantly ($p \leq 0.05$) lower than controls at all dose levels, the decrease ranging from approximately 19% at 0.4 mg/kg-day to 60% at 9.7

mg/kg-day. Weight gain was significantly reduced in the 9.7 mg/kg-day group after the first week of exposure; body weight was approximately 10% less than controls at the end of the study (food consumption was not measured). There were no clear DBCP-related changes in any of the liver and kidney endpoints; the only apparent effect in these tissues was a subtle and non-statistically significant increase in the mean number of nuclei per renal proximal tubule cross-section at 9.7 mg/kg-day. Absolute weights of full and fluid-expelled seminal vesicles were significantly increased in two of the three dose groups at ≥ 3.3 mg/kg-day, but the toxicological significance is unclear because the changes were not dose-related (no increase at 5.4 mg/kg-day) or pronounced (full and empty seminal vesicle weights were only 4.7 and 13.6% higher than controls at 9.7 mg/kg-day at the end of the study), and relative seminal vesicle weights were not reported. Paired absolute testes weight was approximately 10% lower than controls ($p < 0.05$) at 9.7 mg/kg-day, but the toxicological significance is unclear because the decrease was small and there was no significant change in relative testes weight. Based on the decreases in body weight gain and water consumption, and a possible increase in turnover of renal proximal tubular cells, this study identifies a NOAEL of 5.4 mg/kg-day and LOAEL of 9.7 mg/kg-day for systemic toxicity in rats.

Groups of 10 male and 10 female Sprague-Dawley rats were exposed to DBCP intake levels of 0, 0.015, 0.26, 2.96 or 19.43 mg/kg-day in the drinking water for 60 days before mating in a one-generation reproduction study (Johnston et al., 1986) detailed in the *Reproductive and Developmental Toxicity* section. Effects on non-reproductive endpoints included significantly reduced food and water consumption and body weight gain (60-70% less weight gain than controls) in the adult (F_0) males and females at 19.43 mg/kg-day. Relative liver weight was significantly increased in the adult males at 19.43 mg/kg-day, but is unlikely to be toxicologically relevant because the magnitude of increase was small (11%) and there were no exposure-related gross or histopathological changes in the liver or other tissues in either sex. Based on the decreases in body weight gain and water and food consumption, this study identifies a NOAEL of 2.96 mg/kg-day and a LOAEL of 19.43 mg/kg-day for systemic toxicity in rats.

A limited amount of information on chronic non-neoplastic effects of orally-administered DBCP is available from an NCI (1978) carcinogenesis bioassay in rats and mice. In the NCI rat study, groups of 50 male and 50 female Osborne-Mendel rats were gavaged with DBCP in corn oil at reported time-weighted average doses of 15 or 29 mg/kg-day on 5 days/week for up to 64-78 weeks, as detailed in the *Carcinogenicity and Genotoxicity* section. Groups of 20 males and 20 females were used as vehicle-treated and untreated controls and observed for up to 83-109 weeks. Endpoints that were evaluated included clinical signs, body weight, and gross pathology and histopathology of major tissues, organs and gross lesions. Effects that were observed in all treated groups included abdominal urine stains and hunched appearance as early as weeks 6 and 18, respectively, and dose-related decreases in body weight gain after approximately the first 20 weeks of treatment. The high-dose male and low- and high-dose female groups were terminated early because of high mortality that was likely related to development of forestomach tumors.

The main treatment-related nonneoplastic lesion was dose-related toxic nephropathy, which occurred in essentially all of the dosed rats (50/50 low-dose males, 49/50 high-dose males, 42/42 low-dose females and 44/44 high-dose females), but not in controls of either sex. The toxic nephropathy was histologically characterized by degenerative changes in the proximal convoluted tubules at the junction of the cortex and medulla, with cloudy swelling, fatty degeneration and necrosis of the tubular epithelium. The damaged tubules often had infiltration of inflammatory cells, fibrosis and calcium deposition, and occasionally contained large basophilic regenerative cells. Incidences of testicular atrophy (histology not otherwise specified) were increased in the treated males (11/20 untreated controls, 4/19 vehicle controls, 38/50 low-dose, 47/49 high-dose). This study identified a chronic LOAEL of 15 mg/kg-day for testicular and kidney pathology, but no NOAEL, in rats.

In the NCI (1978) chronic mouse study, groups of 50 B6C3F1 mice of each sex were gavaged with DBCP in corn oil in reported time-weighted average doses of 114 or 219 mg/kg-day (males), or 110 or 209 mg/kg-day (females), on 5 days/week for up to 47-60 weeks, as detailed in the *Carcinogenicity and Genotoxicity* section. Groups of 20 males and 20 females were used as vehicle-treated and untreated controls and observed for up to 78-90 weeks. Endpoints that were evaluated included clinical signs, body weight, and gross pathology and histopathology of major tissues, organs and gross lesions. Study endpoints are the same as in the NCI (1978) rat study. There were no apparent effects of treatment on body weight gain, appearance or behavior. Clinical signs characterized by a hunched or thin appearance and apparent compound-related deaths were observed in the high-dose animals beginning in week 38 of the study. All treated groups were terminated early because of high mortality that was likely related to the development of forestomach tumors. The only treatment-related nonneoplastic lesion was dose-related toxic nephropathy, which occurred in 11/46 (24%) low-dose males, 45/48 (94%) high-dose males, 14/50 (28%) low-dose females and 43/46 (93%) high-dose females, but in no male or female control mice. The toxic nephropathy occurred primarily in the proximal tubules and was comparable in appearance to the renal lesions in the rats. No increased incidence of male reproductive lesions was reported in an appended summary table of nonneoplastic lesions. This study identified a chronic LOAEL of 110 mg/kg-day for kidney pathology, but no NOAEL, in mice.

Unpublished chronic studies of dietary DBCP were conducted in rats and mice (Hazelton Laboratories, 1977, 1978; Shell Oil Company, 1986). In the rat study (Hazelton Laboratories, 1977), groups of 60 male and 60 female Charles River rats were exposed to nominal DBCP intake levels of 0, 0.3, 1.0 or 3.0 mg/kg-day in the diet for 104 weeks. The actual dosage intakes were estimated to be 0, 0.24, 0.80 and 2.39 mg/kg-day (Shell Oil Company, 1986), as detailed in the *Carcinogenicity and Genotoxicity* section. An interim kill of 10 rats/sex/group was performed at 52 weeks. There were no treatment-related effects on behavior or other outward signs of toxicity, hematological parameters, clinical chemistry values, or urinalysis results. There were no statistically significant ($p < 0.05$) changes in survival at 104 weeks. Mean body gain was

significantly (52%) lower than controls in the high-dose males. Evaluation of selected organ weights (liver, kidneys, spleen, heart, thyroid, adrenals, testes and epididymis) showed significantly increased absolute kidney weight (12% higher than controls) at week 52, and increased relative weight of heart (15%) and adrenals (42%) at week 104. Histological examinations showed that the only treatment-related non-neoplastic lesion was an increased severity of interstitial nephritis, accompanied by cytomegaly of the renal tubular epithelial cells, in 6/10 high-dose females (apparently at the interim sacrifice). Incidences of these lesions were not reported for the other dose groups or after 104 weeks. Increased incidences of tumors in the kidneys, stomach and liver were found, as detailed in the *Carcinogenicity and Genotoxicity* section. Additional information on the experimental design and results were not provided in the available summaries of this study (Shell Oil Company, 1986; U.S. EPA, 1979, 1988a). This study identified a chronic LOAEL of 2.39 mg/kg-day for reduced body weight gain in rats. The reporting inadequacies in the available summaries of the study precluded identification of a reliable NOAEL.

In the unpublished chronic dietary study in mice (Hazelton Laboratories, 1978), groups of 50 male and 50 female HaM/ICR Swiss mice were exposed to nominal DBCP intake levels of 0, 0.3, 1.0 or 3.0 mg/kg-day in the diet for 78 weeks. The actual dosage intakes were estimated to be 0, 0.28, 0.91 and 2.7 mg/kg-day (Shell Oil Company, 1986), as detailed in the *Carcinogenicity and Genotoxicity* section. There were no treatment-related clinical signs of toxicity or effects on body weight gain, food consumption, survival, clinical chemistry values, or urinalysis results. Hematological evaluation showed significantly ($p < 0.05$) reduced red blood cell counts, hematocrit and hemoglobin concentration in the males at 2.7 mg/kg-day compared to controls. Necropsies found a dose-related increased incidence of white nodules in the nonglandular mucosa of the stomach in both sexes (additional data not available). Histological examinations were only conducted in the control and 2.7 mg/kg-day groups and showed effects in the nonglandular stomach that included acanthosis, hyperkeratosis, and increased basal cell activity, as well as increased incidences of tumors, as detailed in the *Carcinogenicity and Genotoxicity* section. Additional information on experimental design and results, including incidences of the nonneoplastic lesions, was not reported in the available summaries of this study (Shell Oil Company, 1986; U.S. EPA, 1979, 1988a). This study identified a chronic LOAEL of 2.7 mg/kg-day for histopathology in the nonglandular stomach in mice. A reliable NOAEL cannot be identified because histological examinations were not performed at doses lower than the LOAEL.

Inhalation Systemic Toxicity Studies:

Information on the subchronic inhalation toxicity of DBCP is available from a study in which groups of 30 male and 30 female Sprague-Dawley rats were exposed to 0, 0.1, 1, or 10 ppm (0, 0.97, 9.7, or 97 mg/m³) of DBCP for 6 hours/day, 5 days/week for 14 weeks, and observed for the following 32-weeks, for a total study duration of 46 weeks (Rao et al., 1983). There were no DBCP-related clinical signs or changes in body weight. Histopathological

changes in the adrenal gland were observed that included: foci of altered cells in the cortex at 14 weeks; cortical hyperplasia in females at ≥ 1 ppm and males at 10 ppm at 46 weeks (terminal sacrifice); and cortical hematocysts in females at ≥ 1 ppm at 46 weeks (incidences of 1/20 at 0.1 ppm, 4/19 at 1 ppm and 16/17 at 10 ppm). Other effects observed at 10 ppm after 46 weeks included ovarian cysts in females (7/17) and mineralized deposits in the cerebrum of the brain of both sexes (15/18 males and 6/17 females). Testicular atrophy and decreased spermatogenesis occurred in males at concentrations as low as 1 ppm, as detailed in the *Reproductive and Developmental Toxicity* section. The respiratory tract was not examined, precluding identification of an effect level(s) for respiratory toxicity. Based on testicular and adrenal histopathological effects that were slight at 1 ppm and marked at 10 ppm, this study identifies a LOAEL of 1 ppm and NOAEL of 0.1 ppm for subchronic toxicity in rats.

Fischer 344 rats (5/sex/group) were exposed to 0, 1, 5 or 25 ppm (0, 9.66, 48.3 or 241.6 mg/m³) of DBCP for 6 hours/day, 5 days/week for 13 weeks (NTP, 1982; Reznik et al., 1980a, 1980b). The duration adjusted concentrations are 1.7, 8.6 or 43 mg/m³. Both sexes in the 25 ppm group exhibited blood stains around the nasal orifice throughout the study. Two females at this level died during weeks 10 and 11 of exposure, and another two females and one male were sacrificed during weeks 10-12 due to moribund conditions. Other effects in the 25-ppm rats included a 60% decrease in body weight gain compared with controls, severe hair loss, and inflammation and severe necrosis of the respiratory and olfactory epithelium in the dorsal part of the nasal cavity. The incidences of the nasal cavity lesions appears to have been concentration-related (incidence data not reported). The 25-ppm rats also had respiratory effects in the tracheal epithelium (necrosis in 7/10) and lungs (squamous metaplasia of the bronchial epithelium, with hyperplasia and partial regeneration of the bronchial and bronchiolar epithelium; incidence data not reported). Testicular atrophy with hypospermatogenesis occurred in 5/5 of the 25-ppm males. Histopathological changes were induced in the liver (focal necrosis, hepatocytic hydropic changes, cytomegaly) and kidneys (toxic tubular nephrosis) of the 1- and 5- ppm rats, indicating that 1 ppm is a subchronic LOAEL for hepatic and renal effects and that no NOAEL was identified. The 1-ppm level may also be a LOAEL for respiratory effects because the nasal cavity lesions appear to have been concentration-related.

B6C3F1 mice (10/sex/group) were exposed to 0, 1, 5 or 25 ppm (9.66, 48.3 or 241.6 mg/m³) of DBCP for 6 hours/day, 5 days/week for 13 weeks (NTP, 1982; Reznik et al., 1980c). Effects were observed at 25 ppm that included mortality (4/5 males died before the end of the study) and histopathological changes in liver (hydropic hepatocyte changes in males), kidneys (nephrosis in males), and respiratory tract, including inflammatory, necrotizing and proliferative lesions in the nasal cavity epithelium and necrosis of the bronchiolar epithelium. Effects observed at ≥ 5 ppm included regeneration and hyperplasia of the bronchiolar epithelium and megalocytic epithelial cells (20/20 mice at 5 ppm), and dose-related decreased body weight gain (69% in males and 19% in females in the high-dose group). No additional incidence data or information on effects in non-respiratory tract tissues were reported. This study identifies a

NOAEL of 1 ppm and LOAEL of 5 ppm for systemic effects based on decreased body weight gain, as well as for respiratory effects based on histopathology in the nasal cavity and bronchiolar epithelium.

Information on the chronic toxicity of inhaled DBCP is available from an NTP bioassay in rats and mice. In the rat study (NTP, 1982; Reznik et al., 1980a), groups of 50 male and 50 female F344 rats were exposed to DBCP by whole body inhalation in concentrations of 0, 0.6 or 3 ppm (0, 5.8 or 29 mg/m³) for 6 hours/day, 5 days/week for 105-107 weeks (controls), 103 weeks followed by observation for 1 week (low-dose), or 84 weeks followed by observation for 0-1 weeks (high-dose). Clinical signs and body weight were evaluated during the study, and gross and histological examinations on all major tissues, including the nasal cavity and testes, were performed at the time of sacrifice and in animals that died early. Increasing numbers of treated rats had respiratory signs that began to be detected at week 46 and included wheezing/sneezing and bloody crust on nose and eyes; palpable masses were also noted on the face or nasal areas. Mean body weight gain was decreased in the high-dose males and females after approximately week 65. The male and female high-dose groups were terminated early because of early deaths due to respiratory tract tumors (interference with breathing and metastasis to the brain); increased incidences of tumors occurred in the nasal cavity, tongue, pharynx and other tissues, as detailed in the *Carcinogenicity and Genotoxicity* section. A concentration-related respiratory effect observed in the male rats was focal hyperplasia of the nasal cavity (0/50 controls, 31/50 low-exposed, 1/49 high-exposed) that was not accompanied by an increased incidence in hyperplasia in either the bronchioles or the alveolar epithelium. In the female rats, incidences of nasal cavity abscesses were increased in both exposed groups (1/50, 5/50, 12/50). Hyperplasia of the nasal cavity also occurred in the females (0/50, 24/50, 23/50); this was not accompanied by increased incidences of hyperplasia in either the bronchioles or the alveolar epithelium. The decrease in focal hyperplasia of the nasal cavity in both sexes at the highest exposure level was concomitant with an increase in neoplastic lesions at this exposure. Incidences of other lesions in female rats were only increased in the high-exposed group; these included chronic inflammation, hyperkeratosis, and squamous metaplasia of the nasal cavity, hyperkeratosis of the esophagus, stomach hyperkeratosis and acanthosis, toxic nephropathy and necrosis of the cerebrum. Pigmentation of the spleen (10/50, 28/50, 34/48) and degeneration of adrenal cortex (4/50, 19/50, 13/48) were increased at both levels in female rats. Incidences of testicular lesions, including hyperplasia of the interstitial cells and testicular degeneration, were inversely related to exposure concentration (e.g., interstitial cell hyperplasia occurred in 41/50 control, 18/50 low-exposed and 6/48 high-exposed animals). Other systemic effects that were increased in the high-exposed males included splenic pigmentation and atrophy, hyperkeratosis of the esophagus, and toxic nephropathy. This study identifies a chronic LOAEL of 0.6 ppm for both systemic effects (spleen and adrenal histopathology) and respiratory effects (nasal cavity histopathology) in rats.

In the chronic mouse inhalation study (NTP, 1982; Reznik et al., 1980c), groups of 50 male and 50 female B6C3F1 mice were whole-body exposed to 0, 0.6 or 3 ppm (0, 5.8 or 29 mg/m³) of DBCP for 6 hours/day, 5 days/week for 80 weeks (male controls), 76 weeks followed by observation for 0-1 weeks (low- and high-dose males), 105-107 weeks (female controls), 103 weeks followed by observation for 1 week (low-dose females) or 76 weeks followed by observation for 0-1 weeks (high-dose females). Body weight, clinical observations, and gross and histopathology were evaluated as described for the NTP (1982) rat study. The male and female high-dose groups were terminated early because of early deaths related to respiratory tract tumors (interference with breathing and metastasis to the brain); increased incidences of tumors occurred in the nasal cavity and lungs, as detailed in the *Carcinogenicity and Genotoxicity* section. Early mortality also occurred in low-dose and control mice, but appeared to be associated with urogenital infection, rather than tumor development (NTP, 1982). No clinical signs were reported, but mean body weight gain was depressed by 17-28% in the high-exposed males after week 60 and by 25% in high-exposed females after week 76. Other effects in the male mice included concentration-dependent respiratory effects, including hyperplasia of the nasal cavity (2/42 low-dose and 12/48 high-dose), bronchioles (7/40 and 29/45), and alveolar epithelium (2/40 and 7/45); these effects were not found in controls. The 3 ppm males also had increased incidences of suppurative inflammation in the nasal cavity and focal hyperplasia of the bronchi. Other effects in the high-exposed males included splenic atrophy and toxic nephropathy. Effects observed in males at ≥ 0.6 ppm included hyperkeratosis (0/37 controls, 10/41 low-dose, 17/44 high-dose) and acanthosis (0/37, 6/41, 11/44) in the stomach, and kidney inflammation (0/40, 9/42 and 7/46). There were no concentration-related histological changes in the testes, seminal vesicles, or epididymides. Effects also occurred in female mice at ≥ 0.6 ppm, including suppurative inflammation (0/50, 5/50, 13/50) and hyperplasia of the nasal cavity (0/50, 17/50, 3/50), bronchioles (0/50, 5/49, 11/47) and alveolar epithelium (0/50, 5/49, 11/47). The decreased incidences of nasal cavity hyperplasia (focal) at 3 ppm in both sexes was concomitant with increases in neoplastic lesions at this exposure level. Other findings in the 3 ppm female mice included increased incidences of splenic atrophy and endometrial cyst. Other findings in the ≥ 0.6 ppm females included hyperkeratosis (0/50, 20/48, 24/46) and acanthosis (0/50, 12/48, 18/46) of the stomach. A LOAEL of 0.6 ppm is identified for both systemic effects (gastrointestinal and kidney histopathology) and respiratory effects (nasal cavity histopathology).

Reproductive and Developmental Toxicity Studies:

Groups of 6 male Dutch rabbits were exposed to drinking water that provided reported DBCP intakes of 0, 0.94, 1.88, 3.75, 7.5 or 15.0 mg/kg on 5 days/week (0, 0.7, 1.3, 2.7, 5.4 or 10.7 mg/kg-day) for 10 weeks (Foote et al., 1986a, 1986b). General health, body weight, semen quality, and libido were evaluated throughout the study. Assessments of fertility (mated with untreated females) and serum levels of reproductive hormones (follicle stimulation hormone, luteinizing hormone and testosterone) were performed during the last week of the study. Endpoints evaluated following sacrifice at the end of the study included organ weights (liver,

kidneys, testes, epididymides, accessory sex glands), quantitative histology of testes and epididymides, and sperm morphology and forward motility and morphology. There were no statistically significant ($p \leq 0.05$) changes in any of the study endpoints at 0.7 mg/kg-day. Effects observed at higher doses included dose-related reductions in numbers of all germ cell types within Stage I seminiferous tubular cross sections (significantly reduced numbers of spermatogonia and preleptotene spermatocytes at ≥ 1.3 mg/kg-day, pachytene spermatocytes at ≥ 2.7 mg/kg-day, and round spermatids at ≥ 5.4 mg/kg-day) (Table 1). Other effects included dose-related significantly reduced numbers of leptotene primary spermatocytes per Sertoli cell at ≥ 2.7 mg/kg-day, and significantly reduced mean diameter of seminiferous tubules and increased percentage of sperm with abnormal tails at ≥ 5.4 mg/kg-day (Table 2). Testis weight and volume, and sperm production (number of seminiferous tubules with round or elongating spermatids), output (ejaculate volume times sperm concentration) and motility were reduced, and serum FSH level was increased, at 10.7 mg/kg-day (Table 3). Fertility was not affected at any dose level, as assessed by number of males producing young, number or percentage of live births, total number of young, average litter size, and gestation length. The results summarized above are based on comparisons of mean data from the treated and control groups. Regression analyses showed highly significant correlations between DBCP dosage and essentially all of the testicular responses. The findings of this study indicate that rabbits are more sensitive than rats to testicular effects of DBCP. This study identified a NOAEL of 0.7 mg/kg-day and LOAEL of 1.3 mg/kg-day for reproductive toxicity in male rabbits.

In rabbits, reproductive toxicity was evaluated in groups of 10 New Zealand white males (age 6 months) that were exposed to 0, 0.1, 1 or 10 ppm (0, 0.94, 9.4 or 94 mg/m³) vapor for 6 hours/day, 5 days/week for 14 weeks, and observed for the following 32 weeks (0, 0.1 and 1 ppm groups) or 38 weeks (10 ppm group) (Rao et al., 1982). The 10 ppm rabbits were exposed for only 8 weeks due to high mortality (apparently from pneumonia). Body weight and hematological and clinical chemistry parameters were evaluated, but no exposure related changes were found. No gross lesions were found in the lungs or upper respiratory tract, but these tissues were not examined histologically. Semen was collected during the exposure and recovery periods to assess sperm motility, viability and counts. The average sperm count of the 10-ppm rabbits was significantly less than that of the controls after 7 weeks of exposure, and remained decreased for the duration of the exposure and observation periods. At 1 ppm (9.4 mg/m³), sperm counts were significantly reduced, compared with controls, from weeks 11 to 13 of exposure. At 0.1 ppm (0.94 mg/m³), sperm counts were sporadically lower than control values (significantly reduced at only one interim time point). The percentage of live sperm in the semen of the 10 ppm (94 mg/m³) rabbits was also significantly reduced compared to controls during weeks 8-26. Rabbits exposed to 1 ppm (9.4 mg/m³), but not those exposed to 0.1 ppm (0.94 mg/m³), exhibited significant decreases in the percentage of live sperm during weeks 6, 12 and 13. From the 8th week of exposure onward, the 10-ppm (94 mg/m³) rabbits had a marked decrease in the percentage of progressively motile sperm; no consistent statistically significant decreases in this endpoint were found at ≤ 1 ppm (9.4 mg/m³) (Table 4). Abnormal spermatozoa

Table 1				
Mean (\pm SE) Numbers of Germ Cells per Stage I Seminiferous Tubular Cross Section (Foote et al., 1986b)				
DBCP (mg/kg-day)	Spermatogonia	Primary Spermatocytes		Round Spermatids
		Preleptotene	Pachytene	
0.00	2.3 ^a \pm 0.13	42.5 ^a \pm 2.4	39.3 ^a \pm 3.0	141.3 ^a \pm 11.0
0.7	2.0 ^{a,b} \pm 0.13	41.9 ^{a,b} \pm 2.4	39.7 ^a \pm 3.0	128.5 ^a \pm 11.0
1.3	1.8 ^{b,c,d} \pm 0.12	35.0 ^{b,c} \pm 2.2	36.8 ^{a,b} \pm 2.8	121.8 ^a \pm 10.1
2.7	1.6 ^{c,d} \pm 0.12	29.3 ^{c,d} \pm 2.2	30.0 ^{b,c} \pm 2.8	84.8 ^{a,b} \pm 10.1
5.4	1.5 ^d \pm 0.12	26.0 ^d \pm 2.2	20.9 ^{c,d} \pm 2.8	55.2 ^{b,c} \pm 10.1
10.7 ^f	1.0 ^e \pm 0.15	13.6 ^e \pm 2.6	11.2 ^d \pm 3.4	36.6 ^c \pm 12.3
Mean	1.7 \pm 0.05	31.8 \pm 0.93	30.2 \pm 1.19	95.8 \pm 4.4

^{a-c} Column means with different superscripts differ, $p < 0.01$.

^f Four rabbits only in this group.

Table 2						
Influence of DBCP on Selected Variables Expressed as a Percentage of Control Values (Foote et al., 1986b)						
Variable Measured	% of Controls at Each Level of DBCP (mg/kg-day)					
	0.00	0.7	1.3	2.7	5.4	10.7
Paired testis weight	100	87	96	89	71	45
Diameter of seminiferous tubules	100	99	95	92	85	71
Leptotene spermatocytes per Sertoli cell	100	93	89	68	57	29*
Number of germ cells per Stage I tubule						
Spermatogonia	100	87	78	70	65	43*
Preleptotene spermatocytes	100	98	82	69	61	32*
Pachytene spermatocytes	100	101	93	76	53	28*
Round spermatids	100	91	86	60	39	26*

* Two animals with testicular damage too severe to classify stages are omitted.

DBCP Dosage (mg/kg-day)	Hormone Concentration (ng/ml)		
	FSH	LH	Testosterone
0.00	2.60 ^a	0.25 ^a	4.33 ^a
0.7	2.92 ^a	0.16 ^a	1.37 ^a
1.3	1.78 ^a	0.48 ^a	1.76 ^a
2.7	1.90 ^a	0.54 ^a	2.66 ^a
5.4	4.62 ^a	0.41 ^a	2.59 ^a
10.7	7.33 ^b	1.02 ^a	2.63 ^a
Overall means	3.57	0.49	2.54
Standard error	0.55	0.13	0.44

^{a-b} Treatment means within columns with different superscripts are significantly different ($p < 0.01$).

Table 4				
Viability of Sperm from Control and DBCP Exposed Rabbits (Rao et al., 1982)				
Week of Study	% Live Sperm, Mean \pm S.D.			
	ppm DBCP (mg/m ³)			
	0	0.1 (0.94)	1.0 (9.4)	10 (94)
Pre-Exposure				
-2	87 \pm 9	88 \pm 28	92 \pm 6	87 \pm 6
-1	93 \pm 3	88 \pm 8	89 \pm 6	89 \pm 5
Exposure				
1	88 \pm 15	71 \pm 25	81 \pm 25	89 \pm 6
2	92 \pm 5	87 \pm 9	84 \pm 18	90 \pm 6
3	76 \pm 21	84 \pm 12	79 \pm 15	86 \pm 10
4	88 \pm 6	80 \pm 14	83 \pm 5	76 \pm 20 ^a
5	78 \pm 12	81 \pm 5	80 \pm 15	78 \pm 8
6	89 \pm 5	86 \pm 6	80 \pm 13 ^a	82 \pm 8
7	88 \pm 5	87 \pm 10	86 \pm 10	81 \pm 11
8	88 \pm 5	85 \pm 8	73 \pm 25	49 \pm 26 ^a
9	87 \pm 11	88 \pm 12	83 \pm 9	53 \pm 36 ^a
10	83 \pm 16	90 \pm 6	82 \pm 11	49 \pm 17 ^a
11	88 \pm 5	84 \pm 18	79 \pm 13	31 \pm 14 ^a
12	84 \pm 10	89 \pm 6	69 \pm 22 ^a	-/b
13	86 \pm 14	78 \pm 18	72 \pm 12 ^a	5 ^c
14	83 \pm 13	87 \pm 6	74 \pm 17	-/b

Table 4 cont.				
Week of Study	% Live Sperm, Mean \pm S.D.			
	Ppm DBCP (mg/m ³)			
	0	0.1 (0.94)	1.0 (9.4)	10 (94)
Post-Exposure				
16	90 \pm 5	93 \pm 4	84 \pm 8	0 ^c
19	85 \pm 22	92 \pm 3	92 \pm 8	-/b
24	79 \pm 8	86 \pm 7	87 \pm 16	-/b
26	96 \pm 4	96 \pm 2	92 \pm 5	36 ^c
27	88 \pm 12	86 \pm 24	95 \pm 4	51 ^c
28	96 \pm 2	76 \pm 29	89 \pm 11	47 ^c
30	86 \pm 15	82 \pm 15	82 \pm 17	46 \pm 65 ^a
32	82 \pm 15	89 \pm 6	86 \pm 13	34 ^c
34	82 \pm 11	79 \pm 23	62 \pm 31	47 \pm 17 ^a
36	85 \pm 11	81 \pm 22	89 \pm 8	47 \pm 23 ^a
38	86 \pm 12	53 \pm 20 ^a	77 \pm 30	53 \pm 10 ^a
40	84 \pm 14	89 \pm 6	84 \pm 4	59 \pm 17 ^a
42	67 \pm 22	82 \pm 7	81 \pm 3	66 \pm 8
44	81 \pm 16	91 \pm 4	92 \pm 3	72 \pm 1
46	90 \pm 6	78 \pm 5	68 \pm 36	70 \pm 13

^a Significantly different from the control value by Dunnett's test, $p < 0.05$.

^b Insufficient number of sperm for determination of the percentage of live sperm.

^c Single value.

within the seminiferous tubules were counted in 3-4 rabbits per group; the percentage of abnormal sperm at 14 weeks was 5% in controls, 10% at 0.1 ppm (0.94 mg/m³), and 18% at 1 ppm (9.4 mg/m³).

To assess the effects of DBCP on fertility in the rabbits, exposed males were mated to unexposed females at study weeks 14 and 41 (Rao et al., 1982) (Tables 5, 6). There were no effects on the libido of the exposed male rabbits during week 14, based on percentages of males (78-100%) that copulated with unexposed females. Five of the 10 males exposed to 10 ppm were infertile (none of the females that they were mated with became pregnant). The mean number of implantations/litter in the 1 ppm (9.4 mg/m³) group was significantly less than that of the control group. During week 41 (27 weeks post-exposure), all rabbits exposed to 0.1 or 1 ppm (0.94 or 9.4 mg/m³) DBCP produced normal litters, and 2 of the 5 infertile males exposed to 10 ppm (94 mg/m³) recovered (sperm counts increased) and produced normal litters. Serum levels of follicle stimulating hormone (FSH) were significantly elevated at 14 weeks in the males exposed to 1 ppm (9.4 mg/m³) and at 46 weeks in the males exposed to 10 ppm (94 mg/m³), but serum levels of testosterone were unchanged (Table 7). The increases in serum FSH were consistent with the decreases in sperm count. Gross pathologic examinations showed small testes size in rabbits exposed to 1 or 10 ppm. Testes weight was significantly decreased to 50% of control values (week 14) in the group exposed to 1 ppm (9.4 mg/m³) and to 75% of control values (week 8) in the group exposed to 10 ppm. Histological examinations showed reproductive system effects that included atrophy of the testes, epididymides, and accessory sex glands, including the prostate. The testicular atrophy was severe, as characterized by nearly complete or complete loss of spermatogenic elements in nearly all seminiferous tubules. Following the recovery period, tubular regeneration was observed in the testes of some 10 ppm (94 mg/m³) rabbits (3/5 had regeneration such that 25% of the seminiferous tubules appeared normal). At 1 ppm, testicular recovery was reported to be nearly complete in some rabbits (incidences not given). The testes of the 0.1 ppm rabbits appeared normal. The lack of exposure-related adverse testicular and fertility effects at 0.1 ppm indicates that this study identified a NOAEL of 0.1 ppm (0.94 mg/m³) and LOAEL of 1 ppm (9.4 mg/m³) for reproductive effects in rabbits.

Groups of 15 Sprague-Dawley male rats were administered DBCP in corn oil by gavage in daily doses of 0, 0.94, 1.88, 3.75, 7.5 or 15.0 mg/kg for 77 days (Amann and Berndtson, 1986). Body weight and clinical signs were assessed throughout the study. An additional group of 15 males was used as a nongavaged control group. From day 65 to 71, each male was caged with two untreated female rats to assess fertility; determinations included pregnancy rate, numbers of corpora lutea, normal and dead embryos, and implantation sites. Endpoints evaluated in males at the end of the study included selected organ weights (liver, kidneys, testes, epididymis, tunica albuginea, vesicular gland), serum levels of LH and FSH, epididymal sperm reserves, testicular sperm production, quantitative testicular histopathology (including diameter of seminiferous tubules and numbers and distribution of germ cell types), percentage progressive

Table 5				
Fertility of Unexposed Female Rabbits Mated with DBCP-Exposed Males (14 weeks) (Rao et al., 1982)				
	ppm DBCP (mg/m ³)			
	0	0.1 (0.94)	1.0 (9.4)	10 (94)
No. of males	9	10	9	5
% of males which copulated (no.)	100(9)	90(9)	78(7)	100(5)
% of fertile males (no. fertile males/no. bred to females)	67(6/9)	100(9/9)	86(6/7)	0(0/5)
Corpora lutea/dam ^a	10 ± 1	10 ± 3	8 ± 4	
Total implantation/litter ^a	9 ± 1	9 ± 1	6 ± 2 ^b	
Resorptions/litter ^a	0.7 ± 0.8	0.9 ± 0.8	0.3 ± 0.5	
% pre-implantation loss ^a	9 ± 15	14 ± 13	21 ± 20	
% post-implantation loss ^a	8 ± 9	10 ± 9	4 ± 6	

^a Mean ± S.D.

^b Significantly different from the control value by the appropriate statistical test, $p < 0.05$.

Table 6				
Fertility of Unexposed Female Rabbits Mated with DBCP-exposed Males (41 weeks) (Rao et al., 1982)				
	ppm DBCP (mg/m ³)			
	0	0.1 (0.94)	1.0 (9.4)	10 (94)
No. of males	6	6	5	5
% of males which copulated (no.)	83(5)	100(6)	80(4)	100(5)
% of fertile males (no. males impregnating one female/number bred)	100(5/5)	100(6/6)	100(4/4)	40(2/5)
% fertile males (no. males impregnating two females/number bred)	80(4/5)	100(6/6)	100(4/4)	20(1/5)
Corpora lutea/dam*	10 ± 1	10 ± 2	10 ± 1	12 ± 3
Total implantations/litter*	8 ± 2	8 ± 2	8 ± 2	10 ± 6
Resorptions/litter*	0.7 ± 0.7	0.8 ± 0.5	0.2 ± 0.3	1.0 ± 1.4
% pre-implantation loss*	24 ± 20	22 ± 17	19 ± 13	22 ± 26
% post-implantation loss*	16 ± 24	15 ± 12	3 ± 3	7 ± 10

* Mean ± S.D.

No value differed significantly from the control value by the appropriate statistical test.

Week of Study	ppm DBCP (mg/m ³)	n	FSH (ng/ml)	Testosterone (ng/ml)
14	0	3	6.9 ± 2.7	1.2 ± 1.1
	0.1 (0.94)	4	5.6 ± 3.4	1.4 ± 1.6
	1.0 (9.4)	4	15.1 ± 4.6*	3.4 ± 2.7
46	0	6	3.5 ± 1.7	1.4 ± 0.9
	0.1 (0.94)	6	4.5 ± 3.8	3.4 ± 2.7
	1.0 (9.4)	5	3.7 ± 3.7	0.8 ± 0.6
	10.0 (94)	5	12.9 ± 2.0*	1.0 ± 1.0

* Significantly different from the control value by Dunnett's test, $p < 0.05$.

n = number of animals.

sperm, and sperm morphology. Comparison of group mean values showed no clear effects of DBCP at doses below 15 mg/kg-day. Mean final body weight in the 15 mg/kg-day group was 11.6% lower than the gavaged controls and 15.1% lower than the nongavaged controls; the difference was statistically significant ($p < 0.05$) only compared to the nongavaged control group. There were no significant changes in paired testes weight at any dose in comparison to either control group. Evaluation of the left testis (right testicular data not reported) showed statistically significant effects at 15 mg/kg-day that included reduced testis weight compared to nongavaged controls, reduced left testicular parenchymal weight compared to either control group, reduced daily sperm production per testis compared to nongavaged controls, reduced number of epididymal sperm in cauda compared to gavaged controls, and reduced total number of epididymal sperm compared to either control group. The histological evaluations showed significantly reduced seminiferous tubule diameter and ratio of leptotene spermatocytes/Sertoli cells at 15 mg/kg-day compared to either control group. There was no treatment-related effect on fertility (pregnancy rate), although pregnant females had significantly more dead embryos and a higher ratio of dead embryos to corpora lutea at 15 mg/kg-day compared to either control group. Regression analyses showed that there were highly significant correlations between increasing DBCP dosage and adverse effects for a number of endpoints (e.g., body weight, total number of epididymal sperm, paired testicular weight, left cauda epididymis weight, daily sperm production by left testis, seminiferous tubule diameter, and ratio of leptotene spermatocytes to Sertoli cells). Discriminant analysis using two endpoints of spermatogenesis suppression (daily sperm production per testis and ratio of leptotene spermatocytes to Sertoli cells) indicated that values for the three higher dose groups (≤ 3.75 mg/kg-day) were greater than those for the nongavaged control group. This study identified a NOAEL of 7.5 mg/kg-day and LOAEL of 15 mg/kg-day for reproductive effects, including decreased fertility, in rats.

Groups of 20 male Sprague-Dawley rats were exposed to 0, 0.4, 3.3, 5.4 or 9.7 mg DBCP/kg-day in drinking water for 64 days in a subchronic study (Heindel et al., 1989) detailed in the *Oral Systemic Toxicity* section. There were no clear effects on sperm development or numbers, or weights of the testes or seminal vesicles, indicating that a LOAEL for reproductive toxicity was not identified and that the reproductive NOAEL was 9.7 mg/kg-day.

A one-generation oral reproduction study was conducted in which groups of 10 male and 10 female Sprague-Dawley rats were exposed to drinking water that provided reported average DBCP intakes of 0, 0.015, 0.26, 2.96 or 19.43 mg/kg-day (Johnston et al., 1986). Both sexes were exposed for 60 days before mating and through mating, and subsequently continuing in females through gestation and the first five days of lactation. Each male was paired with a female of the same dose level for a mating period of 5 days; the male was then rested for 2 days before being paired with another female of the same dose level for a second mating period of 5 days. Adult males were killed 2 days following the second mating period, and adult females and pups were killed on postnatal day 4. Endpoints that were evaluated before or at parturition included clinical signs, food and water consumption, body weight, gestation length, litter size,

number of live and dead births, and resorbed implantation sites. Endpoints that were evaluated postpartum included number of live pups, litter and maternal weights, and pup sex ratio. Fertility, gestation, gestation survival, and pup viability indices were calculated. Sacrificed adults (both sexes) and pups were examined for gross pathological changes, and selected organ weights (including liver, kidneys, testes, epididymides, male accessory sex organs, ovaries and uterus) were measured in adults. Histological examinations were performed in control and high-dose adults (including liver, kidneys, stomach, testes, coagulating glands, seminal vesicles, epididymides, prostate, ovaries, oviduct, uterus, cervix) and male pups (limited to testes and epididymides). Effects were observed in adults of both sexes at 19.43 mg/kg-day that included significantly reduced food and water consumption and body weight gain (e.g., weight gain through day 69 was 63.5 and 70.1% less than controls in males and females, respectively). Relative liver weight was significantly increased in adult males at 19.43 mg/kg-day (10.6% higher than controls), but there were no exposure-related gross or histopathological changes in the liver, reproductive organs, or any other tissues in either sex. The only effects in offspring occurred at 19.43 mg/kg-day, consisting of significantly reduced mean pup body weight on postnatal days 1 and 4 (both $p \leq 0.05$) and a non-significant decrease in day 4 survival index (percentage of live pups surviving to day 4; 75% compared to 99% in controls, $p > 0.05$). The decreases in pup growth and survival were considered to be secondary effects resulting from the maternal toxicity. This study identified a NOAEL of 2.96 mg/kg-day and LOAEL of 19.43 mg/kg-day for developmental toxicity (reduced pup body weight) in rats. There were no effects on fertility or any other reproductive endpoints, indicating that a LOAEL for reproductive toxicity was not identified and that the reproductive NOAEL is 19.43 mg/kg-day.

Reproductive toxicity in Swiss CD-1 mice was tested using the NTP continuous breeding protocol in which groups of 20 males and 20 females were orally treated with DBCP in corn oil by gavage in dose levels of 25, 50 or 100 mg/kg-day (Reel et al., 1984; Lamb et al., 1997). Groups of 40 males and 40 females were used as vehicle controls. The mice were treated for 7 days before mating and then during a 98-day cohabitation/continuous breeding period and a subsequent 21-day segregation period, for a total study duration of 126 days. The last litters produced by control and high-dose mice were raised until weaning when one or two female and male pups from each litter were selected for eventual breeding. These F1 mice received the same 0, 25, 50 or 100 mg/kg-day treatments as their parents and were paired at 90 ± 10 days of age for up to 7 days, after which females were allowed to deliver. Reproductive endpoints were evaluated in both generations and included number of litters/pair, number of live pups/litter, pup body weight/litter, cumulative days to litter, absolute testis weight, relative epididymis, prostate and seminal vesicle weights, epididymal sperm parameters (number, motility, morphology), and estrous cycle length. Other endpoints that were evaluated in both generations included body weight, food and water consumption, clinical signs and mortality, and relative liver and kidney weights. The mean number of litters per pair was significantly reduced in the F0 mice at 25 and 100 mg/kg-day; this effect was not observed at 50 mg/kg-day. None of the other reproductive or systemic toxicity endpoints were significantly affected in the F0 generation. Effects in the F1

generation only occurred at 100 mg/kg-day and were essentially limited to significantly reduced relative epididymis and prostate weights in males and significantly increased relative liver weight in both sexes. Analysis of covariance showed an indication of reduced pup weights in the offspring of the 100 mg/kg-day F1 mice. The tendencies for decreased numbers of litters in the F0 generation and reduced pup weights in the F1 generation suggest that 100 mg/kg-day is a LOAEL and 50 mg/kg-day is a NOAEL for reproductive and developmental toxicity in mice.

Developmental toxicity was evaluated in groups of 15 Wistar rats that were treated with 0, 12.5, 25.0 or 50.0 mg/kg-day doses of DBCP in corn oil by gavage on days 6-15 of gestation (Ruddick and Newsome, 1979). Dams were sacrificed on gestation day 22, and maternal weight gain, litter size, litter weight, and fetal skeletal and visceral abnormalities were evaluated. Maternal weight gain was significantly ($p < 0.05$) reduced at 25 and 50 mg/kg-day (33 and 69% less than controls, respectively), and fetal weight was significantly reduced at 50 mg/kg-day (12% less than controls). There was a non-statistically significant reduction in the number of litters at 50 mg/kg-day (10, compared to 13 in controls and 13 or 14 in the other treated groups). Additionally, although 4/15 females in the 50 mg/kg-day group were recorded as not pregnant at necropsy, 3 of the 4 non-pregnant females had uteri that were edematous and contained a pinkish fluid suggestive of embryoletality. The skeletal and visceral examinations showed no evidence of malformations or variations at any dose level. The authors concluded that the fetotoxic effects were secondary to the maternal toxicity. This study identifies a NOAEL of 12.5 mg/kg-day and LOAEL of 25 mg/kg-day for maternal effects, as well as a NOAEL of 25 mg/kg-day and LOAEL of 50 mg/kg-day for embryoletality, in rats.

DBCP induced dominant lethal mutations in orally-exposed rats. Groups of 15 male SD rats were administered DBCP in olive oil by gavage in doses of 0, 10 or 50 mg/kg-day for 5 days and then mated with untreated females in the pro-estrus stage (Teramoto et al., 1980). The male rats were mated to one female each week for 10 weeks. Groups of 6 male BDF1 mice were similarly treated with 0, 50 or 150 mg DBCP/kg-day for 5 days. After treatment, each male mouse was allowed to mate with two untreated females for 7 days, and new female mice were mated with the treated males weekly for 6 weeks. Mated female rats and mice were sacrificed 12-14 days after mating, and numbers of corpora lutea, implants, live embryos, and early and late embryonic deaths were determined. DBCP induced dominant lethal mutations in the male rats at ≥ 10 mg/kg-day, as indicated by a significantly increased incidence of dead implants in rats mated at weeks 4 and 5 (early spermatid stage) in the 10 mg/kg-day group and weeks 1-6 (peaked at weeks 4 and 5) in the 50 mg/kg-day group. DBCP did not reduce the frequency of fertile mating in the mice, indicating that there was no dominant lethal effect in this species at ≤ 150 mg/kg-day. The dominant lethal effect in rats was confirmed in similarly-designed studies in which males were orally treated with ≥ 10 mg DBCP/kg-day for 5 days before mating (Au et al., 1990; Saito-Suzuki et al., 1982).

No inhalation developmental toxicity studies of DBCP were located, although reproductive toxicity was tested by inhalation in rats and rabbits (Rao et al., 1982, 1983).

Reproductive toxicity in male and female rats was evaluated as part of a subchronic inhalation study in which Sprague-Dawley rats (30/sex/group) were exposed to 0, 0.1, 1, or 10 ppm (0, 0.97, 9.7, or 97 mg/m³) of DBCP vapor, 6 hours/day, 5 days/week for 14 weeks, followed by a 32-week recovery period, for a total study duration of 46 weeks (Rao et al., 1983). Non-reproductive effects of exposure are summarized in the *Inhalation Systemic Toxicity* section. Absolute and relative testes and epididymides weights were significantly reduced compared to controls at 10 ppm after 14 weeks (interim sacrifice), and relative testes weight remained significantly reduced in 10 ppm males after 46 weeks (final sacrifice). No significant differences were seen in organ weights of exposed female rats compared with their controls. Gross and histopathologic changes occurred in testes (decreased size and dark color, decreased spermatogenesis in individual seminiferous tubules, lack of germinal cells in 5/5 males) at 10 ppm after 14 weeks. At the 46-week terminal sacrifice, concentration-related testicular atrophy was observed in all male groups (12/18 at 10 ppm, 5/20 at 1 ppm, 3/19 at 0.1 ppm, and 2/17 at 0 ppm). To assess fertility in the male rats, groups of 20 males were mated with unexposed females during weeks 2, 4, 6, 10, 12, 14, 16, 20, 24, 28, and 42. The percentage of males that impregnated at least one female was at least 85% at all exposure levels, and there were no significant differences between the exposed and control groups. A significant increase ($p < 0.05$) in post-implantation loss was observed in the 10 ppm group during the fourth week of exposure and remainder of the exposure period; this appears to be a treatment-related dominant lethal effect. By the tenth week of recovery, the average number of resorptions in the 10-ppm group was similar to that of the controls. To assess fertility in the female rats, 20 females per group were mated with unexposed males for a 5-day period during weeks 14, 18 and 20. Fertility of the exposed female rats was not significantly different from that of the controls except for a higher incidence of 10-ppm dams having litters of 4 or fewer pups. No exposure-related major gross alterations were observed in pups resulting from the matings with the exposed rats of either sex. This study identified a NOAEL of 1 ppm and LOAEL of 10 ppm for male reproductive toxicity in rats.

Carcinogenicity Studies:

Information on the carcinogenicity of orally administered DBCP is available from an NCI (1978) gavage bioassay in rats and mice. In the rat study, groups of 50 male and 50 female Osborne-Mendel rats were treated with DBCP in corn oil by gavage at reported time-weighted average doses of 15 or 29 mg/kg-day on 5 days/week. Groups of 20 males and 20 females were used as vehicle-treated and untreated controls. The low-dose males were treated for 78 weeks and observed untreated for the following 5 weeks. The high-dose males were treated for 64 weeks and then killed. The low- and high-dose females were treated for 73 and 64 weeks, respectively, and then killed. The high-dose male and low- and high-dose female groups were

terminated early because of high mortality that was likely tumor-related. Both sexes of vehicle and untreated control rats were killed after 83 and 109 weeks, respectively. Clinical signs and body weight were evaluated during the study. Gross pathology and histopathology of major tissues, organs and gross lesions were evaluated at the time of sacrifice and, when possible, in animals that died early. Abdominal urine stains and hunched appearance were prevalent clinical signs and observed as early as weeks 6 and 18, respectively. Dose-related decreases in body weight gain occurred in both sexes after approximately the first 20 weeks of treatment. Ninety percent of the low-dose males died by week 83 and 80% of the high-dose males died by week 62. High incidences of squamous-cell carcinomas of the forestomach occurred in low- and high-dose male and female rats as detailed in Table 8. This lesion occurred with frequent metastases to the abdominal viscera and lungs, and was not found in either vehicle or untreated controls. Increased incidences of mammary gland adenocarcinomas also occurred in both groups of treated female rats (2/20 untreated controls, 0/20 vehicle controls, 24/50 low-dose, 31/50 high-dose). Treatment-related nonneoplastic lesions were found in the kidneys and testes, as detailed in the *Oral Systemic Toxicity* section.

Table 8								
Incidences of Rat Forestomach Tumors in the NCI (1978) Chronic Gavage Study								
	Male Rats				Female Rats			
Dose (mg/kg-day)	0 ^a	0 ^b	15	29	0 ^a	0 ^b	15	29
Tumor Incidence ^c	0/20	0/19	47/50	47/50	0/20	0/20	38/50	29/49
^a Untreated control group ^b Vehicle (corn oil) control group ^c Forestomach squamous-cell carcinoma								

A study was conducted to identify early forestomach lesions in rats exposed to the same dose levels that induced the forestomach tumors in the NCI (1978) bioassay (Ghanayem et al., 1986). Groups of 8 male F344 rats were gavaged with 15 or 29 mg DBCP/kg-day in corn oil on 5 days/week for 2 weeks. Sixteen males were used as vehicle controls. Histological examination of the entire stomach was performed on all treated and control animals 24 hours after the last dose. The high-dose rats developed significantly ($p < 0.001$) increased 100% incidences of forestomach epithelial (mucosal) cell proliferation (0/16 controls, 1/8 low-dose, 8/8 high-dose) and hyperkeratosis (0/16, 0/8, 8/8). It appears that the proliferative changes in the mucosa were generally more pronounced toward the proximal (esophageal) end of the forestomach, with a gradual decrease in severity in more distal areas. Although the results of this study suggest that forestomach epithelial cell proliferation may precede tumor development in the forestomach in

rats, the authors note that there is insufficient evidence to conclude that proliferation is necessarily related to the carcinogenic process at this tissue site.

In the NCI (1978) oral mouse study, groups of 50 B6C3F1 mice of each sex were treated with DBCP in corn oil by gavage at reported time-weighted average doses of 114 or 219 mg/kg-day (males), or 110 or 209 mg/kg-day (females), on 5 days/week. Groups of 20 males and 20 females were used as vehicle-treated and untreated controls. The untreated control, vehicle control, low-dose and high-dose male mice were treated for 78, 59, 60 and 47 weeks, respectively, and then killed. The untreated control, vehicle control, low-dose and high-dose female mice were treated for 90, 60, 60 and 47 weeks, respectively, and then killed. The treated male and female groups (both dose levels) were terminated early because of high mortality that was likely tumor-related. Body weight, clinical observations, and gross and histopathology were evaluated as described for the NCI (1978) rat study. There were no apparent effects of treatment on body weight gain, appearance or behavior. Clinical signs characterized by a hunched or thin appearance and apparent compound-related deaths were observed in the high-dose animals beginning in week 38 of the study. Eighty-four percent of the low-dose males died by week 59 and 80% of the high-dose males died by week 47. Over 90% of each dosed group was found to have squamous-cell carcinomas of the forestomach, as detailed in Table 9. These lesions were histologically similar to the squamous-cell carcinomas induced in the rats and also occurred with frequent metastases to the abdominal viscera and lungs. No forestomach neoplasms occurred in either vehicle or untreated control mice. The only treatment-related nonneoplastic lesion was dose-related toxic nephropathy, as detailed in the *Oral Systemic Toxicity* section.

Table 9								
Incidences of Mouse Forestomach Tumors in the NCI (1978) Gavage Study								
	Male Mice				Female Mice			
Dose (mg/kg-day)	0 ^a	0 ^b	114	219	0 ^a	0 ^b	110	209
Tumor Incidence ^c	0/20	0/20	43/46	47/49	0/20	0/20	50/50	47/48
^a Untreated control group ^b Vehicle (corn oil) control group ^c Forestomach squamous-cell carcinoma								

Unpublished chronic studies of dietary DBCP were conducted in rats and mice (Hazelton Laboratories, 1977, 1978; Shell Oil Company, 1986). In the rat study (Hazelton Laboratories, 1977), groups of 60 male and 60 female Charles River rats were exposed to nominal DBCP intake levels of 0, 0.3, 1.0 or 3.0 mg/kg-day in the diet for 104 weeks. In an adjustment for

evaporative losses of DBCP from the feed, U.S. EPA (1979) estimated that the actual dosage intakes were 0, 0.20, 0.68 and 2.0 mg/kg-day. Shell Oil Company (1986) more recently estimated that the actual intakes were 0, 0.24, 0.80 and 2.39 mg/kg-day; these estimates appear to have adjusted for a food consumption calculation error, as well as for evaporative losses. An interim kill of 10 rats/sex/group was performed at 52 weeks. There were no treatment-related clinical signs of toxicity or effects on survival, although mean body gain was significantly (52%) lower than controls in the high-dose males. Histological examinations showed significantly increased incidences of stomach, kidney and liver tumors in the high-dose rats, as summarized in Table 10. The only treatment-related non-neoplastic lesion was an increased severity of interstitial nephritis, as detailed in the *Oral Systemic Toxicity* section. Additional information on experimental design and results was not reported in the available summaries of this study (Shell Oil Company, 1986; U.S. EPA, 1979, 1988a).

Table 10								
Rat Tumor Incidences in the Hazelton Laboratories (1977) Diet Study of Dibromochloropropane (U.S. EPA, 1979; Shell Oil Company, 1986)								
	Male Rats				Female Rats			
(mg/kg-day)	0	0.24	0.80	2.39	0	0.24	0.80	2.39
Stomach, carcinoma ^a	0/48	0/46	3/46	21/41 ^g	0/48	0/45	0/47	8/43 ^g
Stomach, total ^b	0/48	0/46	3/46	21/41 ^g	0/48	0/45	0/47	10/43 ^g
Kidney, carcinoma ^c	0/48	1/46	3/46	9/41 ^g	0/48	0/45	0/47	8/43 ^g
Kidney, total ^d	0/48	1/46	4/46	15/41 ^g	0/48	0/45	0/47	10/43 ^g
Liver, carcinoma ^e	0/48	1/46	2/46	5/41 ^g	0/48	1/45	3/47	0/43
Liver, total ^f	0/48	5/46	4/46	8/41 ^g	0/48	3/45	5/47 ^g	3/43
^a Squamous cell carcinoma ^b Squamous cell papilloma or carcinoma ^c Renal tubular cell carcinoma ^d Renal tubular cell adenoma or carcinoma ^e Hepatocellular carcinoma ^f Neoplastic nodules or hepatocellular carcinoma ^g Significantly greater than control group.								

In the unpublished chronic dietary study in mice (Hazelton Laboratories, 1978), groups of 50 male and 50 female HaM/ICR Swiss mice were exposed to nominal DBCP intake levels of 0, 0.3, 1.0 or 3.0 mg/kg-day in the diet for 78 weeks. In an adjustment for evaporative losses of DBCP from the feed, U.S. EPA (1979) estimated that the actual dosage intakes were 0, 0.28, 0.91 and 2.7 mg/kg-day. Shell Oil Company (1986) more recently estimated that the actual dosage intakes were 0, 0.3, 1.6 and 4.8 mg/kg-day; these estimates appear to have adjusted for a food consumption calculation error as well as for evaporative losses. The mice may have received some inhalation exposure because it was common for the mice to sleep in the feed cup. Interim sacrifices were not performed. There were no treatment-related clinical signs of toxicity or effects on body weight gain, food consumption or survival. Histological examinations were only conducted in the 0 and 2.7 mg/kg-day groups and showed significantly increased incidences of tumors in the nonglandular stomach in both sexes. The predominant type of tumor in the nonglandular stomach was squamous cell carcinoma, which occurred in 26/49 high-dose males and 19/50 high-dose females, and in none of the controls (0/50 males, 0/50 females). Stomach papillomas occurred in 6/49 high-dose males, 6/50 high-dose females (6/50) and no controls. Non-neoplastic lesions were also observed in the stomach of the high-dose mice, as discussed in the *Oral Systemic Toxicity* section. Additional information on the experimental design and results was not reported in the available summaries of this study (Shell Oil Company, 1986; U.S. EPA, 1979, 1988a).

Information on the carcinogenicity of inhaled DBCP is available from an NTP (1982) carcinogenesis bioassay in rats and mice. In the NTP rat study, groups of 50 male and 50 female F344 rats were exposed to DBCP by whole body inhalation in concentrations of 0, 0.6 or 3 ppm (0, 4 or 29 mg/m³) for 6 hours/day, 5 days/week for 105-107 weeks (controls), 103 weeks followed by observation for 1 week (low-dose), or 84 weeks followed by observation for 0-1 weeks (high-dose). The male and female high-dose groups were terminated early because of accelerated mortality associated with respiratory tract tumors. Clinical signs and body weight were evaluated during the study, and gross and histological examinations on all major tissues, including the nasal cavity, were performed at the time of sacrifice and, when possible, in animals that died early. Increasing numbers of treated rats of both sexes had severe respiratory signs and palpable masses on the face or nasal areas that began to be detected at week 46, and body weight gain was decreased in high-dose males and females after approximately week 65. Statistically significant increased incidences of tumors occurred in the nasal cavity (both sexes), tongue (both sexes), pharynx (females), adrenal cortex (females), and mammary gland (females), as summarized in Table 11. The respiratory tract tumors were major contributing factors in the early deaths, due to interference with breathing and metastasis to the brain. Exposure-related non-neoplastic lesions occurred in the nasal cavity and other tissues as summarized in the *Inhalation Systemic Toxicity* section. Inflammation, hyperplasia, hyperkeratosis of the nasal mucosa and adjacent structures were found in the exposed rats. In addition, an increased incidence of hyperkeratosis, acanthosis, and chronic inflammation of the stomach were observed in high-dosed animals. Toxic changes related to DBCP exposure also included toxic tubular

nephropathy in the high-dose male and female animals which was characterized by cytomegalic nuclei in tubule cells, especially those of the pars recta. A few dosed animals had focal hyperplasia of the renal tubular cells characterized by the presence of pale, very large cells. These unusual changes may be related to exposure, since morphology of these cells was similar to those observed in the tubular-cell adenomas and adenocarcinomas.

In the NTP (1982) mouse inhalation study, groups of 50 male and 50 female B6C3F1 mice were whole-body exposed to 0, 0.6 or 3 ppm (0, 4 or 29 mg/m³) of DBCP for 6 hours/day, 5 days/week for 80 weeks (male controls), 76 weeks followed by observation for 0-1 weeks (low- and high-dose males), 105-107 weeks (female controls), 103 weeks followed by observation for 1 week (low-dose females) or 76 weeks followed by observation for 0-1 weeks (high-dose females). The male and female high-dose groups were terminated early because of early mortality associated with respiratory tract tumors. Early mortality also occurred in low-dose and control mice but appeared to be associated with urogenital infection rather than tumor development. Body weight, clinical observations, and gross and histopathology were evaluated as described for the NTP (1982) rat study. No clinical signs were reported, but mean body

	Male Rats			Female Rats		
	0 ppm	0.6 ppm	3.0 ppm	0 ppm	0.6 ppm	3.0 ppm
Nasal cavity ^a	0/50 ^b	32/50 ^c	39/49 ^c	1/50 ^b	21/50 ^c	42/50 ^c
Tongue, squamous-cell papilloma and/or carcinoma	0/50 ^b	1/50	11/49 ^c	0/50 ^b	4/50	9/50 ^c
Pharynx, squamous-cell papilloma and/or carcinoma	0/50	3/50	1/49	0/50 ^b	0/50	6/50 ^c
Adrenal, cortical adenoma	1/49	6/49	3/48	0/50	7/50 ^c	5/48 ^c
Mammary gland, fibroadenoma	0/50	0/50	0/49	4/50	13/50 ^c	4/50

^aIncludes unspecified carcinoma, squamous-cell carcinoma, squamous-cell papilloma, unspecified adenoma, unspecified adenocarcinoma, adenomatous polyp and carcinosarcoma.
^bSignificant dose-related trend.
^cSignificantly greater than control group.

weight gain was decreased in the high-dose males and females after approximately week 60. Statistically significant increased incidences of tumors occurred in the nasal cavity and lungs of both sexes, as summarized in Table 12. The respiratory tract tumors were major contributing factors in the early deaths, due to interference with breathing and metastasis to the brain. Exposure-related non-neoplastic lesions occurred in the nasal cavity, lungs and other tissues, as summarized in the *Inhalation Systemic Toxicity* section. In mice inflammation and hyperplasia of the nasal mucosa and related structures, multifocal hyperplasia of the lung, hyperkeratosis and acanthosis in the forestomach, and minimal toxic tubular nephropathy in the kidney characterized by cytomegaly of the occasional tubular epithelial cells were observed.

Table 12						
Mouse Tumor Incidences in the NTP (1982) Inhalation Bioassay of Dibromochloropropane						
	Male Mice			Female Mice		
	0 ppm	0.6 ppm	3.0 ppm	0 ppm	0.6 ppm	3.0 ppm
Nasal cavity ^a	0/45 ^b	1/42	21/48 ^c	0/50 ^b	11/50 ^c	38/50 ^c
Lung ^d	0/41 ^b	3/40	11/45 ^c	4/50 ^b	12/50 ^c	18/50 ^c
^a Unspecified/squamous-cell carcinoma, unspecified adenocarcinoma and adenomatous polyp were most prevalent. Also observed were squamous-cell papilloma, unspecified malignant neoplasm, carcinosarcoma, fibrosarcoma, unspecified sarcoma, keranthoacanthoma and hemangiosarcoma. ^b Significant dose-related trend. ^c Significantly greater than control group. ^d The most commonly occurring lung neoplasms were alveolar/bronchiolar adenoma and carcinoma and bronchus/bronchiole papillary carcinoma. Also observed were bronchus/bronchiole papillary adenoma and unspecified/bronchus squamous-cell carcinoma.						

Mode of Action:

DBCP is metabolized via oxidation by cytochrome P450 enzymes and conjugation with glutathione to form reactive products that can bind to cellular DNA and proteins (IARC, 1999). The principal adduct appears to be *S*-[1-hydroxymethyl)-2-(*N*7-guanyl)-ethyl]glutathione, which has been detected in rat and mouse tissues following *in vivo* administration, and several studies suggest that cytochrome P450-mediated metabolism is of minor importance for organ toxicity (IARC, 1999). *In vitro* genotoxicity studies found that DBCP induced reverse mutation in *Salmonella typhimurium* TA100, in the presence of metabolic activation, forward mutation (Ara

test) in *S. typhimurium* BA13, as well as reverse mutation in *S. typhimurium* TA100 expressing human GST-A1-1 or P1-1, DNA strand breaks, sister chromatid exchanges, chromosomal aberrations and neoplastic transformation in mammalian cells. *In vivo* studies have shown that DBCP induces sex-linked recessive lethal mutations, mitotic recombinations and heritable translocations in *Drosophila melanogaster*, as well as various genotoxic effects in mammals, including DNA strand breaks in cells from various tissues (including testicular) in rats and guinea pigs, unscheduled DNA synthesis in rat spermatocytes, micronuclei in bone marrow cells of rats and mice, and micronuclei in forestomach cells and dominant lethal effects in orally-dosed rats (IARC, 1999). Rats are more sensitive than mice to the *in vivo* genotoxicity of DBCP. These observations clearly demonstrate that DBCP is a potent mutagen.

DERIVATION OF PROVISIONAL SUBCHRONIC AND CHRONIC ORAL RfD VALUES FOR DIBROMOCHLOROPROPANE

Pertinent data on effects of repeated oral exposures to DBCP are available from subchronic, chronic, reproductive and developmental toxicity studies in rats, mice and rabbits exposed by gavage, drinking water or diet. The gavage studies identified LOAELs of 9.7 mg/kg-day for reduced body weight gain in rats exposed for 64 days (Heindel et al., 1989), 15 mg/kg-day for testicular pathology and reduced male fertility in rats exposed for 77 days (Amann and Berndtson, 1986), 15 mg/kg-day for testicular and kidney pathology in rats exposed to 15 mg/kg-day for 73-78 weeks (NCI, 1978), 110 mg/kg-day for kidney pathology in mice exposed for 60 weeks (NCI, 1978), and 25 mg/kg-day for maternal toxicity (reduced body weight gain) and 50 mg/kg-day for developmental toxicity (reduced fetal body weight gain and possible embryoletality) in rats exposed for 8 days during gestation (Ruddick and Newsome, 1979). The dietary studies identified LOAELs of 2.39 mg/kg-day for decreased body weight gain in rats exposed for 104 weeks and 2.7 mg/kg-day for stomach pathology in mice exposed for 78 weeks (Hazelton Laboratories, 1977, 1978; Shell Oil Company, 1986). The drinking water studies identified LOAELs of 19.4 mg/kg-day for maternal and developmental toxicity (reduced body weight gain) in rats exposed for 60 days (Johnston et al., 1986) and 1.3 mg/kg-day for male reproductive toxicity in rabbits exposed for 10 weeks (Foote et al., 1986a, 1986b).

The 1.3 mg/kg-day drinking water LOAEL for testicular effects in rabbits (Foote et al., 1986a, 1986b) is the lowest observed adverse effect level for subchronic oral exposure to DBCP. Effects at the LOAEL and higher doses in the rabbits included dose-related reduced numbers of spermatogonia and preleptotene spermatocytes at ≥ 1.3 mg/kg-day, reduced numbers of leptotene spermatocytes at ≥ 2.7 mg/kg-day, reduced seminiferous tubule diameter and increased abnormal sperm morphology at ≥ 5.4 mg/kg-day, and testicular atrophy and reduced sperm production at 10.7 mg/kg-day. LOAELs for effects of subchronic exposure in other species are distinctly higher than in rabbits, including 9.7 mg/kg-day for reduced body weight gain and 15 mg/kg-day for reduced male fertility in rats exposed by gavage (Amann and Berndtson, 1986; Heindel et al.,

1989). The NOAEL for reproductive effects in rabbits is 0.7 mg/kg-day (Foote et al., 1986a, 1986b), and there are no NOAELs in other species below the 1.3 mg/kg-day rabbit LOAEL. The 0.7 mg/kg-day NOAEL in rabbits therefore is the most appropriate basis for subchronic p-RfD derivation. Additional support for the testes as the critical target for p-RfD derivation is provided by evidence that DBCP is a known testicular toxicant in occupationally exposed humans. Support for the rabbit as the most sensitive species for oral exposure is provided by subchronic inhalation studies that have similarly shown that the rabbit is more sensitive than rats and mice to testicular effects of DBCP (Rao et al., 1982, 1983), as well as evidence that the rabbit is generally more sensitive to testicular effects than other animal species (Pease et al., 1991). The 0.7 mg/kg-day NOAEL for testicular effects in rabbits (Foote et al., 1986a, 1986b), therefore, is the most appropriate basis for derivation of a subchronic p-RfD for DBCP.

A **subchronic p-RfD of 0.002 mg/kg-day** is derived by applying to the NOAEL of 0.7 mg/kg-day an uncertainty factor of 300 (10 for extrapolation from animals to humans, 10 to protect sensitive individuals, and 3 for database limitations, including lack of a multigeneration reproduction study), as follows:

$$\begin{aligned} \text{subchronic p-RfD} &= \text{NOAEL/ UF} \\ &= 0.7 \text{ mg/kg-day} / 300 \\ &= 0.002 \text{ mg/kg-day or } 2\text{E-}3 \text{ mg/kg-day} \end{aligned}$$

The lowest LOAELs for chronic oral exposure to DBCP are 2.39 mg/kg-day for reduced body weight gain in rats and 2.7 mg/kg-day for stomach pathology in mice in the unpublished dietary studies (Hazelton Laboratories, 1977, 1978). Non-neoplastic effects in the only other chronic oral studies of DBCP occurred at higher doses of 15 mg/kg-day in rats (testicular and kidney pathology) and 110 mg/kg-day in mice (kidney pathology) exposed by gavage (NCI, 1978). All of the chronic LOAELs are higher than the 1.3 mg/kg-day subchronic LOAEL for testicular effects (reduced spermatogenesis) in rabbits (Foote et al., 1986a, 1986b) used to derive the subchronic p-RfD, indicating that the NOAEL from the subchronic study (0.7 mg/kg-day) is also the most appropriate basis for derivation of a chronic p-RfD.

A **chronic p-RfD of 0.0002 mg/kg-day** is derived by applying to the 0.7 mg/kg-day subchronic NOAEL an uncertainty factor of 3000 (10 for extrapolating from subchronic to chronic exposure, 10 for extrapolation from animals to humans, 10 to protect sensitive individuals, and 3 for database limitations). Database limitations include the lack of a multigeneration reproduction study, as well as no longer-duration study designed to assess cumulative effects of decreases in spermatogenesis on sperm counts. The p-RfD is calculated as follows:

$$\begin{aligned}
 \text{p-RfD} &= \text{NOAEL/ UF} \\
 &= 0.7 \text{ mg/kg-day} / 3000 \\
 &= 0.0002 \text{ mg/kg-day or } 2\text{E-4 mg/kg-day}
 \end{aligned}$$

Confidence in the key study is medium. The study featured careful evaluation of a broad array of endpoints relating to male reproductive toxicity in a sensitive species, and showed a clear dose-response, including identification of both NOAEL and LOAEL values. However, group sizes were relatively small, and the relatively short exposure duration may have limited the opportunity to detect a functional effect on male fertility. Confidence in the database is medium. Subchronic, chronic, reproductive and developmental studies are available by oral exposure. However, multigeneration studies of reproductive function have not been conducted, and might be expected to be particularly sensitive to DBCP, since the male reproductive system is the most sensitive target identified. Also, the existing chronic studies were not designed to examine the sensitive reproductive targets at low doses, limiting the usefulness of these studies for risk assessment. As a result, there is a medium confidence in the subchronic and chronic p-RfDs.

DERIVATION OF PROVISIONAL SUBCHRONIC AND CHRONIC INHALATION RfC VALUES FOR DIBROMOCHLOROPROPANE

Information on the toxicity of subchronic inhalation exposures to DBCP is available from studies in rats, mice and rabbits (Rao et al., 1982, 1983; NTP, 1982; Reznik et al., 1980a, 1980b, 1980c). One of the studies in rats (Rao et al., 1983) and the only study in rabbits (Rao et al., 1982) were specifically designed to assess reproductive effects. The testes and other male reproductive tissues were a consistent and particularly sensitive target of toxicity in the rats and rabbits, although effects in several other tissues (e.g., adrenal gland, respiratory tract, liver and kidneys) occurred at similar levels of exposure in all three species. The studies identified LOAELs of 1 ppm for testicular and adrenal histopathology in rats exposed for 14 weeks (Rao et al., 1983), 1 ppm for liver, kidney and nasal cavity histopathology in rats exposed for 13 weeks (NTP, 1982; Reznik et al., 1980a, 1980b), 5 ppm for nasal cavity and bronchiolar histopathology and decreased body weight gain in mice exposed for 13 weeks (NTP, 1982; Reznik et al., 1980c), and 1 ppm for testicular histopathology in rabbits exposed for 14 weeks (Rao et al., 1982).

The lowest LOAEL of 1 ppm occurred in rats (Rao et al., 1983) and rabbits (Rao et al., 1982), and both of the subchronic studies that identified this LOAEL also identified a NOAEL of 0.1 ppm. Although exposures to 1 ppm induced adverse effects at several sites (testis, adrenal, liver, kidney and respiratory tract) in rats and rabbits, the testes is judged to be the critical target for p-RfC derivation because DBCP has also been demonstrated to be a testicular toxicant in occupationally exposed humans. There are no studies evaluating the potential for DBCP to produce effects in the respiratory tract and other non-testicular sites in humans. Testicular histopathology and related male reproductive tissue effects were induced in both rats and rabbits

at 1 ppm, but the effects were more severe in rabbits, indicating that this is the more sensitive species. Oral subchronic toxicity studies have similarly shown that the rabbit is more sensitive than rats to testicular effects of DBCP (Foote et al., 1986a, 1986b), and it is known that the rabbit is generally more sensitive to testicular effects than other species (Pease et al., 1991). The 0.1 ppm NOAEL for testicular effects in rabbits (Rao et al., 1982), therefore, is the most appropriate basis for derivation of a subchronic p-RfC for DBCP.

To calculate the p-RfC, the 0.1 ppm (0.94 mg/m³) NOAEL for testicular effects in rabbits is first duration-adjusted for intermittent exposure (6 hours/day, 5 days/week), as follows (U.S. EPA, 1994b):

$$\begin{aligned} \text{NOAEL}_{\text{ADJ}} &= (\text{NOAEL}_{\text{RABBIT}}) (\text{hours}/24 \text{ hours}) (\text{days}/7 \text{ days}) \\ &= (0.94 \text{ mg}/\text{m}^3) (6/24) (5/7) \\ &= 0.17 \text{ mg}/\text{m}^3 \end{aligned}$$

DBCP is treated as a category 3 gas for purposes of deriving a p-RfC based on extrarrespiratory effects. The human equivalent concentration (HEC) for extrarrespiratory effects produced by a category 3 gas is calculated by multiplying the duration-adjusted NOAEL by the ratio of blood:gas partition coefficients ($H_{b/g}$) in animals and humans (U.S. EPA, 1994b). A blood:gas partition coefficient is not available for DBCP in humans or in rabbits, so a unity value is assumed for the $(H_{b/g})_A/(H_{b/g})_H$ ratio (U.S. EPA, 1994b), yielding a $\text{NOAEL}_{\text{HEC}}$ equal to the $\text{NOAEL}_{\text{ADJ}}$ of 0.17 mg/m³.

A **subchronic p-RfC of 0.002 mg/m³** is derived by applying to the $\text{NOAEL}_{\text{HEC}}$ an uncertainty factor of 100 (3 for extrapolation from animals to humans using the dosimetric adjustments, 10 to protect sensitive individuals, and 3 for database limitations, including lack of a multigeneration reproductive study and inhalation developmental toxicity studies), as follows:

$$\begin{aligned} \text{subchronic p-RfC} &= \text{NOAEL}_{\text{HEC}} / \text{UF} \\ &= 0.17 \text{ mg}/\text{m}^3 / 100 \\ &= 0.002 \text{ mg}/\text{m}^3 \text{ or } 2\text{E-}3 \text{ mg}/\text{m}^3 \end{aligned}$$

Confidence in the critical study is medium. The study featured careful evaluation of a broad array of endpoints relating to male reproductive toxicity in a sensitive species, and showed a clear dose-response, including identification of both NOAEL and LOAEL values. However, the respiratory tract, which is also a sensitive target for DBCP by inhalation exposure, was not examined for histopathology. Confidence in the database is medium. Subchronic, chronic, and reproductive inhalation studies are available, but no multigeneration reproduction studies (expected to be a sensitive test for DBCP) and no developmental toxicity studies. Also, there is uncertainty about the occurrence of respiratory tract effects relative to testicular effects. As a result, there is a medium confidence in the subchronic p-RfC.

A chronic RfC of 0.0002 mg/m³ (2E-4 mg/m³) was similarly derived by U.S. EPA (2005) by using the subchronic rabbit study (Rao et al., 1982), critical endpoint (testicular effects) and NOAEL_{HEC} (0.17 mg/m³) on which the subchronic p-RfC is based, and applying an uncertainty factor of 1000 (10 for use of a subchronic study, 3 for extrapolation from animals to humans using the dosimetric adjustments, 10 to protect sensitive individuals, and 3 for database limitations). Confidence in the critical study, database, and p-RfC were rated as medium.

DERIVATION OF A PROVISIONAL CARCINOGENICITY ASSESSMENT FOR DIBROMOCHLOROPROPANE

Weight-of Evidence Classification

The carcinogenicity of DBCP in humans has been assessed in several cohort mortality and case-control studies. Excesses of lung, liver, biliary tract and/or cervical cancers were observed in occupational cohort mortality studies (Amoateng-Adjepong et al., 1995; Brown, 1992; IARC, 1987; Olsen et al., 1995; Wesseling et al., 1996), but the findings cannot be clearly attributed to DBCP, due to small numbers of cases and/or exposures to other chemicals (IARC, 1999). Case-control studies of the general population found no significant associations between gastric cancer or leukemia and DBCP in drinking water (Wong et al., 1989).

The carcinogenicity of DBCP in animals has been tested by oral and inhalation exposure in rats and mice. Chronic exposure by gavage induced squamous cell carcinomas of the forestomach in rats and mice, as well as adenocarcinomas of the mammary gland in rats (NCI, 1978). Chronic exposure in the diet induced squamous cell carcinomas and papillomas in the stomach of rats and mice, as well as tumors in the kidneys (renal tubular adenoma and carcinoma) and liver (hepatocellular carcinoma and neoplastic nodules) of rats (Hazelton Laboratories, 1977, 1978; Shell Oil Company, 1986). Chronic inhalation exposure induced tumors in the nasal cavity in rats and mice, other parts of the respiratory tract in rats (tongue and pharynx) and mice (lungs), and adrenal cortex of rats (NTP, 1982).

DBCP is metabolized via oxidation by cytochrome P450 enzymes and conjugation with glutathione to form reactive products that can bind to cellular DNA and proteins (IARC, 1999). *In vitro* genotoxicity studies found that DBCP induced mutations in bacteria in the presence of metabolic activation, as well as mutations, DNA strand breaks, sister chromatid exchanges, chromosomal aberrations and neoplastic transformation in mammalian cells (IARC, 1999). The metabolites of DBCP induce reverse and forward mutations in bacterial assays suggesting that DBCP is a proximate carcinogen. *In vivo* genotoxicity studies showed that DBCP induced sex-linked recessive lethal mutations and other effects in *Drosophila*, as well as various effects in mammals, including DNA strand breaks in testicular and other tissues, unscheduled DNA synthesis in rat spermatocytes, micronuclei in bone marrow cells of rats and mice and

forestomach cells of rats, and dominant lethal effects in orally-dosed rats (IARC, 1999). EPA has concluded, by a weight of evidence evaluation, that DBCP is carcinogenic by a mutagenic mode of action.

Under the U.S. EPA (2005) cancer guidelines, DBCP is considered likely to be carcinogenic to humans.

Quantitative Estimates of Carcinogenic Risk

Both oral and inhalation tumor incidence data in rats and mice can be used to assess cancer risks for DBCP. For oral exposure, the diet studies (Hazelton Laboratories, 1977, 1978) are more appropriate than the gavage study (NCI, 1978) for dose-response modeling, because diet is the more relevant route for human exposure, there was high tumor-related early mortality at all dose levels in the gavage study, stomach tumors were induced at much lower doses and with better dose-response in the diet study, and dietary exposure induced tumors systemically as well as locally in the stomach. The rat diet study (Hazelton Laboratories, 1977) is more suitable than the mouse diet study (Hazelton Laboratories, 1978), because three treatment groups were evaluated in the rats compared to only one (high-dose) in the mice. Therefore, for oral exposure, the most appropriate basis for cancer dose-response assessment is tumor incidence data from the diet study in rats (Hazelton Laboratories, 1977). For inhalation exposure, in the only cancer study by this route (NTP, 1982), data in both rats and mice are suitable for analysis.

The derivations of quantitative estimates of cancer risks from oral and inhalation exposure used the methodology in the U.S. EPA (2005) guidelines for carcinogen risk assessment. The mode of action (MOA) evidence of DBCP is analyzed under the carcinogenic MOA framework in EPA's *Guidelines for Carcinogen Risk Assessment* (U.S. EPA, 2005, Section 2.4.3). The hypothesis that DBCP carcinogenicity has a mutagenic MOA is sufficiently supported in animals. DBCP has been found to be carcinogenic in animals (Hazelton Laboratories, 1977, 1978; Shell Oil Company, 1986; NTP, 1982) and short term mutagenicity testing indicates its mutagenic MOA (IARC, 1999). Occupational exposure to DBCP has been found to be associated with increased mortality from lung, liver, biliary tract and/or cervical cancers, but the findings cannot be clearly attributed to DBCP due to small number of cases and/or exposures to other chemicals (IARC, 1999). Under the U.S. EPA (2005) cancer guidelines, DBCP is considered likely to be carcinogenic to humans. In accordance with the 2005 cancer guidelines (U.S. EPA, 2005), the BMDL₁₀ or BMCL₁₀ (lower bound on dose or concentration estimated to produce a 10% increase in tumor incidence over background) was estimated using the U.S. EPA (1996, 2000) benchmark dose methodology, and a linear extrapolation to the origin was performed by dividing the BMDL₁₀ into 0.1 (10%) (U.S. EPA, 1999, 2000) (Tables 13 and 14) (Figures 1 and 2). For oral exposure, the unadjusted values based directly on the oral animal tumor data are adjusted to human values by correcting for

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Table 13. Risk Values Based on Stomach, Kidney and Liver Tumor Incidences in Rats Exposed to DBCP in Diet (Hazelton Laboratories, 1977)

Tumor Type, Species and Sex	Dose ^a (mg/kg-day) and Incidence				Rat BMD ₁₀ ^b (mg/kg-day)	Rat BMDL ₁₀ ^b (mg/kg-day)	Rat 0.1/BMDL ₁₀ (mg/kg-day) ⁻¹	OSF Human 0.1/BMDL ₁₀ ^c (mg/kg-day) ⁻¹
	0	0.24	0.80	2.39				
Stomach carcinoma or papilloma, Male Rat	0/48	0/46	3/46	21/41	0.93	0.73	0.14	0.52
Stomach carcinoma or papilloma, Female Rat	0/48	0/45	0/47	10/43	1.46	0.91	0.11	0.46
Renal adenoma or carcinoma, Male Rat	0/48	1/46	4/46	15/41	0.66	0.46	0.22	0.81
Renal adenoma or carcinoma, Female Rat	0/48	0/45	0/47	10/43	1.46	0.91	0.11	0.46
Hepatocellular carcinoma, Male Rat	0/48	1/46	2/46	5/41	1.83	1.08	0.09	0.33

^a Doses are reported rat average daily doses (Hazelton Laboratories, 1977; Shell Oil Company, 1986).

^b Rat BMD₁₀ and BMDL₁₀ values were calculated (extra risk) from the lowest-degree polynomial model that gave an adequate fit (chi-square goodness-of-fit statistic p value >0.05), as per the U.S. EPA (1996b) Benchmark Dose Technical Guidance Document. Models with more than 2 parameters were not considered for selection (degrees of freedom = # dose groups - 2 = 4 - 2 = 2). Models selected were: Stomach tumor incidence: males, 2-degree model; females, 1-degree model. Kidney tumor incidence: males, 1-degree model; females, 1-degree model. Liver tumor incidence: males, 1-degree model.

^c Human values were calculated as: rat value (0.1/BMDL₁₀) multiplied by $(W_H / W_R)^{1/4}$, where W_H = 70 kg (human reference body weight) and W_R = 0.38 kg (male) or 0.229 kg (female) based on U.S. EPA (1988c) rat reference body weights.

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Table 14. Risk Values Based on Nasal, Lung and Adrenal Tumor Incidences in Rats and Mice Exposed to DBCP by Inhalation (NTP, 1982)

Tumor Type, Species and Sex	HEC ^a (mg/m ³) and Incidence			Human BMC ₁₀ ^b (mg/m ³)	Human BMCL ₁₀ ^b (mg/m ³)	IUR Human 0.1/BMCL ₁₀ (mg/m ³) ⁻¹
	0 mg/m ³	0.23 mg/m ³	1.13 mg/m ³			
Nasal Cavity, Male Rats	0/50	32/50	39/49	0.040	0.018	5.6 ^c
Nasal Cavity, Female Rats	1/50	21/50	42/50	0.043	0.034	2.9
Nasal Cavity, Male Mice	0/45	1/42	21/48	0.23	0.16	0.63
Nasal Cavity, Female Mice	0/50	11/50	38/50	0.069	0.055	1.8
Lung, Male Mice	0/41	3/40	11/45	6.43	4.3	0.023
Lung, Female Mice	4/50	12/50	18/50	4.27	2.6	0.038
Adrenal Cortex, Female Rats	0/50	7/50	5/48	0.81	0.41	0.24 ^c

^a Concentrations are human equivalent concentrations (HECs). The HECs for the nasal cavity tumors (extrathoracic region effect) and lung tumors (pulmonary region effect) were calculated using estimated average body weights from the NTP (1982) study (male rats 0.325 kg, female rats 0.225 kg, male mice 0.035 kg, female mice 0.030 kg) and U.S. EPA (1994b) algorithms by treating DBCP as a category 1 (reactive) gas for these portal of entry effects. The HECs for the adrenal tumors (extrarespiratory effect) were calculated by treating DBCP as a category 3 gas that accumulates in the blood and multiplying the duration-adjusted animal exposure level by the ratio of blood:gas partition coefficients in animals and humans (U.S. EPA, 1994b); the ratio was assumed to be unity because a blood:gas partition coefficient is not available for DBCP in rats or humans.

^b Human BMC₁₀ and BMCL₁₀ values were calculated (extra risk) from the 1-degree polynomial models that gave an adequate fit (chi-square goodness-of-fit statistic p value >0.05). Higher degree polynomial models were not considered for selection (degrees of freedom = # dose groups - 2 = 3 - 2 = 1).

^c To obtain an adequate fit to the data for male rat nasal tumors and female mouse adrenal tumors, the high dose groups were dropped, leaving only controls and one dose group for each of these endpoints.

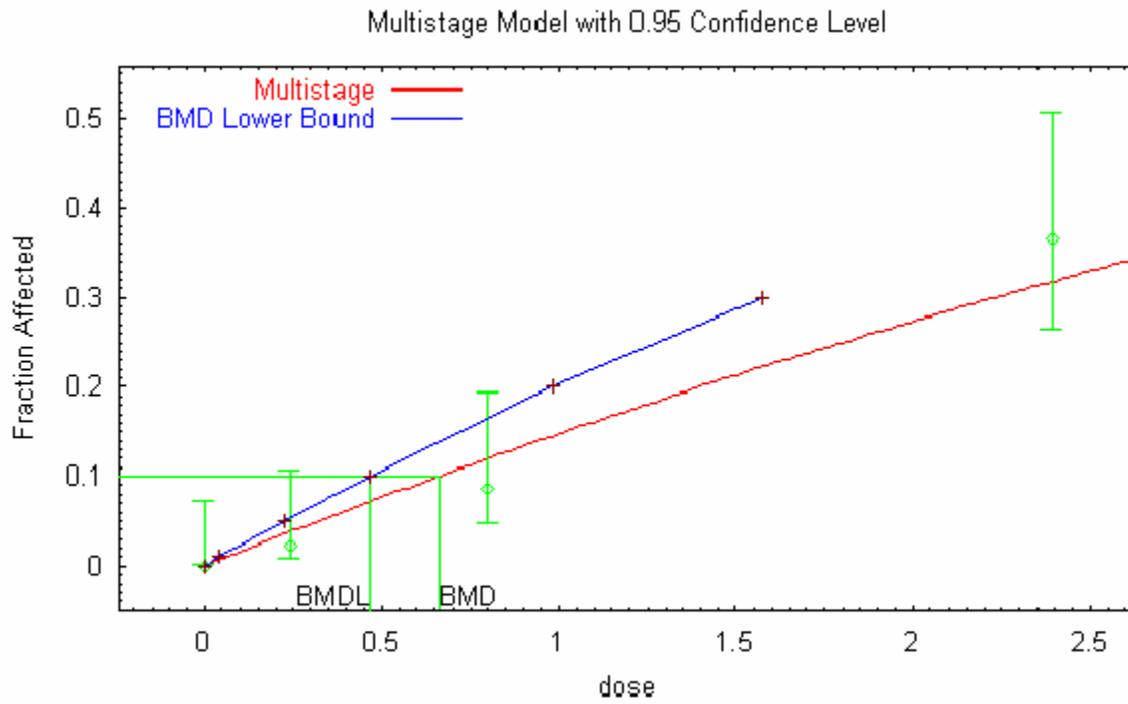


Figure 1. DBCP - Oral Renal Tumors in Male Rats
Dose units are mg/kg-day

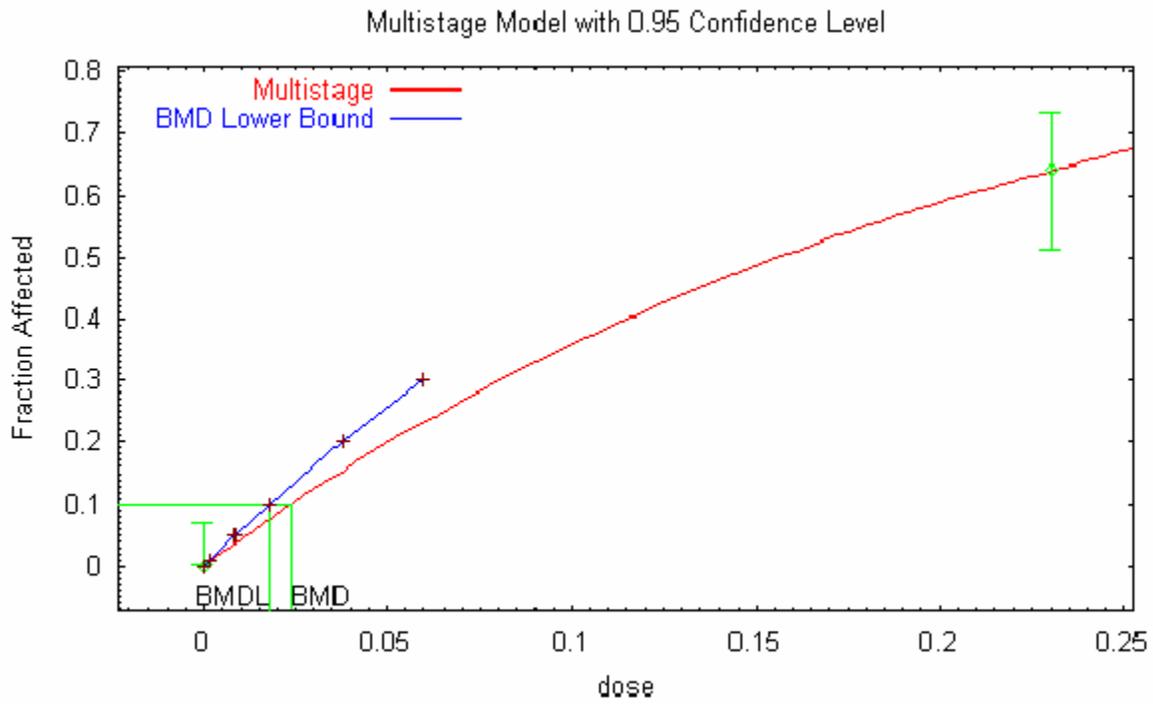


Figure 2. DBCP - Inhalational Nasal tumors in Male Rats
Dose units are mg/kg-day

differences in body weight between humans and rodents. U.S. EPA uses a cross-species scaling factor of body weight raised to the 3/4 power (U.S. EPA, 2005). Adjustment from animal to human slope factor is performed by multiplying the animal value by the ratio of human to animal body weight raised to the 1/4 power. For inhalation exposure, animal to human adjustment is accomplished in the calculation of the HECs (U.S. EPA, 1994b) prior to modeling. No adjustment was used for shorter-than-lifetime observation periods in the NTP (1982) lifetime inhalation bioassays. Although the high-dose groups were terminated after only 84 weeks in the rats and 76 weeks in the mice (compared to reference lifespans of 104 weeks in both species), this was due to early mortality associated with tumor formation. Because the short duration of observation was imposed by the development of tumors, a sufficient period of time had elapsed to evaluate the carcinogenicity of DBCP.

For oral exposure, dose-response modeling was performed using data from the rat diet study (Hazelton Laboratories, 1977) for stomach tumors (squamous cell carcinoma or papilloma) in both sexes, kidney tumors (renal tubular cell adenoma or carcinoma) in both sexes, and hepatocellular carcinoma in males (Table 13). Hepatocellular carcinoma was not modeled in the females because incidences were not significantly increased compared to controls (Table 10), and combined incidences of hepatocellular carcinoma and neoplastic nodules (Table 10) was not modeled in either sex because it is not known if the neoplastic nodule classification included pre-neoplastic lesions. The modeling results are shown in Table 13. The highest estimate of human oral slope factor was based on the combined incidence of renal tubular cell adenomas and carcinomas in male rats, **human slope factor, 0.1/BMDL₁₀ = 0.8 mg/kg-day**, rounded from 0.81.

EPA has concluded, by a weight of evidence evaluation, that DBCP is carcinogenic by a mutagenic mode of action. According to the *Supplemental Guidance for Assessing Susceptibility from Early-Life Exposure to Carcinogens (Supplemental Guidance)* (U.S. EPA, 2005) those exposed to carcinogens with a mutagenic mode of action are assumed to have increased early-life susceptibility. Data for DBCP are not sufficient to develop separate risk estimates for childhood exposure. The **oral slope factor of 8×10^{-1}** per mg/kg-day, calculated from data from adult exposure, does not reflect presumed early-life susceptibility for this chemical and age dependent adjustment factors (ADAFs) should be applied to this slope factor when assessing cancer risks. Example evaluations of cancer risks based on age at exposure are given in Section 6 of the Supplemental Guidance.

Risk Assessment Considerations: The Supplemental Guidance establishes ADAFs for three specific age groups. The current ADAFs and their age groupings are 10 for <2 years, 3 for 2 to <16 years, and 1 for 16 years and above (U.S. EPA, 2005). The 10-fold and 3-fold adjustments in slope factor are to be combined with age specific exposure estimates when estimating cancer risks from early life (<16 years age) exposure to DBCP. These ADAFs and their age groups were derived from the 2005 Supplemental Guidance, and they may be revised

over time. The most current information on the application of ADAFs for cancer risk assessment can be found at www.epa.gov/cancerguidelines/. In estimating risk, EPA recommends using age-specific values for both exposure and cancer potency; for DBCP, age-specific values for cancer potency are calculated using the appropriate ADAFs. A cancer risk is derived for each age group, and these are summed across age groups to obtain the total risk for the exposure period of interest (see Section 6 of the Supplemental Guidance).

The oral slope factor, calculated from adult exposure, is derived from the $BMDL_{10}$, the 95% lower bound on the exposure associated with an 10% extra cancer risk, by dividing the risk (as a fraction) by the $BMDL_{10}$ 0.1 mg/kg-day, and represents an upper bound risk estimate for continuous lifetime exposure without consideration of increased early-life susceptibility due to DBCP's mutagenic mode of action:

The slope factor for DBCP should not be used with exposures exceeding the point of departure ($BMDL_{10}$), because above this level the fitted dose-response model better characterizes what is known about the carcinogenicity of DBCP. For exposures greater than the $BMDL_{10}$, contact the Superfund Technical Support Center. Additionally, age dependent adjustment factors (ADAFs) should be applied to this slope factor when assessing cancer risks to individuals <16 years old as discussed above (U.S. EPA, 2005).

The slope of the linear extrapolation from the central estimate, human BMD_{10} , is $0.1/BMD_{10}$ (human) $(\text{mg/kg-day})^{-1} = (0.6 \text{ mg/kg-day})^{-1}$. The BMD_{10} for humans was calculated from the BMD_{10} for rats (Table 13) according to the same procedure for conversion of the $BMDL_{10}$ for rats to humans.

For inhalation exposure, dose-response modeling was performed using data from the inhalation study (NTP, 1982) for nasal cavity tumors in rats and mice of both sexes, lung tumors in mice of both sexes, and adrenal cortical tumors in female mice (Table 14). Tumors in the rat tongue and pharynx (Table 11) were not modeled because these sites were less sensitive than the others (fewer tumors and only increased at the high exposure level). Mammary gland tumors in the female rats (Table 11) were not modeled because incidences were significantly increased only in the low dose group (no evidence of dose response). The modeling results are shown in Table 14. The highest estimate of human inhalation unit risk was based on the combined incidence of various types of nasal cavity tumors in male rats, **human slope factor, $0.1/BMCL_{10} = 6 \times 10^0$ per mg/m^3** , rounded from 5.6.

EPA has concluded, by a weight of evidence evaluation, that DBCP is carcinogenic by a mutagenic mode of action. According to the Supplemental Guidance for Assessing Susceptibility from Early-Life Exposure to Carcinogens (Supplemental Guidance) (U.S. EPA, 2005) those exposed to carcinogens with a mutagenic mode of action are assumed to have increased early-life susceptibility. Data for DBCP are not sufficient to develop separate risk

estimates for childhood exposure. The **inhalation unit risk of 6×10^0 per mg/m^3** , calculated from data from adult exposure, does not reflect presumed early-life susceptibility for this chemical and age dependent adjustment factors (ADAFs) should be applied to this unit risk when assessing cancer risks. Example evaluations of cancer risks based on age at exposure are given in Section 6 of the Supplemental Guidance.

Risk Assessment Considerations: The Supplemental Guidance establishes ADAFs for three specific age groups. The current ADAFs and their age groupings are 10 for <2 years, 3 for 2 to <16 years, and 1 for 16 years and above (U.S. EPA, 2005). The 10 fold and 3 fold adjustments in unit risk are to be combined with age specific exposure estimates when estimating cancer risks from early life (<16 years age) exposure to DBCP. These ADAFs and their age groups were derived from the Supplemental Guidance, and they may be revised over time. The most current information on the application of ADAFs for cancer risk assessment can be found at www.epa.gov/cancerguidelines/. In estimating risk, EPA recommends using age-specific values for both exposure and cancer potency; for DBCP, age-specific values for cancer potency are calculated using the appropriate ADAFs. A cancer risk is derived for each age group, and these are summed across age groups to obtain the total risk for the exposure period of interest (see Section 6 of the Supplemental Guidance).

The inhalation unit risk, calculated from adult exposure, is derived from the BMCL_{10} , the 95% lower bound on the exposure associated with a 10% extra cancer risk, by dividing the risk (as a fraction) by the BMCL_{10} , and represents an upper bound risk estimate for continuous lifetime exposure without consideration of increased early-life susceptibility due to DBCP's mutagenic mode of action:

The unit risk for DBCP should not be used with exposures exceeding the point of departure (BMCL_{10}) $0.02 \text{ mg}/\text{m}^3$, because above this level the fitted dose-response model better characterizes what is known about the carcinogenicity of DBCP. For exposures greater than the BMCL_{10} , contact the Superfund Technical Support Center. Additionally, age dependent adjustment factors (ADAFs) should be applied to this slope factor when assessing cancer risks to individuals <16 years old as discussed above (U.S. EPA, 2005).

The slope factor of the linear extrapolation from the central estimate, human BMC_{10} , is $0.1/\text{BMC}_{10}$ (humans) $(\text{mg}/\text{m}^3)^{-1} = 2.5 (\text{mg}/\text{m}^3)^{-1}$. No correction is made for differences in body weight for inhalation exposure.

REFERENCES

- ACGIH (American Conference of Governmental Industrial Hygienists). 2002. 2002 Threshold Limit Values for Chemical Substances and Physical Agents and Biological Exposure Indices. ACGIH, Cincinnati, OH.
- Amann, R.P. and W.E. Berndtson. 1986. Assessment of procedures for screening agents for effects on male reproduction: Effects of dibromochloropropane (DBCP) on the rat. *Fund. Appl. Toxicol.* 7: 244-255.
- Amoateng-Adjepong, Y., N. Sathiakumar, E. Delzell and P. Cole. 1995. Mortality among workers at a pesticide manufacturing plant. *J. Occup. Med.* 37: 471-178.
- ATSDR (Agency for Toxic Substances and Disease Registry). 1992. Toxicological Profile for 1,2-Dibromo-3-chloropropane. U.S. Department of Health and Human Services, Public Health Service. Atlanta, GA. TP-91/12.
- Au, W.W., G. Cantelli-Forti, P. Hrelia and M.S. Legator. 1990. Cytogenetic assays in genotoxic studies: somatic cell effects of benzene and germinal cell effects of dibromochloropropane. *Teratog. Carcinog. Mutag.* 10: 125-134.
- Brown, D.P. 1992. Mortality of workers employed at organochlorine pesticide manufacturing plants—an update. *Scand. J. Work Environ. Health.* 18: 155-161.
- Foote, R.H., E.C. Schermerhorn and M.E. Simkin. 1986a. Measurement of semen quality, fertility, and reproductive hormones to assess dibromochloropropane effects in live rabbits. *Fund. Appl. Toxicol.* 6: 628-637.
- Foote, R.H., W.E. Berndtson and T.R. Rounsaville. 1986b. Use of quantitative testicular histology to assess the effect of dibromochloropropane on reproduction in rabbits. *Fund. Appl. Toxicol.* 6: 638-647.
- Ghanayem, B.I., R.R. Maronpot and H.B. Matthews. 1986. Association of chemically induced forestomach cell proliferation and carcinogenesis. *Cancer Lett.* 32: 271-278.
- Goldsmith, J.R. 1997. Dibromochloropropane: Epidemiological findings and current questions. *Ann. NY Acad. Sci.* 837: 300-306.
- Hazelton Laboratories. 1977. 104-Week dietary study in rats, 1,2-dibromo-3-chloropropane (DBCP). Final Report. Unpublished report submitted to Dow Chemical Co., Midland, MI. October 29, 1977. (Cited in U.S. EPA, 1979, 1988a, 1989a)

Hazelton Laboratories. 1978. 78-Week toxicity and carcinogenicity study in mice. Final Report. Project No. 174-125. Unpublished report submitted to Dow Chemical Co., November 3, 1978 (Cited in U.S. EPA, 1979, 1988a, 1989a)

Heindel, J.J., A.S. Berkowitz, G. Kyle et al. 1989. Assessment in rats of the gonadotoxic and hepatorenal toxic potential of dibromochloropropane (DBCP) in drinking water. *Fundam. Appl. Toxicol.* 13: 804-815.

IARC (International Agency for Research on Cancer). 1987. 1,2-Dibromo-3-chloropropane. Overall Evaluations of Carcinogenicity: An updating of IARC Monographs Volumes 1 to 42. IARC Monographs on the Evaluation of Carcinogenic Risks to Humans, Lyon, France. Supplement 7: 191-192.

IARC (International Agency for Research on Cancer). 1999. 1,2-Dibromo-3-chloropropane. In IARC Monographs on the Evaluation of Carcinogenic Risks to Humans. Re-evaluation of some organic chemicals, hydrazine and hydrogen peroxide, Lyon, France. Volume 71, Part Two: 479-500.

Johnston, R.V., D.C. Mensik, H.W. Taylor et al. 1986. Single-generation drinking water reproduction study of 1,2-dibromo-3-chloropropane in Sprague-Dawley rats. *Bull. Environ. Contam. Toxicol.* 37: 531-537.

Lamb, J.C., R. Tyl and A. Davis Lawton. 1997. Dibromochloropropane. Summary of NTP Reproductive Assessment by Continuous Breeding Study. *Environ. Health Persp.* 105: 299-300.

NCI (National Cancer Institute). 1978. Bioassay of dibromochloropropane for possible carcinogenicity. NCI Carcinogenesis Testing Program Technical Report No. 28. NTIS PB277472.

NIOSH (National Institute for Occupational Safety and Health). 2002. Online NIOSH Pocket Guide to Chemical Hazards. Index by CASRN. Online. <http://www.cdc.gov/niosh/npg/npgdcas.html>

NTP (National Toxicology Program). 1982. Carcinogenesis bioassay of 1,2-dibromo-3-chloropropane (CAS No. 96-12-8) in F344 rats and B6C3F1 mice (inhalation study). *Tech. Rep. Ser. No. 206. Pub. No. 82-1762.* p. 188.

Olsen, G.W., K.M. Bodner, B.A. Stafford et al. 1995. Update of the mortality experience of employees with occupational exposure to 1,2-dibromo-3-chloropropane (DBCP). *Am. J. Ind. Med.* 28: 399-410.

OSHA (Occupational Safety and Health Administration). 2002. OSHA Standard 1910.1000 Table Z-1. Part Z, Toxic and Hazardous Substances. Online. http://www.osha-slc.gov/OshStd_data/1910_1000_TABLE_Z_1.html

Pease, W., J. Vandenburg and K. Hooper. 1991. Comparing alternative approaches to establishing regulatory levels for reproductive toxicants: DBCP as a case study. *Environ. Health Perspect.* 91: 141-155.

Potashnik, G. and A. Porath. 1995. Dibromochloropropane (DBCP): A 17-year reassessment of testicular function and reproductive performance. *J. Am. Coll. Occup. Environ. Med.* 37: 1287-1292.

Rao, K.S., J.D. Burek, F. Murray et al. 1982. Toxicologic and reproductive effects of inhaled 1,2-dibromo-3-chloropropane in male rabbits. *Fund. Appl. Toxicol.* 2(5): 241-251.

Rao, K.S., J. Burek, F. Murray et al. 1983. Toxicologic and reproductive effects of inhaled 1,2-dibromo-3-chloropropane in rats. *Fund. Appl. Toxicol.* 3(2): 104-110.

Reel, J.R., T. Wolkowski-Tyl and A.D. Lawton. 1984. Dibromochloropropane: reproduction and fertility assessment in CD-1 mice when administered by gavage. National Toxicology Program, Research Triangle Park, NC. NTIS PB-85-118644. NTP 84-263.

Reznik, G., H. Reznik-Schuller, J.M. Ward and S.F. Stinson. 1980a. Morphology of nasal-cavity tumours in rats after chronic inhalation of 1,2-dibromo-3-chloropropane. *Br. J. Cancer.* 42(5): 772-781.

Reznik, G., S. Stinson and J. Ward. 1980b. Respiratory pathology in rats and mice after inhalation of 1,2-dibromo-3-chloropropane or 1,2-dibromoethane for 13 weeks. *Arch. Toxicol.* 46(3-4): 233-240.

Reznik, G., S. Stinson and J. Ward. 1980c. Lung tumors induced by chronic inhalation of 1,2-dibromo-3-chloropropane in B6C3F1 mice. *Cancer Lett.* 10(4): 339-342.

Ruddick, J.A. and W.H. Newsome. 1979. A teratogenicity and tissue distribution study on dibromochloropropane in the rat. *Bull. Environ. Contam. Toxicol.* 21: 483-487.

Saito-Suzuki, R., S. Teramoto and Y. Shirasu. 1982. Dominant-lethal studies in rats with 1,2-dibromo-3-chloropropane and its structurally related compounds. *Mutat. Res.* 101(4): 321-328.

Shell Oil Company. 1986. Risk Assessment of Dibromochloropropane. NTIS Document No. AD-A271-278.

Teramoto, S., R. Saito, H. Aoyama and Y. Shirasu. 1980. Dominant lethal mutation induced in male rats by 1,2-dibromo-3-chloropropane (DBCP). *Mutat. Res.* 77: 71-78.

U.S. EPA. 1979. Dibromochloropropane (DBCP) suspension order and notice of intent to cancer. *Fed. Reg.* 44(219): 65135-65139.

U.S. EPA. 1988a. Drinking Water Criteria Document for Dibromochloropropane (DBCP). Prepared by the Office of Health and Environmental Assessment, Environmental Criteria and Assessment Office, Cincinnati, OH for the Office of Drinking Water, Washington, DC. EPA 600/X-84/209-2.

U.S. EPA. 1988b. Assessment of the carcinogenic potential of 1,2-dibromo-3-chloropropane. Prepared by the Carcinogen Assessment Group. Available through RCRA/Superfund docket (Cited in U.S. EPA, 1997)

U.S. EPA. 1988c. Recommendations for and Documentation of Biological Values for Use in Risk Assessment. Environmental Criteria and Assessment Office, Office of Health and Environmental Assessment, Office of Research and Development, Cincinnati, OH. PB88-17874. EPA/600/6-87/008.

U.S. EPA. 1989a. Health Effects Assessment for Dibromochloropropane. Prepared by the Office of Health and Environmental Assessment, Environmental Criteria and Assessment Office, Cincinnati, OH for the Office of Solid Waste and Emergency Response, Washington, DC.

U.S. EPA. 1989b. Carcinogen Risk Assessment Verification Endeavor (CRAVE). (Cited in U.S. EPA, 1997)

U.S. EPA. 1991. Chemical Assessments and Related Activities (CARA). Office of Health and Environmental Assessment, Washington, DC. April.

U.S. EPA. 1993. Crave Work Group meeting notes of 6/2-3/92. Summary of disposition for 1,2-dibromo-3-chloropropane. Memo preparation date July 27, 1993. Office of Research and Development, Environmental Criteria and Assessment Office, Washington, DC.

U.S. EPA. 1994a. Chemical Assessments and Related Activities (CARA). Office of Health and Environmental Assessment, Washington, DC. December.

U.S. EPA. 1994b. Methods for Derivation of Inhalation Reference Concentrations and Application of Inhalation Dosimetry, Office of Research and Development, Office of Health and Environmental Assessment, Environmental Criteria and Assessment Office, Washington, DC. EPA/600/8-90/066F.

U.S. EPA. 1996. Benchmark Dose Technical Guidance Document. Risk Assessment Forum, National Center for Environmental Assessment, Office of Research and Development, Washington, DC. EPA/600/P-96/002A.

U.S. EPA. 1997. Health Effects Assessment Summary Tables (HEAST). FY-1997 Update. Prepared by the Office of Research and Development, National Center for Environmental Assessment, Cincinnati, OH for the Office of Emergency and Remedial Response, Washington, DC. July. EPA-540-R-97-036. NTIS PB97-921199.

U.S. EPA. 2000. Benchmark Dose Technical Guidance Document. Risk Assessment Forum, National Center for Environmental Assessment, Office of Research and Development, Washington, DC. EPA/630/R-00/001.

U.S. EPA. 2002. Drinking Water Standards and Health Advisories. Summer 2002. Office of Water, Washington, DC. Online. <http://www.epa.gov/ost/drinking/standards/>

U.S. EPA. 2005. Guidelines for Carcinogen Risk Assessment. Office of Research and Development, National Center for Environmental Assessment, Washington, DC. EPA/630/P-03/001F.

U.S. EPA. 2006. Integrated Risk Information System (IRIS). Office of Research and Development, National Center for Environmental Assessment, Washington, DC. Online. <http://www.epa.gov/iris/>

Wesseling, C., A. Ahlbom, D. Antich et al. 1996. Cancer in banana plantation workers in Costa Rica. *Int. J. Epidemiol.* 25: 1125-1131.

WHO (World Health Organization). 2002. Online catalogs for the Environmental Health Criteria Series. Online. <http://www.who.int/dsa/cat98/chemtox8.htm#>

Wong, O., R.W. Morgan, M.D. Whorton et al. 1989. Ecological analyses and case-control studies of gastric cancer and leukaemia in relation to DBCP in drinking water in Fresno County, California. *Br. J. Ind. Med.* 46: 521-528.