EPA United States
Environmental Protection
Agency

Provisional Peer-Reviewed Toxicity Values for

Commercial or Practical Grade Hexane

Superfund Health Risk Technical Support Center
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U.S. Environmental Protection Agency
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COMMONLY USED ABBREVIATIONS

BMD	Benchmark Dose
IRIS	Integrated Risk Information System
IUR	inhalation unit risk
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
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
RfC	inhalation reference concentration
RfD	oral reference dose
UF	uncertainty factor
UF$_A$	animal to human uncertainty factor
UF$_C$	composite uncertainty factor
UF$_D$	incomplete to complete database uncertainty factor
UF$_H$	interhuman uncertainty factor
UF$_L$	LOAEL to NOAEL uncertainty factor
UF$_S$	subchronic to chronic uncertainty factor

**PROVISIONAL PEER-REVIEWED TOXICITY VALUES FOR
COMMERCIAL OR PRACTICAL GRADE HEXANE**

Background

On December 5, 2003, the U.S. Environmental Protection Agency's (U.S. EPA) 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) U.S. EPA's Integrated Risk Information System (IRIS).
2) Provisional Peer-Reviewed Toxicity Values (PPRTVs) used in U.S. 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 U.S. EPA's 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 U.S. EPA IRIS Program. All provisional toxicity values receive internal review by two U.S. EPA scientists and external peer review by three independently selected scientific experts. PPRTVs differ from IRIS values in that PPRTVs do not receive the multiprogram consensus review provided for IRIS values. This is because IRIS values are generally intended to be used in all U.S. EPA programs, while PPRTVs are developed specifically for the Superfund Program.

Because new information becomes available and scientific methods improve over time, PPRTVs are reviewed on a 5-year basis and updated into the active database. 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 documents 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 Resource Conservation and Recovery Act (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 document and understand the strengths

and limitations of the derived provisional values. PPRTVs are developed by the U.S. EPA Office of Research and Development's National Center for Environmental Assessment, Superfund Health Risk Technical Support Center for OSRTI. Other U.S. 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 U.S. EPA Office of Research and Development's National Center for Environmental Assessment, Superfund Health Risk Technical Support Center (513-569-7300), or OSRTI.

INTRODUCTION

Commercial hexane is a mixture of aliphatic hydrocarbons used as a solvent for adhesives or to clean machinery (U.S. EPA, 2005a). Although the precise amount of each constituent varies, slightly more than half (about 52%) of commercial hexane consists of *n*-hexane. The remaining portion is a mixture of isomers and structurally related chemicals, such as 3-methylpentane (16%), methylcyclopentane (16%), and 2-methylpentane (13%), as well as some minor components such as cyclohexane and 2,4-dimethylpentane (U.S. EPA 2005a). In order to ensure the comparability of the data included in this review, only studies of hexane mixtures with similar composition were reviewed. Studies of mixtures with *n*-hexane content less than 45% or greater than 55% were excluded from consideration.

No chronic or subchronic RfDs or RfCs or cancer assessment for commercial hexane are available on IRIS (U.S. EPA, 2008), the Drinking Water Standards and Health Advisory list (U.S. EPA, 2006), or in the Health Effects Assessment Summary Tables (HEAST; U.S. EPA, 1997). No documents on commercial hexane are listed in the Chemical Assessments and Related Activities (CARA) list (U.S. EPA 1991a, 1994a). The Occupational Safety and Health Administration (OSHA), National Institute of Occupational Safety and Health (NIOSH), and American Conference of Governmental Industrial Hygienists (ACGIH) have not derived occupational exposure limits for commercial hexane (OSHA, 2008; NIOSH, 2008; ACGIH, 2007). The ATSDR, World Health Organization (WHO), and the International Agency for Research on Cancer (IARC) have not published documents on commercial hexane (ATSDR, 2008; WHO, 2008; IARC, 2008). The National Toxicology Program (NTP, 2008) has not performed toxicity or carcinogenicity assessments for *n*-hexane or commercial hexane, and these compounds a not on the *11th Report on Carcinogens* (NTP, 2005).

IRIS (U.S. EPA, 2008) reports a chronic RfC and cancer assessment for *n*-hexane. The IRIS toxicological review for *n*-hexane (U.S. EPA, 2005a) includes a review of data on commercial hexane; the IRIS review was used extensively for this report. Toxicological reviews of this mixture by the Massachusetts Department of Environmental Protection (MADEP, 2003) and the Total Petroleum Hydrocarbon Criteria Working Group (TPHCWG, 1997) were also consulted for relevant information.

To identify toxicological information pertinent to the derivation of provisional toxicity values for commercial hexane, the IRIS toxicological review for *n*-hexane (U.S. EPA, 2005a) was consulted for pertinent studies, as were MADEP (2003) and TPHCWG (1997). Update literature searches to June 2008 were conducted using the following databases: MEDLINE, TOXLINE, BIOSIS, TSCATS, CCRIS, DART/ETIC, GENETOX, HSDB, and Current Contents to identify any studies of commercial hexane published since the IRIS review (U.S. EPA, 2005a). Appendix A provides additional detail on the literature search process.

REVIEW OF PERTINENT DATA

Human Studies
Oral Exposure
No studies examining the health effects of oral exposure to commercial hexane in humans were identified.

Inhalation Exposure
U.S. EPA (2005a) reviewed the epidemiological data on human inhalation exposure to mixtures containing *n*-hexane. The *n*-hexane content of the mixtures varied widely. A few of the studies explicitly examined the effects of human exposure to commercial or technical grade hexane. The summaries of these studies as reported by U.S. EPA (2005a) are represented below.

> *Passero et al. (1983) screened 654 workers in 44 shoe factories and 86 home shops during a period from 1973-1981. Evaluation by clinical and electrodiagnostic examination identified 184 workers with some degree of neurological abnormality. Of these 184 subjects, 9 had other neurological disorders (the authors reported that the most common was radiculopathy due to intervertebral disc disease), 77 displayed minimal changes and were considered normal following repeated examination by the study authors and 98 manifested overt polyneuropathy. The majority of the workplace solvent samples collected contained commercial hexane. The commercial hexane was determined to contain greater than 60% of total mass as hydrocarbons such as pentane, 2-methyl-pentane, 3-methyl-pentane, n-hexane, heptane, cyclopentane, cyclohexane and methyl-cyclopentane. In 7/12 samples taken from workplaces of individuals with the most severe polyneuropathy, over 99% of the total solvent was composed of these hydrocarbons. No relationship was found between length of exposure and severity of disease. In the cases of polyneuropathy, the neurological pattern showed an insidious onset of loss of distal motor and sensory function with marked reflex loss. General symptoms, such as nausea or vomiting, epigastric pain and insomnia, preceded or accompanied the neuropathy. Clinical symptoms were weakness, paresthesia (burning or tingling sensation in limbs) and cramp-like pain with related motor impairment, hypoesthesia (partial loss of sensation and/or diminished sensibility), changes in tendon reflexes and muscle trophism and tone. These symptoms were usually confined to distal portions of the limbs and occurred with varying degrees of intensity depending on the extent of exposure. All 98 polyneuropathy cases exhibited abnormal motor nerve action potentials (MAPs), regardless of severity.*

The occurrence of fibrillations, positive waves, fasciculations and slowing of motor nerve conduction velocity (MCV) increased with disease severity. Several of the most affected cases exhibited CNS involvement with alterations in electroencephalogram or spasticity in the lower limbs and increased deep tendon reflexes. The clinical course of these 98 cases was followed for up to 8 years. Except for the most severe cases, patients improved slowly when removed from the affected environment. However, deterioration continued for some even after exposure ceased.

Seppalainen et al. (1979) compared the visual evoked potentials (VEPs) and electroretinograms (ERGs) of 15 workers to those of 10 healthy subjects with no occupational exposure to solvents or other neurotoxic chemicals. The highest recorded n-hexane levels in the two factories where the workers were exposed ranged from 2000 to 3250 ppm. In both factories, exposure was to technical grade hexane, which contains other aliphatic hydrocarbons with no known neurotoxic effects. Maculopathy, color discrimination deficits, flatter VEPs and diminished peak-to-peak amplitudes of the ERGs were more common among cases than controls.

An earlier study by the same researchers described visual defects in this same group of 15 workers (Raitta et al., 1978). The Farnsworth-Munsell (FM)-100 hue test showed 12 of the 15 subjects to have impaired color vision, one of which was probably due to a congenital abnormality. The other cases of color vision impairment were acquired, mostly in the blue-yellow axis. In 11/15 subjects there was evidence of associated maculopathy (damage of vessels in the eye that leak fluid into the center of the retina causing loss of central vision), in most cases characterized by pigment dispersion.

Animal Studies

The summaries of available studies of commercial hexane contained in the IRIS Toxicological Review of *n*-hexane (U.S. EPA, 2005) are provided below with additional information.

Oral Exposure

Krasavage et al. (1980) reported the results of a 90-day study in rats exposed to commercial grade hexane via oral gavage. The *n*-hexane content of the mixture was reported to be 40%. This study was not included in the review, as studies of mixtures with *n*-hexane content less than 45% or greater than 55% were excluded from consideration.

Inhalation Exposure

MADEP (2003) identified two studies of commercial hexane (Miyagaki, 1967 and IRDC, 1986) with *n*-hexane concentrations outside the limits established for this review (45−55%). The *n*-hexane content in the mixture studied by Miyagaki (1967) was reported to range from 60−75%, while the *n*-hexane content in the commercial hexane used by IRDC (1986) was 37−39%. These studies were not included in the review, as the results could not be reliably considered comparable to mixtures containing ~50% *n*-hexane.

Subchronic Studies—Biodynamics (1989; also published as an abstract by Duffy et al., 1991) conducted a 13-week inhalation toxicity study of commercial hexane in F344 rats and B6C3F1 mice. Animals (10/sex/group) were exposed to target concentrations of 0-, 900-, 3,000-, or 9,000-ppm commercial hexane (51.7−53.5% *n*-hexane) for 6 hours/day, 5 days/week for 13 weeks. Animals were observed twice daily for mortality and clinical signs, with weekly detailed clinical examinations. Body weights and food consumption were recorded weekly. Ophthalmoscopic examinations were made before exposure began and just prior to termination. Blood was collected for hematology (erythrocyte count [RBC], total and differential leukocyte counts, platelet count, hematocrit [Hct], hemoglobin [Hgb], mean corpuscular volume, mean corpuscular hemoglobin [MCH], mean corpuscular hemoglobin concentration [MCHC]), and clinical chemistry (aspartate aminotransferase, alanine aminotransferase, creatinine, blood urea nitrogen [BUN], glucose, total protein, alkaline phosphatase [ALP], albumin, electrolytes, inorganic phosphorus, gamma glutamyl transferase [GGT], total bilirubin, creatine phosphokinase, and lactic acid dehydrogenase [LDH]). In mice, the clinical chemistry parameters were limited by the small quantities of serum. Gross necropsy was performed on all animals at study termination, and selected organ weights (adrenals, ovaries, testes with epididymides, kidneys, liver, brain, lungs, heart, and spleen) were recorded. Comprehensive histopathology examinations (35 tissues) were made on all animals.

In rats, there were no changes in body weight, food consumption, or water intake and no treatment-related mortality at any concentration (Biodynamics, 1989; Duffy et al., 1991). The only treatment-related clinical sign of toxicity was lacrimation in high-concentration female rats (in only 2/10 rats). Corneal dystrophic changes were observed in two high-concentration males; no other ophthalmoscopic changes were reported. In high concentration-males, increased platelet count (8% higher than controls) was observed. Clinical chemistry evaluation indicated increases in creatinine, total protein, and albumin and decreased chloride in high-concentration males. It was noted that these changes were within reference ranges for untreated rats (Wolford et al., 1986). No statistically significant changes in hematology or clinical chemistry were observed in female rats. At the high concentration, there were increases in relative kidney and adrenal weights in males and an increase in relative adrenal weight in females. Histopathological changes in these organs were limited to hydrocarbon nephropathy in the male rats, as discussed in more detail below. Male rats exposed to 9,000 ppm displayed an increase in absolute and relative liver weights (16 and 19% higher than controls, $p < 0.01$). Slight hemorrhage was observed in the livers of 3/10 high-concentration male rats, and acute/subacute inflammation of the liver was noted in 2/10 high-concentration males. These effects were not observed in female rats or control or lower-concentration males. No histopathology was observed in the nasopharyngeal tissues or larynx.

Adverse histopathological findings typical of hydrocarbon nephropathy were observed in the kidneys of high-dose male rats, as described in the experimental pathology report of the study (EPL, 1989). All male rats (controls and exposed) showed some evidence of hyaline droplet formation and related nephropathy. However, this effect was more severe in male rats exposed to commercial hexane compared with controls. The kidneys of high-concentration males showed mild tubular dilatation, with granular material in the lumen and signs of epithelial regeneration compared with controls. High-concentration males displayed mild-to-moderate degrees of epithelial regeneration, a response that was minimal in controls and in animals receiving lower concentrations of commercial hexane.

A minimal LOAEL of 9,000 ppm was identified for rats based on liver effects (increased liver weight, slight hemorrhage, and inflammation) in males, and a NOAEL of 3,000 ppm was identified. Although the incidences of histopathological findings were not statistically significant, the effects, especially hemorrhage, were clearly adverse, were not observed at lower concentrations or in controls, and were correlated with liver-weight increases.

As with rats, there were no effects of treatment on body weight, food or water consumption, or mortality in mice (Biodynamics, 1989; Duffy et al., 1991). Excessive lacrimation was observed in both sexes of high-concentration mice (up to 7/10 animals at one time point at 3,000 ppm and 10/10 at 9,000 ppm in males and 7/10, 9/10, and 9/10 in low-, mid-, and high-concentration females, with increases over time); no control mice displayed this effect. There were no treatment-related ophthalmoscopic findings in mice. Mean corpuscular volume was increased relative to controls in high-concentration males, but no other hematology changes were observed. Absolute and relative liver weights were significantly increased (8% and 9%, $p < 0.05$) in high-concentration female mice, and relative liver weight was increased in high-concentration males (12%, $p < 0.01$). There were no treatment-related histopathology findings in mice. A minimal LOAEL of 900 ppm was identified for these data based on excessive lacrimation, an indication of eye irritation, in female mice. The incidence of this effect increased with time and concentration, was observed in males at higher concentrations, and was not observed in control mice at any time. No NOAEL can be identified.

Bio-Research Laboratories (1990) conducted a 13-week study of the effects of commercial hexane (51.5−53.3% *n*-hexane) in Sprague-Dawley rats (also reported in an abstract by Soiefer et al., 1991). Groups of 12 rats/sex were exposed to 0-, 900-, 3,000-, or 9,000-ppm commercial hexane for 6 hours/day, 5 days/week for 13 weeks. Clinical signs were monitored twice daily, while body weight and food consumption were determined weekly. The animals were evaluated in a functional observational battery (FOB) approximately 1−2 hours after the first exposure and prior to exposure on Days 1, 7, 14, 35, 63, and 91. The FOB included qualitative observations (in the chamber, during handling, and in an arena designed for this purpose) as well as quantitative measures of grip strength (forelimb and hindlimb) and landing foot splay. Motor activity was tested prestudy and on Days 34, 62, and 90. From each exposure group, four rats/sex were given complete gross pathological examinations upon sacrifice. The remaining eight animals/group were subjected to whole body perfusion and set aside for neuropathology examination. In total, six animals/sex in each of the control and high-concentration groups were assessed for histological signs of neuropathology in a large number of nervous system tissues; tissues in other groups were not assessed because effects were not noted in the high-concentration group.

Treatment with commercial hexane did not affect survival, body weight, or food consumption (Bio-Research Laboratories, 1990; Soifer et al., 1991). A higher incidence of staining of the muzzle/head and/or periorbital region was reported in treated animals; the authors suggested that this reflected stress-induced porphyrhinitis. No treatment-related effects were observed in the FOB or motor activity assessments of any treatment groups, nor was evidence of neuropathology observed on examination of nervous system tissues in the high-concentration group. A NOAEL of 9,000 ppm was identified for neurotoxicity/neuropathology in rats based on these data.

The International Research and Development Corporation (IRDC), sponsored by Phillips Petroleum Co., continuously exposed male Sprague-Dawley rats to *n*-hexane and a C6-isomer mixture consisting of *n*-hexane, methylcyclopentane, 3-methylpentane, and 2-methylpentane for 22 hours/day, 7 days/week for 6 months (IRDC, 1992a,b). This study was conducted in two phases; Table 1 shows the exposure groups.

Table 1. Experimental Protocols for Phases I and II of a 6-Month Inhalation Study of *n*-Hexane and a Mixture Containing Hydrocarbon Isomers Plus *n*-Hexane in Male Sprague-Dawley Rats[a]			
Phase	**Group**	**Treatment**	**Number of Animals Treated**
I	I	Controls	24
	II	125-ppm *n*-hexane	14
	III	125-ppm *n*-hexane + 125 ppm C6 isomers[b]	14
	IV	125-ppm *n*-hexane + 375 ppm C6 isomers	14
	V	125-ppm *n*-hexane + 1,375 ppm C6 isomers	14
	VI	500-ppm *n*-hexane	24
II	VII	Controls	20
	VIII	500-ppm C6 isomers	20
	IX	500-ppm *n*-hexane + 500 ppm C6 isomers	20
	X	500-ppm *n*-hexane	20

[a]IRDC (1992a,b).
[b]C6 isomers were a mixture of *n*-hexane-depleted C6 hydrocarbons containing methylcyclopentane, 3-methylpentane, and 2-methylpentane as major components.

For the purposes of this review, the relevant results are from Groups III and IX of the 10 treatment groups used in this study. These groups, but not the other treatment groups, were exposed to C6 mixtures consisting of 50% *n*-hexane. This is the mixture most similar to the composition of commercial hexane, which is 51−53% *n*-hexane. The other major components of the C6 mixture were methylcyclopentane, 2-methylpentane, and 3-methylpentane (IRDC, 1992a, b). For groups III and IX, the proportions of these three components were approximately 15% each; these proportions are also similar to that of commercial hexane.

In both phases of this study, animals were examined daily for signs of clinical toxicity, and body weights were monitored weekly (IRDC, 1992a,b). In Phase I, two controls and four rats from Group VI (see Table 1) were taken from their exposure groups every month for the first 5 months. These animals, plus four from all groups exposed for 6 months, were examined histopathologically for changes to the cervical spinal cord. All surviving animals (10/group) were necropsied at study termination, and the weights of their major organs were recorded. Excised pieces of tissue from a variety of organs and tissues were fixed for histopathological examination, including all abnormal masses, adrenal gland, abdominal aorta, bone marrow, brain, Zymbal's gland, esophagus, epididymides, eye and optic nerve, tongue, Harderian gland, neuroganglia, liver, kidney, lung, lymph nodes, mammary gland, pancreas, parathyroid, pituitary, prostate, salivary gland, skeletal muscle, skin, nasal turbinates, gonads, lacrimal gland, heart,

thymus, thyroid, peripheral nerve, small intestine, large intestine, spinal cord, spleen, seminal vesicle, stomach, and urinary bladder.

No treatment-related differences in survival, clinical signs, or body weight were noted in Group III (125-ppm *n*-hexane and 125-ppm C6 isomers; see Table 1) compared with controls (although body weights were slightly higher than controls) (IRDC, 1992a). None of the mixed hexane treatment groups, including Group III, exhibited neuropathologic or myopathic changes in Phase I of the study. Slight reductions in relative organ weights (compared with controls) were attributable to the higher body weights in Group III animals. Gross necropsy findings in Group III consisted of "tan or red raised, soft areas" in the livers of 2/10 rats, characterized as mild-to-moderate in severity; there were no such findings in controls. Microscopic findings of hepatocellular necrosis in 2/10 Group III rats were consistent with the gross findings. Necrosis was also observed in the livers of rats of other treatment groups but was not observed in any controls. Although the increased incidence was not statistically significant, the authors considered the effect to be treatment-related based on the severity of the effect.

In Phase II of the study (see Table 1), five rats/group were sacrificed after 2 and 6 months for neuropathology evaluation (IRDC, 1992b). The surviving animals (10/group) were sacrificed after 6 months for complete necropsy, organ weight determinations, and histopathology evaluations (see Phase I above for description). There were no treatment-related deaths in Phase II. Beginning at Week 17, abnormal gait was observed in animals of Group IX (500-ppm *n*-hexane and 500-ppm C6 isomers; see Table 1); the incidence and severity of this effect increased with time. Body weight was significantly reduced in Group IX animals beginning in Week 5; at study termination, the average body weight was 25% less than controls ($p < 0.01$). Absolute and relative kidney weights were significantly increased (19% and 61%, respectively; $p < 0.01$) in Group IX animals when compared with controls; other organ weight changes were attributable to reductions in body weight. There were no gross necropsy findings attributable to treatment. Histopathology findings in Group IX animals included axonal degeneration, atrophy, and mononuclear cell infiltration in the tibial and/or sciatic nerves, mild skeletal muscle atrophy, and chronic nephritis. Table 2 shows the incidences and severity of these findings.

Table 2. Incidences of Histopathology Findings for Phase II of a 6-Month Inhalation Study of *n*-Hexane and a Mixture Containing Hydrocarbon Isomers Plus *n*-Hexane in Male Sprague-Dawley Rats[a]		
Target Organ/Cellular Response	**Control**	**Group IX (500-ppm *n*-hexane + 500-ppm C6 Mixture)**
Tibial and Sciatic Nerves (Combined)		
Atrophy (Trace/Mild)	0/9	8/17
Axonal Degeneration (Trace)	0/9	2/17
Mononuclear Cell Infiltration (Trace/Mild)	0/9	3/17
Skeletal Muscle		
Atrophy (Mild)	0/10	1/10
Kidney		
Chronic Nephritis: Trace Mild Moderate	6/10 0/10 0/10	3/10 3/10 1/10

[a]IRDC (1992b).

In contrast to Phase I, neither gross liver abnormalities nor necrosis of hepatocytes was observed in Phase II, despite the higher concentration of test material used (IRDC, 1992a,b). As a result, the liver findings in Phase I are considered anomalous and not related to treatment. Thus, these data are consistent with a NOAEL of 250-ppm mixed hexanes (containing 50% *n*-hexane) and a LOAEL of 1,000 ppm based on neuropathology, muscle atrophy, body weight reductions, and increased severity of chronic nephritis in male rats. The authors indicated that during exposure a brown oily material collected on the glass beads of the inhalation system for each exposure group. Samples of this brown material were subjected to infrared spectroscopy, which confirmed the presence of a phthalate ester-type compound. Although the toxicological effects noted were consistent with the toxicity of *n*-hexane, some uncertainties related to potential co-exposure exist.

Although other groups in this study (IRDC, 1992a,b) were exposed to pure *n*-hexane or to mixtures that are not representative of commercial hexane, examination of the findings in these groups is instructive in helping to distinguish effects that are attributable to *n*-hexane versus mixed hexanes without *n*-hexane, as well as to give an indication of dose-response relationships. To this end, Table 3 compares the effects observed in the different groups. In the table, observed changes are shown under the three primary types of effects (neuropathy/myopathy, hepatic effects, and renal effects) reported in the studies. The comparison of effects among the groups shows the scattered nature of the liver changes (lack of dose-response relationship) in Phase I as well as the absence of liver findings in Phase II, despite higher exposures. The table also provides support for the suggestion that *n*-hexane may be largely responsible for the neuropathy and myopathy findings, as groups exposed to mixtures with low (<500 ppm) or no *n*-hexane did not exhibit evidence of these effects. Interpretation of the kidney findings is not as clear, as groups exposed to pure *n*-hexane (Groups VI and X) or to hexanes without *n*-hexane (Group VIII) exhibited varying degrees of kidney histopathology and/or weight changes.

Chronic Studies—The American Petroleum Institute (API) sponsored two 2-year carcinogenicity studies with commercial hexane: one in F344 rats (Biodynamics, 1993a) and the other in B6C3F1 mice (Biodynamics, 1993b). The principal features and key findings of these studies have been compiled into a single research report that was published in the peer-reviewed literature (Daughtrey et al., 1999). In both studies, 50 animals/sex/group were exposed 6 hours/day, 5 days/week, to a commercial hexane preparation at targeted inhalation concentrations of 0, 900, 3,000, or 9,000 ppm for 2 years. The commercial hexane preparation used in the experiments consisted of 51.5% *n*-hexane, 16% methylcyclopentane, 16.1% 3-methylpentane, 12.9% 2-methylpentane, 3.3% cyclohexane, and trace amounts of other hydrocarbons. Detailed physical examinations were given weekly. Body weight was recorded weekly through Week 13 and monthly for the remainder of the study. All animals were given ophthalmoscopic examinations before the study and at study termination. Differential leukocyte count and erythrocyte morphology were evaluated on blood collected at Months 12, 18, and at termination. Complete necropsies were performed on all animals, and histopathology of a comprehensive list of tissues (>30) was evaluated in control and high-concentration animals, as well as any animals that died prior to terminal sacrifice. Organ weights were not recorded.

There were no statistically significant differences in survival rates between control and exposed rats of either sex (Biodynamics, 1993a; Daughtrey et al., 1999). Exposed animals showed few clinical signs of toxicity in response to exposure to commercial hexane other than lacrimation; this effect was observed in control animals as well, but at an increased incidence in male rats of the mid- and high-concentration groups (incidence varied over time, with peak incidences of 16/50, 26/50, and 30/50 in control, mid-, and high-concentration groups, respectively). Body weights were significantly decreased ($p \leq 0.05$) in mid- and high-concentration rats of both sexes. Terminal body weights were 7 and 11% lower than controls in high-concentration males and females, respectively, with smaller reductions in the mid-concentration group. Ophthalmoscopic findings were unremarkable, as were the limited hematology analyses and gross necropsy observations. Histopathological lesions in the respiratory passages were noted, especially in the nasal turbinates and larynx (see Table 4). Specific findings consisted of hyperplasia of epithelial and goblet cells, chronic inflammation, and increased incidence of intracytoplasmic eosinophilic material in all groups exposed to commercial hexane. Chronic inflammation was also seen to some extent in controls. Low-, mid-, and high-dose males and females displayed squamous metaplasia/hyperplasia of the columnar epithelium in the larynges. Table 4 does not reflect the histologic examinations of the larynx performed only on animals that died prior to terminal sacrifice in the low- and mid-dose groups. There were no treatment-related necropsy findings in tissues located away from the site-of-entry, and no treatment-related histopathological abnormalities in the sciatic nerves were observed in any group of F344 rats exposed to commercial hexane in this study. There was no treatment-related tumor formation at any tissue site in F344 rats. The histopathological lesions of the respiratory tract that were evident, even in low-dose rats of both sexes, suggest that a NOAEL cannot be derived from this study. A LOAEL of 900 ppm (lowest dose tested) is identified based on the nasal and laryngeal lesions.

		Phase I						Phase II			
Group	**I**	**II**	**III**	**IV**	**V**	**VI**	**VII**	**VIII**	**IX**	**X**
n-hexane **(ppm)**	**0**	**125**	**125**	**125**	**125**	**500**	**0**	**0**	**500**	**500**
Mixed hexanes (ppm)	**0**	**0**	**125**	**375**	**1375**	**0**	**0**	**500**	**500**	**0**
Body weight (g)	535	602	609 [b]	572	528	444[b]	581	568	436[c]	407[c]
Neuropathic/myopathic Effects										
Abnormal gait[d]	0/14	0/14	0/14	0/14	0/14	7/14	0/15	0/15	8/15	8/15
Skeletal muscle atrophy	0/10	0/10	0/10	0/9	0/10	9/10	0/10	0/10	1/10	3/10
Tibial or sciatic nerve atrophy (trace/mild)	-[e]	-	-	-	-	-	0/9	0/16	8/17	14/16
Tibial or sciatic nerve axonal degeneration	0/10	0/10	0/10	0/10	0/10	7/10	0/9	0/16	2/17	0/16
Tibial or sciatic nerve mononuclear cell infiltration (trace/mild)	-	-	-	-	-	-	0/9	0/16	3/17	3/16
Spinal cord (thoracic/lumbar/sacral) axonal degeneration	0/10	0/10	0/10	0/10	0/10	8/10	-	-	-	-
Spinal cord (thoracic/lumbar/sacral) vacuolar change	0/10	0/10	0/10	0/10	0/10	1/10	-	-	-	-
Hepatic Effects										
Gross liver discoloration	0/10	3/10	2/10	0/10	1/10	5/10	-	-	-	-
Mean absolute liver weight (g)	15.45	18.11	18.26	17.70	19.81[c]	14.65	-	-	-	-
Mean liver: body weight (%)	2.88	3.01	3.02	3.08	3.74[c]	3.31[c]	-	-	-	-
Panlobular necrosis (trace)	0/10	0/10	2/10	0/10	0/10	0/10	-	-	-	-
Panlobular necrosis (mild)	0/10	1/10	0/10	0/10	1/10	2/10	-	-	-	-
Panlobular necrosis (moderate)	0/10	2/10	0/10	0/10	0/10	0/10	-	-	-	-

Table 3. Comparison of Effects Among Groups of Rats Treated for 6 Months with Various Combinations of *n*-Hexane and Mixed Hexanes[a]

Table 3. Comparison of Effects Among Groups of Rats Treated for 6 Months with Various Combinations of *n*-Hexane and Mixed Hexanes[a]										
	Phase I						Phase II			
Group	I	II	III	IV	V	VI	VII	VIII	IX	X
Renal Effects										
Mean absolute kidney weight (g)	2.98	3.32	3.32	3.65[c]	4.14[c]	3.15	3.08	3.49[b]	3.68[c]	3.40
Mean kidney: body weight (%)	0.56	0.55	0.55	0.65[b]	0.78[c]	0.71[c]	5.32	6.16	8.56[c]	8.44[c]
Mean kidney: brain weight (%)	-	-	-	-	-	-	1.46	1.69[b]	1.99[c]	1.75[c]
Degeneration/regeneration (trace) (mild)	0/10 0/10	0/10 0/10	0/10 0/10	6/10 2/10	5/10 5/10	4/10 0/10	-	-	-	-
Chronic nephritis (trace) (mild) (moderate)	-	-	-	-	-	-	6/11 0/11 0/11	5/10 3/10 0/10	3/10 3/10 1/10	2/10 7/10 1/10

[a]IRDC (1992a,b).
[b]Significantly different from controls ($p < 0.05$).
[c]$p < 0.01$.
[d]Incidence at Week 25.
[e]Effect not present (for quantal endpoints) or not changed by exposure (organ weights).

<table>
<tr><td colspan="5" align="center">Table 4. Incidence of Nasal and Laryngeal Lesions in Male and Female F344 Rats
Exposed to Commercial Hexane for 2 Years[a]</td></tr>
</table>

Target Organ/Cellular Response	Target Concentration of Commercial Hexane (ppm)			
	0	900	3,000	9,000
Nasal Mucosa				
Males				
Goblet Cell Hypertrophy/Hyperplasia	29/48	37/50[b]	43/50[c]	41/50[c]
Epithelial Hyperplasia	2/48	19/50[c]	36/50[c]	43/50[c]
Intracytoplasmic Eosinophilic Material	21/48	49/50[c]	46/50[c]	46/50[c]
Inflammation	9/48	8/50	10/50	23/50
Females				
Goblet Cell Hypertrophy/Hyperplasia	33/50	43/50[c]	43/50[c]	46/50[c]
Epithelial Hyperplasia	6/50	34/50[c]	38/50[c]	42/50[c]
Intracytoplasmic Eosinophilic Material	41/50	47/50[c]	48/50[c]	49/50[c]
Inflammation	8/50	6/50	4/50	13/50
Larynx				
Males				
Columnar Epithelial Hyperplasia/Metaplasia	4/49	-[d]	-	11/50
Females				
Columnar Epithelial Hyperplasia/Metaplasia	1/48	-	-	7/48

[a]Daughtrey et al. (1999); Biodynamics (1993a).
[b]Significantly different ($p < 0.05$) from controls, as calculated by the authors using ordinal logistic regression.
[c]$p < 0.01$.
[d]Laryngeal tissue not examined in all animals of these groups.

There were no statistically significant differences in survival between controls and exposed mice of either sex (Biodynamics, 1993b; Daughtrey et al., 1999). There were no differences in clinical signs of toxicity, ophthalmoloscopic findings, or hematology. Body weight changes in commercial hexane-exposed mice were generally similar to those in controls. Statistically significant ($p \leq 0.05$) body weight depression was noted in females exposed to 9,000 ppm after Week 29. On week 53 body weights were decreased by 14% below controls, however by study termination, body weights in this group only differed from controls by 3%. No nonneoplastic histopathology findings were affected by treatment; however, it should be noted that the nasal turbinates were not examined for histopathology in mice. A minimal LOAEL of 9,000 ppm (lowest dose tested) is identified from these data based on body weight reductions in females; although body weights returned to normal by study termination, the decrease compared with controls persisted for nearly half of the 2-year study. The NOAEL is 3,000 ppm.

In female mice, there was a dose-related increase in the incidence of hepatocellular neoplasms (Biodynamics, 1993b; Daughtrey et al., 1999). When benign and malignant tumors were combined, the incidence reached statistical significance in the high-concentration group. There was also an increased incidence of pituitary adenomas and adenocarcinomas in exposed females (see Table 5). For these tumors there was a significantly elevated incidence at each exposure concentration. Commercial hexane was associated with decreased severity and incidence of cystic endometrial hyperplasia of the uterus among high-dose females compared with controls.

Table 5. Incidence of Liver and Pituitary Tumors in Male and Female B6C3F1 Mice Exposed to Commercial Hexane for 2 Years[a]

Target Organ/Cellular Response	Target Concentration of Commercial Hexane (ppm)			
	0	**900**	**3,000**	**9,000**
Liver				
Males				
Adenomas	10/49	5/50	7/50	10/50
Carcinomas	7/49	11/50	10/50	3/50
Combined adenomas and carcinomas	17/49	16/50	17/50	13/50
Females				
Adenomas	4/50	6/50	4/49	10/50
Carcinomas	3/50	2/50	5/49	6/50
Combined adenomas and carcinomas	7/50	8/50	9/49	16/50[b, c]
Pituitary				
Males				
Hyperplasia	0/46	0/11	0/6	1/46
Adenomas	1/46	0/11	0/6	0/46
Adenocarcinomas	0/46	0/11	0/6	0/46
Total neoplasms	1/46	0/11	0/6	0/46
Females				
Hyperplasia	2/45	4/48	4/48	6/49
Adenomas	0/45	6/48[b]	7/48[d]	5/49[b]
Adenocarcinomas	0/45	0/48	1/48	0/49
Total neoplasms	0/45	6/48[b]	8/48[d]	5/49[b]

[a]Daughtrey et al. (1999); Biodynamics (1993b).
[b]Significantly different ($p < 0.05$) from controls, as calculated by the authors using Fisher's Exact test.
[c]Significant dose-related trend; Cochrane-Armitage test, $p < 0.05$.
[d]Significantly different ($p < 0.01$) from controls, as calculated by the authors using Fisher's Exact test.

Reproductive/Developmental Studies—API sponsored two reproductive studies in laboratory rats and mice exposed to commercial hexane (BRRC, 1989a,b). The first study was a range-finding study in which pregnant Sprague-Dawley rats (8/group) and CD-1 mice (8/group) were exposed to inhaled commercial hexane for 6 hours/day at target concentrations of 0, 900, 3,000, or 9,000 ppm on Gestation Days (GDs) 6−15 (BRRC, 1989a). The mixture composition was not reported. Pregnant rats were terminated on GD 21 and pregnant mice on GD 18. Maternal body weight gain was monitored intermittently and at termination. Uterine weights, number of ovarian corpora lutea, implantation sites, and viable and nonviable implants were evaluated. All live fetuses were weighed, sexed, and examined for external and visceral malformations and skeletal variations. None of the dams of either species displayed overt maternal toxicity during the course of the experiment. There appeared to be a slight increase in body weight gain in the high-dose rats in parallel with increased food and water consumption in this group. The only sign of reproductive or developmental toxicity was a reduction in mean fetal weight per litter (11% below controls, $p < 0.05$) in the progeny of pregnant mice exposed to 9,000-ppm commercial hexane. No treatment-related malformations or variations were observed in either the rat or mouse fetuses. This study identifies a developmental LOAEL of 9,000 ppm for reduced fetal weight in mice, with a developmental NOAEL of 3,000 ppm and maternal NOAEL of 9,000 ppm in mice. In rats, a developmental and maternal NOAEL of 9,000 ppm (the highest dose tested) is identified.

In the full study, BRRC (1989b) exposed pregnant Sprague Dawley rats (25/group) to 0-, 900-, 3,000-, or 9,000-ppm commercial hexane (containing 53% *n*-hexane) for 6 hours/day on GDs 6−15 and sacrificed the animals on GD 21. Maternal body weights and food and water consumption were recorded on GDs 0, 6, 9, 12, 15, 18, and 21, and the weights of liver, kidney, and uterus were measured at sacrifice. As in the range finding study, numbers of ovarian corpora lutea, implantation sites, resorptions, and live and dead fetuses were evaluated. Fetuses were examined for external and visceral abnormalities and for skeletal variations. There were no treatment-related effects on reproductive, developmental, or teratological parameters in any of the groups of rats in the study. Among maternal effects, body weight gain was reduced in high-concentration dams throughout exposure (19% below controls, $p < 0.05$, during GDs 6−15) and in the mid-concentration group for a portion of the exposure period (29% below controls, $p < 0.05$, during GDs 9−12). Food consumption was reduced in the high-concentration dams but not in the mid-concentration dams. In dams exposed to the high concentration, an increased incidence of pulmonary color change (6/21, compared with 0/23 controls; $p < 0.05$) was observed at necropsy. A minimal maternal LOAEL of 3,000 ppm is identified based on body weight reductions during gestation, with a maternal NOAEL of 900 ppm in rats. The reduction in body-weight gain at this concentration was transient, occurring during GDs 9−12 only, and there was no statistically significant reduction in body-weight gain over the entire exposure period or gestational period. No reduction in body weight gain was reported in the range-finding study in rats (BRRC, 1989a), or in the F_0 generation in the two-generation reproductive toxicity study (BRRC, 1991; Daughtrey et al., 1994, described below). Decreased body weight was observed, however, in the F_1 and F_2 pups in the two-generation study (BBRC, 1991; Daughtrey et al., 1994), in rats in a subchronic study (IRDC, 1992b), and female mice in the chronic inhalation study (Biodynamics, 1993b; Daughtrey et al., 1999) discussed previously. Taking into consideration that U.S. EPA regards body weight depression >10% to be adverse, that the effect was transient in the 3,000-ppm dams but more pronounced in the 9,000-ppm dams in the BBRC

(1989b) study, and that a body weight effect also was seen in some (but not other) studies, the LOAEL of 3,000 ppm is considered close to the threshold for this effect. Thus, a developmental NOAEL of 9,000 ppm applies to these data.

In addition, pregnant CD-1 mice (30/group) were exposed to the same regimen as that described for the Sprague-Dawley rats (BRRC, 1989b), and the same evaluations were performed. There were no treatment-related effects on maternal body-weight gain, no changes in food and water consumption, and no other clinical signs of toxicity among the exposed groups compared with controls. Gestational parameters, including the numbers of viable and nonviable implantations/litter and sex ratio, were unaffected by exposure to commercial hexane. However, gross necropsy revealed a dose-dependent increase in the incidence of discoloration of the lungs (0/27, 0/27, 2/25, and 12/29 in control through high-concentration groups). The incidence at 9,000 ppm was significantly different from controls ($p < 0.01$). In addition, dark brown foci were evident in the lungs of 4/29 high-dose and 2/25 mid-dose dams; these incidences were not statistically different from controls.

Fetal body weights were unchanged among the groups, and there were no significant changes in the incidence of individual malformations or pooled external, visceral, or skeletal malformations (BRRC, 1989b). However, there were treatment-related increases in the incidences of two individual skeletal variations in high-dose pups. Comparing the incidences of these effects between controls and high-dose groups by litter, the numbers were 0/26 versus 6/26 for bilateral bone islands at the first lumbar arch and 20/26 versus 26/26 for all unossified intermediate phalanges (statistically significant at $p < 0.05$, Fisher's Exact test as calculated by the study authors). Based on these skeletal variations, a developmental NOAEL of 3,000 ppm and LOAEL of 9,000 ppm were identified. The only effect observed in dams was discoloration of the lungs in mid- and high-dose animals, some of which also had dark brown foci on the lungs. These effects were not observed in mice exposed to the same concentrations in the range-finding study (BRRC, 1989a) and for a much longer duration in a chronic study (Biodynamics, 1993b; Daughtrey et al., 1999). Pulmonary effects were not observed in rats exposed to the same concentrations in subchronic, chronic, reproductive, or developmental toxicity studies. Finally, it is not clear that discoloration of the lungs represents an adverse effect. Thus, the high concentration (9,000 ppm) (the highest dose tested) is considered a NOAEL for maternal effects.

BRRC (1991) carried out a two-generation reproductive/developmental toxicity study in which, prior to breeding, 25 Sprague-Dawley rats/sex/group (F_0 generation) were exposed to concentrations of 0-, 900-, 3,000-, or 9,000-ppm inhaled commercial hexane for 6 hours/day, 5 days/week for 10 weeks. The study was published in the peer-reviewed literature by Daughtrey et al. (1994). Clinical signs of toxicity were monitored daily, and food consumption and body-weight data were recorded weekly. After 10 weeks, males and females were mated, and these mating pairs were exposed to commercial hexane at the same doses for 6 hours/day, 7 days/week for 21 days. Cohabitation was maintained only long enough for pregnancy to be achieved (copulation plug present). For the dams, exposure was continued through GD 19, discontinued until Postnatal Day (PND) 4, and then reinstituted until weaning on PND 28, at which point the F_0 animals were sacrificed. On PND 4, the pups were culled to 4/sex/litter; then, on PND 28, 25 F_1 rats/sex/group were randomly selected for exposure to commercial hexane for 8−11 weeks. Subjects were then mated as described for the F_0 generation. All F_2 rats were sacrificed on PND 28.

Among the reproductive indices evaluated were survival, mating, fertility, gestation, live births, and lactation (BRRC, 1991). All subjects were necropsied, and excised pieces of liver, kidney, pituitary, and upper and lower respiratory tract, and any obvious lesions were examined for histopathology. Reproductive organs and tissues taken for histopathology included the vagina, uterus, ovary, testis, epididymis, seminal vesicles, and prostate.

In the F_0 generation, there were no dose-related changes in body weight gain and no clinical signs of toxicity resulting from exposure to commercial hexane at any concentration (BRRC, 1991). Hyaline droplet nephropathy was visible histopathologically in the high-dose F_0 males. There were no changes in any of the mating indices, fertility, gestation, live pups/litter, or pup viability at PND 28. A treatment-related effect of commercial hexane was a reduction of mean body weight in the F_1 pups of the high-dose dams, an effect that became apparent at PND 14 and beyond. The mean body weight of the F_1 pups remained lower than controls throughout their prebreeding period. The group-specific means were significantly decreased (by approximately 7%) on PND 21 (38.9 ± 4.0 g in high-dose pups versus 41.9 ± 3.95 g in control pups).

There were no overt signs of clinical toxicity and no other signs of reproductive performance deficits in the F_1 generation (BRRC, 1991). Similarly, no lesions in male reproductive organs were apparent at necropsy and histopathological examination. Hyaline droplet nephropathy was observed in F_1 high-dose males (statistically significant). The numbers of pups born to exposed F_1 rats were not statistically different compared with controls. F_2 pup body weights in the high-concentration group were reduced by 6−9% compared with controls after PND 7. The viability of F_2 pups did not differ between the groups. A LOAEL of 9,000 ppm ($31,579$ mg/m^3) is identified based on reduced body weights in the F_1 and F_2 pups after PNDs 14 and 7, respectively. The NOAEL is 3,000 ppm ($10,526$ mg/m^3). The high concentration (9,000 ppm or $31,579$ mg/m^3) is a NOAEL for effects on reproduction.

Other Studies
Genotoxicity
In the few studies that have addressed the genotoxicity/mutagenicity of a mixture containing approximately 50% *n*-hexane, no gene reversion or chromosomal aberrations in Chinese hamster ovary (CHO) cells (with or without activation) or chromosomal aberrations in Chinese hamster lung (CHL) cells were seen in vitro (Microbiological Associates, 1989, 1990). In addition, in vivo, no chromosomal aberrations were induced in male and female Sprague-Dawley rat bone marrow cells after nose-only inhalation exposure to commercial hexane for 6 hours/day on 5 consecutive days at concentrations of 876, 3,249, and 8,715 ppm (Microbiological Associates, 1990).

DERIVATION OF PROVISIONAL SUBCHRONIC AND CHRONIC ORAL RfD VALUES FOR COMMERCIAL HEXANE

No usable information was obtained to develop oral toxicity values (subchronic or chronic p-RfDs) for commercial hexane.

**DERIVATION OF PROVISIONAL SUBCHRONIC AND CHRONIC INHALATION
p-RfC VALUES FOR COMMERCIAL HEXANE**

A total of three epidemiological studies of human inhalation exposure to commercial or practical grade hexane (Passero et al., 1983; Seppalainen et al., 1979; Raitta et al., 1978) were reviewed by U.S. EPA (2005a). None of these studies reported exposure in terms of concentration of commercial hexane which rendered them useless to derive inhalation toxicity values for commercial hexane.

Inhalation studies of commercial hexane in laboratory animals of potential use in developing subchronic p-RfCs are presented in Table 6.

In the subchronic mouse studies (Biodynamics, 1989; Duffy et al., 1991), increased lacrimation occurred in a dose-related fashion at concentrations ≥900 ppm and was selected by the authors as the basis for the LOAEL. However, increased lacrimation did not occur during chronic exposure of mice to the same or higher concentrations (up to 9,000 ppm; Biodynamics, 1993b; Daughtrey et al., 1999). The subchronic mouse study was excluded as a potential basis for the subchronic p-RfC because the biological significance of the lacrimation is uncertain given this inconsistency. In addition to the subchronic, reproductive, and developmental toxicity studies, chronic studies in rats and mice are available (Biodynamics, 1993a,b; Daughtrey et al., 1999).

To facilitate comparison of the studies, effect levels were first adjusted to equivalent continuous exposure concentrations. The NOAELs and LOAELs from each of the studies were converted to human equivalent concentrations (NOAEL$_{HEC}$ and LOAEL$_{HEC}$) based on U.S. EPA (1994b). Available data on commercial hexane indicate that chronic exposure results in irritation effects (specifically, nasal histopathology in rats) at low concentrations (900 ppm; Biodynamics, 1993a; Daughtrey et al., 1999), while subchronic exposure to concentrations as high as 9,000 ppm does not (Biodynamics, 1989; IRDC, 1992a,b) suggesting that exposure duration is an important factor in the genesis of the nasal lesions. Thus, the effect levels for nasal lesions in the chronic rat study were converted to equivalent continuous exposure. The U.S. EPA (2002) also recommends adjusting for continuous exposure in developmental toxicity studies. Thus, the effect levels in the developmental toxicity studies were also adjusted for continuous exposure.

The human equivalent concentration was then calculated using the dosimetric adjustment appropriate to the observed effect, either airway or systemic toxicant (U.S. EPA, 1994b).

With the exception of studies by Biodynamics (1993a); Daughtrey et al, 1999), discussed below, for all other potentially relevant studies, commercial hexane was treated as a Category 3 gas (systemic toxicant) since significant extrarespiratory effects but no significant airway effects were observed in these studies. The ratio of blood:gas partition coefficients were used to make this dosimetric adjustment. For *n*-hexane, the major constituent of commercial hexane, values reported in the literature for blood:gas partition coefficients are 2.29 in F344 rats (Gargas et al., 1989) and 0.8 in humans (Perbellini et al., 1985). However, the U.S. EPA (1994b) recommends using a value of one as a maximum which is utilized in this case. The blood:gas partition coefficient for mice was not located; the default value of one was also used for the mouse studies.

				Table 6.	Available Inhalation Noncancer Dose-Response Information for Commercial Hexane			
Mixture or Compound	**Species and Sex**	**Exposure Concentration (ppm)**	**Exposure**	**NOAEL (ppm)**	**LOAEL (ppm)**	**Responses**	**Comments**	**Reference**
Subchronic								
Commercial hexane (51.5−53.3% *n*-hexane)	SD Rat (M/F)	0, 900, 3,000, 9,000	6 hr/d, 5 d/wk, for 13 wks	9,000	NA	No effect on mortality, clinical condition, body weight, food intake, gross pathology, FOB, motor activity, or histology of nervous system tissues		Soiefer et al., 1991; Bio-Research Laboratories, 1989
Commercial hexane (51.7−53.5% *n*-hexane)	F344 Rat (M/F)	0, 900, 3,000, 9,000	6 hr/d, 5 d/wk, for 13 wks	3,000 (M)	9,000 (M)	Slight hemorrhage and inflammation of liver and kidney in few males (not significantly different from controls)	Increased severity of hyaline droplet nephropathy in treated males	Duffy et al., 1991; Biodynamics, 1989; EPL, 1989
Mixed hexanes (50% *n*-hexane)	SD Rat (M)	0, 250, 1,000	22 hr/d, 7 d/wk, for 6 mos	250	1,000	Abnormal gait; decreased body weight; mild atrophy of sciatic and/or tibial nerve and skeletal muscle	Slight increase in incidence and severity of chronic nephritis; potentially confounded by coexposure to phthalate-ester compound	IRDC, 1992a,b
Chronic								
Commercial hexane (51.5% *n*-hexane)	F344 Rat (M/F)	0, 900, 3,000, 9,000	6 hr/d, 5 d/wk, for 2 yrs	NA	900	Histologic evidence of mucosal irritation in nasal turbinates and larynx in both sexes	No histopathological abnormalities of sciatic nerve	Biodynamics, 1993a; Daughtrey et al., 1999
Commercial hexane (51.5% *n*-hexane)	B6C3F1 Mouse (M/F)	0, 900, 3,000, 9,000	6 hr/d, 5 d/wk, for 2 yrs	3,000	9,000	Body weight depression in females. Minimal LOAEL	Dose-related increase in incidence of liver and pituitary tumors in females. Nasal turbinates not examined in mice	Biodynamics, 1993b; Daughtrey et al., 1999

Table 6. Available Inhalation Noncancer Dose-Response Information for Commercial Hexane								
Mixture or Compound	**Species and Sex**	**Exposure Concentration (ppm)**	**Exposure**	**NOAEL (ppm)**	**LOAEL (ppm)**	**Responses**	**Comments**	**Reference**
Reproductive/Developmental								
Commercial hexane	SD Rat (M/F)	0, 900, 3,000, 9,000	6 hr/d, 5 d/wk, for 2 generations	3,000 (offspring) 9,000 (reproductive)	9,000 (offspring) NA (reproductive)	Reduced body weight in F_1 weanlings and F_2 pups after PND 7	Hyaline droplet nephropathy in 9,000-ppm F_0 and F_1 males	Daughtrey et al., 1994; BRRC, 1991
Commercial hexane	SD Rat (F)	0, 900, 3,000, 9,000	6 hr/d on GDs 6−15	9,000 (maternal) 9,000 (develop-mental)	NA (maternal) NA (develop-mental)	None	Range-finding study	BRRC, 1989a
Commercial hexane	SD Rat (F)	0, 900, 3,000, 9,000	6 hr/d on GDs 6−15	900 (maternal) 9,000 (develop-mental)	3,000 (maternal) NA (develop-mental)	Reduced body weight gain during GD 9−12		BRRC, 1989b
Commercial hexane	CD-1 Mouse (F)	0, 900, 3,000, 9,000	6 hr/d on GDs 6−15	9,000 (maternal) 3,000 (develop-mental)	NA (maternal) 9,000 (develop-mental)	Reduced fetal weights	Range-finding study	BRRC, 1989a
Commercial hexane	CD-1 Mouse (F)	0, 900, 3,000, 9,000	6 hr/d on GDs 6−15	9,000 (maternal) 3,000 (develop-mental)	NA (maternal) 9,000 (develop-mental)	Increased incidence of two skeletal variations		BRRC, 1989b

For the rat study reported by Biodynamics (1993a; Daughtrey et al., 1999), respiratory effects (nasal irritation) were observed. As recommended by U.S. EPA (1994b), commercial hexane was treated as a Category 1 gas, and the Regional Gas Dose Ratio (RGDR) was calculated in order to determine the HEC for respiratory effects from this study. For nasal effects reported in Biodynamics (1993a; Daughtrey et al., 1999), $RGDR_{ET}$ (extrathoracic) values of 0.24 (for males) and 0.16 (for females) were calculated as indicated below (U.S. EPA, 1994b).

$$RGDR_{ET} = \frac{(V_E/SA_{ET})_{rat}}{(V_E/SA_{ET})_{human}}$$
$$= 0.24 \text{ for males}$$
$$= 0.16 \text{ for females}$$

Where

V_E = Minute volume (L/min)

= 0.254 L/min for male F344 rats, 0.167 L/min for female F344 rats, and 13.8 L/min for humans

SA_{ET} = Surface area of the extrathoracic region (cm^2)

= 15 cm^2 for rats, 200 cm^2 for humans

Table 7 shows the $LOAEL_{HEC}$ and $NOAEL_{HEC}$ values.

Subchronic p-RfC Derivation

As Table 7 indicates, the lowest $LOAEL_{HEC}$ values from the subchronic, reproductive, and developmental toxicity studies (not chronic studies) were obtained in the IRDC (1992a,b) subchronic rat study (3,217 mg/m^3) and the BRRC (1989b) developmental toxicity study in rats (2,632 mg/m^3). Effects observed by IRDC (1992a,b) included clinical and histopathological signs of neuropathy as well as decreased body weight gain. Histopathological findings consistent with the mode of action of *n*-hexane were observed in axons (peripheral nervous tissue); these findings were consistent with the observed clinical signs that included altered gait. The IRDC (1992a,b) study included two exposure groups, but the study was conducted in two phases with different experimental protocols; thus, benchmark dose modeling of the data is not practical. The NOAEL/LOAEL method was applied to derive the subchronic p-RfC. The $NOAEL_{HEC}$ (804 mg/m^3) associated with the lowest $LOAEL_{HEC}$ (3,217 mg/m^3) identified in the 13-week subchronic study in rats (IRDC, 1992a,b) was selected as the point of departure (POD) for derivation of the subchronic p-RfC.

Table 7. Calculation of LOAEL[HEC] and NOAEL[HEC] Values for Subchronic p-RfC Derivation

Study	Species	Effect	Effect Level (ppm)	Effect Level[a] (mg/m^3)	Duration-Adjusted Effect Level[b] (mg/m^3)	Dosimetric Adjustment[c]	Human Equivalent Concentration[d] (mg/m^3)
Subchronic							
Duffy et al., 1991; Biodynamics, 1989; EPL, 1989	Rat	Slight hemorrhage and inflammation of liver and kidney in few males	LOAEL = 9,000 NOAEL = 3,000	LOAEL = 31,579 NOAEL = 10,526	LOAEL[ADJ] = 5,639 NOAEL[ADJ] = 1,880	1.0	LOAEL[HEC] = 5,639 NOAEL[HEC] = 1,880
IRDC, 1992a,b	Rat	Abnormal gait; decreased body weight; mild atrophy of sciatic and/or tibial nerve and skeletal muscle	LOAEL = 1,000 NOAEL = 250	LOAEL = 3,510 NOAEL = 877	LOAEL[ADJ] = 3,217 NOAEL[ADJ] = 804	1.0	LOAEL[HEC] = 3,217 NOAEL[HEC] = 804
Chronic							
Biodynamics, 1993a; Daughtrey et al., 1999	Rat	Histologic evidence of mucosal irritation in nasal turbinates and larynx in both sexes	LOAEL = 900 No NOAEL	LOAEL = 3,158	LOAEL[ADJ] = 564	0.24 (M), 0.16 (F) (extrathoracic RGDR for F344 rats)	LOAEL[HEC] = 135 (M) LOAEL[HEC] = 90 (F)
Biodynamics, 1993b; Daughtrey et al., 1999	Mouse	Body weight depression in females. Minimal LOAEL	LOAEL = 9,000 NOAEL = 3,000	LOAEL = 31,579 NOAEL = 10,526	LOAEL[ADJ] = 5,639 NOAEL[ADJ] = 1,880	1.0	LOAEL[HEC] = 5,639 NOAEL[HEC] = 1,880
Reproductive/Developmental							
Daughtrey et al., 1994; BRRC, 1991	Rat	Reduced body weight in F$_1$ weanlings and F$_2$ pups after LD 7	LOAEL = 9,000 NOAEL = 3,000	LOAEL = 31,579 NOAEL = 10,526	LOAEL[ADJ] = 5,639 NOAEL[ADJ] =1,880	1.0	LOAEL[HEC] = 5,639 NOAEL[HEC] = 1,880
BRRC, 1989a	Mouse	Reduced fetal weights	LOAEL = 9,000 (developmental) NOAEL = 3,000 (developmental)	LOAEL = 31,579 NOAEL = 10,526	LOAEL[ADJ] = 7,895 NOAEL[ADJ] = 2,632	1.0	LOAEL[HEC] = 7895 NOAEL[HEC] = 2,632

					Duration-Adjusted		Human Equivalent
			Effect Level	Effect Level[a]	Effect Level[b]	Dosimetric	Concentration[d]
Study	Species	Effect	(ppm)	(mg/m³)	(mg/m³)	Adjustment[c]	(mg/m³)
BRRC, 1989b	Rat	Reduced body weight gain during GDs 9−12	LOAEL = 3,000 (maternal)	LOAEL = 10,526	LOAEL$_{ADJ}$ = 2,632	1.0	LOAEL$_{HEC}$ = 2,632
			NOAEL = 900 (maternal)	NOAEL = 3,158	NOAEL$_{ADJ}$ = 789		NOAEL$_{HEC}$ = 789
BRRC, 1989b	Mouse	Increased incidence of two skeletal variations	LOAEL = 9,000 (developmental)	LOAEL = 31,579	LOAEL$_{ADJ}$ = 7,895	1.0	LOAEL$_{HEC}$ = 7,895
			NOAEL = 3,000 (developmental)	NOAEL = 10,526	NOAEL$_{ADJ}$ = 2,632		NOAEL$_{HEC}$ = 2,632

Table 7. Calculation of LOAEL$_{HEC}$ and NOAEL$_{HEC}$ Values for Subchronic p-RfC Derivation

[a]Effect level converted from ppm to mg/m³ according to Equation 4-1b (mg/m³ = ppm × MW/24.45) of U.S. EPA (1994b). A weighted average (weighted by the proportions of each constituent) MW of 85.79 g/mol was used for commercial hexane. A weighted average MW of 85.81 was used for IRDC (1992a,b) because the proportions in the hexane mixture differed slightly from those of commercial hexane.
[b]Table 6 shows adjusted to equivalent continuous exposure concentration based on exposure regimen, using this equation:
  NOAEL$_{ADJ}$ = NOAEL × exposure hours/24 hours × exposure days/7 days.
[c]Except where noted, the dosimetric adjustment is the ratio of blood:gas partition coefficients; see text for additional information on dosimetric adjustments.
[d]Calculated as shown in this equation: NOAEL$_{HEC}$ = NOAEL × dosimetric adjustment.

LD = luteinizing day.

To derive the **subchronic p-RfC** for commercial hexane, a composite uncertainty factor (UF) of 30 is applied to the NOAEL$_{HEC}$ as follows:

$$\textbf{Subchronic p-RfC} \quad = \quad \text{NOAEL}_{HEC} \div \text{UF}$$
$$= \quad 804 \text{ mg/m}^3 \div 30$$
$$= \quad 26.8 \textbf{ or } \textbf{27} \times \textbf{10}^{\textbf{0}} \textbf{ mg/m}^{\textbf{3}}$$

The composite UF of 30 was composed of the following:

- A default UF of 10 for intraspecies differences is used to account for potentially susceptible individuals in the absence of information on the variability of response in humans.
- A partial UF of 3 ($10^{0.5}$) is used to account for interspecies extrapolation (toxicodynamic portion only) because a dosimetric adjustment was made.
- An UF of 1 for database uncertainty is applied. The database for commercial hexane includes three subchronic toxicity studies in two species, two chronic studies in two species, four developmental toxicity studies in two species, and a two-generation reproductive toxicity study in rats. In addition, the database for commercial hexane is supported in part by a large body of toxicity data for *n*-hexane, the primary component of commercial hexane.

Confidence in the principal study used to derive the subchronic p-RfC (IRDC, 1992a,b) is medium. The principal study (IRDC, 1992a,b) was designed and performed according to standards for these types of studies at two exposure levels, and appropriate neurotoxicological endpoints were evaluated. However, confidence in the database is reduced due to the lack of an adequate <u>continuous</u> exposure study of neurotoxicity. Specifically, the differences between intermittent and continuous exposure raises the possibility that the adjustment of intermittent exposure concentrations to equivalent continuous exposure concentrations does not fully account for neurotoxic potency during continuous exposure. This may be due to saturation of metabolism during high-concentration intermittent exposure such that the potentially neurotoxic metabolite(s) of *n*-hexane and other components of this mixture do not accumulate to the level that may occur during lower-concentration continuous exposure. Thus, the lack of an exposure study produces some uncertainty. Medium confidence in the subchronic p-RfC follows.

Chronic p-RfC Derivation

Among the studies available for use in deriving a chronic p-RfC for commercial hexane (see Tables 6 and 7), the chronic study in rats (Biodynamics, 1993a; Daughtrey et al., 1999) has the lowest LOAEL$_{HEC}$ (135 mg/m^3 for males and 90 mg/m^3 for females) for histologic evidence of nasal/laryngeal irritation). The next highest LOAEL$_{HEC}$ (2,632 mg/m^3) is more than 20-fold higher

In addition, the NOAEL$_{HEC}$ values for all of the remaining studies exceeded the LOAEL$_{HEC}$ values from the chronic rat study (see Table 7). Because nasal irritation appears to be the most sensitive effect in the available studies, the chronic study in rats was selected as the basis for the chronic p-RfC. Table 4 presents data on the incidence of the critical effect (nasal and laryngeal irritation) as reported by the authors (Biodynamics, 1993a; Daughtrey et al., 1999). The incidence of laryngeal lesions, while increased at the high concentration, was not statistically distinguishable from controls. In contrast, as the Table 4 shows, the incidences of

three nasal lesions (goblet cell hyperplasia, epithelial hyperplasia, and intracytoplasmic eosinophilic material) were significantly increased in both male and female rats at all exposure concentrations. The authors did not report the cumulative incidence of nasal lesions. To identify a POD for chronic p-RfC derivation, benchmark dose modeling was conducted on the incidences of goblet cell and epithelial cell hyperplasia in both male and female rats. As shown in Table 4, the incidence of intracytoplasmic eosinophilic material was high in the control groups (44% in males and 82% in females) and increased to a near-maximal response at the lowest exposure concentration; consequently, this endpoint was not considered suitable for benchmark dose modeling.

Because the exposure regimen used by Biodynamics (1993a; Daughtrey et al., 1999) was not continuous, BMD modeling was performed using doses adjusted for continuous exposure followed by conversion to HECs. Available information indicates that duration of exposure is an important factor in the development of nasal lesions; these effects were observed with chronic exposure to 900 ppm (Biodynamics, 1993a; Daughtrey et al., 1999), but not with subchronic exposure to concentrations up to 9,000 ppm (Biodynamics, 1989). Appendix B provides details of the modeling results and exposure duration adjustments. All available dichotomous models in the U.S. EPA BMDS (version 2.1) were fit to the incidence data on goblet cell and epithelial cell hyperplasia in male and female rats (see Table 4). As assessed by the χ^2 goodness-of-fit test, the log-logistic model provided the best fit to the data on each of these two endpoints in male rats (χ^2 $p \geq 0.1$). The BMC_{10HEC} and $BMCL_{10HEC}$ associated with goblet cell hyperplasia in males were 81.86 and 31.43 mg/m^3, respectively. The BMC_{10HEC} and $BMCL_{10HEC}$ associated with epithelial cell hyperplasia in males were 28.20 and 17.59 mg/m^3, respectively. Efforts to model the data on goblet cell and epithelial cell hyperplasia in <u>females</u> were unsuccessful, even when the high dose group was dropped. The lower $BMCL_{10HEC}$ 17.59 mg/m^3), estimated for epithelial cell hyperplasia in male rats, was selected as the POD for the chronic p-RfC.

The **chronic p-RfC** for commercial hexane was calculated as the $BMCL_{10HEC}$ of 17.59 mg/m^3 (See Appendix B) divided by a composite UF of 30 as follows:

$$\begin{aligned} \textbf{Chronic p-RfC} \quad &= \quad BMCL_{10HEC} \div UF \\ &= \quad 17.59 \text{ mg/m}^3 \div 30 \\ &= \quad \textbf{0.58 or } 6 \times 10^{-1} \textbf{ mg/m}^3 \end{aligned}$$

The composite UF of 30 was composed of the following:

- An UF of 10 for intraspecies differences is used to account for potentially susceptible individuals in the absence of information on the variability of response in humans.
- A partial UF of 3 ($10^{0.5}$) is used to account for interspecies extrapolation (toxicodynamic portion only) because a dosimetric adjustment was made.
- An UF of 1 for database uncertainty is applied. The database for commercial hexane includes three subchronic toxicity studies in two species, two chronic studies in two species, four developmental toxicity studies in two species, and a two-generation reproductive toxicity study in rats. In addition, the database for commercial hexane is supported in part by a large body of toxicity data for *n*-hexane, the primary component of commercial hexane.

Confidence in the principal study used to derive the chronic p-RfC (Biodynamics 1993a; Daughtrey et al., 1999) is high. The study was adequate in terms of standards of these types of animal studies and an appropriate range of exposure levels. Toxicological evaluations are consistent with current practices and included comprehensive histopathology examinations, including respiratory tract tissues and the sciatic nerve. Confidence in the database is medium. As noted previously, the toxicological database for inhalation of commercial hexane includes chronic toxicity studies in two species, subchronic toxicity studies in rats, developmental toxicity studies in two species, and a multigeneration reproductive toxicity study in rats. Confidence in the database is reduced due to uncertainty in the relevance of neuropathy observed in rats in the IRDC (1992a,b) study, when other studies did not observe neuropathy at higher concentrations. This inconsistency contributes to the database uncertainty. Medium confidence in the chronic p-RfC follows.

PROVISIONAL CARCINOGENICITY ASSESSMENT
FOR COMMERCIAL HEXANE

Weight-of-Evidence Descriptor

Under the 2005 *Guidelines for Carcinogen Risk Assessment* (U.S. EPA, 2005b), there is **"*Suggestive Evidence for [the] Carcinogenic Potential*"** of commercial hexane in humans. There are no data on carcinogenicity of commercial hexane in humans. A 2-year carcinogenicity bioassay in mice and rats exposed to commercial hexane showed an increased incidence of liver tumors (combined hepatocellular adenomas and carcinomas) in female mice (Daughtrey et al., 1999; Biodynamics, 1993a,b). No increase in liver tumor incidence was observed in treated male mice or in either sex of F344 rats exposed to commercial hexane under the same conditions. The study authors also identified a statistically significant increase in the incidence of pituitary tumors in female mice. Available data on the genotoxicity of commercial hexane are limited; no gene reversion or chromosomal aberrations in mammalian cells and no chromosomal aberrations in the bone marrow of rats exposed in vivo were observed in the only tests conducted.

Mode of Action Information

The U.S. EPA (2005b) *Guidelines for Carcinogen Risk Assessment* defines mode of action as a sequence of key events and processes, starting with the interaction of an agent with a cell, proceeding through operational and anatomical changes and resulting in cancer formation. Toxicokinetic processes leading to the formation or distribution of the active agent (i.e., parent material or metabolite) to the target tissue are not part of the mode of action. Examples of possible modes of carcinogenic action include mutagenic, mitogenic, anti-apoptotic (inhibition of programmed cell death), cytotoxic with reparative cell proliferation, and immunologic suppression.

There are few mechanistic data to support a mode of action determination for commercial hexane. Available genotoxicity data are inadequate to determine whether commercial hexane interacts directly with DNA. No significant liver histopathology has been observed in mice after subchronic or chronic exposure to commercial hexane (Biodynamics, 1989; Biodynamics, 1993b; Daughtrey et al., 1999), indicating that it probably does not cause liver cell toxicity at doses that were tumorigenic in the females studied by Daughtrey et al. (1999). Evidence for

liver weight increases in rats and mice exposed subchronically to commercial hexane (Biodynamics, 1989) raises the possibility that commercial hexane may induce cell proliferation in the livers of female mice; however, no mechanistic data are available to support this hypothesis. Organ weights were not measured in the chronic study (Biodynamics, 1993b; Daughtrey et al., 1999).

Quantitative Estimates of Carcinogenic Risk
Oral Exposure
　　　　No oral quantitative estimate is derived because there are no oral carcinogenicity studies of commercial hexane.

Quantitative Estimates of Carcinogenic Risk
Inhalation Exposure
　　　　Because the cancer descriptor designated here is "Suggestive Evidence for Carcinogenic Potential," a quantitative IUR is provided as a screening value in Appendix C.

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APPENDIX A. DESCRIPTION OF LITERATURE SEARCH PROCESS

The IRIS toxicological review (U.S. EPA, 2005a) contained a thorough review of toxicity data on commercial hexane, so searches were limited to studies published since 2002. Studies included in the U.S. EPA (2005a) review that pertained to commercial hexane were obtained. The search for more recent studies of commercial hexane was combined with searches for *n*-hexane and included terms to identify human exposure studies (epidemiologic, occupational), animal studies, toxicodynamic and toxicokinetic studies, mode-of-action studies, and in vitro and in vivo studies for all relevant endpoints (cancer and noncancer) and durations. The search included health effects and toxicity information available from the U.S. EPA (IRIS), ATSDR, and other relevant federal, state, or international governmental or quasi-governmental agencies, including, but not limited to ACGIH, NIOSH, OSHA, NTP, IARC, WHO, and CalEPA. In addition, electronic databases, including: CURRENT CONTENTS, MEDLINE, TOXLINE, BIOSIS/TOXCENTER, TSCATS/TSCATS2, CCRIS, DART/ETIC, GENETOX, HSDB, and RTECS were searched. Table A-1 shows results of the electronic searches of these databases. An electronic listing of all results of the gross literature review (including titles, references, and abstracts) and a tabular summary of the search results were provided to U.S. EPA.

A toxicologist screened the literature searches based on review of abstracts and titles for studies pertaining to the health effects from exposure to commercial hexane in humans and animals. Decisions about whether to further consider a particular citation were based on the scientific judgment of the toxicologist, based on reading the abstract provided in the literature search output. Studies that were not considered pertinent were not retrieved. Citations may also have been excluded after retrieval and review of the article by the toxicologist. A study may have been excluded if its scope was outside the scope of the use under consideration, if it was not relevant or appropriate, if its study design was inadequate, or if the study showed inadequacy of quality control or flaws in the interpretation of results.

Following the literature search and screening process, a table of studies considered likely to have data suitable for derivation of provisional toxicity values was prepared for U.S. EPA review. The table identified each reference, title, a brief description of the study and findings, and a conclusion as to whether the study was likely to be useful for provisional toxicity value derivation. The initial determination of relevance was based on readily available information (i.e., titles and abstracts, if available). U.S. EPA approval of the selected studies based on review of the table preceded development of the PPRTV document.

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Table A-1. Summary of Electronic Database Searches for *n*-Hexane and Commercial Hexane											
Chemical/ CASRN	**PUBMED**	**TOXLINE Special (on TOXNET)**	**BIOSIS (STN) update**	**TSCATS 2**	**CCRIS**	**DART/ ETIC (not Pub Med)**	**GENE-TOX**	**HSDB**	**RTECS**	**Current Contents**
Dates Searched	Entry date from 2003 on	200212:200709 [em]	UP >19991231 AND PY > 2002	**TSCATS 2** only >01/01/2000 receipt date	Not date limited	2003 on	Not date limited	Not date limited	Not date limited	Last 6 months
Hexane **110-54-3**	**62** (45 + 17 in process) with hexane in title	**51** records (33 + 18 NTIS)	**5 full cites** downloaded limited to animal with hexane in title **80 titles** downloaded limited to human and removed cites with EXTRACT*	**0 records**	1	**0** since 2003	**0**	1	1	8 titles downloaded

APPENDIX B. DETAILS OF BENCHMARK DOSE MODELING FOR THE PROVISIONAL CHRONIC RfC

Model Fitting Procedure for Dichotomous Data

All available dichotomous models in the EPA BMDS (version 2.1) are fit to the data using the extra risk option. The multistage model is run for all polynomial degrees up to n-1 (where n is the number of dose groups including control); the lowest degree polynomial providing adequate fit is used for comparison with the other models, per U.S. EPA (2000) guidance. Adequacy of model fit is judged based on the χ^2 goodness-of-fit p-value ($p > 0.1$), magnitude of scaled residuals in the vicinity of the benchmark response (BMR), and visual inspection of the model fit. Among all the models providing adequate fit, the BMDL from the model with the lowest Akaike's Information Criterion (AIC) is selected as a potential POD from which to derive the p-RfC. When several models have the same AIC, the model resulting in the lowest BMDL is selected. In accordance with U.S. EPA (2000) guidance, benchmark doses (BMDs) and lower bounds on the BMD (BMDLs) associated with a BMR of 10% extra risk are calculated for all models.

Results of Model Fitting

All available dichotomous models in the EPA BMDS (version 2.1) were fit to the incidence data on goblet cell and epithelial cell hyperplasia in male and female rats (Biodynamics, 1993a; Daughtrey et al., 1999; see Table 4). Because the exposure regimen used by Biodynamics (1993a; Daughtrey et al., 1999) was not continuous, BMD modeling was performed using doses adjusted for continuous exposure followed by conversion to HECs. As assessed by the χ^2 goodness-of-fit test, the log-logistic model provided the best fit to the data for either endpoint in males (χ^2 $p \geq 0.1$; see Tables B-1 and B-2 and Figures B-1 and B-2). The BMC_{10HEC} and $BMCL_{10HEC}$ associated with goblet cell hyperplasia in males are 81.86 and 31.43 mg/m^3, respectively. The BMC_{10HEC} and $BMCL_{10HEC}$ associated with epithelial cell hyperplasia in males are 28.20 and 17.59 mg/m^3, respectively (Table B-5).

Efforts to model the data on goblet cell and epithelial cell hyperplasia in females were unsuccessful, even when the high dose group was dropped from the analysis. The software failed to execute completely with any model.

Table B-1. Input Data Used for Chronic p-RfC Derivation for Commercial Hexane (51.1% *n*-Hexane) for the Incidence of Goblet Cell Hypertrophy/Hyperplasia in Male Rats[a]					
PPM	$(mg/m^3)^a$	Daily Average Concentration $(mg/m^3)^c$	HEC $(mg/m^3)^d$	Total Neoplasms Response	Number of Subjects
0	0	0	0	29	48
900	3168	565.7	136	37	50
3000	10560	1885.7	453	43	50
9000	31680	5657.1	1358	41	50

[a]Daughtrey et al. (1999); Biodynamics (1993a).
[b]PPM conversion: $C(mg/m^3) = PPM \times MW/24.45 = (PPM \times 86.177 \text{ g/mol})/24.45 = 3.52 \times PPM$.
[c]Average daily concentration = $C(mg/m^3) \times$ (hours exposure/24hours) $\times$ (days exposure/7 days a week) = $C(mg/m^3) \times (6/24 \times 5/7)$.
[d]HEC = (PPM conversion) $\times$ (average daily concentration) $\times$ RGDR. The critical effect: respiratory effects (nasal irritation), Category 1 gas, extrathoracic (ET) and the **$RGDR_{ET} = (V_E/SA_{ET})_{rat}/(V_E/SA_{ET})_{human} = 0.24$ for males.**

Table B-2. BMD Modeling Results Based on Goblet Cell Hypertrophy/Hyperplasia in Male Rats[a]				
Model	χ^2 *p*-Value	AIC	BMC_{10HEC} (mg/m^3)	$BMCL_{10HEC}$ (mg/m^3)
Gamma (power ≥1)	0.0769	218.736	179.571	94.6424
Logistic	0.0677	219.007	211.022	119.216
Log-logistic	**0.1348**	**217.388**	**81.8615**	**31.43**
Log-probit (slope ≥1)	0.0380	220.163	345.465	167.467
Multistage (degree = 3)	0.0769	220.268	167.663	90.9906
Probit	0.0641	219.125	226.245	132.60
Weibull	0.0769	218.736	179.573	94.6424
Quantal linear	0.0769	218.736	179.573	94.6424

[a]Daughtrey et al. (1999); Biodynamics (1993a).
[b]Degree of polynomial initially set to (*n*-1) where *n* = number of dose groups including control; no model provided adequate fit. Betas restricted to ≥0.
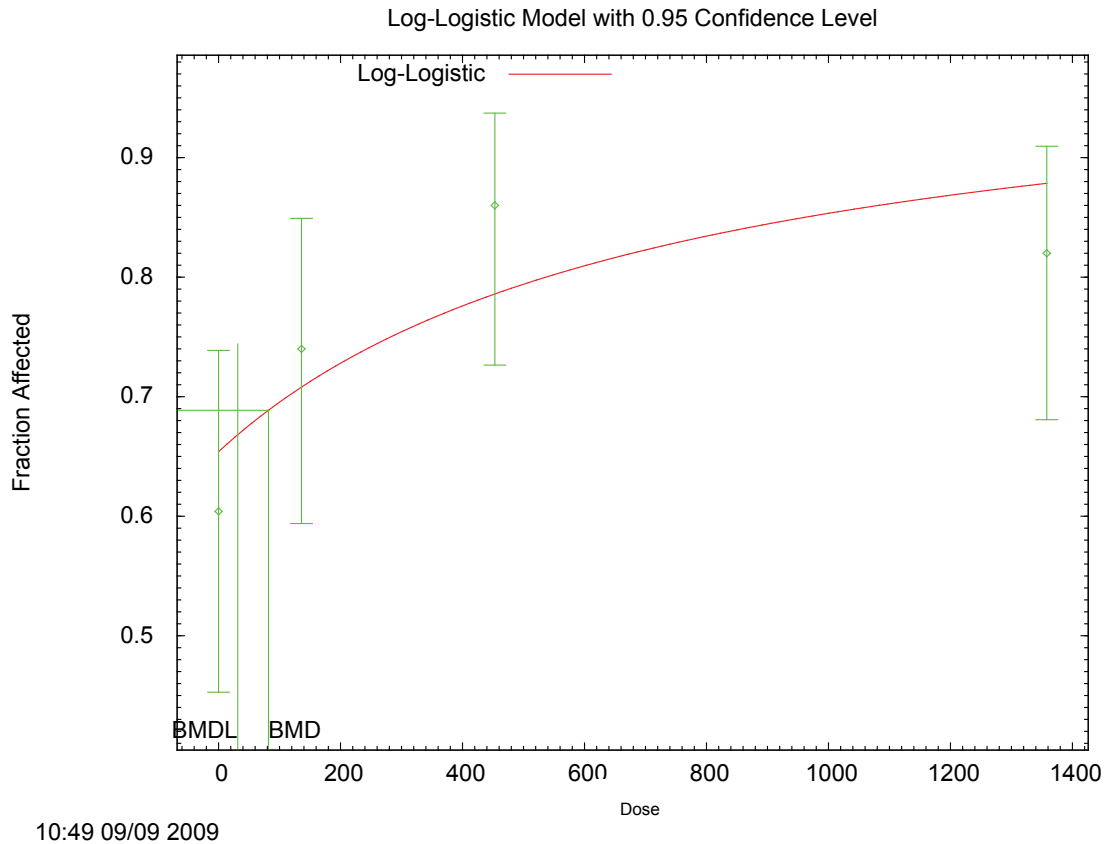
**Figure B-1. Dose-Response Modeling for Incidence of Goblet Cell
Hypertrophy/Hyperplasia in Male Rats**

```
====================================================================
        Logistic Model. (Version: 2.12; Date: 05/16/2008)
        Input Data File:
C:\USEPA\BMDS21\Data\lnlGoblethypetrophyhyperplastiammaleloglostic.(d)
        Gnuplot Plotting File:
C:\USEPA\BMDS21\Data\lnlGoblethypetrophyhyperplastiammaleloglostic.plt
                                        Wed Sep 09 10:49:49 2009
====================================================================

 BMDS Model Run
~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~

   The form of the probability function is:

   P[response] = background+(1-background)/[1+EXP(-intercept-slope*Log(dose))]


   Dependent variable = Percent
   Independent variable = Dose
   Slope parameter is restricted as slope >= 1

   Total number of observations = 4
   Total number of records with missing values = 0
   Maximum number of iterations = 250
   Relative Function Convergence has been set to: 1e-008
   Parameter Convergence has been set to: 1e-008
```

36

User has chosen the log transformed model

```
                   Default Initial Parameter Values
                      background =          0.604
                       intercept =       -6.31119
                           slope =              1
```

```
            Asymptotic Correlation Matrix of Parameter Estimates

            ( *** The model parameter(s)  -slope
                  have been estimated at a boundary point, or have been specified by
the user,
                  and do not appear in the correlation matrix )

                    background      intercept

background                  1          -0.75

 intercept              -0.75              1
```

```
                            Parameter Estimates

                                              95.0% Wald Confidence
Interval
        Variable         Estimate        Std. Err.     Lower Conf. Limit     Upper Conf.
Limit
        background        0.654013              *               *                    *
         intercept        -6.60225              *               *                    *
             slope               1              *               *                    *

* - Indicates that this value is not calculated.
```

```
                          Analysis of Deviance Table

        Model       Log(likelihood)  # Param's  Deviance  Test d.f.   P-value
       Full model       -104.696          4
      Fitted model      -106.694          2       3.9964      2         0.1356
      Reduced model     -109.673          1       9.95448     3         0.01896

           AIC:            217.388
```

```
                              Goodness  of  Fit
                                                                 Scaled
      Dose     Est._Prob.   Expected   Observed      Size       Residual
    -------------------------------------------------------------------
      0.0000     0.6540       31.393     28.992        48        -0.728
    136.0000     0.7079       35.396     37.000        50         0.499
    453.0000     0.7857       39.287     43.000        50         1.280
   1358.0000     0.8783       43.916     41.000        50        -1.261

   Chi^2 = 4.01      d.f. = 2        P-value = 0.1348
```

Benchmark Dose Computation

```
Specified effect =          0.1

Risk Type        =      Extra risk

Confidence level =          0.95

         BMD =          81.8615

        BMDL =          31.43
```

Table B-3. Input Data Used for Chroninc p-RfC Derivation for Commercial Hexane (51.1% _n_-Hexane) for the Incidence of Epithelial Hyperplasia in Males Rats[a]					
PPM	**(mg/m^3)[a]**	**Daily Average Concentration (mg/m^3)[c]**	**HEC (mg/m^3)[d]**	**Total Neoplasms Response**	**Number of Subjects**
0	0	0	0	2	48
900	3168	565.7	136	19	50
3000	10560	1885.7	453	36	50
9000	31680	5657.1	1358	43	50

[a]Daughtrey et al. (1999); Biodynamics (1993a).
[b]PPM conversion: C(mg/m^3) = PPM × MW/24.45 = (PPM × 86.177 g/mol)/24.45 = 3.52 × PPM.
[c]Average daily concentration = C(mg/m^3) × (hours exposure/24hours) × (days exposure/7 days a week) = C(mg/m^3) × (6/24 × 5/7).
[d]HEC = (PPM conversion) × (average daily concentration) × RGDR. The critical effect: respiratory effects (nasal irritation), Category 1 gas, extrathoracic (ET) and the $RGDR_{ET}$ = $(V_E/SA_{ET})_{rat}/(V_E/SA_{ET})_{human}$ = 0.24 for males.

Table B-4. BMD Modeling Results Based on Epithelial Hyperplasia in Male Rats[a]				
Model	**AIC**	**χ^2 _p_-Value**	**BMC$_{10HEC}$ (mg/m^3)**	**BMCL$_{10HEC}$ (mg/m^3)**
Gamma (power ≥1)	194.941	0.0072	52.6794	42.3999
Logistic	212.945	0.0000	137.593	111.172
Log-logistic[c]	**188.687**	**0.5456**	**28.2021**	**17.5851**
Log-probit (slope ≥1)	188.843	0.4713	29.7984	8.05051
Multistage (degree = 3)	189.538	0.0041	50.6871	41.2042
Probit	214.179	0.0000	144.716	121.314
Weibull	194.941	0.0072	52.6794	42.3999
Quantal linear	194.941	0.0072	52.6794	42.3999

[a]Daughtrey et al. (1999); Biodynamics (1993a).
[b]Degree of polynomial initially set to (_n_-1) where _n_ = number of dose groups including control; no model provided adequate fit. Betas restricted to ≥0.
The lowest AIC, with $p ≥ 0.1$ and lowest residual.

Log-Logistic Model with 0.95 Confidence Level

11:19 09/09 2009
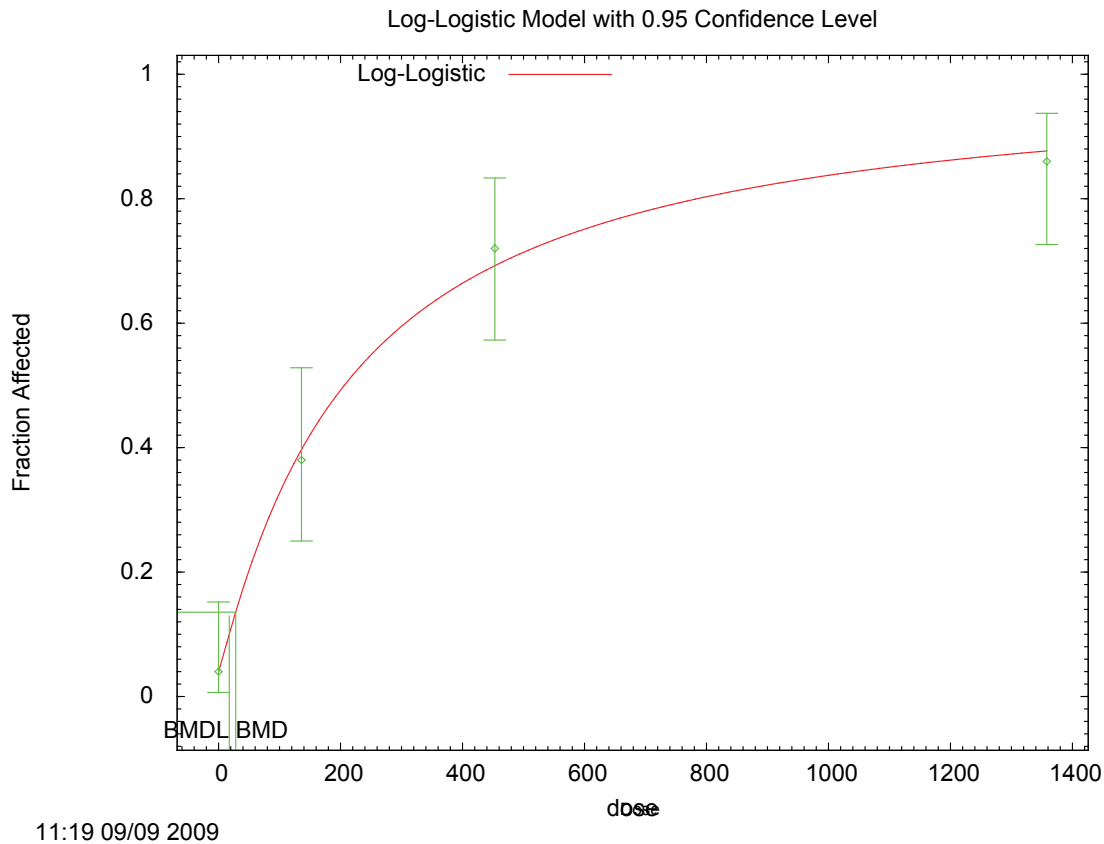
Figure B-2. Dose-Response Modeling for Incidence of Epithelial Hyperplasia in Male Rats

```
=====================================================================
        Logistic Model. (Version: 2.12; Date: 05/16/2008)
        Input Data File:
C:\USEPA\BMDS21\Data\lnlEpithelialhyhyperplastiamEpLoglogm.(d)
        Gnuplot Plotting File:
C:\USEPA\BMDS21\Data\lnlEpithelialhyhyperplastiamEpLoglogm.plt
                                    Wed Sep 09 11:19:54 2009
 =====================================================================

 BMDS Model Run
~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~

   The form of the probability function is:

   P[response] = background+(1-background)/[1+EXP(-intercept-slope*Log(dose))]


   Dependent variable = Percent
   Independent variable = Dose
   Slope parameter is restricted as slope >= 1

   Total number of observations = 4
   Total number of records with missing values = 0
   Maximum number of iterations = 250
   Relative Function Convergence has been set to: 1e-008
   Parameter Convergence has been set to: 1e-008
```

User has chosen the log transformed model

```
          Default Initial Parameter Values
                background =        0.04
                intercept =     -5.59407
                    slope =      1.03257
```

Asymptotic Correlation Matrix of Parameter Estimates

```
              background     intercept        slope

background            1         -0.13        0.087

 intercept        -0.13             1        -0.99

     slope        0.087         -0.99            1
```

Parameter Estimates

```
                                          95.0% Wald Confidence
Interval
       Variable       Estimate      Std. Err.    Lower Conf. Limit    Upper Conf.
Limit
      background      0.0395001            *               *                 *
       intercept       -5.73192            *               *                 *
           slope        1.05848            *               *                 *
```

* - Indicates that this value is not calculated.

Analysis of Deviance Table

```
     Model      Log(likelihood)   # Param's   Deviance   Test d.f.   P-value
   Full model       -91.1604          4
  Fitted model      -91.3433          3      0.365824        1        0.5453
 Reduced model      -137.235          1      92.1485         3        <.0001

        AIC:           188.687
```

Goodness of Fit

```
                                                        Scaled
    Dose    Est._Prob.    Expected    Observed    Size    Residual
 -------------------------------------------------------------------
   0.0000     0.0395        1.896       1.920       48      0.018
 136.0000     0.3949       19.747      19.000       50     -0.216
 453.0000     0.6901       34.505      36.000       50      0.457
1358.0000     0.8754       43.772      43.000       50     -0.331
```

Chi^2 = 0.37   d.f. = 1   P-value = 0.5456

Benchmark Dose Computation

Specified effect =   0.1

Risk Type   =   Extra risk

```
Confidence level =          0.95

        BMD =          28.2021

       BMDL =          17.5851
```

Selection of Model and POD

Table B-5. BMD Models with Acceptable Fit					
Effect	**Model**	χ^2 *p*-Value	**AIC**	**BMC$_{10}$ (mg/m^3)**	**BMCL$_{10}$ (mg/m^3)**
Goblet cell	Log logistic	0.138	217.388	81.8615	31.43
Epithelial cell	**Log logistic**	**0.5456**	**188.687**	**28.2021**	**17.5851**
Epithelial cell	Log probit	0.4713	188.843	29.7984	8.0 5051

For the epithelial cell data, the log-logistic model was selected based on the lowest AIC (17.5851 mg/m^3). In comparison to the goblet cell data of 31.43 mg/m^3, this lower value **was selected for the POD (BMCL$_{HEC10}$ = 17.5851 mg/m^3)**

APPENDIX C. DETAILS OF BENCHMARK DOSE MODELING FOR THE SCREENING PROVISIONAL INHALATION UNIT RISK (IUR)

For the reasons noted in the main document, it is inappropriate to derive a provisional IUR. However, information is available which, although insufficient to support derivation of a provisional toxicity value, under current guidelines, may be of limited use to risk assessors. In such cases, the Superfund Health Risk Technical Support Center summarizes available information in an Appendix and develops a "Screening Value." Appendices receive the same level of internal and external scientific peer review as the main document to ensure their appropriateness within the limitations detailed in the document. Users of screening toxicity values in an appendix to a PPRTV assessment should understand that there is considerably more uncertainty associated with the derivation of an appendix screening toxicity value than for a value presented in the body of the assessment. Questions or concerns about the appropriate use of Screening Values should be directed to the Superfund Health Risk Technical Support center.

Model Fitting Procedure for Cancer Data
The Multistage cancer model for dichotomous data (EPA BMDS (version 2.1) was fit to the incidence data using the extra risk option according to U.S. EPA (2000). Goodness-of-fit is assessed by the χ^2 goodness of fit test (required ≥ 0.1).

Selection of Data for BMD Input:
According to the 2005 Cancer Guidelines, with the data from Biodynamics, 1993b; Daughtrey et al., 1999 in **Table 5**, the appropriate input of tumor data for BMD analysis is:
1) Combined adenoma and adenocarcinoma in pituitary of <u>male mice</u>. The data, even with the highest dose removed, would not produce an acceptable fit.
2) Combined adenoma and adenocarcinoma in pituitary of <u>female mice</u> with all doses included. This data, did not satisfy the χ^2 goodness of fit test (required ≥ 0.1) test. See Table C-1 and Figure C-1
3) Combined adenoma and adenocarcinoma in pituitary of <u>female mice</u> with the highest dose removed. See Table C-2 and Figure C-2
4) Combined hepatocellular adenomas and carcinomas in <u>male mice</u>. The data failed to produce an acceptable fit.
5) Combined hepatocellular adenomas and carcinomas in female mice (Biodynamics, 1993b; Daughtrey et al., 1999) See Table C-3 and Figure C-3.

Data used for development of the screening p-IUR are represented in Table 5 (Biodynamics, 1993b; Daughtrey et al., 1999). According to the 2005 Cancer Guidelines (U.S. EPA, 2005), BMD modeling was performed using the BMD Cancer Multistage Model on combined data for both pituitary and hepatic tumors for both males and females. Only the female data, without the high dose, provided an adequate fit for the pituitary tumors. For the liver tumors, only the full data set for females only provided an adequate fit.

Table C-1. Input Data for Combined <u>Pituitary Adenomas and Adenocarcinomas</u> in Female B6C3F1 Mice[a]

PPM	(mg/m³)[b]	Daily Average Concentration (mg/m³)[c]	Multiplier[d]	HEC (mg/m³)[e]	Total Neoplasms Response	Number of Subjects
0	0	0	1	0	0	45
900	3168	565.7	1	565.7	6	48
3000	10560	1885.7	1	1885.7	8	48
9000	31680	5657.1	1	5657.1	5	49

[a]Daughtrey et al. (1999); Biodynamics (1993b).
[b]PPM conversion: $C(mg/m^3) = PPM \times MW/24.45 = (PPM \times 86.177 \text{ g/mol})/24.45 = 3.52 \times PPM$.
[c]Average daily concentration = $C(mg/m^3) \times$ (hours exposure/24hours) $\times$ (days exposure/7 days a week) = $C(mg/m^3) \times (6/24 \times 5/7)$.
[d]Blood gas partition coefficient = $[(H_{(b/g)})_A] / [(H_{(b/g)})_H]$. A default value of 1.0 was used because both partition coefficients were not available.
[e]HEC: Human equivalent concentration (HEC) for Extra-respiratory effects (Cat 3 Gas) = Daily average concentration $\times$ Blood gas partition coefficient.

BMD analysis of combined <u>pituitary</u> adenomas and adenocarcinomas in female mice.

1) **Full data set:**

The complete data set (4 values) for combined adenoma and adenocarcinoma in pituitary of female mice did not adequately fit the data (χ^2 *p*-value <0.1).

Table C-1. BMD Modeling Results for Combined Pituitary Adenomas and Adenocarcinomas in Female Mice B6C3F1 Mice for 2 Years (Full Set of Points)[a]

Model	χ^2 *p*-Value	AIC	BMC_{10} (mg/m³)	$BMCL_{10HEC}$ (mg/m³)
Multistage cancer	0.0136	122.643	16038.6	3162.62

[a]Daughtrey et al. (1999); Biodynamics (1993b).

Multistage Cancer Model with 0.95 Confidence Level
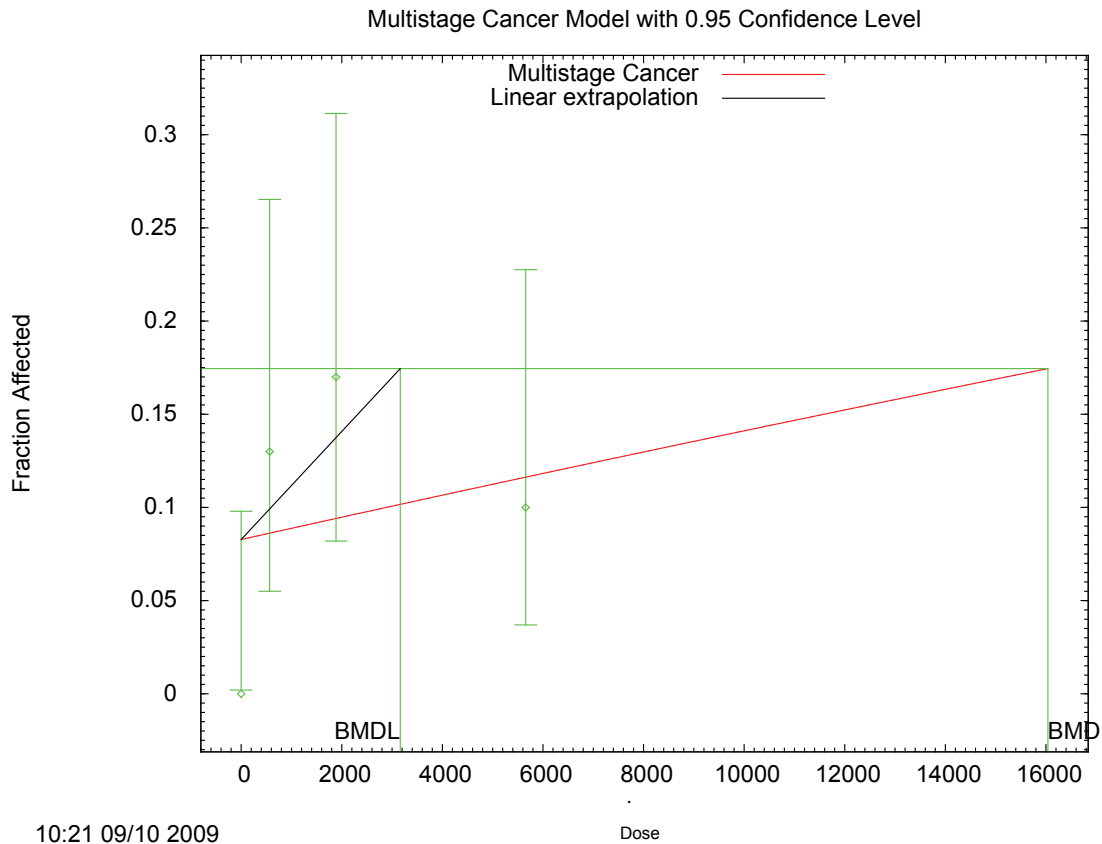
Figure C-1. Dose-Response Modeling for Combined Pituitary Adenomas and Adenocarcinoma of Female B6C3F1 Mice for 2 Years (Full Set of Points)

```
========================================================================
         Multistage Cancer Model. (Version: 1.7;  Date: 05/16/2008)
         Input Data File:
C:\USEPA\BMDS21\Data\mschexaneunitriskwithlastpointhexaneunitriskwithlastpoint.(d)
         Gnuplot Plotting File:
C:\USEPA\BMDS21\Data\mschexaneunitriskwithlastpointhexaneunitriskwithlastpoint.plt
                                        Thu Sep 10 09:40:32 2009
========================================================================

 BMDS Model Run
~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~

   The form of the probability function is:

   P[response] = background + (1-background)*[1-EXP(
                 -beta1*dose^1-beta2*dose^2)]

   The parameter betas are restricted to be positive


   Dependent variable = Percent
   Independent variable = Dose

 Total number of observations = 4
 Total number of records with missing values = 0
 Total number of parameters in model = 3
```

```
Total number of specified parameters = 0
Degree of polynomial = 2



Maximum number of iterations = 250
Relative Function Convergence has been set to: 1e-008
Parameter Convergence has been set to: 1e-008



                  Default Initial Parameter Values
                      Background =    0.0877332
                         Beta(1) = 7.85063e-006
                         Beta(2) =            0


          Asymptotic Correlation Matrix of Parameter Estimates

          ( *** The model parameter(s)  -Beta(2)
                have been estimated at a boundary point, or have been specified by
the user,
                and do not appear in the correlation matrix )

                Background        Beta(1)

Background             1          -0.76

   Beta(1)          -0.76              1



                            Parameter Estimates

                                                 95.0% Wald Confidence
Interval
        Variable        Estimate        Std. Err.     Lower Conf. Limit    Upper Conf.
Limit
       Background        0.0828151           *                 *                  *
         Beta(1)      6.5692e-006            *                 *                  *
         Beta(2)                0            *                 *                  *

* - Indicates that this value is not calculated.



                         Analysis of Deviance Table

       Model        Log(likelihood)  # Param's  Deviance  Test d.f.   P-value
     Full model         -56.3582          4
    Fitted model        -59.3215          2        5.92663      2        0.05165
   Reduced model        -62.4223          1       12.1283       3        0.006956

        AIC:          122.643



                           Goodness  of  Fit
                                                        Scaled
      Dose      Est._Prob.    Expected    Observed    Size     Residual
    -------------------------------------------------------------------
      0.0000     0.0828        3.727       0.000        45      -2.016
    566.0000     0.0862        4.139       6.240        48       1.081
   1886.0000     0.0941        4.517       8.160        48       1.801
   5657.0000     0.1163        5.697       4.900        49      -0.355
```

```
 Chi^2 = 8.60       d.f. = 2          P-value = 0.0136


   Benchmark Dose Computation

Specified effect =           0.1

Risk Type        =      Extra risk

Confidence level =          0.95

          BMD =        16038.6

         BMDL =        3162.62


BMDU did not converge for BMR = 0.100000
BMDU calculation failed
          BMDU = Inf
```

2) Limited Data Set:

According to BMD Guidance (2000) the model fit was next tested with the highest concentration point omitted. Results are shown in Table C-2, and the curve in Figure C-2.

Table C-2. BMD Modeling Results for Incidence of <u>Combined Pituitary Adenomas and Adenocarcinomas</u> in Female B6C3F1 Mice for 2 Years (with High Dose Omitted)[a]				
Model	χ^2 **p-Value**	**AIC**	**BMC_{10HEC} (mg/m^3)**	**$BMCL_{10HEC}$ (mg/m^3)**
Multistage cancer	0.2048	83.8129	809.461	536.985

[a]Daughtrey et al. (1999); Biodynamics (1993b).

**The Inhalation unit risk for combined pituitary tumor is provided by the BMDS
p-IUR** = Multistage Cancer slope factor from BMDS = **0.000186255 or <u>1.9 E-4 mg/m^3</u>**

Note: Default BMR is 0.1, or p-IUR = BMR ÷ $BMCL_{10HEC}$ = 0.1 ÷ 536.985 = 2×10^{-4} mg/m^3
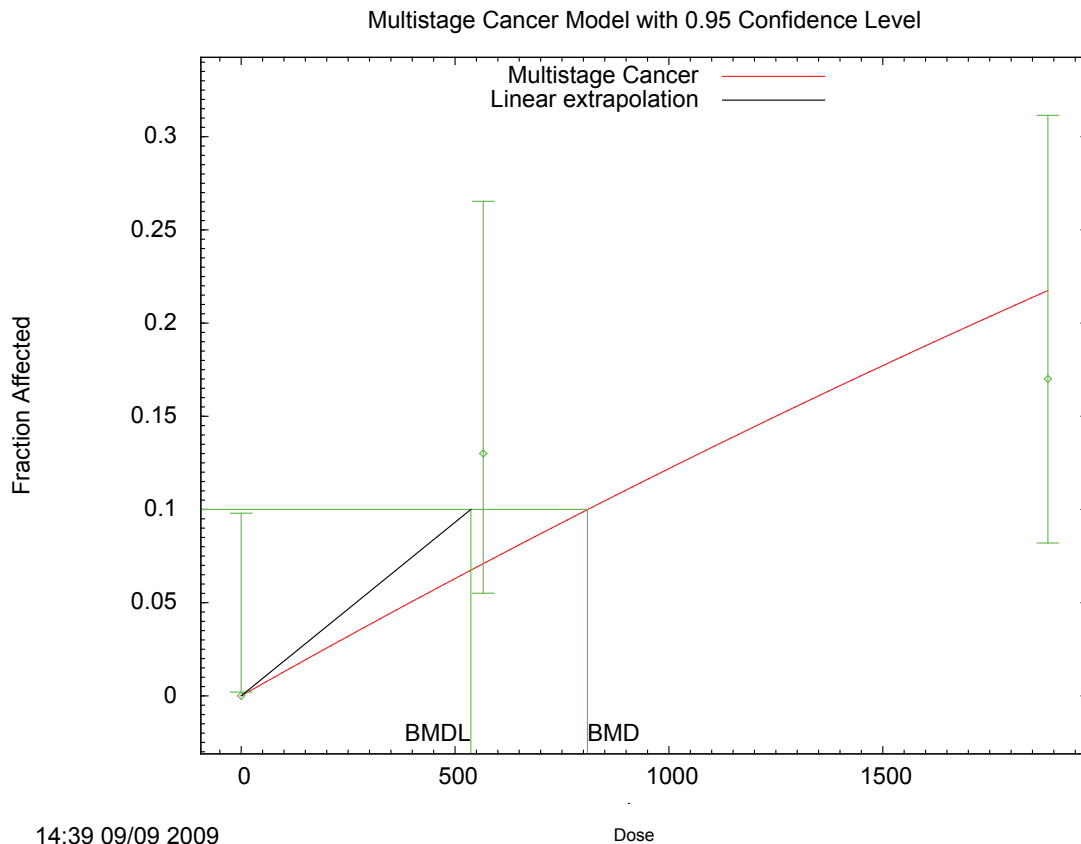
Figure C-2. Dose-Response Modeling for Combined Pituitary Adenomas and Adenocarcinoma of Female B6C3F1 Mice for 2 Years (with High Dose Omitted)

```
====================================================================
        Multistage Cancer Model. (Version: 1.7;  Date: 05/16/2008)
        Input Data File:
C:\USEPA\BMDS21\Data\mschexaneunitriskhexaneunitriskwithlastpointout.(d)
        Gnuplot Plotting File:
C:\USEPA\BMDS21\Data\mschexaneunitriskhexaneunitriskwithlastpointout.plt
                                    Wed Sep 09 14:39:21 2009
====================================================================

 BMDS Model Run
~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~

   The form of the probability function is:

   P[response] = background + (1-background)*[1-EXP(
                 -beta1*dose^1-beta2*dose^2)]

   The parameter betas are restricted to be positive


   Dependent variable = Percent
   Independent variable = Dose

Total number of observations = 3
Total number of records with missing values = 0
Total number of parameters in model = 3
Total number of specified parameters = 0
```

```
   Degree of polynomial = 2


   Maximum number of iterations = 250
   Relative Function Convergence has been set to: 1e-008
   Parameter Convergence has been set to: 1e-008



                   Default Initial Parameter Values
                     Background =    0.0362474
                        Beta(1) =  8.7614e-005
                        Beta(2) =           0



           Asymptotic Correlation Matrix of Parameter Estimates

           ( *** The model parameter(s)  -Background    -Beta(2)
                 have been estimated at a boundary point, or have been specified by
   the user,
                 and do not appear in the correlation matrix )

                  Beta(1)

     Beta(1)             1



                             Parameter Estimates

                                                 95.0% Wald Confidence
   Interval
          Variable        Estimate        Std. Err.    Lower Conf. Limit    Upper Conf.
   Limit
          Background              0            *                *                    *
            Beta(1)    0.000130161            *                *                    *
            Beta(2)              0            *                *                    *

   * - Indicates that this value is not calculated.



                            Analysis of Deviance Table

          Model       Log(likelihood)  # Param's  Deviance  Test d.f.   P-value
          Full model        -40.4291         3
        Fitted model        -40.9064         1     0.954683      2         0.6204
       Reduced model        -46.4923         1    12.1265        2         0.002327

            AIC:          83.8129



                              Goodness  of  Fit
                                                          Scaled
       Dose     Est._Prob.    Expected    Observed    Size     Residual
   --------------------------------------------------------------------
      0.0000     0.0000        0.000       0.000        45      0.000
    566.0000     0.0710        3.409       6.240        48      1.591
   1886.0000     0.2177       10.448       8.160        48     -0.800

   Chi^2 = 3.17     d.f. = 2        P-value = 0.2048


     Benchmark Dose Computation
```

49

Specified effect =   0.1

Risk Type   =   Extra risk

Confidence level =   0.95

            BMD =   809.461

          BMDL =   536.985

          BMDU =   1632.44

Taken together, (536.985, 1632.44) is a 90   % two-sided confidence
interval for the BMD

Multistage Cancer Slope Factor =   0.000186225

BMD Analysis of combined <u>liver</u> adenomas and adenocarcinomas in female mice.

The complete data set (4 values) for combined hepatocellular adenomas and carcinomas in female mice was adequately simulated by the BMD multistage model

<table>
<tr><th colspan="7">Table C-3. Input Data for Combined <u>Liver</u> Adenomas and Adenocarcinomas in <u>Female</u> B6C3F1 Mice Exposed to Commercial n-Hexane for 2 Years[a]</th></tr>
<tr>
<th>PPM</th>
<th>(mg/m^3)[a]</th>
<th>Daily Average Concentration (mg/m^3)[c]</th>
<th>Mutiplier[d] for HECs</th>
<th>HEC (mg/m^3)[e]</th>
<th>Total Neoplasms Response</th>
<th>Number of Subjects</th>
</tr>
<tr><td>0</td><td>0</td><td>0</td><td>1</td><td>0</td><td>7</td><td>50</td></tr>
<tr><td>900</td><td>3168</td><td>565.7</td><td>1</td><td>362.5</td><td>8</td><td>50</td></tr>
<tr><td>3000</td><td>10560</td><td>1885.7</td><td>1</td><td>1875</td><td>9</td><td>49</td></tr>
<tr><td>9000</td><td>31568</td><td>5657.1</td><td>1</td><td>5625</td><td>16</td><td>50</td></tr>
</table>

[a]Daughtrey et al. (1999); Biodynamics (1993b).
[b]PPM conversion: $C(mg/m^3) = PPM \times MW/24.45 = (PPM \times 86.18 \ g/mol)/24.45 = 3.52 \times PPM$
[c]Average daily concentration = $C(mg/m^3) \times$ (hours exposure/24hours) $\times$ (days exposure/7 days a week) = $C(mg/m^3) \times (6/24 \times 5/7)$.
[d]Blood gas partition coefficient = $[(H_{(b/g)})_A] / [(H_{(b/g)})_H]$. A default value of 1.0 was used because both partition coefficients were not available.
[e]HEC: Human equivalent concentration (HEC) for Extra-respiratory effects (Cat 3 Gas) = Daily average concentration × Blood gas partition coefficient.

<table>
<tr><th colspan="5">Table C-4. BMD Modeling Results for Incidence of <u>Combined</u> <u>Liver Adenomas and Adenocarcinomas</u> in Female B6C3F1 Mice Exposed to Commercial n-Hexane for 2 Years.[a]</th></tr>
<tr>
<th>Model</th>
<th>χ^2 p-Value</th>
<th>AIC</th>
<th>BMC$_{10HEC}$ (mg/m^3)</th>
<th>BMCL$_{10HEC}$ (mg/m^3)</th>
</tr>
<tr><td>Multistage cancer</td><td>0.8902</td><td>199.908</td><td>3263.62</td><td>1447.45</td></tr>
</table>

[a]Daughtrey et al. (1999); Biodynamics (1993b).

The Inhalation unit risk for combined liver <u>adenomas and adenocarcinomas is provided by the BMDS</u>

p-IUR = Multistage Cancer slope factor from BMDS = $\mathbf{7 \times 10^{-5} \ mg/m^3}$

Note: Default BMR is 0.1, thus p-IUR = BMR ÷ BMCL$_{10HEC}$ = 0.1 ÷ 1447.45 = 7×10^{-5} mg/m^3
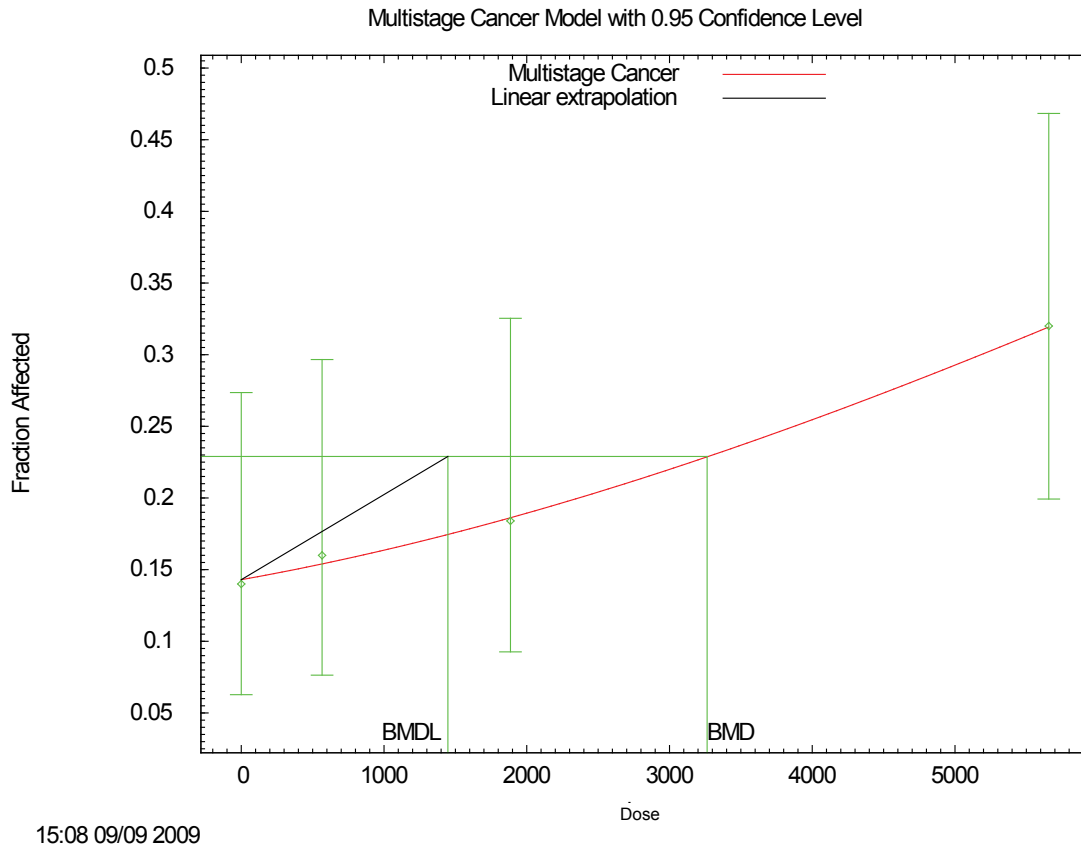
Figure C-3. Dose-Response Modeling for Incidence of <u>Combined Liver Adenomas and Adenocarcinomas</u> in Female B6C3F1 Mice Exposed to Commercial *n*-Hexane for 2 Years[a]

```
====================================================================
       Multistage Cancer Model. (Version: 1.7;  Date: 05/16/2008)
       Input Data File:
C:\USEPA\BMDS21\Data\mscHexaneunitrisklivertumorHexaneunitrisklivertumor.(d)
       Gnuplot Plotting File:
C:\USEPA\BMDS21\Data\mscHexaneunitrisklivertumorHexaneunitrisklivertumor.plt
                                    Wed Sep 09 15:08:06 2009
====================================================================

 BMDS Model Run
~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~

   The form of the probability function is:

   P[response] = background + (1-background)*[1-EXP(
                 -beta1*dose^1-beta2*dose^2)]

   The parameter betas are restricted to be positive


   Dependent variable = Percent
   Independent variable = Dose

Total number of observations = 4
Total number of records with missing values = 0
Total number of parameters in model = 3
Total number of specified parameters = 0
```

```
Degree of polynomial = 2


Maximum number of iterations = 250
Relative Function Convergence has been set to: 1e-008
Parameter Convergence has been set to: 1e-008



                    Default Initial Parameter Values
                      Background =     0.143625
                          Beta(1) = 2.02065e-005
                          Beta(2) = 3.62707e-009


            Asymptotic Correlation Matrix of Parameter Estimates

                  Background       Beta(1)       Beta(2)

Background                1         -0.71          0.61

   Beta(1)            -0.71             1         -0.97

   Beta(2)             0.61         -0.97             1



                          Parameter Estimates

                                            95.0% Wald Confidence
Interval
       Variable        Estimate       Std. Err.    Lower Conf. Limit    Upper Conf.
Limit
       Background       0.143332           *              *                    *
         Beta(1)    2.07799e-005           *              *                    *
         Beta(2)    3.52476e-009           *              *                    *

* - Indicates that this value is not calculated.

 Warning: Likelihood for the fitted model larger than the Likelihood for the full
model.
Error in computing chi-square; returning 2


                        Analysis of Deviance Table

      Model       Log(likelihood)  # Param's  Deviance  Test d.f.   P-value
    Full model        -96.968          4
  Fitted model        -96.9539         3      -0.0281741     1              2
  Reduced model       -99.8788         1       5.82161       3         0.1206


           AIC:        199.908



                          Goodness  of  Fit
                                                            Scaled
      Dose     Est._Prob.     Expected     Observed     Size     Residual
   -------------------------------------------------------------------
      0.0000     0.1433         7.167        7.000        50      -0.067
    566.0000     0.1543         7.715        8.000        50       0.111
   1886.0000     0.1865         9.139        9.016        49      -0.045
   5657.0000     0.3196        15.979       16.000        50       0.006

  Chi^2 = 0.02      d.f. = 1       P-value = 0.8902
```

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```
   Benchmark Dose Computation

Specified effect =            0.1

Risk Type        =      Extra risk

Confidence level =           0.95

         BMD =        3263.62

        BMDL =        1447.45

        BMDU =        8572.34

Taken together, (1447.45, 8572.34) is a 90     % two-sided confidence
interval for the BMD
```

Multistage Cancer Slope Factor =   6.9087e-005

Selection of IUR for Commercial Hexane

Table C-5. p-IURs for Pituitary and Liver Tumors in Female B6C3F1 Mice for 2 Years	
Tumor Database	**p-IUR**
Combined pituitary adenomas and adenocarcinomas	2×10^{-4} mg/m^3
Combined liver adenomas and adenocarcinomas	7×10^{-5} mg/m^3

The screening provisional IUR was selected from the greatest slope:

Screening p-IUR = 2×10^{-4} per mg/m^3 or 2×10^{-7} per μg/m^3