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1,3-Butadiene

CAS Registry Number: 106-99-0

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TEXAS COMMISSION ON ENVIRONMENTAL QUALITY

Revision History

Original Development Support Document (DSD) posted as final on August 7, 2008.

Revised DSD September 14, 2015: the 24 hour reference value (ReV) (Final, June 16, 2014) was added to the Summary Tables and the derivation of the 24-hour ReV was added as Appendix 9. Refer to TCEQ (2015) for guidelines on deriving 24-hour ReVs.

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Chapter 1 Summary Tables and Figure

Table 1 provides a summary of health- and welfare-based values based on an acute and chronic evaluation of 1,3-butadiene (BD). Table 2 provides summary information on BD's physical/chemical data.

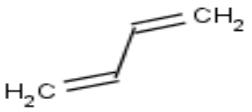
Table 1. Health- and Welfare-Based Values

Short-Term Values	**Concentration**	**Notes**
^{acute}ESL [6 h] (HQ = 0.3)	1,100 µg/m^3 (510 ppb)	**Critical Effect:** Developmental toxicity; reduction in extragestational weight gain and in fetal body weight in CD-1 mice
Acute ReV [6 h] (HQ = 1.0)	3,700 µg/m^3 (1,700 ppb) [a]	Same as above
acute ReV [24 h] (HQ =1)	950 µg/m^3 (430 ppb) [a, b]	Same as above
^{acute}ESL$_{odor}$	510 µg/m^3 (230 ppb) [a] **Short-Term ESL for Air Permit Reviews**	50% detection threshold, mild aromatic odor
^{acute}ESL$_{veg}$	---	Concentrations producing vegetative effects were significantly above other ESLs
Long-Term Values	**Concentration**	**Notes**
^{chronic}ESL$_{nonlinear(nc)}$ (HQ = 0.3)	9.9 µg/m^3 (4.5 ppb) **Long-Term ESL for Air Permit Reviews**	**Critical Effect:** Reproductive toxicity: ovarian atrophy in B6C3F1 mice
Chronic ReV (HQ = 1.0)	33 µg/m^3 (15 ppb) [a]	Same as above
^{chronic}ESL$_{linear(c)}$	20 µg/m^3 (9.1 ppb) [a, c]	**Cancer Endpoint:** Leukemia in occupational exposure study of styrene-butadiene synthetic rubber production workers
^{chronic}ESL$_{veg}$	---	No data found

[a] Values that may be used for evaluation of air monitoring data
[b] Appendix 9 provides the derivation of the 24 hour ReV for BD based on TCEQ (2015)
[c] Based on unit risk factor (URF) = 5.0E-07 per µg/m^3 (1.1E-06 per ppb) and a risk level of 1 in 100,000 excess cancer risk

Abbreviations used: HQ, hazard quotient; ppb, part per billion; mg/m3, milligrams per cubic meter; µg/m3, micrograms per cubic meter; h, hour; ESL, Effects Screening Levels; ReV, Reference Value; acuteESL, acute health-based ESL; acuteESLodor, acute odor-based ESL; acuteESLveg, acute vegetation-based ESL; chronicESL linear(c), chronic health-based ESL for linear dose-response cancer effect; chronicESLnonlinear(nc), chronic health-based ESL for nonlinear dose-response noncancer effects; and chronicESLveg, chronic vegetation-based ESL

Table 2. Chemical and Physical Data

Parameter	Value	Reference
Molecular Formula	C_4H_6 or $H_2C{:}CHHC{:}CH_2$	Lewis 1993
Chemical Structure		ChemIDplus Lite
Molecular Weight	54.1	TRRP 2006
Physical State	gas/organic	TRRP 2006
Color	colorless	Lewis 1993
Odor	mild aromatic odor	ACGIH 2001
CAS Registry Number	106-99-0	TRRP 2006
Synonyms	vinylethylene; erythrene; bivinyl; divinyl; biethylene; pyrrolylene; a,g-butadiene	Lewis 1993 NTP 1993
Solubility in water	735 mg/L	TRRP 2006
Log K_{ow}	2.03	TRRP 2006
Vapor Pressure	2,100 mm Hg at 20 ° C	TRRP 2006
Vapor Density (air = 1)	1.87	Lewis 1992
Density (water = 1)	0.6211 (liquid at 20 ° C)	Lewis 1993
Melting Point	-113° C	Lewis 1992
Boiling Point	-4.41 ° C	Lewis 1993
Conversion Factors	1 µg/m^3 = 0.45 ppb @ 25°C 1 ppb = 2.21 µg/m^3	NTP 1993

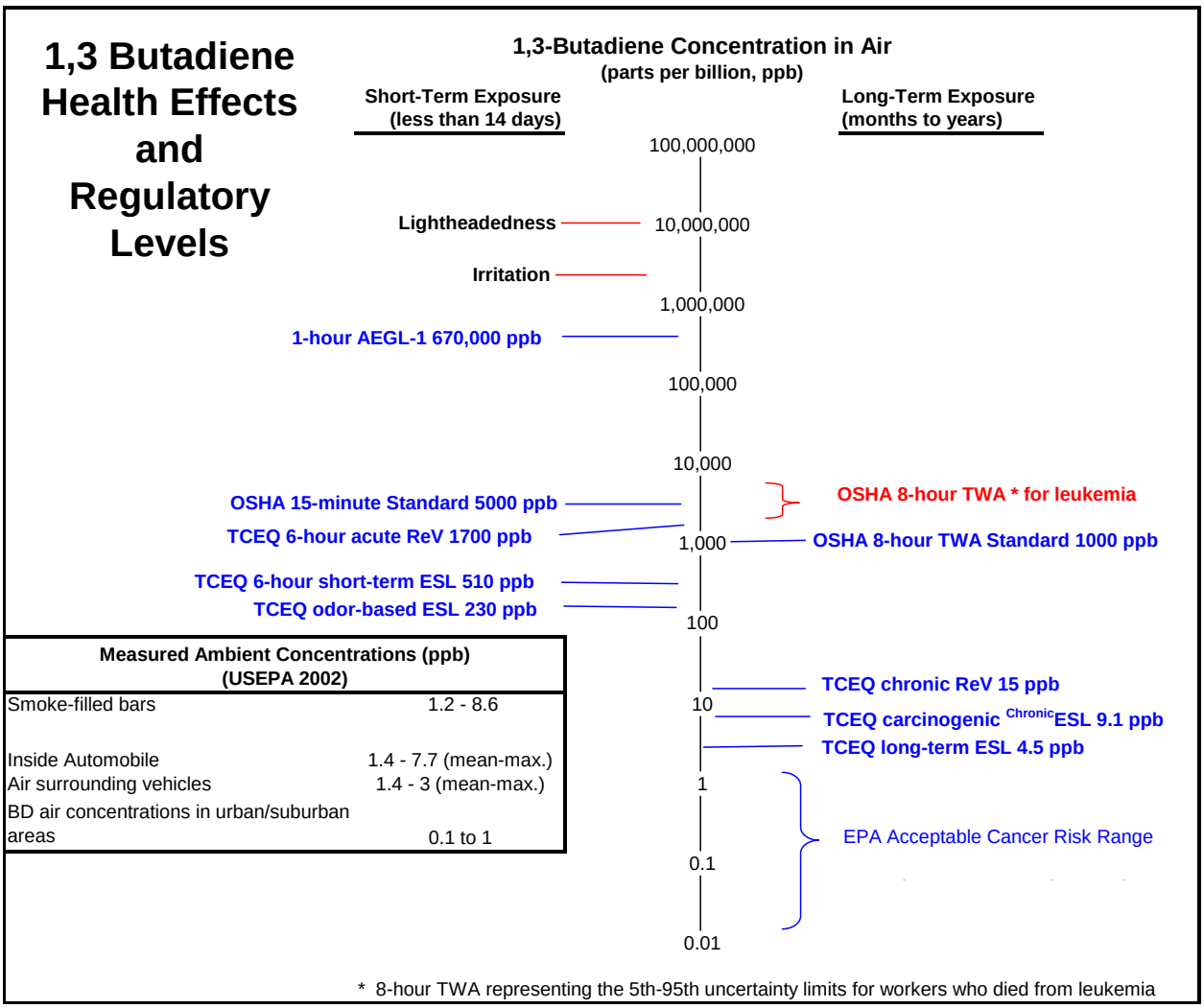


Figure 1 BD Health Effects and Regulatory Levels.

This figure compares BD's acute toxicity values (acute ReV, odor-based ESL, and health-based, short-term ESL) and chronic toxicity values (chronic ReV and long-term ESL) found in Table 1 to USEPA's acceptable cancer risk range (USEPA 2002), OSHA's occupational values, and the AEGL-1 value (AEGL 2005). USEPA's (2002) acceptable cancer risk range is based on an older epidemiology study that has recently been updated to include additional information with validated, more accurate BD exposure estimates.

Abbreviations used: **BD**, 1,3-butadiene; **TCEQ**, Texas Commission on Environmental Quality; **TWA**, Time-Weighted Average; **ESL**, Effects Screening Level; **ReV**, Reference Value; **OSHA**, Occupational Safety and Health Administration; **USEPA**, United State Environmental Protection Agency; and **AEGL-1**, Level 1-Acute Exposure Guideline Levels.

Chapter 2 Major Sources or Uses

BD is used as an intermediate in the production of polymers, elastomers, and other chemicals. Its major uses are in the manufacture of styrene-butadiene rubber (SBR) (synthetic rubber) and thermoplastic resins. Elastomers of BD are used in the manufacture of tires, footwear, sponges, hoses and piping, luggage, packaging, and a variety of other molded products. In addition, BD is used as an intermediate to produce a variety of industrial chemicals, including the fungicides captan and captfol. The primary way that BD is released into the environment is via emissions from gasoline- and diesel-powered vehicles and equipment. Lesser releases occur from the combustion of other fossil fuels and biomass. Minor releases occur in production processes, tobacco smoke, gasoline vapors, and vapors from the burning of plastics as well as rubber (Miller 1978; USEPA 2002). United States Environmental Protection Agency's (USEPA) (2001) National-Scale Air Toxics Assessment of emissions from the 1996 National Toxics Inventory indicates that statewide BD emissions from mobile sources (onroad and nonroad) accounted for approximately 54% of the National Toxics Inventory BD emissions in Texas, with major facility sources and area/other sources (e.g., smaller facilities) comprising the remainder of 46%.

Chapter 3 Acute Evaluation

3.1 Health-Based Acute ReV and ^{acute}ESL

3.1.1 Physical/Chemical Properties and Key Studies

3.1.1.1 Physical/Chemical Properties

BD is a highly volatile, colorless gas with a mildly aromatic odor. The main chemical and physical properties of BD are summarized in Table 2. It is soluble in ethanol, diethyl ether, and organic solvents, and only slightly soluble in water.

3.1.1.2 Key Studies

This section is based on USEPA (2002) and AEGL (2005). Both of these sources state "The acute toxicity of BD is of low order." (USEPA 2002; AEGL 2005). A review of the scientific literature since 2002 indicates that a subchronic inhalation study in rats conducted by the American Chemistry Council (ACC 2003) is a new animal study that was not considered by USEPA (2002), and the findings of Spencer *et al.* (2001) and Chi *et al.* (2002) on the possible reproductive/developmental mode of action of BD were not considered. Therefore, these studies are discussed in Sections 3.1.1.2.2 and 3.1.2.2, respectively. Animal data show BD is a potential reproductive/developmental hazard to humans. Since the reproductive/developmental effects of BD in rats and mice are among the effects observed at the lowest exposure levels following acute inhalation exposure, the following sections focus on these health effects. Chapter 5 of *Health Assessment of 1,3-Butadiene* (USEPA 2002) provides a detailed discussion on potential

reproductive/developmental effects in humans and animals, and AEGL (2005) discusses other types of acute toxicity data.

3.1.1.2.1 Human Studies

Albertini *et al.* (2007) conducted a molecular epidemiological study of BD-exposed Czech workers to compare female to male responses. The focus of the study was to collect data on urine concentrations of BD metabolites and blood concentrations of BD-metabolite hemoglobin adducts. However, questionnaire responses for female-specific adverse health questions in control and exposed females were also obtained. There were 26 female control workers and 23 female BD-exposed workers. The years of employment were 17.6 $\pm$ 9.3 years for control and 19.4 $\pm$ 9.9 years for exposed females (mean $\pm$ S.D.). Multiple external exposure measurements were obtained (10 full 8-hour (h) shift measures by personal monitoring per worker) over a 4-month period before biological samples were collected. Mean 8-h time-weighted average (TWA) exposure levels were 0.008 milligram per cubic meter (mg/m^3) (0.0035 parts per million (ppm)) for controls and 0.397 mg/m^3 (0.180 ppm) for exposed. Individual single 8-h TWA values were as high as 9.793 mg/m^3 (4.45 ppm). Analysis of questionnaire responses for female-specific adverse health questions showed no significant differences between controls and exposed for miscarriages, still births, ectopic pregnancies, molar pregnancies, low birth weight (<2,500 g) babies, or pre-term births, based on information collected on all pregnancies. The ability of the study to detect differences in the evaluated endpoints may be limited because there were few subjects evaluated.

The health effects observed in humans occur at high concentrations and include the following: odor perception (ACGIH 2001; Ruth 1986; and Nagata 2003); slight smarting of the eyes and difficulty in focusing on instrument scales (Carpenter *et al.* 1944); and tingling sensation and dryness of the nose and throat (Larionov *et al.* 1934) (Table 3). A poorly reported study conducted by Ripp (1967) in human volunteers reported effects of olfactory perception at 4.0 mg/m^3 (1.8 ppm) and sensitivity of the eye to light at 3.9 mg/m^3 (1.7 ppm). There were no effects on the occurrence of an electrocortical conditioned reflex at 3 mg/m^3 (1.4 ppm). Khalil *et al.* (2007) reported that BD produced increased neurological risks in a random cohort of 310 patients who had been exposed to accidental leakage and release of BD due to an explosion. The environmental contamination persisted for a few hours to several days in the atmosphere of the areas surrounding the plant. Exposure concentrations of BD or information on other chemicals that may have been released during the explosion were not provided.

Table 3 Acute Effects of BD in Humans

Study	Concentration (Exposure Duration)	Subjective Symptoms	Differences Observed
Carpenter *et al.* 1944 2 males 1-hour (h) lunch break Nominal Concentrations	2,000 ppm [1] (7 h)	Slight smarting of the eyes; difficulty in focusing on instrument scales	Results of tapping test and steadiness test – no differences
Same as above	4,000 ppm (6 h)	Slight smarting of the eyes; difficulty in focusing on instrument scales	Results of tapping test and steadiness test – no differences
Same as above	8,000 ppm (8 h)	No subjective complaints [2]	Results of tapping test and steadiness test – no differences
Larionov *et al.* (1934) No details on number of subjects and gender	1% (10,000 ppm) 5 minute (min)	Tingling sensation and dryness of the nose and throat.	Slight increase in pulse rate. No effects on blood pressure or respiration

1 Difficulty in focusing on instrument scales was the basis of the AEGL-1 value. The 1-h AEGL-1 value of 670 ppm = 2,000 ppm divided by an intraspecies uncertainty factor of 3.

2 No subjective complaints because of slight anxiety of subjects concerning the possibility of an explosion.

3.1.1.2.2 Animal Studies

3.1.1.2.2.1 Reproductive/Developmental Toxicity in Rats

In 1982, Hackett *et al.* (International Institute of Synthetic Rubber Producers (IISRP) 1982) conducted a reproductive/developmental study that included exposure of pregnant rats at 0, 200, 1,000, and 8,000 ppm 6 hours/day (h/day) on gestation day (GD) 6-15 and then sacrifice on GD 20. The most sensitive endpoints were a significant decrease in maternal body weight gain on GD 6-9 and extragestational weight gain (lowest observed adverse effect level (LOAEL) of 1,000 ppm and no observed adverse effect level (NOAEL) of 200 ppm for both endpoints). Minor skeletal defects were found to be significantly elevated at the lowest concentration, and the percentage of fetuses with major skeletal defects was significantly elevated at 1,000 ppm and above. The incidence of marked-to-severe wavy ribs and the total number of abnormal ossifications and irregular ossification of the ribs were elevated at 8,000 ppm.

In 1987, Hackett *et al.* (1987a) repeated the IISRP (1982) study at slightly lower concentrations

to confirm the 1982 findings in rats and to compare the effects of similar BD exposures in mice (Hackett *et al.* 1987b). The results of the Hackett *et al.* (1987b) study in mice are discussed in the next section. Pregnant rats (Hackett *et al.* 1987a) were exposed for 10-days via inhalation to 0, 40, 200, and 1,000 ppm on GD 6-15 for 6 h/day (Hackett *et al.* 1987a). For rats, the most sensitive short-term endpoints were decreases in maternal body weight gain on GD 6-11 and decreases in extragestational weight gain (NOAEL of 200 ppm and LOAEL of 1,000 ppm for both endpoints). Effects from BD exposure for fetal measures were not observed (i.e., no developmental toxicity was observed).

In 2003, a subchronic reproductive/developmental study in rats sponsored by the American Chemistry Council was conducted by WIL Research Laboratories, Inc (ACC 2003). Since this study was not available for USEPA's BD assessment (USEPA 2002), the major findings of the study are discussed below. The study was conducted using the following guidelines:

- USEPA TSCA Good Laboratory Practice Standards;
- The protocol met or exceeded applicable regulations of the Organisation for Economic Cooperation and Development (OECD) Guideline for Testing of Chemicals, Guideline 421, Reproduction/Development Toxicity Screening Test (July 27, 1995) and Office of Prevention, Pesticides & Toxic Substances (USEPA) 870.3550 (July 2000) requirements.

This study was conducted to provide information on the potential adverse effects of BD on male and female reproduction within the scope of a screening study. Assessments of gonadal function, mating behavior, conception, gestation, parturition, lactation of the F_0 generation, and the development of F_1 offspring from conception through weaning and post-weaning exposure were included. Three groups of F_0 animals, each consisting of 12 male and 12 female Crl:CD®(Sprague-Dawley) IGS BR rats, were exposed to 300, 1,500, and 6,000 ppm BD via whole-body inhalation exposure 6 h/day for 14 days prior to the breeding period and continuing throughout the gestation and lactation periods. A control group was exposed to clean, filtered air on a comparable regimen. For F_0 dams, the daily inhalation exposures were suspended on GD 21 through lactation day 4, to avoid any confounding effects of exposure on nesting or nursing behavior. Exposures were resumed for these dams on lactation day 5. The F_1 generation pups were potentially exposed to BD *in utero* and through nursing during lactation until weaning. Beginning on postnatal day (PND) 21, one male and one female from each litter were exposed for seven consecutive days to the same concentration of the BD concentration as its dam. Beginning on PND 28, one previously unexposed male and one previously unexposed female per litter were exposed for seven consecutive days to the same BD concentration as its dam.

Under the conditions of the current study, there were no adverse BD-related effects on any parameter measured in either the F_0 or F_1 animals at the exposure level of 300 ppm. Adverse BD-related effects were noted at 1,500 and 6,000 ppm and consisted of persistent reductions in body weight parameters in F_0 and F_1 males and females and transient reductions in food consumption (week 0-1) for F_0 males and females.

Adverse BD-related effects noted exclusively at 6,000 ppm consisted of clinical observations indicative of chromodacryorrhea, chromorhinorrhea, and salivation in F_0 males and females as well as infrequent occurrences of dried red material in the perioral and perinasal regions of four exposed F_1 pups (three males and one female).

Based on the results of this study, an exposure level of 300 ppm was considered to be the NOAEL in rats for F_0 parental systemic toxicity and for systemic toxicity for F_1 animals following post-weaning 6-h daily exposures (PND 21-27 or PND 28-34). The NOAEL for effects on gonadal function, mating behavior, conception, gestation, parturition, lactation of the F_0 generation, and the development of F_1 offspring from conception through weaning was considered to be 6,000 ppm.

The findings of this subchronic reproductive/developmental study showed effects of reduction in body weight parameters as the most sensitive endpoint in male and female rats with a NOAEL of 300 ppm. Developmental effects were not observed. This study is included in the acute toxicity section because it is a well-conducted, high-quality study with a NOAEL of 300 ppm, which is slightly higher than the NOAEL of 200 ppm determined in previous rat studies (IISRP 1982; Hackett *et al.* 1987a).

3.1.1.2.2.2 Reproductive/Developmental Toxicity in Mice

Hackett *et al.* (1987b) exposed pregnant mice for 10 days via inhalation at 0, 40, 200, and 1,000 ppm (analytical concentrations of 0, 39.9, 200, and 1,000 ppm) on GD 6-15 for 6 h/day. Maternal toxicity manifested as reduced body weight gain (GD 11-16) and extragestational weight gain was observed at 200 and 1,000 ppm. Total body weight at GD 18 was decreased at 1,000 ppm. Therefore, the NOAEL for maternal toxicity was 40 ppm. Hackett *et al.* (1987b) reported the most sensitive short-term developmental endpoint was decreased fetal body weight in male mice at 40 ppm. BD caused reduced fetal body weight and increased frequency of skeletal variations at 200 and 1,000 ppm which are concentrations corresponding to maternal toxicity expressed as reduced body weight. Major malformations in the mouse fetus were not detected although the potential for altered development was indicated by a dose-related increase in supernumerary ribs and reduced ossifications, particularly of the sternebrae.

Hackett *et al.* (1987b) reported that statistical differences were observed at the lowest exposure concentration of 40 ppm for male fetal body weight. Therefore, a NOAEL was not identified for this effect. However, Hackett *et al.* (1987b) conducted analyses of variance (ANOVA) on the average pup weight followed-up by Student's t-tests comparing the average pup weight for different treatment groups. Their pairwise comparisons using Student's t-test did not adjust significance levels for the number of multiple tests. In addition, their analyses did not adjust for well-known important covariate effects such as litter size. Christian (1996) noted that the apparent significant decrease in male fetal body weight in the 40 ppm group was the result of the statistical analysis used, which was considered to be inappropriate.

Data reported by Hackett *et al.* (1987b) were reanalyzed by Green (2003). The Green (2003) reanalysis was based on analysis of covariance (ANCOVA) on the average pup weight adjusted for covariates and used the Dunnett-Hsu test to compare the mean weights for each of the exposed groups to the mean weight for the control group. Application of the statistical analysis indicates that the 40 ppm exposure concentration is a NOAEL in this study. Other previously analyzed endpoints were also analyzed by more appropriate methodology (Green 2003). In each instance, the NOAEL was at least as high as previously reported. For a few endpoints, a higher NOAEL was found. The overall NOAEL for this study is 40 ppm, based on the fetal body weights.

In order to assess the Green (2003) reanalysis, Sielken *et al.* (Appendix 1) conducted a review of the Hackett *et al.* (1987b) study and the Green (2003) reanalysis, concentrating on male fetal body weight. The Sielken *et al.* review (Appendix 1) indicates that Green's (2003) conclusions are reasonable and based on standard statistical analyses practices that were overlooked by Hackett *et al.* (1987b). Green used the Dunnett-Hsu test to compare the mean weights for each of the exposed groups to the mean weight for the control group after both were adjusted for the effects of the covariates. The Dunnett-Hsu test was specifically designed for this situation. In addition to reviewing the statistical methodology used in the Hackett *et al.* (1987b) and Green (2003) studies, Sielken *et al.* (Appendix 1) re-analyzed the fetal body weight data to confirm the numerical results obtained by Green (2003). Sielken *et al.* (Appendix 1) also performed a sensitivity analysis with respect to the effects of covariates and determined the outcome of the more powerful statistical analyses where the individual pup weights were analyzed and the dams were treated as random effects. These analyses support the finding that the NOAEL based on either male or female fetal body weight for this study is 40 ppm (Sielken *et al.* (Appendix 1)).

Table 4 is similar to Table 5-6 in USEPA (2002) but only contains parameters that were significantly different from controls. There were no statistical differences in number of pregnant dams, litters with live fetuses, implantations per dam, resorptions per litter, dead fetuses per litter, fetuses per number of litters examined, or sex ratio (% males) between treated mice and control mice (data not shown). The highlighted cells in Table 4 have been corrected based on the Hackett *et al.* (1987b) study reanalyses by Green (2003) and Sielken *et al.* (Appendix 1). The appropriate NOAEL for early resorptions is 1,000 ppm (not 200 ppm as reported by Hackett *et al.* (1987b)), and the LOAEL for decreases in male fetal body weight is 200 ppm (not 40 ppm). Decreases in male fetal body weight occur at the same concentrations as decreases in maternal weight gain (Table 6).

Table 5 is similar to Table 5-7 in USEPA (2002) but only contains parameters that were significantly different from controls. There were no results contrary to those of the Hackett *et al.* (1987b) after the reanalysis by Green (2003). The only fetal effects noted were significant increases in minor skeletal abnormalities at 200 and/or 1,000 ppm indicative of growth retardation (i.e., increases in supernumerary ribs and reduced ossification in the sternebrae). These effects occurred at the same concentrations as decreases in maternal weight gain (Table 6).

Table 4 Developmental Toxicity in CD-1 Mice Exposed to BD by Inhalation a

Parameters	0 ppm	40 ppm	200 ppm	1,000 ppm
Early resorptions	1.00 ± 0.23	0.58 ± 0.21	0.43 ± 0.13 [c, g]	0.75 ± 0.16
Fetal body weight (gram (gm)) (Mean per litter)	1.34 ± 0.03 [b]	1.28 ± 0.01	1.13 ± 0.02 [c]	1.04 ± 0.03 [c]
Females	1.30 ± 0.03 [b]	1.25 ± 0.01	1.10 ± 0.02 [c]	1.06 ± 0.02 [c, f]
Males	1.38 ± 0.03 [b]	1.31 ± 0.02 [c, d]	1.13 ± 0.02 [c]	1.06 ± 0.02 [c]
Placental weight (mg) (Mean per litter)	86.8 ± 2.99 [b]	85.4 ± 2.29	78.6 ± 3.24 [c]	72.6 ± 1.88 [c]
Females	83.1 ± 3.03 [b]	80.9 ± 2.46	74.7 ± 3.52	70.1 ± 2.33 [c]
Males	89.3 ± 3.03 [b, e]	89.5 ± 2.27	80.1 ± 2.35 [c]	74.5 ± 1.81 [c]

a All values mean ± standard error from USEPA (2002)

b $p \leq 0.05$, significant linear trend

c $p \leq 0.05$, pairwise comparison with corresponding control parameter based on Hackett et al. (1987b)

d $p > 0.05$ based on Green (2003) and Sielken et al. reanalyses(Appendix 1)

e 89.3 + 3.05 (Hackett et al. 1987b)

f 1.02 + 0.02 (Hackett et al. 1987b)

g $p > 0.05$ based on Green (2003)

Source: USEPA (2002)

Table 5 Variations in CD-1 Mice Exposed to BD by Inhalation

Parameters	0 ppm	40 ppm	200 ppm	1,000 ppm
Variations: Abnormal sternebrae a, b	0.6 ± 0.9	0.4 ± 0.7	0.4 ± 0.8	0.8 ± 1.3 c
Variations: Supernumerary ribs a, b	1.7 ± 2.3	1.6 ± 2.1	6.0 ± 3.6 c	9.9 ± 3.0 c
Reduced ossification (all sites combined) a	1.7 ± 1.7	1.2 ± 1.5	2.7 ± 2.7	3.9 ± 2.6 c

a Mean percentage per litter (mean ± SD)

b $p \leq 0.05$, significant linear trend, orthogonal contrast test

c $p \leq 0.05$, Tukey's test

d $p \leq 0.05$, Fisher exact test (fetal incidence)

Source: USEPA (2002) and Hackett et al. (1987b)

Table 6 Maternal Toxicity in Pregnant CD-1 Mice Exposed to BD by Inhalation a

Parameters	0 ppm	40 ppm	200 ppm	1,000 ppm
Whole-body weight (gm)				
Day 0	28.4 ± 0.25	28.3 ± 0.32	28.3 ± 0.32	28.4 ± 0.32
Day 18	54.9 ± 1.21 [b]	55.4 ± 1.09	52.5 ± 1.01	50.8 ± 0.86 [c, f]
Body weight gain (gm)				
Days 0-6	2.7 ± 0.3	3.0 ± 0.3	2.5 ± 0.2	2.3 ± 0.2
Days 6-11	5.5 ± 0.4	5.8 ± 0.3	5.6 ± 0.3	4.8 ± 0.3
Days 11-16	13.3 ± 0.6 [b]	12.7 ± 0.4	11.4 ± 0.5 [c]	10.6 ± 0.4 [c]
Days 16-18	5.5 ± 0.3 [b]	5.7 ± 0.3	4.7 ± 0.4	4.8 ± 0.3
Gravid uterine weight (gm)	19.3 ± 1.00 [b]	20.3 ± 0.80	18.0 ± 0.87	16.8 ± 0.67 [c, g]
Extragestational weight (gm) [d]	35.5 ± 0.48 [b]	35.1 ± 0.44	34.5 ± 0.46	34.1 ± 0.36 [c]
Extragestational weight gain (gm) [e]	7.60 ± 0.48 [b]	6.99 ± 0.38	6.20 ± 0.38 [c]	5.91 ± 0.28 [c]

a All values mean ± standard error from USEPA (2002)

b $p \leq 0.05$, significant linear trend

c $p \leq 0.05$, pairwise comparison with corresponding control parameter

d Body weight on GD 18 minus gravid uterine weight

e Extragestational weight minus body weight on GD 0

f 50.8 + 0.87 (Hackett et al. 1987b)

g 16.7 + 0.67 (Hackett et al. 1987b)

Source: USEPA (2002)

Table 6 is similar to Table 5-5 in USEPA (2002) but only lists data on maternal weight loss measures which are the main parameters that were significantly different from controls. There were no results contrary to those of Hackett *et al.* (1987b) based on the reanalysis of Green (2003). Table 6 indicates that there was a statistical reduction in extragestational weight gain (i.e., maternal weight minus gravid uterine weight) and weight gain (GD 11-16) at 200 ppm. A statistical decrease in gravid uterine weight occurred at 1,000 ppm. These results suggest that BD produces maternal toxicity but little or no intrauterine effects at 200 ppm. For mice and rats, body weight changes and changes in body weight gain in pregnant dams with no change in gravid uterine weight usually indicate maternal toxicity as discussed by Pohl *et al.* (1998):

> "Changes in maternal body weight corrected for gravid uterine weight at sacrifice may indicate whether the effect is primarily maternal or fetal. For example, there may be a significant reduction in weight gain and in gravid uterine weight throughout gestation but

> no change in corrected maternal weight gain, which would generally indicate an intrauterine effect. Conversely, a change in corrected weight gain and no change in gravid uterine weight generally suggest maternal toxicity and little or no intrauterine effect."

Although reduction in maternal body weight gain was an effect that was consistently observed in studies in rats (at higher concentrations) and mice (IISRP 1982; Hackett *et al.* 1987a, 1987b; and ACC 2003), there is experimental evidence that BD exposure causes a reduction in serum progesterone which may result in fetal/placental effects (Section 3.1.2.2 *MOA for Reproductive/ Developmental Effects*). Therefore, the data from the following developmental and maternal toxicity endpoints observed in mice (Hackett *et al.* (1987b) was evaluated using benchmark dose modeling to determine a point of departure (POD) because they had a positive dose-response relationship:

- Developmental endpoints: decreased placental weight and fetal body weight, abnormal sternebrae, reduced ossification for all sites and increased incidence of supernumerary ribs
- Maternal toxicity: decreases in extragestational weight gain, body weight gain (GD 11-16), whole-body weight (day 18), gravid uterine weight, and extragestational weight

3.1.2 Mode-of-Action (MOA) Analysis

It is generally agreed that BD produces toxicity when it is metabolized to its reactive metabolites after animals are exposed to BD. However, there is a difference in the metabolism amongst species. The basis of the species differences between rats and mice may be related to the greater production of toxic intermediates and a lower capacity for detoxification of these intermediates (USEPA 2002).

3.1.2.1 Metabolism

The following chemical terminology, similar to the terminology in USEPA (2002), is used in the DSD. Figure 2 is Figure 3.1 from USEPA (2002):

- 1,2-Epoxy-3-butene (EB). EB is also used for epoxybutene, 1,3-butadiene monoepoxide, 1,3-butadiene monoxide, 1,2-epoxybutene-3, vinyl oxirane, and 3,4-epoxy-1-butene;
- 1,2:3,4-Diepoxybutane (DEB). DEB is also used for diepoxybutane, butadiene diepoxide, and butadiene bisoxide;
- 3-Butene-1,2-diol (butene-diol). Butene-diol is also used for 1,2-dihydroxybut-3-ene; and
- 1,2-Dihydroxy-3,4-epoxybutane (EBD). EBD is also used for epoxybutanediol, 3,4-epoxybutanediol, 3,4-epoxybutane-1,2-diol, and 3,4-epoxy-1,2-butanediol.

The general metabolic scheme of BD, which has been reviewed by Himmelstein *et al.* (1997), is shown in Figure 2. BD is first metabolized to 1,2-epoxy-3-butene (EB), a process that is primarily associated with cytochrome P450 (CYP) 2E1, but can also be accomplished by additional isoforms including CYP 2A6 and 4B1. This electrophilic metabolite can be detoxified by conjugation with glutathione and subsequent excretion in the urine as urinary metabolites 1-hydroxy-2-(N-acetylcysteinyl)-3-butene and 2-hydroxy-1-(N-acetylcysteinyl)-3-butene (collectively known as M2 metabolite). It can also undergo hydrolysis by epoxide hydrolase (EH) to form 3-butene-1,2-diol (butene-diol). Butene-diol can also be conjugated with glutathione and subsequently excreted in the urine as urinary 1,2-dihydroxy-4-(N-acetylcysteinly)-butane (M1 metabolite). It can be further oxidized by cytochrome P450 to the 1,2-dihydroxy-3,4-epoxybutane (EBD). An alternative pathway for the metabolism of EB is oxidation to the 1,2:3,4-diepoxybutane (DEB) which can be further hydrolyzed to EBD or conjugated by glutathione. This series of epoxidation and detoxication steps generates three electrophilic metabolites: EB, DEB, and EBD.

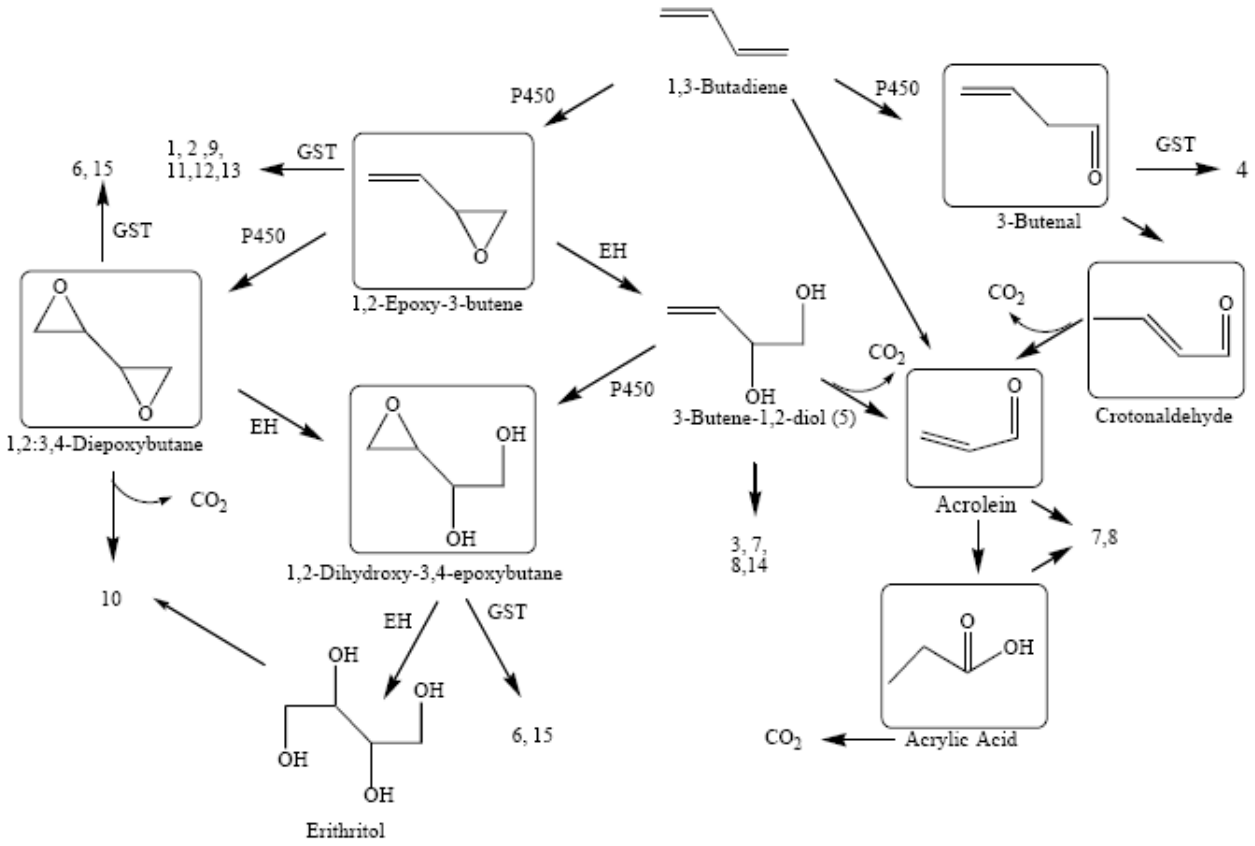


Figure 2 Schematic of BD Metabolism

P450 stands for cytochrome P450, EH stands for epoxide hydrolase, GST stands for glutathione transferase, and GSH stands for glutathione (Figure 3-1 from USEPA (2002)). The reactive metabolites are shown inside boxes. The urinary metabolites are numbered and listed in Table 3-1 of USEPA (2002).

Cochrane and Skopek (1994) have shown that DEB is 100 times more mutagenic than EB and 200 times more mutagenic than EBD in human lymphocytes. Kligerman and Yu (2007) used an *in vitro* system of lymphocytes treated with EB or DEB and measured sister chromatid exchange and chromosome aberrations. DEB-induced damage for both sister chromatid exchange and chromosome aberrations was persistent in G_0 cells and DEB was much more genotoxic than EB. EB did not induce sister chromatid exchange in lymphocytes unless actively cycling cells were treated. The extent to which DEB is produced and reaches target tissues will play a role in the toxicity. The ability of EB to reach actively dividing or repair deficient cells will also contribute somewhat to toxicity (Kligerman and Yu 2007). Mice form more DEB than rats or humans whereas EBD is more readily formed in humans than in rats (Slikker *et al.* 2004; Swenberg *et al.* 2007).

Human genetic polymorphisms are likely to affect individual susceptibility to BD and its metabolites. Metabolic activation rates in humans exhibit a high degree of variability and appear to span the range of activation rates between mice and rats when evaluated with *in vitro* systems measuring enzyme kinetics (greater than ten-fold). Other *in vitro* studies and *in vivo* molecular epidemiological studies indicate the range of increased sensitivity due to human genetic polymorphisms is approximately two- to four-fold (Albertini *et al.* 2001, 2003; Begemann *et al.* 2001; Fustinoni *et al.* 2002; Hayes *et al.* 1996, 2000, 2001; Smith *et al.* 2001; and Zhao *et al.* 2000, 2001). Several genes appear to be important in the BD metabolic pathway. Inherent susceptibilities have been shown for both EB and DEB (Weincke and Kelsey 1993), which may be due to glutathione S-transferase theta (GSTT1) status. Also, glutathione S-transferase GSTM1 appears to be an important detoxifying factor for EB, so that GSTM1 null individuals would be expected to have greater effects following formation of EB. Unfortunately, no data have been published on the effects of GST polymorphisms of EBD. Genetic polymorphisms have also been identified for EH and CYP 2E1 that would be expected to affect susceptibility to BD and its metabolites. The role of these proteins in the toxicokinetics of numerous chemicals is reasonably well known. Three *in vitro* studies (Csanády *et al.* 1992; Seaton *et al.* 1995; and Duescher and Elfarra 1994) using rodent and human tissue samples have demonstrated that CYP 2E1 plays a role in the oxidation of both BD and EB.

Polymorphisms that reduce EH activity may increase susceptibility to BD-induced effects. Likewise, rapid CYP 2E1 metabolizers may potentially be at greater risk. As previously mentioned, mice are much more sensitive to BD's reproductive/developmental effects than rats. The basis of the species differences between rats and mice may be related to the greater production of toxic intermediates, specifically DEB, and a lower capacity for detoxification of these intermediates in mice (USEPA 2002). Conjugation with GSH is an important detoxification route. Himmelstein *et al.* (1997) points out that GSH depletion occurs at longer exposure duration or at higher concentrations leading to higher body burdens of EB and DEB (Himmelstein *et al.* 1997).

3.1.2.2 MOA for Reproductive/Developmental Effects

The most sensitive reproductive effect observed in 2-year chronic exposure studies was ovarian atrophy in female mice (NTP 1993). Ovarian atrophy is discussed in greater detail in Chapter 4. The specific mechanism of action for the reproductive/developmental effects produced by BD is unknown, although the MOA may involve DEB-induced ovarian atrophy and a decrease in

serum progesterone levels (Spencer *et al.* 2001; Chi *et al.* 2002). Both Spencer *et al.* (2001) and Chi *et al.* (2002) hypothesize that DEB inhibits ovarian function, leading to a decrease in progesterone. Both estrogen and progesterone acting together, followed by progesterone postimplantation levels, are required for endocrine support for mammalian gestation. DEB does not appear to alter relative levels of estrogen receptor α mRNA expression (Spencer *et al.* 2001). Spencer *et al.* (2001) demonstrated that four daily intraperitoneal (i.p.) injections of DEB caused a dose-dependent decrease in endometrial weight, protein, and DNA, with decreases in serum progesterone in pseudo-pregnant Sprague-Dawley rats. Inducible nitric oxide synthase, pituitary adenylate cyclase-activating polypeptide (PACAP) mRNA expression, and matrix metalloproteinase-9 (MMP-9) activity were also decreased. These enzymes are important in implantation of the blastocyst and tissue remodeling. These changes lead to an inhibitory effect on uterine deciduoma growth/differentiation. Similar results were obtained when pregnant Sprague-Dawley rats were treated with four daily i.p. doses of DEB (Chi *et al.* 2002). Serum progesterone levels were significantly decreased as well as placental PACAP mRNA expression and MMP-9 activity (Chi *et al.* 2002). Chi *et al.* (2002) concluded:

> "In summary, the reproductive toxicity of diepoxybutane in pregnant rats apparently involved coordinated inhibition of placental molecular mechanisms (PACAP and MMP-9), uterine developmental processes (implantation and fetal metabolism) and progesterone secretion."

Based on the above information and consistent with USEPA (2002), the reproductive/developmental effects in mice are considered to have a threshold (i.e., a nonlinear MOA) and to be concentration and duration dependent.

3.1.3 Dose Metric

For the reproductive/developmental key study (Hackett *et al.* 1987b), data on the exposure concentration of the parent chemical are available. Since the MOA of the toxic response is not fully elucidated and data on other more specific dose metrics are not available (e.g. blood concentration of parent chemical, area under blood concentration curve of parent chemical, or putative metabolite concentrations in blood or target tissue), the exposure concentration of the parent chemical was used as the default dose metric.

3.1.4 Points of Departure (PODs) for Key Studies

The LOAEL for maternal toxicity in rats (1500 ppm) reported from a subchronic study conducted by the American Chemistry Council (ACC 2003) is more than seven times the LOAEL for developmental effects and maternal toxicity observed in mice (200 ppm). In addition, the slope of the rat dose-response curve is not steep, so the data from maternal toxicity in rats will not be considered. Data from mice for the following developmental and maternal toxicity endpoints (Section 3.1.1.2.2.2 R*eproductive/*

Developmental Toxicity in Mice), which are all continuous data, were modeled with Benchmark Dose Modeling (BMDS) Software (Version 1.4.1c) using continuous models:

- Developmental endpoints: decreased placental weight, fetal body weight, abnormal sternebrae, reduced ossification for all sites, and increased incidence of supernumerary ribs
- Maternal toxicity: decreases in extragestational weight gain, body weight gain (GD 11-16), whole-body weight (day 18), gravid uterine weight, and extragestational weight

Since the selected endpoints are from a single study (Hackett *et al.* 1987b) and the same dosimetric adjustments and uncertainty factors will be applied to each endpoint, the endpoint with the lowest POD determined with BMD modeling may be the critical effect, if the endpoint is considered adverse, biologically plausible, and consistent with the proposed MOA.

3.1.4.1 Critical Effect Size

If there is an accepted level of change in the endpoint that is considered to be biologically significant, then that amount of change is chosen for evaluation (USEPA 2000). For dichotomous data, this level is typically expressed as a certain increase in the incidence of adverse outcomes and is referred to as the benchmark response (BMR). In order to distinguish continuous data from dichotomous data, Dekkers *et al.* (2001) recommended the term "critical effect size" (CES) be used instead of the term "BMR," since for continuous data, the effect measure is expressed on a continuous scale. A CES defines the demarcation between non-adverse and adverse changes in toxicological effect parameters for continuous data (Dekkers *et al.* 2001). For example, a CES of 10% or CES_{10} for continuous data (i.e., a 10% change in the mean of a treated group compared to the control mean) is not the same as a BMR of 10% or BMR_{10} (i.e., 10% of total animals responding for dichotomous data).

3.1.4.1.1 Critical Effect Size for Developmental Endpoints – Linear Model

Changes in fetal and placental weight were analyzed using the average fetal or placental weight for each litter. For a decrease in fetal body weight, a CES was defined in terms of a prespecified level of response, corresponding to a 5% relative decrease in the mean when compared to controls (CES_{05}) (Kavlock *et al.* 1995; Allen *et al.* 1996). It was also assumed that a CES_{05} for placental weight was the demarcation between non-adverse and adverse changes, although empirical data are not available for this endpoint. For abnormal sternebrae, reduced ossification for all sites, and increased incidence of supernumerary ribs (usually associated with maternal stress/weight loss), a 5% relative decrease in the mean when compared to controls (CES_{05}) was used based on the findings by Allen *et al.* (1994) that indicated the CES_{05} for malformed fetuses was similar to study NOAELs. The CES results for one standard deviation (SD) ($CES_{1\ SD}$) were calculated and are presented in Table 7 for comparison purposes as suggested by USEPA (2000).

3.1.4.1.2 Critical Effect Size for Maternal Endpoints – Linear Model

A 10% reduction in body weight or organ weight relative to the mean body weight in the control animals (CES_{10}) is typically considered an adverse affect (USEPA 2000; Dekkers *et al.* 2001). It was assumed that a CES_{10} for decreased maternal extragestational weight gain, decreased maternal body weight gain (GD 11-16), whole-body weight (day 18), gravid uterine weight, and extragestational weight was adverse. The $CES_{1\ SD}$ was calculated and is presented in Table 7 for comparison purposes, as suggested by USEPA (2000).

3.1.4.1.3 Unrestricted Power Model and $CES_{1\ SD}$

As shown in Table 7, the differences between BMC_{05} and $BMCL_{05}$ values for fetal/placental endpoints or BMC_{10} and $BMCL_{10}$ values for maternal endpoints using the unrestricted power model ranged from approximately 20- to 100,000-fold (Table 7) which may be due to the unrealistically high slope in the low dose region at the level of the CES_{05} or CES_{10}. Therefore, the $CES_{1\ SD}$ was a more relevant choice for the unrestricted power model because it avoids the steep-slope region (Appendix 2 *Benchmark Modeling Results Using the Power Model (11/19/07 Email from Bruce Allen*) and corresponds to USEPA guidance (2000). A CES of 1 SD from control mean corresponds to an approximately 10% excess risk for individuals below the 2nd percentile or above the 95th percentile of the control distribution for normally distributed effects (USEPA 2000). The BMC_{05} and $BMCL_{05}$ values or BMC_{10} and $BMCL_{10}$ values are presented in Table 7 and in Appendix 2, but are not discussed in the following sections.

3.1.4.2 Benchmark Concentration Modeling

Appendix 2 contains the dose-response data (i.e., dose, mean, SD, number of litters, percent control response, and coefficient of variation) (Tables 2A and 2B) and summary tables of modeling results from BMDS Software (Version 1.4.1c) (Tables 2C, 2D, 2E) for all ten endpoints. Table 7 and Figures 3 and 4 contain a summary of modeling results for the endpoints that could be adequately modeled. Modeling results using the unrestricted polynomial model (i.e., 2nd degree polynomial) produced a nonmonotonic dose-response curve, which is not considered biologically plausible, so unrestricted polynomial model results were not considered. The Hill model was not used because it is not the best choice for estimating the dose-response in the lower end of the data. The Hill model inherently gives too much weight to the higher doses, compromising the fit to the lower doses. Use of the Hill model with only four concentrations resulted in overparameterization of the data (i.e., model estimates of the dose-response curve artificially passed through every data point). The only models that adequately modeled the experimental data with 95% confidence (i.e., goodness of fit p-value and scaled residual values did not imply rejection at the 5% significance level and the model was not over-parameterized) and visual inspection of the dose-response curve indicated an adequate fit were the linear model (i.e., 1st degree polynomial model) and the unrestricted power model (Table 7 and Appendix 2, Tables 2C and 2D). Results from the restricted power model were identical to the linear model. A discussion of BMC modeling results from the linear model and the unrestricted power model is presented below.

Continuous data were modeled using continuous models in USEPA's BMDS software (version 1.4.1c). The TS did not attempt to change continuous data into dichotomous data and model the resulting dose-response curve with dichotomous models. USEPA (2000) noted that when continuous data were changed into dichotomous data, it potentially resulted in loss of information about the magnitude of response. Other investigators have noted the following when modeling continuous data as dichotomized data:

- Kavlock *et al.* (1995) found evidence that the confidence limits on the maximum likelihood estimates were larger when "quantalizing" continuous fetal body weight data;
- Gaylor (1996) found considerable precision was lost upon explicitly dichotomizing the data, even for moderate sample sizes; and

- West and Kodel (1999) noted the implicit approach (i.e., continuous data) gave substantially better results than modeling explicitly dichotomized data for sample sizes in the range of 10-20 animals per dose group, which is the number of pregnant dams in the Hackett *et al.* study (1987b).

3.1.4.2.1 Data Not Amenable to Modeling

According to guidance in USEPA (2000), if the data for an endpoint are not amenable to modeling, the POD will be the statistically-derived study NOAEL. The following endpoints could not be modeled with confidence in either the linear model (all exposure concentrations), linear model (highest concentration excluded), or the unrestricted power model, because the modeling was not acceptable with respect to either test one (i.e., no significant difference (p value > 0.05) between responses and/or variances among the dose levels, so modeling the data with a dose/response curve may not be appropriate) or test four (i.e., the goodness of fit p value was less than 0.1) (Appendix 2, Tables 2C and 2D). That is, for the following endpoints, none of the three models passed test one or none of the three models passed test four (Appendix 2, Tables 2C and 2D). The coefficient of variations were very large for increased incidence of supernumerary ribs, abnormal sternebrae, and reduced ossification for all sites (Appendix 2, Table 2B). The study NOAEL will be used as the POD for the following toxicity endpoints:

- increased incidence of supernumerary ribs (test four); NOAEL = 40 ppm;
- abnormal sternebrae (test one); NOAEL = 200 ppm;
- reduced ossification for all sites (test four); NOAEL = 200 ppm;
- gravid uterine weight (test one); NOAEL = 200 ppm; and
- extragestational weight (test one); NOAEL = 200 ppm.

3.1.4.2.2 Decreased Placental Weight

Decreased placental weight could be adequately modeled with confidence including all four exposure concentrations with the linear model and the unrestricted power model (Table 7 and Figure 3):

- Linear model:
  - BMC_{05} = 344 ppm, $BMCL_{05}$ = 256 ppm
  - $BMC_{1\ SD}$ = 1,063 ppm, $BMCL_{1\ SD}$ = 734 ppm
- Unrestricted power model:
  - $BMC_{1\ SD}$ = 874 ppm, $BMCL_{1\ SD}$ = 233 ppm.

Both a nonhomogeneous and homogeneous variance were used to model the data. The scaled residuals for a nonhomogeneous variance were slightly smaller in the low-dose region of the dose response curve, so the results from a nonhomogeneous variance are reported. The Akaike's Information Criterion (AIC) for the linear model was smaller than the AIC for the unrestricted power model, indicating the most appropriate POD for decreased placental weight is the $BMCL_{05}$ of 256 ppm based on the linear model.

3.1.4.2.3 Decreased Fetal Body Weight

Fetal body weight could be adequately modeled with confidence with the linear model when the highest concentration of 1,000 ppm was eliminated (Table 7 and Figure 3). Both a nonhomogeneous and homogeneous variance were used to model the data. The scaled residuals for a nonhomogeneous variance were slightly smaller in the low-dose region of the dose response curve, so the results from a nonhomogeneous variance are reported. Decreased fetal body weight had a BMC_{05} of 65.8 ppm and $BMCL_{05}$ of 54.7 ppm and a $BMC_{1\ SD}$ of 94.8 ppm and $BMCL_{1\ SD}$ of 71.8 ppm. The POD for decreased fetal body weight is the $BMCL_{05}$ of 54.7 ppm

3.1.4.2.4 Decreased Maternal Extragestational Weight Gain

Decreased extragestational weight gain could be adequately modeled with confidence including all concentrations with the unrestricted power model (Table 7 and Figure 3): $BMC_{1\ SD}$ = 723 ppm and $BMCL_{1\ SD}$ = 51.3 ppm. The POD for decreased extragestational weight gain is the $BMCL_{1\ SD}$ of 51.3 ppm. (Extragestational weight is maternal body weight on GD 18 minus gravid uterine weight. Extragestational weight gain is extragestational weight minus body weight on GD 0.)

3.1.4.2.5 Decreased Maternal Body Weight Gain (GD11-16)

When the highest exposure concentration of 1,000 ppm was eliminated, decreased maternal body weight gain (GD11-16) could be adequately modeled with confidence with the linear model. Decreased maternal body weight gain (GD11-16) could be adequately modeled with confidence including all exposure concentrations with the unrestricted power model (Table 7 and Figure 4):

- Linear model without the highest dose:
    - BMC_{10} = 145 ppm, $BMCL_{10}$ = 94.3 ppm
    - $BMC_{1\ SD}$ = 238 ppm; $BMCL_{1\ SD}$ = 148 ppm
- Unrestricted power model:
    - $BMC_{1\ SD}$ = 392 ppm; $BMCL_{1\ SD}$ = 63.5 ppm

The AIC for the linear model with three doses cannot be compared to the AIC for the unrestricted power model with four doses because the number of doses differ, so the TS chose the $BMCL_{1\ SD}$ of 63.5 ppm from the unrestricted power model because it was the lowest POD, included all concentrations, and captured the nonlinear characteristics of the dose-response relationship. The POD for decreased maternal body weight gain (GD11-16) is the $BMCL_{1\ SD}$ of 63.5 ppm.

3.1.4.2.6 Decreased Maternal Whole Body Weight

Decreased maternal whole body weight could be adequately modeled with confidence including all concentrations with the linear model and the unrestricted power model (Table 7 and Figure 4):

- Linear model:
    - BMC_{10} = 1,344 ppm, $BMCL_{10}$ = 896 ppm;
    - $BMC_{1\ SD}$ = 1,121 ppm, $BMCL_{1\ SD}$ = 732 ppm
- Unrestricted power model:

- $BMC_{1\ SD}$ = 962 ppm and $BMCL_{1\ SD}$ = 304 ppm

The AIC for the linear model was equal to the AIC for the unrestricted power model, so the TS chose the lowest $BMCL_{1\ SD}$ of 304 ppm from the unrestricted power model. The POD for decreased maternal body weight gain (GD11-16) is the $BMCL_{1\ SD}$ of 304 ppm.

Table 7 BMC Modeling Results for Maternal/Developmental Toxicity

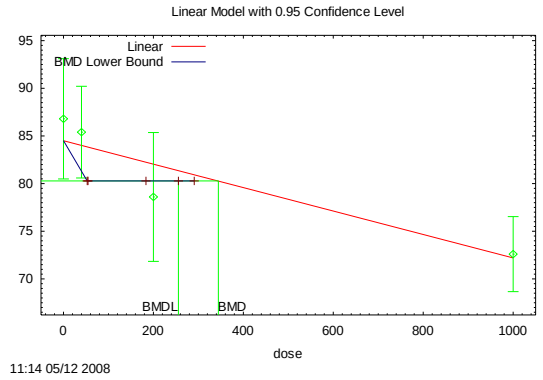
Endpoint	BMD Model / Critical Effect Size	BMC (ppm) / 0.05	BMCL (ppm) / 0.05	BMC (ppm) / 1 SD	BMCL (ppm) / 1 SD	p-value for fit	AIC	Scaled Residual *
Placental weight	Linear **	344	256	1063	734	0.767	466	< |2|
	Power ** (unrestricted)	123	4.17	874	233	0.984	468	< |2|
Fetal body weight	Linear ** without highest dose	65.8	54.7	94.8	71.8	0.350	212	< |2|
	Critical Effect Size	0.10	0.10	1 SD	1 SD			
Maternal whole body weight	Linear	1344	896	1121	732	0.257	321	< |2|
	Power (unrestricted)	1403	599	962	304	0.194	321	< |2|
Maternal body weight gain (GD11-16)	Linear without highest dose	145	94.3	238	148	0.734	153	< |2|
	Power (unrestricted)	108	5.96	392	63.5	0.339	200	< |2|
extra-gestational weight gain	Power (unrestricted)	31.4	0.0000345	723	51.3	0.424	164	< |2|

* All scaled residuals at each concentration were less than an absolute value of 2 (< |2|) (Appendix 2, Table 2E)

** Both a nonhomogeneous and homogeneous variance were used to model the data. The scaled residuals for a nonhomogeneous variance were slightly smaller in the low-dose region of the dose response curve, so the results from a nonhomogeneous variance are reported.

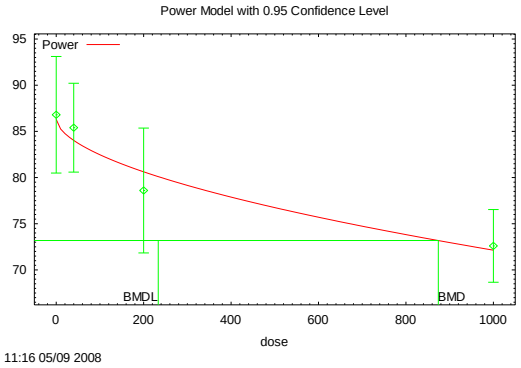
Placental weight

$BMCL_{05}$ = 256 ppm (linear model-four doses)



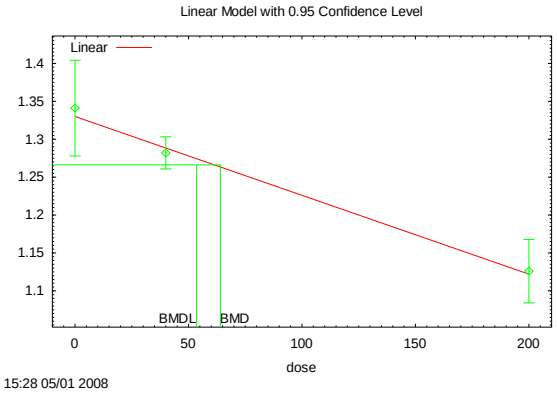
Placental weight

$BMCL_{1\ SD}$ = 233 ppm (unrestricted power model)



Fetal body weight

$BMCL_{05}$ = 54.7 ppm (linear model-three doses)



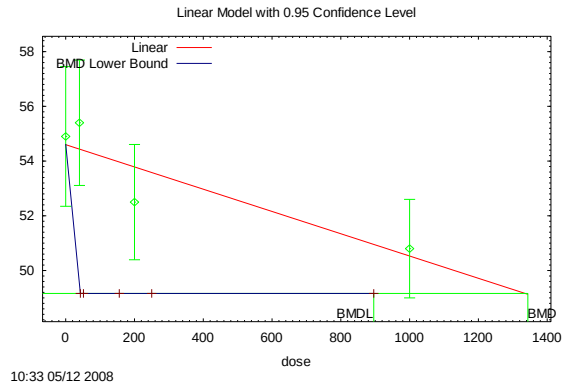
Maternal extragestational weight gain

$BMCL_{1\ SD}$ = 51.3 ppm (unrestricted power model)

Figure 3 BMC Dose-Response Curves for Placental Weight, Fetal Body Weight, and Maternal Extragestational Weight Gain

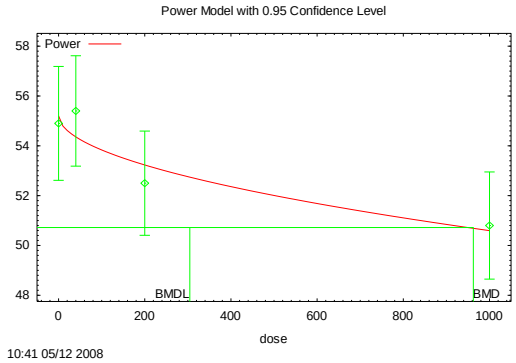
Maternal whole body weight (day 18)

$BMCL_{10}$ = 896 ppm (linear model-four doses)



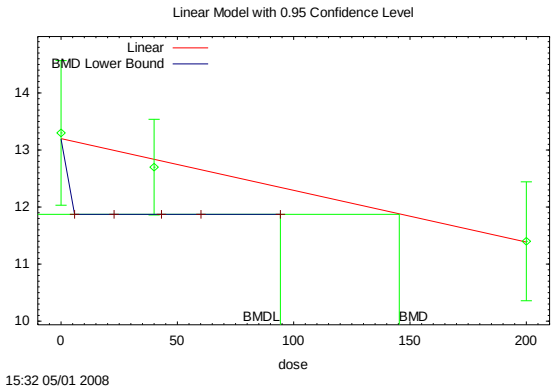
Maternal whole body weight (day 18)

$BMCL_{1\ SD}$ = 304 ppm (unrestricted power model)



Maternal body weight gain (GD11-16)

$BMCL_{10}$ = 94.3 ppm (linear model-three doses)



Maternal whole body weight gain (GD11-16)

$BMCL_{1\ SD}$ = 63.5 ppm (unrestricted power model)

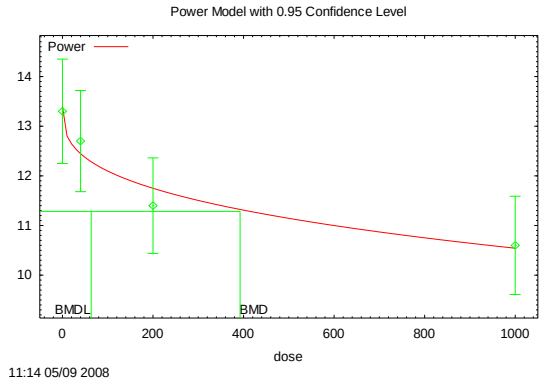


Figure 4 BMC Dose-Response Curves – Maternal Body Weight and Weight Gain

3.1.4.2.7 Summary of Modeling Results

A summary of $BMCL_{05}$ values for developmental effects and $BMCL_{10}$ values for maternal effects from the linear model, and a summary of $BMCL_{1\ SD}$ values from the unrestricted power model, is shown in Table 8 with study NOAELs for comparison. If data from an endpoint cannot be modeled, USEPA (2000) suggests the study NOAEL for that endpoint be used as the POD.

Reduction in maternal extragestational weight gain with a $BMCL_{1\ SD}$ of 51.3 ppm and reduction in fetal body weight with a $BMCL_{05}$ of 54.7 ppm will be the PODs and endpoints selected by the TS to be critical effects. These effects are adverse, relevant PODs to the proposed MOA (i.e., decreased serum progesterone levels) and produced the lowest PODs. Both of these values are comparable to, although slightly higher than, the study NOAEL of 40 ppm.

Table 8 Summary of BMC Modeling

Parameter	**BMCL1 SD Unrestricted power**	**BMCL05 or BMCL10 Linear Model**	**NOAEL**
placental weight	233 ppm	**$BMCL_{05}$= 256 ppm [1, 2]**	40
fetal body weight	--- [4]	**$BMCL_{05}$ = 54.7 ppm [1]**	40
extragestational weight gain	**51.3 ppm [1]**	--- [4]	40
body weight gain (GD 11-16)	**63.5 ppm [1, 3]**	**$BMCL_{10}$ = 94.3 ppm**	40
whole-body weight (day 18)	**304 ppm [1, 3]**	**$BMCL_{10}$= 896 ppm**	200
increased incidence of supernumerary ribs	--- [4]	--- [4]	40 [4]
abnormal sternebrae	--- [4]	--- [4]	200 [4]
reduced ossification for all sites	--- [4]	--- [4]	200 [4]
gravid uterine weight	--- [4]	--- [4]	200 [4]
extragestational weight	--- [4]	--- [4]	200 [4]

1 POD for selected endpoint shown in bold and highlighted cells

2 lowest AIC value

3 lowest BMCL value chosen as POD for selected endpoint

4 toxicity endpoint could not be modeled with confidence (test 4) or trend test failed (test 1)

Increased incidence of supernumerary ribs was a toxicity endpoint that could not be adequately modeled. Hackett *et al.* (1987b) indicated this endpoint is associated with reduced fetal body weight and with maternal toxicity as evidenced by a reduction in maternal weight gain during gestation, which were adequately modeled. The critical effects chosen by the TS are decreased extragestational weight gain with a POD of 51.3 ppm and reduced fetal body weight with a POD of 54.7 ppm, which would also be protective of potential teratogenicity as suggested by increased incidence of supernumerary ribs in mice.

3.1.4.2.8 BMC Modeling Results from USEPA (2002)

USEPA used several different approaches to model fetal body weight dose-response data and reported BMC modeling results adjusted to reflect a 24-h exposure duration (Table 10-13, USEPA 2002). Refer to USEPA (2002) for a complete discussion of the advantages and disadvantages of each model (log-logistic, three-dose continuous power, and hybrid) and cutoff values used by USEPA. The values in Table 10-13 (USEPA 2002) were converted from a 6 h/day exposure to continuous exposure (6/24). In contrast, Table 9 shows the results from Table 10-13 (USEPA 2002), except data are shown for a 6-h/day exposure (i.e., the original duration of the Hackett *et al.* (1987b) study). USEPA's results from the restricted three-dose continuous power model (BMC_{05} = 65.1 ppm and $BMCL_{05}$ = 53.5) (Table 9) are almost identical to results derived by the TS using the three-dose linear model (BMC_{05} = 65.8 and $BMCL_{05}$ = 54.7 ppm) (Table 7) (i.e., modeling results from the restricted continuous power model are the same as the continuous linear model).

Table 9 Fetal Body Weight Modeling (6-h Exposure Duration) *

Model	Response	Cutoff	BMC (ppm)	BMCL (ppm)
Log-logistic (four dose groups)	Individual fetal body weight	BMR = 5th percentile	27.6	11.6
Log-logistic (four dose groups)	Individual fetal body weight	BMR = 10th percentile	40	18.8
Continuous power (three dose groups)	Fetal body weight/litter	CES = 5% relative reduction	65.1	53.5
Continuous power (three dose groups)	Fetal body weight/litter	CES = 25th percentile	5.12	36.7
Continuous power (three dose groups)	Fetal body weight/litter	CES = 0.5 SD absolute reduction	52.4	42.6
Hybrid model (4 dose groups)	Fetal body weight/litter	P_0 = 0.05	28.3	13.3

* Adapted from Table 10-13 (USEPA 2002), except the data are for an exposure duration of 6 h, not 24 h

USEPA (2002) used a 5th percentile BMR and $BMCL_{05}$ of 11.6 ppm as their POD because the log-logistic model:

- fit all four exposure levels adequately;
- accounted for intralitter correlation or litter size; and
- was a more health-protective choice to use for the POD.

For reasons previously discussed in Section 3.1.4.2 *Benchmark Concentration Modeling*, the TS did not consider BMC modeling results from the log-logistic model (which involves converting continuous data to dichotomous data). The advantages and disadvantages of using the hybrid approach to model reduction in fetal body weight after exposure of pregnant dams to BD are discussed by USEPA (2002). For all modeling results, the BMC and BMCL values based on a $CES_{1\ SD}$ are provided (i.e. results equivalent to the hybrid approach*).

3.1.5 Dosimetric Adjustments

The USEPA closely examined the physiologically-based toxicokinetic (PBTK) models for BD to determine if additional modeling could reduce uncertainties in the interspecies scaling between mice and humans for ovarian atrophy and other endpoints (USEPA 2002, Chapter 9). USEPA stated that despite advances in the models over the past decade, the current models are inadequate for this purpose. For example, the PBTK models do not yet accurately describe the distribution of the major metabolites in various compartments, do not yet include the reportedly important epoxydiol metabolites, and have not been adequately validated. A PBTK model not included in USEPA (2002) was developed by Smith *et al.* (2001), who investigated genetic and dietary factors affecting human metabolism of BD. Human volunteers were exposed to 2 ppm BD for a 20-min exposure with a 40-min washout period. Smith *et al.* (2001) fitted a three-compartment PBTK model to investigate BD uptake and estimate model parameters.

Recently, Filser *et al.* (2007) measured and evaluated the BD-dependent blood burden of the following metabolites in rats and mice: EB, DEB, EBD and butene-diol (refer to Figure 2). Brochot *et al.* (2007) conducted a global sensitivity analysis for a proposed PBTK model. However, relevant parameters and a validated PBTK model for extrapolation from animals to humans are still lacking. Therefore, default duration exposure and dosimetric adjustments from animal-to human exposure were used.

3.1.5.1 Critical Effect and Default Exposure Duration Adjustments

Both decreased maternal extragestational weight gain and reduced fetal body weight occurred at similar concentrations and are considered developmental endpoints since they are highly correlated. Since the POD is derived from a developmental endpoint, the exposure duration will not be adjusted to 1 h according to ESL Guidelines (TCEQ 2006) due to potential sensitive windows of exposure. The $BMCL_{1\ SD}$ of 51.3 ppm based on the Hackett *et al.* (1987b) study for reduction in extragestational weight gain is used as the POD since it is slightly lower than the $BMCL_{05}$ of 54.7 ppm for decreased fetal body weight and is adverse, biologically plausible, and consistent with the proposed MOA.

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* A CES of 1 SD from control mean corresponds to an approximately 10% excess risk for individuals below the 2nd percentile or above the 95th percentile of the control distribution for normally distributed effects (USEPA 2000).

3.1.5.2 Default Dosimetry Adjustments from Animal-to-Human Exposure

BD is only slightly soluble in water and is moderately soluble in blood (USEPA 2002). It is readily absorbed from the air into the blood through the lungs. The health effects it produces at lower concentrations are mainly remote effects, so dosimetric adjustments were performed as a

Category 3 gas which is consistent with USEPA (2002) and based on guidance in USEPA (1994). For Category 3 gases, the default dosimetric adjustment from animal-to-human exposure is conducted using the following equation:

$POD_{HEC} = POD_{ADJ} \times [(H_{b/g})_A / (H_{b/g})_H]$

where:

$H_{b/g}$ = ratio of the blood:gas partition coefficient

A = animal

H = human

For BD, the blood:gas partition coefficients for mice range from 1.2 to 3.0 with a mean of 1.67 (Appendix 3 of USEPA 2005a) and for humans 1.22 $\pm$ 0.30 (mean $\pm$ SD) (Brochot *et al.* 2007). When $(H_{b/g})_A / (H_{b/g})_H$, > 1, a default value of 1 is used for $(H_{b/g})_A / (H_{b/g})_H$, the regional gas dose ratio (RGDR) (USEPA 1994).

Reduction in extragestational weight gain

$POD_{HEC} = POD_{6\,h} \times RGDR = 51.3 \text{ ppm} \times 1 = 51.3 \text{ ppm}$

3.1.6 Adjustments of the POD_{HEC}

The MOA by which BD produces maternal/developmental toxicity is assumed to be nonlinear (Section 3.1.2.2), so a POD was determined and uncertainty factors (UFs) were applied to derive a ReV. The following UFs were applied to the 6-h POD_{HEC} of 51.3 ppm: 10 for intraspecies variability (UF_H), 3 for extrapolation from animals to humans (UF_A), and 1 for database uncertainty (UF_D), a total UF = 30:

Reduction in extragestational weight gain

$$\begin{aligned} \text{acute ReV} &= POD_{HEC} / (UF_H \times UF_A \times UF_D) \\ &= 51.3 \text{ ppm} / (10 \times 3 \times 1) \\ &= 1.71 \text{ ppm} \\ &= 1{,}710 \text{ ppb} \end{aligned}$$

A full UF_H of 10 was used to account for intraspecies variability. There is experimental evidence that indicates BD-sensitive human subpopulations may exist due to metabolic genetic polymorphisms (USEPA 2002), although recent studies indicate that variability due to genetic polymorphisms is less than 10 based on metabolism of BD in humans with different genotypes. While the results examining metabolic differences between humans with different genotypes in some cases are inconsistent, overall, the differences between genotypes have been small (i.e., generally a factor of two to four) (Albertini *et al.* 2001, 2003; Begemann *et al.* 2001; Fustinoni *et al.* 2002; Hayes *et al.* 1996, 2000, 2001; Smith *et al.* 2001; and Zhao *et al.* 2000, 2001).

A UF_A of 3 was used for extrapolation from animals to humans* because default dosimetric adjustments from animal-to-human exposure were conducted, which account for toxicokinetic differences but not toxicodynamic differences. This approach is likely conservative, since

existing studies indicate that mice are relatively sensitive laboratory animals in regards to the reproductive effects of BD (e.g., greater production of toxic intermediates and a lower capacity for detoxification of these intermediates (USEPA 2002)).

A database UF_D of 1 was used because the overall acute toxicological database for BD meets the requirements for a high confidence database for an acute ReV (TCEQ 2006):

- acute inhalation studies in humans;
- two inhalation bioassays in different species investigating a wide range of endpoints; and
- two prenatal developmental toxicity studies in different species (USEPA 2002; AEGL 2005).

Both the quality of the studies and the confidence in the acute database is high.

3.1.7 Health-Based Acute ReV and ^{acute}ESL

The 6-h acute ReV value of 1,710 ppb was rounded to two significant figures at the end of all calculations resulting in an acute ReV of 1,700 ppb (3,700 µg/m^3). The rounded acute ReV was then used to calculate the 6-h ^{acute}ESL. At the target hazard quotient of 0.3, the 6-h ^{acute}ESL is 510 ppb (1,100 µg/m^3) (Table 10). This acute ReV and ^{acute}ESL are considered to be conservative since pregnant mice exposed to BD and their offspring develop maternal/developmental toxicity much easier than similarly- exposed rats, available scientific information suggests mice are more sensitive than humans, and BD-induced reproductive/ developmental effects have never been observed in humans.

* For the chronic assessment, as discussed in Section 4.1.5.2 *Toxicokinetic Adjustments from Animal-to-Human Exposure*, the total UF_A for ovarian atrophy was reduced to 1 based on strong MOA evidence that DEB (not EB or BD) causes ovarian atrophy (Doerr *et al.* 1995) and toxicokinetic data that DEB levels in mice are much higher than in humans (Section 4.1.5.2). Doerr *et al.* (1995) investigated ovarian atrophy in both mice and rats after exposure to BD, EB, and DEB. However, for the acute assessment, there is not strong evidence that DEB alone is responsible for reproductive/developmental effects because Spencer *et al.* (2001) and Chi *et al.* (2002) only evaluated the effects of DEB in rats. Therefore, a full toxicodynamic UF_A of 3 was used.

Table 10 Derivation of the Acute ReV and ^{acute}ESL

Parameter	Summary
Study	Hackett *et al.* 1987b
Study population	CD-1 mice (18-21 pregnant mice per dose group)
Study quality	High
Exposure Methods	0, 40, 200, and 1,000 ppm on gestation days (GD) 6-15 for 6 h/day
Critical Effects	Reduction in extragestational weight gain and fetal body weight; developmental toxicity
POD	51.3 ppm ($BMCL_{1\ SD}$)
Exposure Duration	6 h
Extrapolation to 1 h	No adjustment because the critical effect was a maternal/developmental endpoint
POD (6 h)	51.3 ppm
6-h POD_{HEC}	51.3 ppm (gas with systemic effects, based on default RGDR = 1.0)
Total uncertainty factors (UFs)	30
Interspecies UF	3
Intraspecies UF	10
LOAEL UF	Not applicable
Incomplete Database UF	1
Database Quality	High
acute ReV [6 hr] (HQ = 1)	**3,700 µg/m^3 (1,700 ppb)**
^{acute}ESL [6 h] (HQ = 0.3)	**1,100 µg/m^3 (510 ppb)**

3.1.8 Comparison of ^{acute}ESL to Generic ESL

When a subacute study is used to derive a 1-h ^{acute}ESL, Section 3.2.3 of the ESL guidelines (TCEQ 2006) suggests a generic ESL ($^{acute}ESL_{generic}$) be derived using approaches in Section 3.6 for comparison to the 1-h ^{acute}ESL to ensure the derived value is not overly conservative. The Threshold of Concern (TOC) approach utilizes the lowest reported inhaled concentration which produced death in 50% of the study specimens after exposure (LC_{50}). Shugaev (1969) reported the 2-h LC_{50} of BD in mice was 122,000 ppm and the 4-h LC_{50} in rats was 128,000 ppm which would classify BD as a TOC Category 5 gas, and the corresponding $^{acute}ESL_{generic}$ would be 1,000 µg/m^3 for a 1-h exposure duration (Table 3-3 of the ESL guidelines (TCEQ 2006)). The 6-h ^{acute}ESL of 1,100 µg/m^3 based on the subacute study cannot be directly compared to the 1-h $^{acute}ESL_{generic}$ because the exposure durations are different. However, the 6-h ^{acute}ESL is slightly higher than the 1-h $^{acute}ESL_{generic}$ of 1,000 µg/m^3 for a Category 5 gas. This provides confidence that the derived value is not overly conservative.

3.2. Welfare-Based Acute ESLs

3.2.1 Odor Perception

ACGIH (2001) reports BD has a mildly aromatic odor with recognition occurring at 1 to 1.6 ppm. Ruth (1986) states the 50% odor detection threshold is 352 µg/m^3 (160 ppb) and the 100% recognition threshold is 2,860 µg/m^3 (1,300 ppb). The 50% odor detection threshold for BD determined by the triangular odor bag method was 230 ppb (Nagata 2003). Both Ruth (1986) and Nagata (2003) are listed as sources of information for odor thresholds in Appendix B of the ESL Guidelines (TCEQ 2006). However, only the Nagata (2003) study meets the criteria for acceptable odor threshold measurement techniques developed by the American Industrial Hygiene Association (TCEQ 2006). Therefore, the $^{acute}ESL_{odor}$ is 230 ppb (510 µg/m^3). Since odor is a concentration-dependent effect, the same 1-h $^{acute}ESL_{odor}$ is assigned to all averaging times.

3.2.2 Vegetation Effects

BD concentrations that produce vegetative effects, such as abscission and inhibition of growth, are orders of magnitude higher than concentrations of ethylene, propylene, and acetylene that produce similar effects (USDHEW 1970). Since concentrations producing vegetative effects (approximately > 10,000 ppm) are significantly above other health- and odor-based concentrations, an $^{acute}ESL_{veg}$ was not developed for BD.

3.3. Short-Term ESL and Values for Air Monitoring Evaluation

The acute evaluation resulted in the derivation of the following values:

- 6-h $^{acute}ESL$ = 1,100 µg/m^3 (510 ppb)
- 6-h acute ReV = 3,700 µg/m^3 (1,700 ppb)
- 1-h $^{acute}ESL_{odor}$ = 510 µg/m^3 (230 ppb)

The short-term ESL for air permit evaluations is the $^{acute}ESL_{odor}$ = 510 µg/m^3 (230 ppb) as it is lower than the health-based 6-h $^{acute}ESL$ of 1,100 µg/m^3 (510 ppb) (Table 1). If the predicted 1-h maximum ground level concentration (GLC_{max}) is less than the health-based 6-h $^{acute}ESL$, then no acute health effects would be expected. If the GLC_{max} exceeds the health-based 6-h $^{acute}ESL$, then it will be necessary to calculate a 6-h GLC_{max} in order to evaluate potential health effects.

For the evaluation of ambient air monitoring data, the $^{acute}ESL_{odor}$ of 510 µg/m^3 (230 ppb) is lower than the acute ReV of 3,700 µg/m^3 (1,700 ppb), although both values may be used for the evaluation of ambient air monitoring data (Table 1). The $^{acute}ESL$ (HQ = 0.3) is not used to evaluate ambient air monitoring data. If measured 1-h ambient air monitoring data is less than the 6-h acute ReV, then no acute health effects would be expected. If the health-based 6-h acute ReV is exceeded, and it is possible to calculate a 6-h value (i.e., automatic gas chromatographic data), then a 6-h averaged value will be calculated in order to evaluate potential health effects.

3.4 Comparison of TCEQ's Acute ReV versus USEPA's Acute Reference Concentration

USEPA (2002) derived a 24-h acute reference concentration (RfC) of 3.2 µg/m^3 (7 ppb) based on decreased fetal body weight. A value of 2.9 ppm for a 24-h POD_{HEC} is reported in Table 10-25 of USEPA (2002) using the log-logistic model (i.e., continuous data was changed into dichotomous data and modeled with a dichotomous model). USEPA applied UFs of 3 for interspecies variability, 10 for intraspecies variability, 4 for effect level extrapolation factor (to decrease risk to below the benchmark response level; analogous conceptually to the LOAEL-to-NOAEL UF), and 3 for incomplete database because a neurodevelopmental toxicity study had not been completed (total UF = 400) (Table 11).

The TS evaluated ten different toxicity endpoints using BMC modeling. The acute ReV for a 6-h exposure duration is based on decreased extragestational weight gain with a POD being 51.3 ppm, although reduction in fetal body weight had a similar POD of 54.7. A UF of 3 was applied for interspecies extrapolation and 10 for intraspecies variability (total UF = 30). An effect level extrapolation factor (somewhat equivalent to a LOAEL-to-NOAEL UF) was not applied because BMC modeling was used to determine the POD, considered an appropriate NOAEL surrogate. An acute database UF was not applied because the acute database for BD meets the minimum database with high confidence for an acute ReV (TCEQ 2006). Table 11 compares the derivation of the 6-h acute ReV and 6-h ^{acute}ESL to USEPA's 24-h acute RfC (USEPA 2002).

Table 11 Table 11. Acute ReV Compared to USEPA's RfC

POD_{HEC}	**Inter-species**	**Intra-species**	**Effect Level Extrapolation Factor**	**Incomplete Database**	**Total UF**	**Acute Reference Value**
TCEQ 51.3 [6 h] [1] Decreased extragestational weight gain	3	10	---	---	30	acute ReV [6 h] 1,700 ppb ^{acute}ESL [6 h] 510 ppb
USEPA 2.9 ppm [24 h] [2] Decreased fetal body weight	3	10	4	3	400	acute RfC [24 h] 7 ppb

[1] Lowest adverse POD determined from an evaluation of ten toxicity endpoints

[2] The unadjusted 6-h $BMCL_{05}$ for decreased fetal body weight was 11.6 ppm using a log-logistic BMC model (continuous data was converted to dichotomous data)

USEPA's RfC is approximately 240 times lower than TCEQ's ReV due to the following reasons:

- The exposure duration for USEPA's RfC is 24 h, whereas the exposure duration for TCEQ's ReV is 6 h, which makes USEPA's RfC approximately four times lower;
- The TCEQ did not use an effect level extrapolation factor of 4 because BMC modeling was used to determine the POD, considered an appropriate NOAEL surrogate, or a UF_D of 3 which makes USEPA's RfC 12 times lower; and
- The TCEQ used the $BMCL_{1\ SD}$ of 51.3 ppm as a POD for reduction in extragestational weight gain because it was the lowest POD of adverse effects based on BMC analysis of ten toxicity endpoints, whereas USEPA used the $BMCL_{05}$ from a log-logistic model (i.e., continuous data for fetal body weight was converted into dichotomous data), which makes USEPA's RfC approximately 4.4 times lower.

Consideration of the above differences accounts for approximately a 210-fold difference (4 x 12 x 4.4). While an exact partitioning of the 240-fold difference may not be possible, there are science-based and logical explanations accounting for most of the differences.

Chapter 4 Chronic Evaluation

4.1 Noncarcinogenic Potential

4.1.1 Physical/Chemical Properties and Key Studies

Refer to Section 3.1.1.1 for a discussion of physical/chemical properties.

This section is based on USEPA (2002). Chapter 5 of USEPA (2002) discusses the chronic reproductive/developmental effects of BD. Animal data indicate that BD is a potential

reproductive hazard because reproductive effects are observed at the lowest concentrations tested in animals. Chapter 6 of USEPA (2002) discusses other subchronic and chronic health effects observed in animals exposed to BD. Few adverse noncarcinogencic effects have been observed other than reproductive and developmental effects, except for hematological effects in mice exposed to higher concentrations and increases in organ weights in rats (USEPA 2002, Chapter 6). Hematological effects in mice may not be relevant for humans, as demonstrated by Tsai *et al.* (2005). Tsai *et al.* (2005) conducted a hematology surveillance study of petrochemical workers at two Shell facilities and reported there were no significantly increased abnormalities for any hematology parameter among exposed employees (404 exposed employees and 733 comparison employees).

A review of the scientific literature since 2002 did not reveal any other chronic inhalation studies that could be used instead of the 2-year chronic bioassays conducted by the National Toxicology Program (NTP 1993) which are summarized in the following sections but discussed in detail in USEPA (2002).

4.1.1.1 Human Studies

Albertini *et al.* (2007) conducted a molecular epidemiological study of BD-exposed Czech workers to compare female to male responses as discussed previously in Section 3.1.1.2.1. Briefly, there were no significant differences reported between control and exposed groups for miscarriages, still births, ectopic pregnancies, molar pregnancies, low birth weight (< 2,500 g) babies, or pre-term births, based on information collected on all pregnancies. The ability of the study to detect differences in the evaluated endpoints may be limited because there were only a few subjects evaluated.

4.1.1.2 Animal Studies

The most sensitive reproductive effects observed in 2-year chronic exposure studies were ovarian atrophy in female mice and testicular atrophy in male mice (NTP 1993). Testicular atrophy was primarily a high-exposure effect, so this section focuses on ovarian atrophy. In this bioassay, groups of 70 female B6C3F1 mice were exposed by inhalation 6 h/day, 5 days/week to 0, 6.25, 20, 62.5, or 200 ppm BD for up to 103 weeks, and groups of 90 female mice were exposed to 625 ppm. An interim evaluation of ovarian atrophy was conducted at 9 months on ten mice per group and also at 15 months. Significant concentration-related decreases in survival were seen in female mice exposed to concentrations $\geq$ 20 ppm, primarily due to the development of malignant neoplasms. Statistically significant increases in the incidence of ovarian atrophy were observed in all exposure groups following lifetime exposures. The LOAEL for ovarian atrophy was observed at the lowest exposure level (6.25 ppm, 6 h/day, 5 days/week, for 2 years). Uterine atrophy was also observed in the highest exposure groups; however, this is likely to be a secondary effect of ovarian atrophy. Rats exposed to 0, 1,000, and 8,000 ppm did not develop adverse reproductive effects, thus providing further evidence that rats are less sensitive to the effects of BD than mice (HLE 1981; Owen *et al.* 1987; Owen and Glaister 1990).

4.1.2 MOA Analysis

Refer to Section 3.1.2 for a discussion of BD metabolism. There is strong evidence that ovarian atrophy is mediated by the diepoxide metabolite, DEB, the most reactive of BD metabolites (Doerr *et al.* 1995, 1996; USEPA 2002). There are marked species differences in effects seen

between rats, which do not exhibit BD-induced ovarian atrophy, and mice, which do exhibit BD-induced ovarian atrophy. Doerr *et al.* (1995, 1996) evaluated the ovarian effects of the metabolites of BD in mice and rats and also examined 4-vinylcyclohexene, a structurally similar compound. Doerr *et al.* (1995, 1996) showed that the diepoxide of BD or 4-vinylcyclohexene is required for ovarian toxicity to occur in the rat. EB was ovotoxic to mice but not rats. Thus, the resistance of the rat to ovarian toxicity of BD is likely due to the decreased ability of the rat to produce DEB. Filser *et al.* (2007) was unable to detect DEB in venous blood of male Sprague-Dawley rats (detection limit 0.01 µmol/l) when they were exposed to 1,200 ppm for 6-8 h, whereas DEB was detected in B6C3F1 mice at 3.2 µmol/l at 1,280 ppm BD. Humans are similar to rats in that they do not readily produce the diepoxide metabolite (refer to Section 4.1.5.2.2 *Estimate for the Toxicokinetic UF_A Based on Empirical Data).*

Swenberg *et al.* (2007) compared results in Czech Republic occupationally-exposed workers to results in mice and rats for a N,N-(2,3-dihydroxy-1,4-butadiyl) valine (pry-Val) hemoglobin adduct specific for DEB at similar BD concentrations (Table 12). The pry-Val adduct was not detected in human females or males, while female mice were 78 times more likely than human females to produce DEB as evaluated with pry-Val adducts (Table 12). Pry-Val adducts for human females were based on the limit of quantitation (LOQ) because pry-Val adducts were not detected (Swenberg *et al.* 2007). At the 2007 and 2008 Society of Toxicology meetings, Georgieva *et al.* (2007; 2008) presented results using a more sensitive analytical method to measure pry-Val adducts in the Czech Republic workers. Pry-Val adducts were detected at low concentrations in Czech Republic workers. There was not a clear dose-response relationship between pry-Val adducts and BD concentrations from the Georgieva *et al.* (2007) study, and the authors hypothesized that the pry-Val adducts could have been formed from other unknown sources. However, the Georgieva *et al.* (2008) study showed the amount of pyr-Val was significantly higher in the polymerizatrion workers than in the monomer workers and controls.

Table 12 DEB-Specific pyr-Val Hb Adduct in Mouse, Rat, and Human (Swenberg *et al.* 2007)

Concentration	1 ppm BD 6 h/day 4 weeks (4.0 ppm-weeks)	1 ppm BD 6 h/day 4 weeks (4.0 ppm-weeks	1 ppm BD 6 h/day 4 weeks (4.0 ppm-weeks)	1 ppm BD 6 h/day 4 weeks (4.0 ppm-weeks)	Mean 0.18 ppm for 4 months (3.1 ppm-weeks)	Mean 0.37 ppm for 4 months (6.3 ppm-weeks)
Species	Female mice	Male mice	Female rat	Male rat	Female human	Male human
Pyr-VAL Hb adducts (pmol/g in 50 mg globin)	23.5 ± 3.1 female mice have 78 times more pyr-Val adducts than female humans	30.8 ± 4.6 male mice have 103 times more pyr-Val adducts than male humans	0.7 ± 0.1	0.9 ± 0.03	< 0.3 limit of quantitation (LOQ)	< 0.3 LOQ

4.1.3 Dose Metric

For ovarian atrophy, data on the exposure concentration of the parent chemical are available, whereas data on more specific dose metrics, such as the monoepoxide or diepoxide metabolites in blood or target tissue, are not available. As discussed previously in Section 3.1.5, a validated PBTK model for extrapolation from animals to humans is still lacking. Therefore, the exposure concentration of the parent chemical was used as the default dose metric.

4.1.4 PODs for Key Studies and Critical Effect

Using benchmark concentration dose modeling and a Weibull time-to-response model, USEPA (2002) calculated a BMC_{10} of 1.05 ppm and $BMCL_{10}$ of 0.88 ppm based on the 1993 NTP 2-year inhalation bioassay, including interim sacrifice data. In calculating the BMC_{10} and $BMCL_{10}$, lesion severity was not taken into account, and the 625 ppm group was excluded because of high early mortality. In addition, ovarian atrophy was modeled to reflect extra risks only until age 50, because BD-induced ovarian atrophy is believed to result from follicular failure, and after menopause, follicles would no longer be available.

The PODs for all prenatal deaths (dominant lethal effect) ($BMCL_{05}$ = 10 ppm) and for testicular atrophy ($BMCL_{10}$ = 16 ppm) were also determined by USEPA (2002) and were significantly higher than the $BMCL_{10}$ of 0.88 ppm. Therefore, ovarian atrophy was selected as the critical effect (USEPA 2002).

Sielken *et al.* (Appendix 3) repeated the BMC modeling performed by USEPA using the same procedures described above and calculated the BMC_{05} and $BMCL_{05}$ as well as the BMC_{10} and $BMCL_{10}$ (Appendix 3). The $BMCL_{05}$ has generally been considered a conservative NOAEL surrogate (Barnes *et al.* 1995; Fowles *et al.* 1999; Filipsson *et al.* 2003) whereas the $BMCL_{10}$

may be analogous to a NOAEL or LOAEL. The BMC_{10} and $BMCL_{10}$ calculated by Sielken *et al.* (Appendix 3) were 1.15 ppm and 0.881 ppm, respectively, which agreed with the BMC_{10} of 1.05 ppm and $BMCL_{10}$ of 0.88 ppm calculated by USEPA (2002).

USEPA (2002) analyzed ovarian atrophy data excluding the highest dose group and also including all the data. Traditionally, EPA drops the highest dose group when the model does not fit the data well or when quantal data are fit with a quantal model and there is high mortality in the highest dose group. The ovarian atrophy data, however, were modeled with a time-to-response model (i.e., a model that accounts for the time of death) as opposed to a quantal model which does not account for time of death. Furthermore, the model fit to the data that excluded the highest dose group was not better than the model fit to the data that included the highest dose group as shown by Sielken *et al.* (Appendix 3). However, USEPA (2002) excluded the highest dose group because of early mortality. The BMC_{05} and $BMCL_{05}$ were 0.560 ppm and 0.429 ppm, respectively, excluding the highest dose and 0.607 ppm and 0.462 ppm, respectively, including the highest dose. Since a time-to-response model was used, the TS used the $BMCL_{05}$ modeling result of 0.462 ppm as the POD (uses all the data).

Because the Weibull time-to-response model in these analyses is linear in dose, the BMC_{05} and $BMCL_{05}$ values are approximately half the corresponding BMC_{10} and $BMCL_{10}$ values. The values of BMC_{05} and $BMCL_{05}$ can be used if the dose-response relationship below the lowest experimental dose is believed to be the linear Weibull time-to-response model fit to the data. The assumption of linearity below the lowest experimental dose is usually conservative and, therefore, health protective. However, there is less uncertainty behind the benchmark dose methodology when it is used to identify the POD (BMC_{05} and $BMCL_{05}$) within the range of the experimental data (the range of the non-zero doses in the experimental data) and to be a dose whose risk can be reasonably reliably estimated without undue sensitivity to the dose-response model selected or the model estimation. Here, the BMC_{05} and $BMCL_{05}$ are below the range of the experimental data and, hence, introduce an additional element of uncertainty into the POD. However, the $BMCL_{05}$ for ovarian atrophy was used as the POD because the TS preferentially uses a benchmark response level of 5% for more severe effects such as ovarian atrophy, and the $BMCL_{05}$ is considered to be a conservative NOAEL surrogate (TCEQ 2006).

4.1.5 Dosimetric Adjustments

Based on the summary of information in Section 3.1.5 and the detailed discussion in USEPA (2002, Chapter 9), default duration exposure from animal-to-human exposure were not used. Instead, empirical data were used to estimate BD-specific toxicokinetic adjustments from animal-to-human exposure.

4.1.5.1 Default Exposure Duration Adjustments

The $BMCL_{05}$ = 0.462 ppm for ovarian atrophy (Appendix 3) represents exposure concentrations that were already adjusted from discontinuous to continuous exposures.

4.1.5.2 Toxicokinetic Adjustments from Animal-to-Human Exposure

The following sections discuss methods for a toxicokinetic adjustment from animal-to-human exposure as opposed to a toxicodynamic adjustment. The standard toxicokinetic UF is 3 and the toxicodynamic UF is 3 for a total UF_A = 10. If default toxicokinetic dosimetry adjustments from

animal-to-human exposure based on procedures in USEPA (1994) are used, a toxicokinetic UF_A of 1 may be justified as demonstrated in Section 4.1.5.2.2. However, there is empirical evidence to indicate that the toxicokinetic UF is considerably less than 1 because mice metabolize BD to the reactive metabolite DEB much more than humans as discussed in Section 4.1.2 MOA Analysis. Although the experimental data are not sufficient to develop a chemical-specific adjustment factor (CSAF) for BD, it would support a UF_A substantially less than 1. The toxicokinetic UF_A that will be used by the TS is 0.3, although it may be substantially less than 0.3, as discussed below. If a BD-specific toxicokinetic UF = 0.3 is used with the standard toxicodynamic UF = 3, the total UF_A = 1.

4.1.5.2.1 Default Dosimetry Adjustments from Animal-to-Human Exposure

As discussed previously in Section 3.1.5.2, dosimetric adjustments were performed as a Category 3 gas which is consistent with USEPA (2002) and based on guidance in USEPA (1994) with a RGDR = 1:

$$POD_{HEC} = POD_{ADJ} \times RGDR$$
$$= 0.462 \text{ ppm} \times 1$$
$$= 0.462 \text{ ppm}$$

The toxicokinetic UF_A using these default procedures is 1. However, procedures discussed in Section 4.1.5.2.2 were used to justify a toxicokinetic UF_A less than 1.

4.1.5.2.2 Estimate for the Toxicokinetic UF_A Based on Empirical Data

Humans produce much lower levels of DEB than mice as demonstrated by experimental data on DEB-specific pyr-Val Hb adducts (Section 4.1.2) and urinary metabolites (Sabourin *et al.* 1992 as reviewed by Henderson *et al.* 1996 and Henderson 2001)). DEB is the BD metabolite responsible for ovarian atrophy (Section 4.1.2; USEPA 2002). The toxicokinetic UF_A may range from less than 0.2 to 0.01 based on data discussed in Sections 4.1.5.2.2.1 to 4.1.5.2.2.3. There is uncertainty in these estimates since data on a more specific dose metric in humans and mice (i.e., area under the curve DEB blood concentration or tissue DEB concentration) are not available. The TS will use a toxicokinetic UF_A of 0.3 based on the following experimental data:

- comparison of specific pyr-Val Hb adducts in humans and mice (Section 4.1.5.2.2.1);
- comparison of the total butadiene metabolites in blood from monkeys and mice (Section 4.1.5.2.2.2);
- comparison of DEB blood concentrations from rats and mice (Section 4.1.5.2.2.3); and
- comparison of DEB tissue levels from rats and mice (Section 4.1.5.2.2.3).

4.1.5.2.2.1 Human-to-mouse experimental data

Swenberg *et al.* (2007) noted humans form 100-times less pyr-Val adducts than similarly exposed mice, a humans-to-mouse ratio of 0.01. At the present time, procedures for developing a chemical-specific-adjustment factor based on pyr-Val Hb adducts are not available because the rate constant for the association between DEB and Hb adducts is unknown (i.e., the DEB blood concentration area under the curve, the dose metric appropriate for chronic exposure, cannot be estimated, see example from Fennell *et al.* 2005 for acrylamide).

4.1.5.2.2.2 Monkey-to-mouse experimental data

Sabourin *et al.* 1992 (as reviewed by Henderson *et al.* 1996 and Henderson 2001) showed that monkeys and humans had similar urinary excretion of the M1 and MII metabolites of BD. Dahl and Henderson (2000) showed the *in vitro* metabolism of BD by hepatic microsomes from cynomolgus monkeys and humans is similar. This indicates experimental data in monkeys may be applicable to humans since they metabolize BD similarly. Dahl *et al.* (1990; 1991) demonstrated that the uptake of BD as a result of metabolism was much lower in monkeys than in mice or rats. For equivalent inhalation exposures, the concentrations of total BD metabolites in the blood were 5-50 times lower in the monkey than in the mouse, a monkey-to-mouse ratio of 0.2 to 0.02. These results indicate that epoxide levels in monkey tissue would be lower than mouse tissue since blood epoxide concentrations were lower in the monkey than in rats or mice (Dahl *et al.* 1991).

4.1.5.2.2.3 Rat-to-mice experimental data

For the purpose of approximating a bounding estimate of UF_A between mice and humans, a comparison of rat data to mice data may be informative. Primates and humans metabolize BD more similarly to rats than mice (Henderson *et al.* 1996; Henderson 2001). Swenberg *et al.* (2007) demonstrated humans form at least 3-times less pyr-Val than similarly exposed rats, and Dahl *et al.* (1991) showed total BD metabolites in the blood were 4-14 times lower in monkey than in the rat. Several investigators have measured DEB blood levels in rats and mice (reviewed in Filser *et al.* 2007). There was a difference in DEB blood concentrations between mice and rats of more than one order of magnitude based on data from several laboratories, when exposed to around 65 ppm BD, a rat-to-mice ratio < 0.1. Thornton-Manning *et al.* (1995) demonstrated that DEB-tissue levels in mice were 40- to 163-fold greater than those in rats (4-h exposure to around 65 ppm), a rat-to-mice ratio of 0.025 to 0.0006.

4.1.6 Adjustments of the POD_{HEC}

The MOA by which BD produces ovarian atrophy is metabolism of the parent compound to DEB (Section 4.1.2), which is considered a threshold, nonlinear MOA. Therefore, a POD was determined and UFs applied to derive a ReV. The following UFs were applied to the POD_{ADJ} of 0.462 ppm: 10 for intraspecies variability (UF_H), 1 for extrapolation from animals to humans (UF_A), 1 for extrapolation from a LOAEL-to-NOAEL (UF_L) and 3 for database uncertainly (UF_D), a total UF = 30:

$$\text{Chronic ReV} = POD_{ADJ} / (UF_H \times UF_A \times UF_L \times UF_D)$$
$$= 0.462 \text{ ppm} / (10 \times 1 \times 1 \times 3)$$
$$= 0.0154 \text{ ppm}$$

- A full UF_H of 10 was used to account for intraspecies variability. There is experimental evidence to indicate that BD-sensitive human subpopulations may exist due to metabolic genetic polymorphisms (USEPA 2002), although the differences between genotypes have generally been a factor of two to four as previously discussed in Section 3.1.6.1.
- The UF_A is composed of a toxicokinetic and toxicodynamic component. A toxicokinetic UF_A of 0.3 was used for extrapolation from animal to human based on empirical data (Section 4.1.5.2.2). A toxicodynamic UF_A of 3 was used because the key sequence of

events and understanding of how DEB interacts in different species to produce ovarian atrophy is not available. The resulting total UF_A was 1.

- A UF_L of 1 was used because BMC modeling was performed to determine a POD based on the $BMCL_{05}$ (TCEQ 2006).
- The toxicological database for BD is extensive. However, a UF_D of 3 was applied because of the absence of a multigenerational reproductive study, consistent with USEPA (2002).
- Both the quality of the studies and the confidence in the chronic database is high.

4.1.7 Health-Based Chronic ReV and $^{chronic}ESL_{nonlinear(nc)}$

The chronic ReV value based on ovarian atrophy was rounded to two significant figures at the end of all calculations resulting in a chronic ReV of 15 ppb (33 µg/m^3). The rounded chronic ReV was then used to calculate the $^{chronic}ESL_{nonlinear(nc)}$. At the target hazard quotient of 0.3, the $^{chronic}ESL_{nonlinear(nc)}$ is 4.5 ppb (9.9 µg/m^3) (Table 13).

Table 13 Derivation of the Chronic ReV and $^{chronic}ESL_{nonlinear(nc)}$

Parameters	**Summary**
Study	2-year bioassays (NTP 1993)
Study Population	70 female B6C3F1 mice; 90 female mice
Study Quality	high
Exposure Method	103 week exposures via inhalation at 0, 6.25, 20, 62.5, or 200 ppm; 90 female mice exposed to 625 ppm
Critical Effects	ovarian atrophy in female mice
POD (original animal study)	Not available. BMD modeling was conducted on data already adjusted from discontinuous to continuous exposure
Exposure Duration	6 h/day, 5 days/week, for 2 years
Extrapolation to continuous exposure (POD_{ADJ})	0.462 ppm ($BMCL_{05}$)
POD_{HEC}	0.462 ppm Adjustment not applicable; a toxicokinetic UF_A based on empirical data was used
Total UFs	30
Interspecies UF	1
Intraspecies UF	10
LOAEL UF	1
Subchronic to chronic UF	Not applicable
Incomplete Database UF	3
Database Quality	high
Chronic ReV (HQ = 1)	**33 µg/m^3 (15 ppb)**
$^{chronic}ESL_{nonlinear(nc)}$ (HQ = 0.3)	**9.9 µg/m^3 (4.5 ppb)**

4.1.8 Derivation of Chronic ReV versus USEPA's Chronic RfC

Table 14 provides a comparison of the derivation of the chronic ReV of 33 µg/m^3 (15 ppb) versus the chronic RfC of 2 µg/m^3 (0.9 ppb) (USEPA 2002). USEPA's RfC is approximately 17 times lower than TCEQ's ReV due to the following reasons:

- The TCEQ did not use an effect level extrapolation factor of 10 which makes USEPA's RfC 10 times lower;
- The TCEQ used a UF_A of 1 based on data that DEB is the reactive metabolite responsible for ovarian atrophy and empirical data demonstrating DEB is produced in much lower concentrations than humans, whereas USEPA used a UF_A of 3, which makes USEPA's RfC three times lower; and

- The TCEQ used a $BMCL_{05}$ of 462 ppb which included the highest dose whereas USEPA used a $BMCL_{10}$ of 880 ppb which excluded the highest dose, which makes USEPA's RfC approximately 2 times higher.

Consideration of the above differences accounts for approximately a 15-fold difference (10 x 3 x 0.5). While an exact partitioning of the 17-fold difference may not be possible, there are science-based and logical explanations accounting for most of the differences.

Table 14 Comparison of Chronic ReV and Chronic RfC

Chronic Toxicity Value	POD_{HEC}	UF_H	UF_A	UF_L or Effect Level Extrapolation Factor	UF_{Sub}	UF_D	Total UFs	Chronic Toxicity Value
ReV based on ovarian atrophy (TCEQ)	462 ppb $BMCL_{05}$ including highest dose	10	1	1 UF_L	1	3	30	15 ppb
RfC based on ovarian atrophy (USEPA)	880 ppb $BMCL_{10}$ excluding highest dose	10	3	10 Effect Level extrapolation Factor	1	3	1,000	0.88 ppb

4.2 Carcinogenic Potential

4.2.1 Carcinogenic Weight of Evidence and MOA

USEPA has classified BD as known to be carcinogenic to humans by inhalation (DHHS 2000; USEPA 2002) based on the following findings:

- Increased lymphohematopoietic cancers in workers occupationally exposed via inhalation to BD based on epidemiologic studies (leukemias in polymer workers and non-Hodgkin's lymphoma in monomer workers);
- BD causes a variety of tumors in mice and rats by inhalation in various studies;
- Demonstration that BD is metabolized into genotoxic metabolites by experimental animals and humans.

Table 15 provides information on the carcinogenic weight of evidence provided by other organizations. Although the mechanism of action, as opposed to the MOA, by which BD produces tumors is unknown, scientific evidence suggests that carcinogenic effects are mediated by genotoxic metabolites of BD (i.e., EB, DEB, and EBD, Section 3.1.2 and Figure 2). A detailed review of the weight of evidence, carcinogenic hazard assessment, and MOA analysis for lifetime exposure potential is included in USEPA (2002). Preston (2007) recently reviewed the evidence that BD works through a mutagenic MOA and concluded: "For butadiene, the MoA is DNA-reactivity and subsequent mutagenicity and so following the EPA's cancer guidelines, a linear extrapolation is used from the POD, unless additional data support a non-linear extrapolation." Therefore, an inhalation unit risk factor (URF) and $^{chronic}ESL_{linear(c)}$ (i.e., air

concentration at 1 in 100,000 excess cancer risk) was developed for BD.

Although a linear extrapolation from a POD will be used to calculate a URF based on the MOA of BD, a free-standing NOAEL for biomarkers of effect (hypoxanthine-guanine phosphoribosyltransferase (HPRT) mutations and chromosome aberrations) at mean BD exposure concentrations of 0.800 ppm has been demonstrated by Albertini *et al.* (2001) in a small initial study of workers in the Czech Republic.

Table 15 Carcinogenic Weight of Evidence

Institution	**Carcinogenic Ranking**
International Agency for Research on Cancer (IARC 2007)	Group 1, Carcinogenic to humans
National Institute for Occupational Safety and Health 1997	Potential occupational carcinogen
Occupational Safety and Health Administration 1996	"Potential occupational carcinogen" There is strong evidence that workplace exposure to BD poses an increased risk of death from cancers of the lymphohematopoietic system.
ACGIH 2001	A2, Suspected Human Carcinogen
USEPA 2002; DHHS 2000	Carcinogenic to humans by inhalation

4.2.2 Epidemiological Studies and Exposure Estimates

Chapter 7 of USEPA (2002) discusses the epidemiologic studies of carcinogenicity for BD, and Chapter 10 discusses the dose-response assessment of the preferred occupational epidemiological study conducted by researchers at the University of Alabama at Birmingham (UAB) (Delzell *et al.* 1995; 1996). Numerous epidemiology studies were reviewed, but USEPA (2002) concluded the UAB exposure estimates provided the best published set of data to evaluate human cancer risk from BD exposure. USEPA published an inhalation URF of 3.0×10^{-5} per $\mu g/m^3$ or 0.08 per ppm based on leukemia mortality data from the UAB occupational epidemiological study (Delzell *et al.* 1995; 1996).

Delzell *et al.* (1995, 1996) investigated a cohort of synthetic rubber production workers exposed to BD in a retrospective cohort mortality study. The investigators developed a job exposure matrix (JEM) for BD, styrene, and benzene based on industrial hygiene data, which contained estimates of the average daily exposure (in ppm based on the 8-h TWA) and the number of annual peaks (defined as > 100 ppm for BD and >50 ppm for styrene) for each area and job code for each study year. The investigators were then able to estimate cumulative exposures (part-per-million (ppm)-years) and number of peak exposures (peak years) for each individual worker by linking the JEM with the study subjects' work histories.

Recently, the exposure estimates and the UAB epidemiology study of leukemia were updated:

- The UAB butadiene exposure estimates were updated and estimates for styrene and dimethyldithiocarbamate (DMDTC) were calculated (DMDTC is an immune system depressant) (Macaluso *et al.* 2004);
- The UAB epidemiology study and analysis of leukemia data was updated (Sathiakumar *et al.* 2005; Graff *et al.* 2005) (see below for additional details);
- Dr. Delzell and associates finalized a Health Effects Institute (HEI) report that discussed the updated exposure estimates and analysis of leukemia mortality data. Additional analyses requested by the Health Effects Review committee were included in the HEI report (HEI 2006).

The Health Review Committee (HEI 2006) thoroughly reviewed Delzell's findings and concluded

> "An analysis of butadiene that controlled for the possibly carcinogenic coexposures to styrene and DMDTC produced the most important result of the investigation: the clear and consistent exposure-response relation observed between cumulative exposure to butadiene and mortality from leukemia. . . . and support the presence of a linear increase in the relative rate of leukemia mortality with increasing cumulative exposures to butadiene."

After the HEI report was finalized, an exposure estimate validation study was conducted on the updated UAB butadiene exposure estimates (Sathiakumar *et al.* 2007). At lower concentrations, there was reasonably good agreement between measured versus estimated BD exposures; whereas at higher exposures, the estimates tended to be less than the measured values. On average, estimates were about 10% lower than measurements. Based on the validation study of Sathiakumar *et al.* (2007), the updated exposure estimates of Macaluso *et al.* (2004) have a higher confidence than original exposure estimates. Dose-response modeling was conducted based on the updated studies (Cheng *et al.* 2007; Sielken *et al.* 2007). These new, updated studies were used by the TS to update the USEPA (2002) assessment. A review of the scientific literature indicated there were no other epidemiology studies (e.g., Tsai 2005; Alder *et al.* 2006) that would be appropriate to evaluate human cancer risk from BD exposure.

Subjects included in the updated study were 16,579 men classified as having worked (for at least one year before 1 January 1992) at any of six synthetic rubber plants located in Texas (two plants), Louisiana (two plants), Kentucky (one plant) and Canada (one plant). Of the 16,579 subjects in the updated study, 488 subjects were excluded because they dropped out of follow-up at ages younger than the youngest leukemia decedent (age 33 years) (Cheng *et al.* 2007). Thus, results of leukemia analyses were based on 16,091 subjects and 485,732 person-years of observation. The updated study provided seven more years of follow-up (through 1998), a larger number of decedents, and a total of 81 deaths with leukemia as the primary or contributing cause. The association of BD exposure to lymphoid and myeloid neoplasms was investigated. BD-exposure estimates were also updated, and quantitative estimates of each subject's exposure to butadiene, styrene and dimethyldithiocarbamate (DMDTC), an immune system depressant (Irons and Pyatt 1998; Irons *et al.* 2001) were determined.

4.2.3 Dose-Response Assessment

4.2.3.1 Beta coefficient (β) and Standard Error Based on Observed Data

Cheng *et al.* (2007) investigated the dose-response relationship between BD and leukemia rate ratios using a log-linear exponential Cox regression analysis. Three BD exposure indices were evaluated by Cheng *et al.* (2007): (1) continuous, time-dependent BD exposure indices (ppm-years); (2) the total number of exposures to BD concentrations >100 ppm (number of peak exposures) and (3) average intensity of BD. All three BD exposure indices were positively associated with leukemia.The term "peak" is used by the UAB group to refer to the cumulative number of exposures to >100 ppm BD. These exposures were frequently of short duration (several seconds to several minutes). However, the term "peak" or "peak exposures" is misleading and will not be used in this assessment. Instead, the more descriptive term "number of high-intensity tasks" (i.e., number of HITs) is used. The dose metric used by the TS for the dose-response assessment was cumulative BD ppm-years, a dose metric commonly used for dose-response modeling based on epidemiological studies.

The data needed to conduct a detailed mechanism of action analysis were not available, so the use of a biologically-based model was not possible. Rather, standard epidemiological models such as the log-linear Cox proportional hazards models with age included as an index variable and the linear Poisson regression, a conservative linear default model, were selected. Whereas Cheng *et al.* (2007) used the log-linear Cox regression analysis with continuous, untransformed data and mean-scored deciles (grouped data), Sielken *et al.* (2007) used a linear Poisson regression analysis with mean-scored deciles (grouped data) to investigate the relationship between BD and leukemia rate ratios. Cheng *et al.* (2007) and Sielken *et al.* (2007) calculated betas (β) (maximum likelihood estimates (MLEs)) and standard errors (SE) from the updated UAB epidemiological study and updated exposure estimates (Table 16).

The Cox regression analysis using continuous cumulative exposure estimates is preferred to the Cox with mean-scored deciles (grouped data) because the former uses the best estimate of cumulative BD ppm-years. Additionally, Cox regression analyses use individual data and adjust for the effects of age in an optimal way (age is used as the index variable and implicitly a covariate) and are preferred over Poisson regression analyses.

Cheng *et al.* (2007) also determined the β and SE for data restricted to the lower 95% of the exposure range of all subjects since spline regression analysis indicated that above an exposure level of 1,123 BD ppm-year, the data were sparse, and the dose-response relationship was erratic (Figure 5a). Figure 5b shows the dose-response relationship below 1,123 BD ppm-years. Spline regression indicated that the ln hazard ratio for leukemia increased in a fairly linear fashion in the exposure range below the 95% of exposure, although the choice of "knots" may affect the appearance of spline curves. The β estimates obtained from restricted data were higher (i.e., more conservative). Evaluating the β and SE for restricted data, which are more conservative, may address concerns that data were sparse (there were only four leukemia decedents), and exposure-response trends were erratic for cumulative BD above 1,123 ppm-years (Cheng *et al.* 2007; Steenland 2005). Sielken *et al.* (2007) examined the results of progressively restricting the data to lower concentrations (i.e., < 1,338, 1,000, 500, 400, 300, 200, and 100 ppm-years).

Interestingly, these analyses showed the absence of a statistically significant low-dose risk versus cumulative BD ppm-years for restricted data less than 300 ppm-years.

Table 16 Values of Maximum Likelihood Estimate (MLE) of Beta (β), Standard Error (SE), and 95% Upper Confidence Limit (UCL) on β a

Covariates	Model	Source	β (MLE) ± SE p-Value	β (95% UCL) b
Age	Cox log-linear ppm-years continuous [c]	Cheng *et al.* (2007)	2.9E-04 ± 1.0E-04 < 0.01	4.545E-04
Age	Cox log-linear ppm-years mean-scored deciles [e]	Cheng *et al.* (2007)	7.5E-04 ± 2.2E-04 < 0.01	1.112E-03
Age	Cox log-linear (restricted to lower 95% of exposure range) ppm-years continuous [c]	Cheng and Delzell [d]	1.58E-03 ± 3.9E-04 < 0.001	2.221E-03
Age	Poisson linear ppm-years mean-scored deciles [e]	Sielken *et al.* (2007)	1.68E-03 ± 8.21E-04 < 0.001	3.031E-03
Age & Other Covariates[f]	Cox log-linear ppm-years continuous [c]	Cheng *et al.* (2007)	3.0E-04 ± 1.4E-04 0.04	5.303E-04
Age & Other Covariates[f]	Cox log-linear ppm-years mean-scored deciles [e]	Cheng *et al.* (2007)	5.8E-04 ± 2.7E-04 0.03	1.024E-03
Age & Other Covariates[f]	Cox log-linear (restricted to lower 95% of exposure range) ppm-years continuous [c]	Cheng *et al.* (2007)	1.31E-03 ± 4.7E-04 < 0.01	2.083E-03

a units are in ppm-years and based on occupational exposure concentrations

b β (95% UCL) = β(MLE) + (1.645 x SE)

c ppm-years is included as a continuous variable (untransformed) in a parametric model of the effect of ppm-years

d Personal communication, 1/30/2008 email from Cheng and Delzell. Cheng et al. (2007) reported results for Cox log-linear (restricted to lower 95% of exposure range) ppm-years continuous for age & other covariates, but not age only. The 1/30/2008 email provided the values for Cox log-linear (restricted to lower 95% of exposure range) ppm-years continuous for age.

e ppm-years is included as a continuous variable with values grouped into mean-scored deciles (untransformed) in a parametric model of the effect of ppm-years

f other covariates are year of birth, race, DMDTC, years since hire and plant

Table 16 shows results from the BD dose-response relationship conducted by Cheng *et al.* (2007) using log-linear Cox regression procedures, continuous data or mean-scored decile data, adjusted either for age as a covariate or adjusted for other covariates (age, year of birth, race, plant, years since hire and DMDTC) for the full range of exposure data and to data restricted to the lower 95% of the exposure range. The linear Poisson mean-scored decile data adjusted for age as a covariate is also included in Table 16. The TS used these values to calculate 95% upper confidence limit (UCL) values, URFs and corresponding air concentrations at 1 in 100,000 excess cancer risk (Table 17). Cheng *et al.* (2007) results support the presence of a relationship between high cumulative exposure and leukemia and high intensity of exposure and leukemia.

Beta estimates were also calculated by Cheng *et al.* (2007) for unlagged and lagged BD exposure but these β estimates were not used by the TS because lagging BD exposure had little impact on the dose-response relationship between leukemia and BD ppm-years. Sielken & Associates have shown that when windows of exposure were evaluated in the model, there was little impact on the dose-response relationship between leukemia and BD ppm-years (personal communication from Sielken & Associates). The association of BD exposure with leukemia, lymphoid neoplasms, and myeloid neoplasms was investigated by both Cheng *et al.* (2007) and Sielken *et al.* (2007). Lymphoid neoplasms were associated with ppm-years and myeloid neoplasms were associated with number of HITs in models that controlled only for age but not after adjusting for multiple covariates (age, year of birth, race, plant, years since hire and DMDTC). These potency estimates were not used by the TS because evidence of an association between BD and all lymphoid neoplasm or all myeloid neoplasms was not persuasive (Cheng *et al.* 2007; Sielken *et al.* 2007).

Sielken *et al.* (2007) used a linear Poisson regression model to examine the dose-response relationships adjusted for age as a categorical covariate (Table 16), age + number of HITs as covariates, and multiple covariates. Sielken *et al.* (2007) found that if the exposure dosimetric is cumulative ppm-years, the performance of the predictor for leukemia rate ratio is statistically significantly improved if the categorical covariates age + number of HITs are included in the Poisson regression model. If covariates other than age + number of HITs are included, the model fit using cumulative ppm-years was not significantly improved except for styrene. However, if styrene was included as a covariate, the slope was negative, so styrene was not included as a covariate. Although Sielken *et al.* (2007) performed this statistical analyses for covariates using Poisson regression models, their findings are also generally applicable for the Cox proportional hazards models.

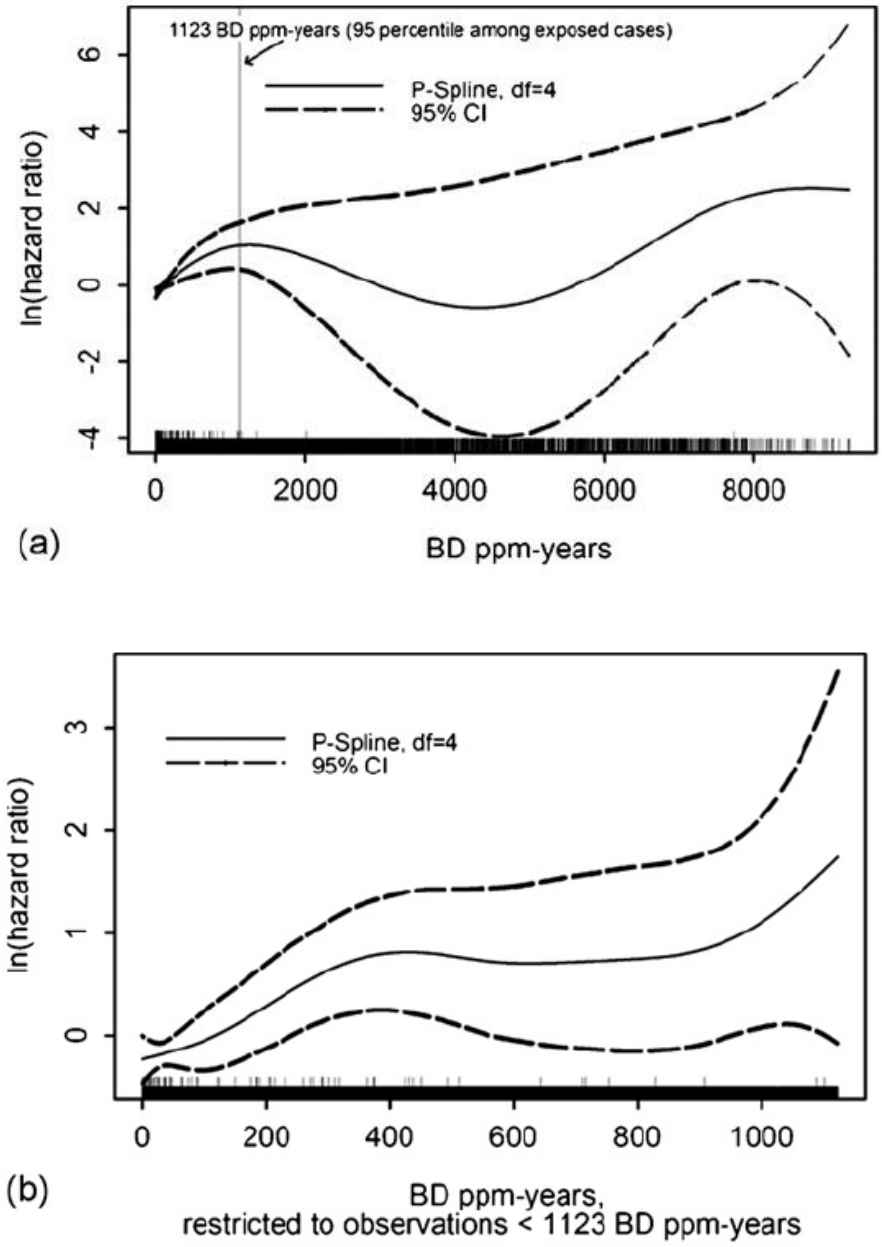

Figure 5 Exposure-Response in Models using Continuous BD Variables and Restricted Data

(Figure 1 (a, b) from Cheng *et al.* (2007), reproduced with permission). Penalized splines for BD ppm-years and leukemia (a and b). Rugs just above the x-axis of each figure depict the frequency of observations (lower rug) and leukemias (upper rug) at corresponding BD variable values. There were only four leukemia decedents above 1,123BD ppm years.

4.2.3.2 Dosimetric Adjustments

Occupational concentrations were converted to environmental concentrations for the general population using the following equation (TCEQ 2006):

$Concentration_{HEC} = Concentration_{OC} \times (VE_{ho}/VE_h) \times (days\ per\ week_{oc}/days\ per\ week_{res})$

where: VE_{ho} = occupational ventilation rate for an 8-h day (10 m^3/day)

VE_h = non-occupational ventilation rate for a 24-h day (20 m^3/day)

$days\ per\ week_{oc}$ = occupational weekly exposure frequency (default of 5 days per week)

$days\ per\ week_{res}$ = residential weekly exposure frequency (7 days per week)

4.2.3.3 Extrapolation to Lower Exposures

4.2.3.3.1 URFs and Air Concentrations at 1 in 100,000 Excess Cancer Risk

Table 17 shows estimates of air concentrations at 1 in 100,000 excess cancer risk (10^{-5}-risk air concentrations) based on βs (column three) and 95% UCLs (column five) using the log-linear Cox regression and linear Poisson regression models. Air concentrations were solved iteratively with life-table analyses using the BEIR IV approach (NRC 1988). Air concentrations based on extra risk were calculated as opposed to added risk. The following mortality and survival rates were used to calculate air concentrations based on a lifetime exposure of 70 years, the default used by TCEQ for exposure analysis (TCEQ 2006):

- US mortality rates for 2000-2003 for all leukemia (Surveillance, Epidemiology, and End Results database (SEER 2006)) (Appendix 4)
- US survival rates for 2000 (Arias 2002) (Appendix 4).

Columns four and six of Table 17 provide URFs calculated using the linear extrapolation default approach (USEPA 2005a; TCEQ 2006). Risk estimates are obtained by first calculating a POD_{HEC} at the low end of the range of observations using appropriate models and then extrapolating to zero by means of a straight line (linear extrapolation default). The air concentration at 0.1% excess risk level (i.e., 1 in 1,000 excess cancer risk) is chosen for determining the POD because it is within the observable response range of leukemia deaths. The URFs in units of ppm^{-1} at the POD_{HEC} (when the POD_{HEC} was set to either the effective concentration (EC_{001}) or the 95% UCL lowest effective concentration (LEC_{001})) were calculated as follows:

$$URF = 0.001/EC_{001}$$

$$URF = 0.001/LEC_{001}$$

Columns four and six of Table 17 also provide 10^{-5}-risk air concentrations based on the corresponding URFs. Air concentrations calculated using the corresponding URFs are more conservative than air concentrations calculated based on the Cox regression model, because this model is a log-linear model. As a health-protective policy decision, 10^{-5}-risk air concentrations calculated with URFs based on the default linear approach were adopted and all subsequent discussions will refer to the URF (MLE) or URF (95%UCL) and their corresponding 10^{-5}-risk air concentration values.

Table 17 URFs and Air Concentrations Corresponding to 1 in 100,000 Extra Leukemia Risk

Covariates	Model type of data	Air Concentration 1 in 100,000 excess cancer risk using model β (MLE)	URF (MLE) [a] Air Concentration 1 in 100,000 excess cancer risk using URF EC001	Air Concentration 1 in 100,000 excess cancer risk using model β (95% UCL)	URF (95% UCL)[b] Air Concentration 1 in 100,000 excess cancer risk using URF LEC001
Age	Cox log-linear ppm-years continuous c Cheng et al. (2007)	87.36 ppb	1.371E-04/ppm 72.93 ppb	55.74 ppb	2.149E-04/ppm 46.53 ppb
Age	Cox log-linear ppm-years mean-scored deciles [d] Cheng et al. (2007)	33.78 ppb	3.546E-04/ppm 28.20 ppb	22.78 ppb	5.258E-04/ppm 19.02
Age	Cox log-linear (restricted to lower 95% of exposure range) ppm-years continuous c Cheng and Delzell f	16.03 ppb	7.471E-04/ppm 13.39 ppb	11.41 ppb	1.050E-03/ppm 9.523 ppb
Age	Poisson linear ppm-years mean-scored deciles [d] Sielken et al. (2007)	15.11 ppb	6.614E-04/ppm 15.12 ppb	8.376 ppb	1.193E-03/ppm 8.381 ppb
Age & Other Covariates[e]	Cox log-linear ppm-years continuous [c] Cheng et al. (2007)	84.45 ppb	1.418E-04/ppm 70.50 ppb	47.77 ppb	2.507E-04/ppm 39.88 ppb
Age & Other Covariates[e]	Cox log-linear ppm-years mean-scored deciles [d] Cheng et al. (2007)	43.68 ppb	2.742E-04/ppm 36.47	24.74 ppb	4.842E-04 20.65
Age & Other Covariates[e]	Cox log-linear (restricted to lower 95% of exposure range) ppm-years continuous c Cheng et al. (2007)	19.34 ppb	6.194E-04/ppm 16.14 ppb	12.16 ppb	9.849E-04/ppm 10.15 ppb

a URF = 0.001/EC001

b URF = 0.001/LEC001

c ppm-years is included as a continuous variable (untransformed) in a parametric model of the effect of ppm-years

d ppm-years is included as a continuous variable with values grouped into mean-scored deciles (untransformed) in a parametric model of the effect of ppm-years

e Other covariates are year of birth, race, DMDTC, years since hire and plant

f Personal communication, 1/30/2008 email from Dr. Cheng and Dr. Delzell. Cheng et al. (2007) reported results for Cox log-linear continuous (restricted to lower 95% of exposure range) ppm-years for age & other covariates, but not age only or age + # HITs. Dr. Cheng and Dr. Delzell provided the β and SE values for Cox log-linear continuous (restricted to lower 95% of exposure range) ppm-years for age and age + # HITs in the 1/30/2008 email.

4.2.3.3.2 Age as a Covariate

Models that only include age as a non-exposure covariate have the advantage of model parsimony (i.e., the model includes as few variables as necessary to explain the relationship when there is not sufficient biological knowledge to justify the inclusion or exclusion of a variable). When age is included as a covariate (Table 18), the 10^{-5}-risk air concentrations using the Poisson linear model were the most conservative: 15.12 ppb (MLE) and 8.381 ppb (95% UCL). However, as stated previously, Cox log-linear analysis using continuous, untransformed data are preferred over the linear Poisson regression analysis with mean-scored deciles (grouped data) because it uses the best estimate of cumulative BD ppm-years, uses individual data, and adjusts for the effects of age in an optimal way. Using the Cox log-linear model and restricted data, the 10^{-5}-risk air concentrations of 13.39 ppb (MLE) and 9.523 ppb (95% UCL) were more conservative than Cox log-linear mean-scored deciles (28.20 ppb MLE and 19.02 ppb 95% UCL) and continuous, untransformed data (72.93 ppb MLE and 46.53 ppb 95% UCL).

Table 18 Age as a Covariate

Model type of data	EC001 URF (MLE) a 10^{-5}-risk air concentration using URF	LEC001 URF (95% UCL) b 10^{-5}-risk air concentration using URF
Cox log-linear Cheng et al. (2007) ppm-years continuous c	1.371E-04/ppm 72.93 ppb	2.149E-04/ppm 46.53 ppb
Cox log-linear Cheng et al. (2007) ppm-years mean-scored deciles d	3.546E-04/ppm 28.20 ppb	5.258E-04/ppm 19.02
Cox log-linear (restricted to lower 95% of exposure range) Cheng et al. (2007) ppm-years continuous c	7.471E-04/ppm 13.39 ppb	1.050E-03/ppm 9.523 ppb
Poisson linear Sielken et al. (2007) ppm-years mean-scored deciles d	6.614E-04/ppm 15.12 ppb	1.193E-03/ppm 8.381 ppb

a URF = 0.001/EC001

b URF = 0.001/LEC001

c ppm-years is included as a continuous variable (untransformed) in a parametric model of the effect of ppm-years

d ppm-years is included as a continuous variable with values grouped into mean-scored deciles (untransformed) in a parametric model of the effect of ppm-years

The 10^{-5}-risk air concentration estimates based on restricted data are preferred because the impact of sparse data and the erratic exposure-response trends above 1,123 ppm-years are reduced. The EC_{001} and LEC_{001} values from the Cox regression models with continuous restricted data are approximately 5-fold smaller than the values from the Cox regression models

with all the data, 2-fold smaller than the values from the Cox regression models with all the data and mean-scored deciles, and within 13% from the values of the Poisson regression model with mean-scored deciles. The use of mean-scored deciles in the Cox log-linear and Poisson linear models may reduce the impact of misclassification because it reduces the influence of data at the extreme exposure estimates (Cheng *et al.* 2007), although it may increase misclassification at the low end, where exposure estimates are likely to be better. Therefore, continuous, untransformed data are preferred (also refer to Section 4.2.3.1 *Beta coefficient (β) and Standard Error Based on Observed Data* for additional reasons).

4.2.3.3.3 Other Covariates

4.2.3.3.3.1 Models that adjusted for multiple covariates

Cheng *et al.* (2007) fit models that adjusted for age, year of birth, race, DMDTC, years since hire and plant. Except for the exposure covariate DMDTC, an immune system depressant (Irons and Pyatt 1998; Irons *et al.* 2001), these covariates are typically evaluated in epidemiology dose-response models. Sielken *et al.* (2007) included statistically-based covariates and determined that if covariates other than age + number of HITs are included, the model fit using cumulative ppm-years was not significantly improved except for styrene. However, if styrene was included as a covariate, the slope was negative, so styrene was not included as a covariate. Although Sielken *et al.* (2007) performed this statistical analysis for covariates using the Poisson regression model, his findings are generally applicable for the Cox regression model. Therefore, URF (MLE) and URF (95% UCL) from models that adjusted for age, year of birth, race, DMDTC, years since hire, and plant were not considered as potency factors by the TS, although these values are provided in Tables 16 and 17 for comparison purposes. There were minor differences between 10^{-5}-risk air concentrations in models that adjusted for age only or models that adjusted for multiple covariates (Table 17).

4.2.3.3.3.2 Models that adjusted for age + number of HITs > 100 ppm

The cumulative number of HITs > 100 ppm may better explain the increased leukemia mortality observed in the BD worker cohort (Cheng *et al.* 2007; Sielken *et al.* 2007). Sielken *et al.* (2007) demonstrated that when the categorical covariates of age + number of HITs are included in the Poisson regression model, the model's ability to predict the leukemia rate ratio was statistically improved. The UAB group evaluated the number of HITs > 100 ppm because BD ppm-years could not by itself adequately explain worker's leukemia risk. Since the USEPA Science Advisory Board (USEPA 1998) recommended that consideration of peak exposures to BD be evaluated during its review of the draft health risk assessment of BD (USEPA 1998b), the TS did evaluate the effect of including number of HITs. However, BD ppm-years and number of HITs are both exposure variables and may be correlated, so it may not be appropriate to include both of them in the same model. Cheng *et al.* (2007) found these BD exposure variables were weakly correlated for continuous values (Pearson correlation coefficient of 0.30) as opposed to grouped (deciles) values (Pearson correlation coefficient of 0.80). Therefore, the TS evaluated Cox regression models using continuous (untransformed) variables that adjusted for age and the continuous (ungrouped) value of cumulative number of HITs > 100 ppm. The 10^{-5}-risk air concentration based on the URF (MLE) increased approximately 18% and the 10^{-5}-risk air concentration based on the URF (95% UCL) increased approximately 6% (Tables 18 and 19 of subsection 4.2.5.2 *Estimating Risks for the General Population from Occupational Workers*)

when number of HITs > 100 ppm was included as a covariate. These models were not the preferred models selected to represent excess leukemia mortality risk, but are useful for evaluating uncertainty of estimating risks to the general population when data are based on occupational workers who are exposed to peak BD exposures > 100 ppm. Section 4.2.5 *Uncertainty Analysis*, subsection 4.2.5.2 *Estimating Risks for the General Population from Occupational Workers* provides a more detailed discussion.

4.2.4 Potency Estimate Selected to Represent Excess Leukemia Mortality Risk

Of the various estimates presented in Table 17, the potency estimate of 1.050E-03 per ppm (10^{-5}-risk air concentrations of 9.523 ppb) from the Cox regression model using restricted continuous data, age as a covariate, the URF based on the 95% UCL, and a linear default approach is selected to represent the excess leukemia mortality risk from the occupational data. However, refer to Section 4.2.4.1 *Evaluating Susceptibility from Early-Life Exposures* and Section 4.2.4.2 *Relevance of Estimated Risk to the Texas General Population* for additional adjustments to the URF (95% UCL) and 10^{-5}-risk air concentrations. The ranges in the cancer potency estimates from the different models were within a factor of five:

- The cancer potency estimates and 10^{-5}-risk air concentrations using URFs (MLE) in Table 17 range from 7.471E-04 per ppm (13.39 ppb) to 1.371E-04 per ppm (72.93 ppb).
- The cancer potency estimates and 10^{-5}-risk air concentrations using URFs (95% UCL) in Table 17 range from 1.193E-03 per ppm (8.381 ppb) to 2.149E-04 per ppm (46.53 ppb).

The UAB group recommended the estimate of the dose-response relationship that is based on the continuous, untransformed form of BD ppm-years, age included as the index variable, and the full range of exposure data (2.9E-04 (β), 1.0E-04 (S.E.)). However, due to the high potential for distortion of the dose-response relationship as a result of exposure misclassification at high exposure concentrations, Cheng *et al.* (2007) also recommended that an uncertainty analysis be incorporated into any risk assessment that uses these data. However, since the purpose of this assessment is to calculate a health-protective 10^{-5}-risk air concentration for evaluation of air permits and ambient air monitoring data, the TS decided as a policy decision to use the results based on restricted data because they are more conservative and to address concerns about sparse data and an erratic exposure-response relationship at high exposure concentrations.

The Cox regression analysis using continuous, untransformed data are preferred over the Cox log-linear and linear Poisson regression analysis with mean-scored deciles (grouped data) because it uses the best estimate of cumulative BD ppm-years, uses individual data, and adjusts for the effects of age in an optimal way. A linear default was used to extrapolate to lower concentrations and the URF (95% UCL) was preferred to account for uncertainties as discussed in the uncertainty section. The confidence intervals are indicators of the variability, and to some extent the uncertainty, in the dose-response curve for mortality. The risk to the general population will be lowered since using the URF (95% UCL) adds conservatism to the estimate. There was only a 1.4 fold difference between estimates using the MLE compared to the 95% UCL, which supports the quality of the epidemiological data.

4.2.4.1 Evaluating Susceptibility from Early-Life Exposures

USEPA (2005b) provides default, age-dependent adjustment factors (ADAFs) to account for potential increased susceptibility in children due to early-life exposure when a chemical has been identified as acting through a mutagenic MOA for carcinogenesis and the cancer assessment did not include exposures at an early age (generally before age 16). This is the case for the epidemiological leukemia data utilized in this evaluation. BD is currently identified by USEPA as having a mutagenic MOA. USEPA (2005b) states:

> "The following adjustments represent a practical approach that reflects the results of the preceding analysis, which concluded that cancer risks generally are higher from early-life exposure than from similar exposure durations later in life:
>
> - For exposures before 2 years of age (i.e., spanning a 2-year time interval from the first day of birth up until a child's second birthday), a 10-fold adjustment.
> - For exposures between 2 and <16 years of age (i.e., spanning a 14-year time interval from a child's second birthday up until their sixteenth birthday), a 3-fold adjustment
> - For exposures after turning 16 years of age, no adjustment."

The ADAF is an adjustment to the slope factor (as opposed to an adjustment to the dose metric). The ADAF is to be applied on an age-specific basis. That is, the ADAFs are applied to each relevant year in a life and the risks for all years summed to get the lifetime risk, as opposed to calculating a lifetime excess risk without ADAFs and then multiplying this calculated value by a constant ADAF.

When the dose metric is cumulative exposure and when using a life-table analysis BEIR IV approach (NRC 1988), an implementation consistent with USEPA guidelines is to calculate the excess risk in each year using the age-specific dose (cumulative dose) for that year and multiply the slope by the age-specific ADAF for that year (age). This is consistent with USEPA's guidelines from the point of view of both excess risk being calculated using age-specific exposures and ADAFs being age-specific modifiers of the slope (potency). That is, the excess risk in year "i" is calculated with the β or 95% UCL multiplied by ADAF(i). Refer to Appendix 5 *Calculating Excess Risk with Age-Dependent Adjustment Factors using a Life-Table Analysis*.

The TS calculated potency factors both with and without ADAFs. When the ADAFs are not applied, the selected potency estimate is 1.050E-03 per ppm (9.523 ppb 10^{-5}-risk air concentration). When the ADAFs are incorporated into the life-table analyses using the BEIR IV approach (NRC 1988), the selected potency estimate is 1.062E-03 per ppm (9.416 ppb 10^{-5}-risk air concentration). There is a minor difference between potency estimates calculated with and without ADAFs, when the URF is rounded to two significant figures at the end of all calculations. Toxicokinetic and toxicodynamic evidence indicates children are not more susceptible to chemical leukemogenesis than adults for acute myeloid leukemia and acute nonlymphocytic leukemia (Johnsrud *et al.* 2003; Levine and Bloomfield 1992; Pyatt *et al.* 2005; Pyatt *et al.* 2007; USEPA 1997), so the application of ADAFs may not be justified. USEPA (1997) provides a detailed discussion of the critical steps that may contribute to BD leukemogenesis.

4.2.4.2 Relevance of Estimated Risks to the Texas General Population

There is uncertainty about whether potency estimates are representative of the mortality risks that might be associated with environmental BD exposures in Texas because potency estimates were developed based on the leukemia mortality experience of predominantly male workers in the styrene-butadiene rubber industry, total US rates of mortality from leukemia and total US survival rates (Appendix 4). In order to address this uncertainty, Texas-specific mortality rates for 1999-2003 for all leukemia and Texas-specific survival rates for 2003 were kindly provided by the Texas Department of State Health Services, Cancer Epidemiology and Surveillance Branch, Texas Cancer Registry. There were minor differences in calculated air concentrations when Texas versus US all leukemia mortality rates and survival rates were used because the Texas-specific rates are very similar to US rates (Appendix 4). The selected potency estimate is 1.062E-03 per ppm (9.416 ppb 10^{-5}-risk air concentration) using US rates of mortality from leukemia and total US survival rates when ADAFs are incorporated (Section 4.2.4.1) and is 1.097E-03 per ppm (9.112 ppb 10^{-5}-risk air concentration) using Texas-specific mortality rates for 1999-2003 for all leukemia and Texas-specific survival rates for 2003 when ADAFs are incorporated. There is no difference between potency estimates calculated with either US rates or Texas rates when the URF is rounded to two significant figures at the end of all calculations (i.e., 1.1E-03 per ppm). The $^{chronic}ESL_{linear(c)}$ or air concentration at 1 in 100,000 excess cancer risk is 9.1 ppb (20 µg/m^3).

4.2.5 Uncertainty Analysis

4.2.5.1 Estimating Risks for other Potentially Sensitive Subpopulations

Leukemia mortality was evaluated based on healthy male workers employed at North American plants that manufactured SBR. Since these workers were healthy, they may underestimate risks to the general population that are comprised of sensitive subpopulations. It is unknown whether workers with genetic polymorphisms as discussed in Section 3.1.2 (i.e., genes that regulate the metabolism of BD to mutagenic intermediates and genes that regulate the detoxification of those metabolites) were represented in the cohort. Populations with certain lifestyle choices may be more sensitive to health effects caused by BD. Children may or may not be more sensitive to mutagenic carcinogens (see Section 4.2.4.1).

Studies in which animals were exposed to high BD concentrations suggest that female animals may be more sensitive than male animals for cancer effects after exposure to BD (USEPA 2002). Initial studies conducted in humans by Albertini *et al.* (2007) indicate that except for lower production of both urine BD metabolites in females, no female-male differences in response to low BD exposures were detected (mean 8-h TWA exposure levels were 0.180 ppm for BD-exposed female workers and 0.370 ppm for BD-exposed male workers as discussed in Section 4.5). A significant finding from Albertini *et al.* (2007) is "females showed lower concentrations of both M1 and M2 metabolites in the urine per unit of BD exposure than did males while exhibiting the same M1/(M1 + M2) ratio, reflecting the same relative utilization of the hydrolytic (producing M1) and the conjugation (producing M2) detoxication pathways as males." This may indicate that females absorb less BD per unit of exposure than male workers.

The UAB group has analyzed mortality results for 4,863 female workers employed in the SBR industry from 1943 to 2002 (Sathiakumar and Delzell 2007a, b). Preliminary results indicate that

standard mortality rates (SMRs) for lung and bladder cancer were elevated in female workers. Both excesses were concentrated among ever-hourly employees and among ever-hourly employees with 20+ years since hire, but neither cancer displayed a pattern of increasing SMRs with increasing duration of employment. For lung cancer, analyses of cumulative exposure indices were conducted. Results for lung cancer indicated a moderately positive association with each agent, without exposure-response. The SMRs for leukemia, non-Hodgkin lymphoma or other forms of lymphohematopoietic cancers, breast cancer, and ovarian cancer were not elevated (Sathiakumar and Delzell 2007b). For lung and bladder cancer, the absence of any trend of increasing SMRs with increasing duration of employment, the lack of any exposure-response trend for cumulative exposure to BD, styrene, or DMDTC and the absence of positive results in studies of male employees indicate that these occupational exposures may not have been responsible for the observed excesses of lung and bladder cancers among women in the industry (Sathiakumar and Delzell 2007b).

Since the UAB cohort was comprised primarily of males, a linear default was used to extrapolate to lower concentrations, and the URF (95% UCL) was used instead of the URF (MLE) to account for the uncertainty of calculating potency estimates for the general population.

4.2.5.2 Estimating Risks for the General Population from Occupational Workers

There is uncertainty regarding the extrapolation of risks from occupational workers exposed to high BD concentrations and to BD HITs > 100 ppm to risks for the general population who are exposed to much lower BD concentrations and not exposed to BD HITs > 100 ppm. Epidemiological studies in Texas, at sites downwind of facilities that produce styrene-butadiene rubber that investigated BD exposures and increased mortality from any cause at low concentrations typical for the general population have not found a significant association between mortality from leukemia and exposure to BD, although there are only a few epidemiology studies that have been conducted (reviewed by Grant *et al.* (2007)). Figure 6 shows the 5^{th}, 50^{th}, and 95^{th} percentiles of the distribution of the cumulative number of BD HITs > 100 ppm in the UAB cohort study indicating SBR workers were frequently exposed to BD HITs > 100 ppm. In contrast, air monitoring data in Texas do not indicate the general population is exposed to BD HITs > 100 ppm. For example, Figure 7 provides 40-min BD concentrations (ppbv) at a monitoring site at Milby Park (2005 thru the first quarter of 2008). Milby Park is located predominantly downwind of nearby major industrial sources of BD emissions. There were only four times in a two-year period that the concentration of BD exceeded 200 ppb and the maximum peak BD concentration was 1,600 ppb. Maximum 40-min BD concentration data from 25 other ambient air monitoring sites in Texas indicate peak concentrations have not approached 1,600 ppb; in fact, maximum concentrations are less than 150 ppb. Other exposure studies indicate that the general population is exposed to concentrations of BD much lower than occupational workers (USEPA 2002, Gordon *et al.* 1999; Sapkota and Buckley 2003; Sapkota *et al.* 2005; Grant *et al.* 2007).

The inclusion of age and number of HITs > 100 ppm BD as covariates in the Cox regression modeling may result in cancer potency estimates that are more relevant to BD exposures experienced by the general population. Once age is in the model, inclusion of number of BD HITs results in a significant improvement in the fit (likelihood) (Sielken *et al.* 2007). The general population is not expected to be exposed to BD concentrations greater than 100 ppm, so

adjusting for BD HITs > 100 ppm as a covariate produces cancer potency estimates more relevant to BD exposures experienced by the general population.

Slikker *et al.* (2004) provides a discussion of the role of dose-dependent transitions in mechanisms of toxicity for BD as well as several other chemicals. Exposure to BD at high concentrations may result in a change from the hydrolytic pathways that are normally used by humans to form EBD to the formation of the more toxic metabolite, DEB (i.e., metabolic enzymes may be saturated) (Figure 1). In addition, DNA repair mechanisms as well as protective enzymes may become saturated and other protective cellular constituents may be depleted which could result in mechanisms of toxic tissue injury that are not relevant at exposures significantly less than 100 ppm. As mentioned previously, Albertini *et al.* (2001) showed a clear NOAEL for biomarkers of effect (hypoxanthine-guanine phosphoribosyltransferase (HPRT) mutations and chromosome aberrations) at mean BD exposure concentrations of 0.800 ppm in a study of workers in the Czech Republic (see Section 4.5 for additional information) and Sielken *et al.* (2007) analyses showed the absence of a statistically significant low-dose risk versus cumulative BD ppm-years for restricted data less than 300 ppm-years.

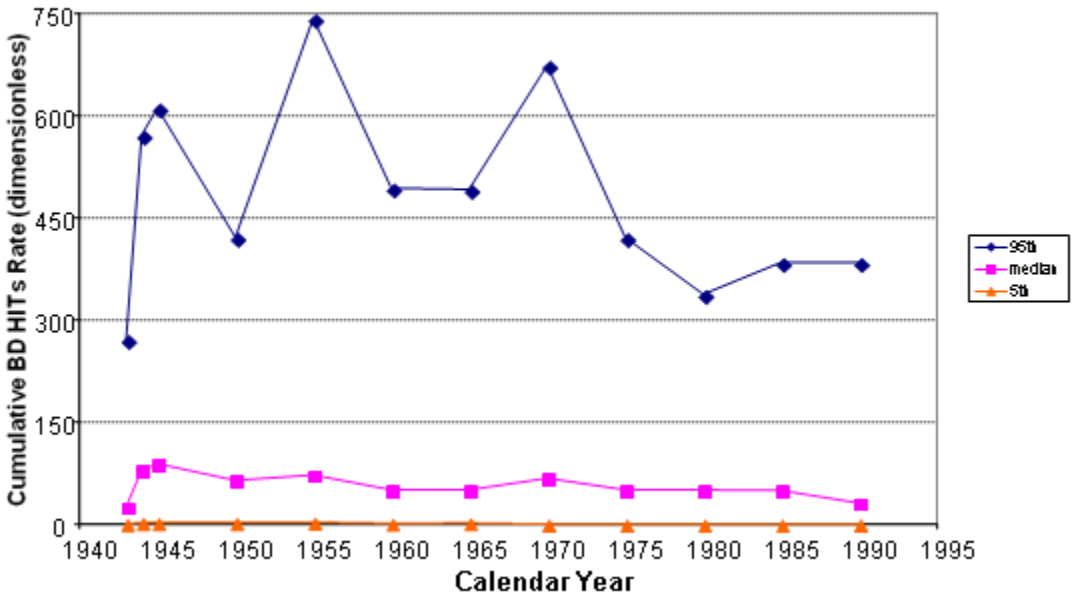

Figure 6 Distribution of BD HITs > 100 ppm among BD-Exposed Workers in a Calendar Year.

The cumulative number of BD HITs rate (dimensionless) versus calendar year is shown for the 5th, 50th, and 95th percentiles of the distribution among BD-exposed workers included in the UAB cohort study.

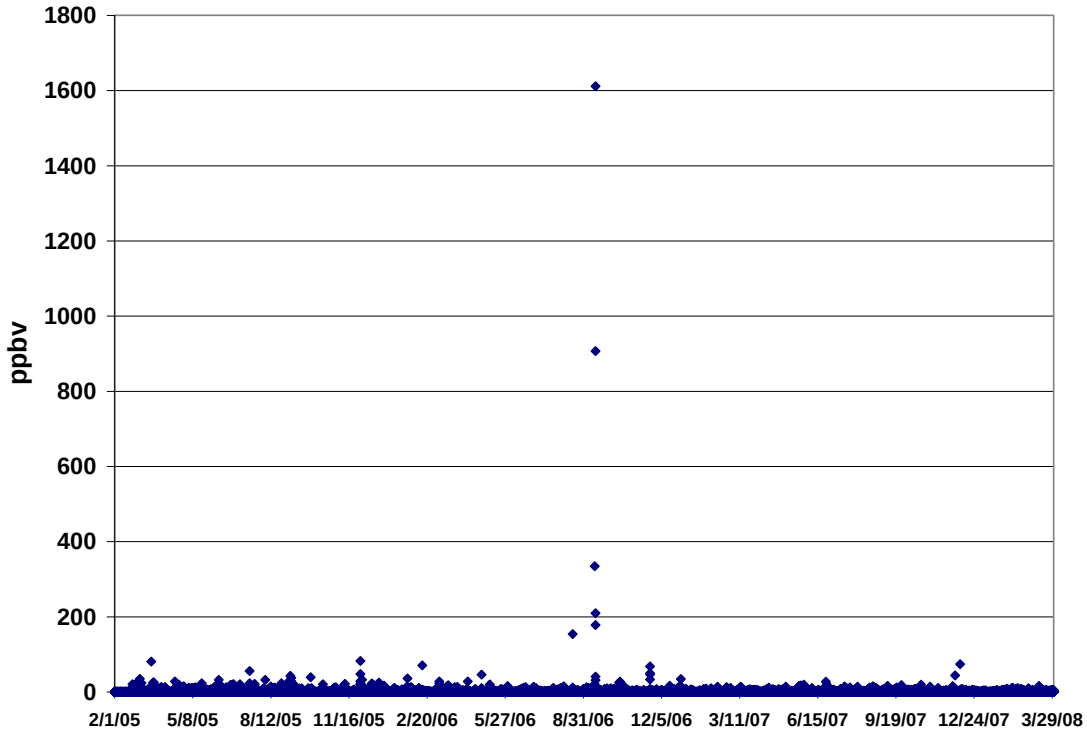

Figure 7 Forty-Minute BD Concentrations (ppbv) at Milby Park (2005 – first quarter of 2008).

Milby Park is located predominantly downwind of nearby major industrial sources of BD emissions (Grant *et al.* 2007). Forty-minute auto gas chromatography data.

Table 19 contains β, SE, and 95% UCL values when age & number of HITs > 100 ppm are included as covariates for the different models in Table 18. Table 20 contains URFs and 10^{-5}-risk air concentrations using Texas-specific mortality rates for 1999-2003 for all leukemia and Texas-specific survival rates for 2003 when ADAFs are incorporated based on the β and 95% UCL values in Table 16 (age only) and Table 19 (age & number of HITs).

Table 19 Age & Number of HITS > 100 ppm [a]

Covariates - Age & Number of HITs > 100 ppm	Model	Source	β (MLE) ± SE	β (95% UCL) [b]
	Cox log-linear ppm-years continuous [c] # of HITS continuous [e]	Cheng *et al.* (2007)	2.5E-04 ± 1.2E-04 [g]	4.474E-04
	Cox log-linear ppm-years mean-scored deciles [h] # of HITS categorical [f]	Sielken *et al.* (Appendix 6)	2.8E-04 ± 2.4E-04	6.748E-04
	Cox regression (restricted to lower 95% of exposure range) ppm-years continuous [c] # of HITS continuous [e]	Cheng and Delzell [d]	1.34E-03 ± 4.6E-04	2.097E-03
	Poisson linear ppm-years mean-scored deciles [h] # of HITS categorical [f]	Sielken *et al.* (2007)	1.89E-04 ± 3.6E-04	7.812E-04

a units are in ppm-years and based on occupational exposure concentrations

b β (95% UCL) = β(MLE) + (1.645 x SE)

c ppm-years is included as a continuous variable (untransformed) in a parametric model of the effect of ppm-years

d Personal communication, 1/30/2008 email from Dr. Cheng and Dr. Delzell. Cheng et al. (2007) reported results for Cox log-linear (restricted to lower 95% of exposure range) ppm-years continuous for age & other covariates, but not age only or age + # HITs. Dr. Cheng and Dr. Delzell provided the β and SE values for Cox log-linear continuous (restricted to lower 95% of exposure range) ppm-years for age and age + # HITs in the 1/30/2008 email.

e number of HITS > 100 ppm is included as a continuous variable (untransformed) in a parametric model of the effect of the number of HITs > 100 ppm

f number of HITS > 100 ppm is included as a categorical variable (based on quintiles) in a nonparametric model of the effect of the number of HITs > 100 ppm

g back calculated from the corresponding p-value in Cheng et al. (2007)

h ppm-years is included as a continuous variable with values grouped into mean-scored deciles (untransformed) in a parametric model of the effect of ppm-years

Table 20 Age & Number of HITS > 100 ppm; URFs and Air Concentrations Corresponding to 1 in 100,000 Extra Leukemia Risk a

Model **type of data**	**EC001** **URF (MLE)[b]** **10-5-risk air concentration using URF**	**EC001** **URF (MLE)[b]** **10-5-risk air concentration using URF**	**LEC001** **URF (95% UCL)[c]** **10-5-risk air concentration using URF**	**LEC001** **URF (95% UCL)[c]** **10-5-risk air concentration using URF**
Cox log-linear Cheng et al. (2007) ppm-years continuous [d]	age	1.433E-04/ppm 69.79 ppb	age	2.246E-04/ppm 44.53 ppb
Same as Above	age & HITs [g] 1.2-fold higher	1.235E-04/ppm 80.95 ppb	age & HITs [g] 1.02-fold higher	2.210E-04/ppm 45.24 ppb
Cox log-linear Cheng et al. (2007) and Sielken et al. (Appendix 6) ppm-years mean-scored deciles[f]	age	3.706E-04/ppm 26.98 ppb	age	5.494E-04/ppm 18.20 ppb
Same as above	age & HITs [h] 2.7-fold higher	1.383E-04/ppm 72.28 ppb	age & HITs [h] 1.6-fold higher	3.334E-04/ppm 29.99 ppb
Cox regression (restricted to lower 95% of exposure range) Cheng and Delzelle ppm-years continuousd	age	7.807E-04/ppm 12.81 ppb	age	1.097E-03/ppm 9.112 ppb
Same as above	age & HITs [g] 1.2-fold higher	6.621E-04/ppm 15.10 ppb	age & HITs [g] 1.1-fold higher	1.036E-03/ppm 9.651 ppb
Poisson linear Sielken et al. (2007) ppm-years mean-scored deciles[f]	age	6.976E-04/ppm 14.33 ppb	age	1.258E-03/ppm 7.946 ppb
Same as above	age & HITs h 8.9-fold higher	7.846E-05/ppm 127.4ppb	age & HITs [h] 3.9-fold higher	3.243E-04/ppm 30.83 ppb

a using Texas-specific mortality rates for 1999-2003 for all leukemia and Texas-specific survival rates for 2003 when ADAFs are incorporated

b URF = 0.001/EC001

c URF = 0.001/LEC001

d ppm-years is included as a continuous variable (untransformed) in a parametric model of the effect of ppm-years

e Personal communication, 1/30/2008 email from Dr. Cheng and Dr. Delzell. Cheng et al. (2007) reported results for Cox log-linear (restricted to lower 95% of exposure range) ppm-years continuous for age & other covariates, but not age only or age + # HITs. Dr. Cheng and Dr. Delzell provided the β and SE values for Cox log-linear continuous (restricted to lower 95% of exposure range) ppm-years for age and age + # HITs in the 1/30/2008 email.

f ppm-years is included as a continuous variable with values grouped into mean-scored deciles (untransformed) in a parametric model of the effect of ppm-years

g number of HITS > 100 ppm is included as a continuous variable (untransformed) in a parametric model of the effect of the number of HITs > 100 ppm

h number of HITS > 100 ppm is included as a categorical variable (based on quintiles) in a nonparametric model of the effect of the number of HITs > 100 ppm

Using URFs (MLE), the 10^{-5}-risk air concentrations for Cox log-linear, restricted continuous data if age + number of HITs are included as covariates, is 15.10 ppb (Table 20) as compared to 12.81 ppb if age is included as a covariate, approximately 1.2-fold higher. Using URFs (95% UCL), the 10^{-5}-risk air concentrations for Cox log-linear, restricted continuous data if age + number of HITs are included as covariates, is 9.651 ppb (Table 20) as compared to 9.112 ppb if age is included as a covariate approximately 1.1-fold higher. Therefore, estimated risks for the model based on the restricted data that adjusts for age only result in estimates of risks that are 6-20% higher than estimates from the same model that also adjusts for the number of BD HITS > 100 ppm. Similar results were obtained when the full data set was examined (i.e., 2% higher for the URF (95% UCL) to 20% higher for the URF (MLE)).

When categorical data were used (i.e., mean-scored deciles), the differences between the 10^{-5}-risk air concentrations for age only and age + # HITs were much greater: 1.6-fold higher for the URF (95% UCL) to 2.7-fold higher for the URF (MLE) for Cox log-linear mean-scored deciles and 3.9-fold higher for the URF (95% UCL) to 8.9-fold higher for the URF (MLE) for Poisson linear mean-scored deciles. As mentioned previously, Cheng *et al.* (2007) found that BD ppm-years and # of HITs > 100 ppm, both exposure variables, were weakly correlated for continuous (ungrouped) values (Pearson correlation coefficient of 0.30) as opposed to deciles (grouped) values (Pearson correlation coefficient of 0.80).

4.2.5.3 Effect of Occupational Exposure Estimation Error

One of the limitations of most epidemiological studies is potential exposure estimation error. Health Canada (2000) and USEPA (2002) expressed concerns about the validity of exposure estimates from the Delzell (1995, 1996) study. In the updated exposure estimates, Macaluso *et al.* (2004) used a more in-depth job, task, and exposure classification for the cohort, and exposure estimates were developed using exposure modeling, historical exposure data, and plant equipment analysis. Recently, Sathiakumar *et al.* (2007) assessed the validity of the BD exposure estimates by measuring the differences and correlations between calendar year- and job-specific estimates and measurements of BD concentrations at the Canadian Sarnia plant (a latex operation), one plant included in the UAB cohort. Sathiakumar *et al.* (2007) stated in their abstract, "Exposure misclassification may have been more severe for subjects from the validation study plant than for subjects from other plants in the mortality study." BD measurements from the late 1970s onward were available. Estimated concentrations were lower than measured concentrations before 1984 by approximately two-fold, whereas after 1984, estimated concentrations were higher than measured concentrations by approximately three-fold. On average, estimates were about 10% lower than measurements.

Macaluso *et al.* (2004) characterized each of the exposures in the JEM by a distribution. The analyses in Sielken *et al.* (2007) and Cheng *et al.* (2007) used the average of this distribution to characterize job exposure in the JEM and calculations of cumulative ppm-years. Sielken and Associates (2008) (Appendix 7) conducted a sensitivity analysis in order to investigate the effects of the exposure estimation errors identified by Sathiakumar *et al.* (2007) on the β and SE using the full data set and log-linear Cox regression modeling. Beta and SE from the following alternative data sets were determined:

1. The first alternative data set altered the exposure estimate (JEM) values so that prior to 1984 the exposure estimate JEM values were increased approximately 2-fold (i.e., 1.98-fold), and in 1984 and later years the exposure estimate JEM values were decreased approximately 3-fold (i.e., (1/0.37)-fold).

2. The second alternative data set altered the JEM values so that the exposure estimates prior to 1977 were left unchanged, the exposure estimate JEM values for 1977 through 1983 were increased approximately 2-fold (1.98-fold), and the exposure estimates JEM values for 1984 through 1991 were decreased approximately 3-fold [(1/0.37)-fold]. This alternative is the same as the first alternative except that the exposure estimates prior to 1977 were left unchanged because these years were not specifically addressed in Sathiakumar *et al.* (2007).
3. The third alternative data set altered the JEM values so that the exposure estimates prior to 1977 were left unchanged and the exposure estimate (JEM values) for each specific year of 1977 through 1991 were multiplied by the calendar-year specific value for "measurement / estimate" as shown by Table 1 in Appendix 7. The third alternative is the same as the second alternative except the calendar-year specific findings for 1977 to 1991 in Sathiakumar *et al.* (2007) were used.
4. The fourth alternative data set altered the JEM values so that these estimates are all divided by 0.90 corresponding to estimate (JEM value) = 0.90 x measurement because Sathiakumar *et al.* (2007) noted that, "On average, estimates were about 10% lower than measurements."

Table 21 Sensitivity analysis on exposure estimate validation study (Sathiakumar *et al.* 2007)

Data Set **Description of JEM Values**	**β + Standard Deviation of Estimate of β**	**95% UCL on β**	**EC001** **10-5-risk air concentration using URF**	**LEC001** **10-5-risk air concentration using URF**
Original Average in Macaluso Distribution	2.911E-04 + 1.03E-04	4.60E-04	72.65 ppb	45.94 ppb
1st Alternate Sathiakumar Average Calendar-Year Correction before 1984 and Average Calendar-Year Correction after 1983	1.469E-04 + 5.21E-05	2.33E-04	143.97 ppb	90.93 ppb
2nd Alternate Sathiakumar Average Calendar-Year Correction for 1977 through 1983 and Average Calendar-Year Correction for 1984 through 1991	2.478E-04 + 8.66E-05	3.90E-04	85.35 ppb	54.19 ppb
3rd Alternate Sathiakumar Calendar-Year Specific Correction for 1977 through 1991	2.468E-04 + 8.62E-05	3.89E-04	85.70 ppb	54.43 ppb
4th Alternate Sathiakumar Overall 10% Correction	2.620E-04 + 9.26E-05	4.14E-04	80.72 ppb	51.05 ppb

Table 21 shows a summary of results from Appendix 7 for the above mentioned alternate data sets. The 10^{-5}-risk air concentration using the URF (LEC_{001}) was 45.94 ppb based on the original JEM values and increased for all alternative data sets. The increased risk-based values ranged from 90.93 ppb for the 1st alternative data set to 51.05 ppb for the 4th alternative data set. The 10^{-5}-risk air concentration using the URF (EC_{001}) was 72.65 ppb based on the original JEM values and increased for all alternative data set. The increases ranged from 143.97 ppb for the 1st alternative data set to 80.72 ppb for the 4th alternative data set.

There was a pattern reversal in exposure estimates before and after 1984. However, exposures before 1984 were higher (in absolute value) and contributed more to the estimation of the slopes in the dose-response models. Increasing the exposure estimates before 1984 tended to decrease the estimated slopes and increase the estimated concentrations (ppb) corresponding to specified risk levels. This indicates that the β and SE calculated by Cheng *et al.* (2007) and Sielken *et al.*

(2007) were conservative and did not underestimate potency estimates based on concerns about exposure estimation error.

Sielken and Associates (Appendix 7) also considered two additional alternative data sets. The 5th and 6th alternative data sets replaced the average exposure estimated JEM values by the 5th or the 95th percentiles of these distributions, respectively. Then the modeling was done as before except the cumulative ppm-years were calculated using these 5th or 95th percentile JEM values instead of the average JEM values. The 10^{-5}-risk air concentration using the URF (LEC_{001}) was 45.94 ppb based on the original average exposure estimate JEM values and ranged from 20.41 ppb for the 5th percentile to 86.69 ppb for the 95th percentile (Appendix 7), only a four-fold difference. The validation study of Sathiakumar *et al.* (2007) on the updated exposure estimates of Macaluso *et al.* (2004) and the sensitivity study conducted by Sielken and Associates (2008) (Appendix 7) demonstrate the potency estimates derived by the TCEQ based on modeling by Cheng *et al.* (2007) and Sielken *et al.* (2007) have a higher confidence than potency estimates determined by USEPA (2002) using the old 1995 exposure estimates, fewer leukemia deaths, and fewer years of follow-up.

4.2.5.4 Dose-Response Modeling

Modeling results from several different models were presented and both β and upper 95% UCL estimates were reported in order to provide information on the residual uncertainty in the relative risk estimates based on different dose-response modeling:

- For the preferred model (Section 4.2.4), there was approximately a 1.4-fold difference between the 10^{-5}-risk air concentrations of 13.39 ppb calculated with URFs (MLE) versus 9.523 ppb for the URFs (95% UCL) (Table 17).
- The cancer potency estimates and 10^{-5}-risk air concentrations from the log-linear Cox regression model and the linear Poisson regression model using URFs (MLE) in Table 17 range from 7.471E-04 per ppm (13.39 ppb) to 1.371E-04 per ppm (72.93 ppb), a 5.4 fold difference.
- The cancer potency estimates and 10^{-5}-risk air concentrations using URFs (95% UCL) in Table 17 range from 1.193E-03 per ppm (8.381 ppb) to 2.149E-04 per ppm (46.53 ppb), a 5.5 fold difference. The preferred potency estimate based on the URF (95% UCL) of 1.050E-03 per ppm (9.523 ppb) (Table 17) is at the lower, conservative end of the range.

Linear Poisson regression and log-linear Cox regression models are commonly used to investigate dose-response relationships derived from occupational cohort epidemiologic studies based on mortality and are generally considered to be biologically-plausible models for cancer. As discussed previously, models using untransformed continuous (ungrouped) data are preferred over models using grouped data, so the potency estimates from the models using mean-scored deciles were not preferred. The Cox regression analysis using data restricted to the lower 95% of the exposure range was used because it is more conservative and to address concerns about sparse data and an erratic exposure-response relationship at high exposure concentrations. The results from the Cox regression model using restricted data are the most conservative among the Cox regression models analyzed here and slightly less conservative than the results from the Poisson regression using mean-scored deciles when only age is included as a covariate.

Cheng *et al.* (2007) also examined the BD ppm-years exposure-response relationships using natural logarithm (ln)-transformed and square-root transformed continuous BD ppm-years. These models each have advantages as discussed by Cheng *et al.* (2007). The models using ln-transformed and square-root transformed continuous BD ppm-years are not standard models and since the mechanism of action of BD is not sufficiently understood to justify the use of these models, the TS preferred the log-linear Cox regression model. In addition, the ln-transformed model may provide an unrealistically high slope in the low dose region and is, therefore, not preferred. One of the advantages of the ln-transformed and square-root transformed data is they may reduce the influence of data at extreme exposure values. The log-linear Cox regression models using restricted data are more conservative and may address concerns for the influence of data at extreme exposure values at the high exposure range (Section 4.2.5.3).

4.2.5.5 Use of Mortality Rates to Predict Incidence

The potency estimate for BD was calculated from mortality data because incidence data were not available. When using the BEIR IV methodology to calculate URFs and corresponding 10^{-5}-risk air concentrations based on mortality potency estimates, total leukemia mortality rates were used. Total leukemia incidence is higher than leukemia mortality because the survival rate for leukemia has improved through the years. In 1996-2003, the overall relative survival rate was nearly 50 percent (Leukemia & Lymphoma Society 2008). USEPA (2002) used leukemia incidence rates instead of mortality rates to calculate air concentrations based on a life-table analyses using the BEIR IV approach (NRC 1988) in an attempt to account for the uncertainty that potency estimates were based on mortality and not incidence.

The BEIR IV methodology for calculating excess risk is mathematically correct when the specified response is mortality and mortality rates are used but not when the specified response is mortality and incidence rates are used as was done by USEPA (2002). This error is demonstrated in Appendix 8 *Issues in Quantitative Epidemiology: Calculating Excess Risk When Specified Response is Mortality versus Incidence.* Appendix 8 shows that if the specified response is incidence, then the BEIR IV methodology for mortality cannot be used correctly. Teta *et al.* (2004) investigated the validity and implications of using a mortality-based leukemia relative rate model with background leukemia incidence rates, rather than mortality rates. They concluded that a biased estimate of excess lifetime risk will result, and the direction of the bias will vary by potency and the type of leukemia being modeled. Therefore, the TS did not use leukemia incidence rates to account for the uncertainty of calculating potency estimates for BD from mortality data. If the specified response is incidence and incidence rates are used, the BEIR IV methodology can be altered to account for incidence as demonstrated in Appendix 8.

Table 22 contains URFs and 10^{-5}-risk air concentrations calculated using restricted data, β (95% UCL) of 2.221E-03, Texas-specific mortality or incidence rates for 1999-2003 for all leukemia and Texas-specific survival rates for 2003 (Appendix 4), and ADAFs. If leukemia mortality rates are used in the Beir IV model for mortality, the 10^{-5}-risk air concentration is 9.112 ppb compared to 5.011 ppb using incidence rates, approximately 1.8 fold higher. Similar results are obtained when the Beir IV model for incidence is used (adjusted to correctly account for incidence dose-response based on equations in Appendix 8) (Table 22). The uncertainty in using mortality potency factors and mortality rates to predict incidence rates (i.e., to protect against developing leukemia) is approximately 1.8 fold, although the amount and direction of the bias may vary

(Teta *et al.* 2004). The TS will not use leukemia incidence rates to calculate air concentrations using mortality potency factors and the BEIR IV approach (NRC 1988) because it is mathematically incorrect. Given the inherent conservatism when calculating potency estimates, a less-biased estimate of risk based on mortality is better than a more-biased estimate based on incidence. The URF is considered to be sufficiently health-protective because the following conservative default procedures were followed in the calculation of the preferred URF of 1.1E-03 per ppm:

- A linear default was used to extrapolate to lower concentrations instead of using the log-linear Cox regression model to calculate the 10^{-5}-risk air concentrations, approximately 1.2 fold more conservative (Table 17);
- The URF (95% UCL) was used instead of the URF (MLE), approximately 1.4 fold more conservative (Table 17). As mentioned previously, the confidence intervals are indicators of the variability, and to some extent the uncertainty, in the dose-response curve for mortality. The risk of incidence will be lowered since using the URF (95% UCL) adds conservatism to the estimate;
- Data restricted to the lower 95% of the exposure range was used, ranging from 4-5 fold more conservative when compared to unrestricted data (Section 4.2.5.3);
- Model did not adjust for the number of HITs > 100 ppm that occur in occupational exposure but not in environmental exposures (Section 4.2.5.2). The URF would be 1.1 fold less conservative if number of HITs > 100 were adjusted for; and
- Model was based on the average BD concentration estimated by Macaluso *et al.* (2004) and did not incorporate the correction to the exposure estimates suggested by Sathiakumar *et al.* (2007) (Section 4.2.5.3 and Appendix 7).

Therefore, the total conservatism is much greater than the possible bias of 1.8-fold.

Table 22 Effects of using Total Leukemia Incidence Rates versus Mortality Rates [a]

	Dose-Response Model (mortality or incidence rates)	URF	10-5-Risk Air Concentration	Effect on 10-5-Risk Air Concentration
BEIR IV methodology for mortality	Mortality potency factors (mortality rates)	1.097E-03/ppm	9.112 ppb	1.8 fold higher 10-5 risk air concentration when using mortality rates, and not incidence rates
BEIR IV methodology for mortality	Mortality potency factors (incidence rates) c	1.996E-03/ppm	5.011 ppb	
BEIR IV methodology for incidence [b]	Mortality potency factors c (mortality rates) c	1.096E-03/ppm	9.126 ppb	1.8 fold higher 10-5 risk air concentration when using mortality rates, and not incidence rates
BEIR IV methodology for incidence [b]	Mortality potency factors c (incidence rates)	1.989E-03/ppm	5.028 ppb	

a Calculations were performed using restricted data, 95% UCL on β of 2.221E-03, Texas-specific mortality or incidence rates for 1999-2003 for all leukemia and Texas-specific survival rates for 2003 (Appendix 4), and ADAFs

b The BEIR IV methodology was altered to account for incidence if the specified response is incidence and incidence rates are used as demonstrated in Appendix 8

c Incorrect use of The BEIR IV methodology (Appendix 8)

4.2.6 Comparison of TCEQ's URF to USEPA's URF

USEPA published an inhalation URF of 0.08 per ppm in 2002. The URF is based on a Health Canada analysis of data from Delzell *et al.* (1995, 1996) using a linear relative rate model and was calculated for up to 85 years. Relative risks were evaluated with leukemia incidence rates, which is not mathematically correct as demonstrated in Appendix 8. Using the LEC_{01} (i.e., the 95% lower confidence limit of the exposure concentration associated with a 1% increased risk) of 0.254 ppm as the POD and a linear extrapolation to zero yielded a URF of 0.04 per ppm. An adjustment factor of 2 was applied to the URF to yield a final URF of 0.08 per ppm. This adjustment was applied to reflect evidence from studies in mice which suggest that extrapolating leukemia risks from a male-only occupational cohort may underestimate the cancer risks for the general public.The TCEQ derived an inhalation URF of 0.0011 per ppm based on the most current exposure estimates and updated epidemiological study conducted by the UAB group (Macaluso *et al.* 2004; Sathiakumar *et al.* 2005; Graff *et al.* 2005; HEI 2006). As mentioned previously, based on the validation study of Sathiakumar *et al.* (2007), the updated exposure

estimates of Macaluso *et al.* (2004) have a higher confidence than original exposure estimates used by USEPA. Relative risks were evaluated with Texas-specific leukemia mortality rates and survival rates and were calculated for up to 70 years, the default used by the TCEQ in an exposure analysis. The URF is based on the 95% UCL estimate derived with a log-linear Cox regression model, age implicitly included as covariate, and data restricted to the lower 95% of the exposure range (a more conservative value was chosen as a policy decision to address concerns about possible exposure misclassification at the high end of the exposure range) (Cheng *et al.* 2007). Using the LEC_{001} (i.e., the 95% lower confidence limit of the exposure concentration associated with a 0.1% increased risk) as the POD, a linear extrapolation to zero, and adjusting for the increased susceptibility of children using a life-table approach and applying ADAFs (Appendix 5) yields a URF of 0.0011 per ppm. USEPA's URF is approximately 70 times higher (i.e., more conservative) due to the following reasons:

- The updated and validated exposure estimates of the UAB group were approximately five times higher than the original estimates. The updated median ppm-years for all employees was 71 ppm-years versus 15 ppm-years for original estimates (Table VII, Macaluso *et al.* 2004) which makes USEPA's URF approximately five times higher;
- The TCEQ used a default exposure duration of 70 years (TCEQ 2006) whereas USEPA used an exposure duration of 85 years, which makes USEPA's URF approximately three times higher. The TCEQ will use the 70-year default to be consistent between evaluations for different chemicals (i.e., the risk from different chemicals will be more comparable if the dose-response was evaluated using a consistent 70-year exposure analysis). The use of 85 years instead of 70 years has been criticized for a variety of reasons. The dose-response modeling was not done based on person-years corresponding to older ages. The dose-response model based on early ages and older ages may be very different. Furthermore, the relevance of the dose metric (cumulative BD ppm-years) may differ for older ages;
- The TCEQ used an LEC_{001} to calculate the URF because it was within the observable range of the data whereas USEPA used an LEC_{01}, which is above the observable range of the data. This makes USEPA's URF approximately two times higher;
- The TCEQ used total leukemia mortality rates to calculate the URF whereas USEPA used total leukemia incidence rates, which makes USEPA's URF approximately 1.8 times higher; and
- The TCEQ did not apply an adjustment factor of two to the URF to reflect evidence from studies in mice which suggest that extrapolating leukemia risks from a male-only occupational cohort may underestimate the cancer risks for the general public because data on females workers exposed to BD did not indicate they were more sensitive. This adjustment makes USEPA's URF two times higher.

Consideration of the above differences accounts for approximately a 110-fold difference (5 x 3 x 2 x 1.8 x 2), more than the 70-fold difference when comparing URFs. This indicates that the TCEQ assessment was more conservative than USEPA's assessment in some regards. Minor differences between the TCEQ values and USEPA's values may relate to the use of the following:

- USEPA used potency estimates from Health Canada *, which appears to be a linear Poisson model with categorical variables, and the full range of the exposure data whereas TCEQ used the log-linear Cox regression model using continuous, untransformed data restricted to the lower 95% of the exposure range. This may account for the TCEQ's URF being only 70-fold lower rather than 110-fold lower.
- TCEQ used a longer follow-up in the current UAB study and TCEQ used 5 days/7 days as opposed to 240 days/364 days to convert from an occupational exposure to the general population.

While an exact partitioning of the 110-fold difference may not be possible, there are science-based and logical explanations accounting for most of the differences.

4.3. Welfare-Based Chronic ESL

No data were found regarding long-term vegetative effects.

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* In the Health Canada analyses that USEPA relied upon, the effects of age, years since hire, calendar year, race, and styrene exposure (ppm-years) were incorporated into the Poisson regression modeling using a flawed non-standard methodology which implicitly ignores most of the available data. The statistical procedure used by Health Canada stratified the data into a large number (29,403) of very fine strata. 29,352 strata (99.8% of the 29,403 strata) contain zero leukemia responses at all dose levels. These 29,352 strata contain 97.83% of the person years (i.e., most of the data in the study). The strata with zero leukemias at every dose had zero slope. The slope estimation procedure used by Health Canada did not include the strata with zero leukemias at every dose and hence did not include the corresponding zero slopes. The result is that the slope estimation procedure used by Health Canada biased the slope estimate for 1,3-butadiene toward higher values (i.e., overestimated the slope).

A general discussion of the problem of overly stratifying the covariates is given by N. E. Breslow and N. E. Day in Chapter 6 in Statistical Methods in Cancer Research, Volume I, The Analysis of Case-Control Studies, IARC, Lyon, France, 1980. Numerical examples are given by N. E. Breslow and N. E. Day in Chapters 4 and 5 in Statistical Methods in Cancer Research, Volume II, The Design and Analysis of Cohort Studies, IARC, Lyon, France, 1987.

4.4 Long-Term ESL and Values for Air Monitoring Evaluation

The chronic evaluation resulted in the derivation of the following values:

- $^{chronic}ESL_{nonlinear(nc)}$ = 9.9 µg/m^3 (4.5 ppb)
- Chronic ReV = 33 µg/m^3 (15 ppb)
- $^{chronic}ESL_{linear(c)}$ = 20 µg/m^3 (9.1 ppb)
- URF= 5.0E-04 per mg/m^3 (1.1E-03 per ppm)
  = 5.0E-07 per µg/m^3 (1.1E-06 per ppb).

The long-term ESL for air permit reviews is the $^{chronic}ESL_{nonlinear(nc)}$ of 9.9 µg/m^3 (4.5 ppb) because it is lower than the $^{chronic}ESL_{linear(c)}$ of 20 µg/m^3 (9.1 ppb) (Table 1). For evaluation of long-term ambient air monitoring data, the $^{chronic}ESL_{linear(c)}$ of 20 µg/m^3 (9.1 ppb) is lower than the chronic ReV of 33 µg/m^3 (15 ppb), although both values may be used for the evaluation of air data as well as the URF of 5.0E-04 per mg/m^3 (1.1E-03 per ppm) or 5.0E-07 per µg/m^3 (1.1E-06 per ppb). The $^{chronic}ESL_{nonlinear(nc)}$ (HQ = 0.3) is not used to evaluate ambient air monitoring data.

4.5 Other Relevant Information

The proceedings of the International Symposium on Evaluation of Butadiene and Chloroprene Health Risks, held in Charleston, South Carolina on September 20-22, 2005 have recently been published, and the findings and results from many of these articles have been cited in the Development Support Document (DSD). Refer to Himmelstein et al. (2007), which provides an excellent summary of the main findings of the symposium. A summary of the molecular epidemiology findings from Albertini et al. (2007) as summarized by Himmelstein et al. (2007) is reproduced here because of the significance of their findings. The references, which are in numerical format in the journal, have been supplemented with the author(s) names and year of publication.

“1.1.3. Molecular epidemiology

Albertini [9 *(Albertini et al. 2001)]* reported that the initial study of workers in the Czech Republic demonstrated a clear no-observed-adverse-effect level (NOAEL) for biomarkers of effect (hypoxanthine-guanine phosphoribosyltransferase (HPRT) mutations and chromosome aberrations) at mean BD exposure concentrations of 0.800 ppm.

This NOAEL reflects the maximum average exposure level experienced by these workers and was based on extensive external exposure assessments and a comprehensive series of biomarker responses, which included urine metabolites (M1 and M2) and hemoglobin adducts of epoxybutene and EBD (N-[2-dihydroxy-3-butenyl]valine = HB-Val and N- [2,3,4-trihydroxybutyl]valine = THB-Val, respectively), HPRT mutations, sister-chromatid-exchange frequencies and chromosomal aberrations determined by traditional methods and chromosome painting (fluorescence in situ hybridization). Both the urine metabolite and hemoglobin adduct concentrations proved to be excellent biomarkers of exposure. A second study of Czech workers was conducted at this same facility to compare biomarker responses in female and male employees [10 *(Albertini et al. 2007)*]. Mean BD exposure concentrations were lower in this second study than in the first, being 0.180 ppm and 0.370 ppm for females and males,

respectively. Again, there were no BD-associated elevations of HPRT mutation or chromosome aberration frequencies above background in either sex. Similarly, there was no difference between genders in the pattern of BD detoxification, as evidenced by urinary M1 and M2 levels. Females, however, appeared to absorb less BD per unit of exposure, as reflected by urine metabolite concentrations. Concentrations of the N,N-(2,3-dihydroxy-1,4-butadiyl)valine (pyr-Val) hemoglobin adduct, which is specific for the highly genotoxic 1,2:3,4-diepoxybutane (DEB) metabolite of BD, were measured in this second study and found to be below the level of quantification for all workers. Later presentations by Swenberg [11 *(Swenberg et al. 2007)*] and Boysen [12 (*Boysen et al. 2007)*] in this Symposium described extensive studies of pyr-Val concentrations in BD exposed rodents that, coupled with the results of this Czech worker study, indicate that DEB production in humans is below levels produced in mice or rats exposed to as little as 1.0 ppm BD by inhalation."

Chapter 5. References

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Appendix 1. Statistical Analyses of Developmental Endpoints

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The analyses performed by Hackett *et al.* (1987) did have some important statistical flaws that needed to be corrected. The statistical analyses reported by Green (2003) are valid and correct the flaws of Hackett *et al.* analyses. We have focused on the analyses of fetal body weights. The NOAEL based on the fetal body weights for this study is 40 ppm.

Hackett *et al.* (1987) conducted analyses of variance (ANOVA) on the average pup weight followed up by Student's t-tests comparing the average pup weight for different treatment groups. Their pairwise comparisons using Student's t-test do not adjust significance levels that occur for the number of multiple tests. In addition, their analyses did not adjust for well-known important covariate effects such as litter size. Hackett *et al.* analyses were based on dam's average pup weights instead of analyzing the individual pup weights and treating the dam as a random effect, which would result in a more powerful statistical test.

The Green (2003) reanalysis was based on analysis of covariance (ANCOVA) on the average pup weight and adjusting for covariates. In this context, Green used the Dunnett-Hsu test to compare the mean weights for each of the exposed groups to the mean weight for the control group after both are adjusted for the effects of the covariates. This is the specific situation for which the Dunnett-Hsu test was designed. Furthermore, the Dunnett-Hsu test is the appropriate test to use here to determine a NOAEL. Green considered the p-values in the Dunnett-Hsu test to draw his conclusions of significant effects. Green's discussion in A. Evaluation of Earlier Methods and B. Method of Re-Analysis is appropriate.

Green's analyses were based on dam's average pup weights instead of analyzing the individual pup weights and treating the dam as a random effect, which would result in a more powerful statistical test. The statistical conclusions reached by Green (2003) hold even when the more powerful statistical analyses where the individual pup weights are analyzed and the dams are treated as random effects.

Thus, the Green (2003) conclusions are reasonable and based on standard statistical analyses practices that were overlooked by Hackett *et al.* (2003). The NOAEL based on the fetal body weights for this study is 40 ppm.

Statistical Analyses Performed by Sielken & Associates

In addition to reviewing the methodology used in Hackett *et al.* (1987) and Green (2003), Sielken & Associates re-analyzed the fetal body weight data. This was to confirm the numerical results obtained by Green, do a sensitivity analysis with respect to the effects of covariates, and determine the outcome of the

more powerful statistical analyses where the individual pup weights are analyzed and the dams are treated as random effects. These analyses support the finding that the NOAEL based on the fetal body weights for this study is 40 ppm.

Table 1 contains an overview of the results in Tables 2 to 10 which contain the detailed analyses. The raw data used are given in Table 12. The statistical analyses were done in SAS Ver. 9. In the overview in Table 1, all comparisons to control were based on Dunnett-Hsu tests and were one-sided tests for a decrease in fetal body weight compared to control. The outcomes of the more powerful statistical analyses where the individual pup weights are analyzed and the dams are treated as random effects were comparable to the outcomes obtained with the Green ANCOVA model. The results for 1 Covariate (Litter Size) are highlighted since this covariate was always statistically significant at the 5% significance level – the 2nd Covariate (% Males in Litter) was significant for the Males Only analyses.

Table 1. Overview of Statistical Analyses of Fetal body weight Data: The results for 1 Covariate (Litter Size) are highlighted since this covariate was always statistically significant at the 5% significance level – the 2nd Covariate (% Males in Litter) was significant for the Males Only analyses

Table #	Model: Mixed Model: (1) Based on Mean Data (2) Based on Individual Data and Dam as Random Effect	Sex	# of Covariates	Covariates (1) Litter Size (2) % Males in Litter	p-value in Dunnett-Hsu one-sided comparison to control		
					dose=40	200	1,000
2	(1)	M&F	2	(1) & (2)	0.1354	<0.0001	<0.0001
2	(2)	M&F	2	(1) & (2)	0.1383	<0.0001	<0.0001
3	(1)	M&F	1	(1)	0.1120	<0.0001	<0.0001
3	(2)	M&F	1	(1)	0.1184	<0.0001	<0.0001
4	(1)	M&F	0	None	0.0832	<0.0001	<0.0001
4	(2)	M&F	0	None	0.0849	<0.0001	<0.0001
5	(1)	F	2	(1) & (2)	0.2091	<0.0001	<0.0001
5	(2)	F	2	(1) & (2)	0.2373	<0.0001	<0.0001
6	(1)	F	1	(1)	0.1919	<0.0001	<0.0001
6	(2)	F	1	(1)	0.2298	<0.0001	<0.0001
7	(1)	F	0	None	0.1427	<0.0001	<0.0001
7	(2)	F	0	None	0.1854	<0.0001	<0.0001
8	(1)	M	2	(1) & (2)	0.0687	<0.0001	<0.0001
8	(2)	M	2	(1) & (2)	0.0795	<0.0001	<0.0001
9	(1)	M	1	(1)	0.0603	<0.0001	<0.0001
9	(2)	M	1	(1)	0.0695	<0.0001	<0.0001
10	(1)	M	0	None	0.0408	<0.0001	<0.0001
10	(2)	M	0	None	0.0479	<0.0001	<0.0001

In order to obtain a copy of Tables 2-10 (Benchmark Dose Modeling Output) or Table 11. Fetal Body Weight Data of Appendix 1, please send an email to the Toxicology Division providing the name of the DSD and the requested data to the following email address: tox@tceq.texas.gov.

Appendix 2. BMC Modeling for Acute ReV

Table 2A. Dose-Response Data for Maternal Toxicity Endpoints

Dose (ppm)	Mean	Number of Litters	Calculated Standard Deviation **	Standard Error	% Control response	Coefficient of Variation (CV)
Whole-body weight (gm) day 18						
0	54.90	18	5.134	1.21	100%	0.09
40	55.40	19	4.751	1.09	101%	0.09
200	52.50	21	4.628	1.01	96%	0.09
1000	50.80	20	3.846	0.86	93%	0.08
Extragestational weight gain (gm)						
0	7.60	18	2.036	0.48	100%	0.27
40	6.99	19	1.656	0.38	92%	0.24
200	6.20	21	1.741	0.38	82%	0.28
1000	5.91	20	1.252	0.28	78%	0.21
Body weight gain (gm) gestation days 11-16						
0	13.30	18	2.546	0.60	100%	0.19
40	12.70	19	1.744	0.40	95%	0.14
200	11.40	21	2.291	0.50	86%	0.20
1000	10.60	20	1.789	0.40	80%	0.17
Gravid uterine weight (gm)						
0	19.30	18	4.243	1.00	100%	0.22
40	20.30	19	3.487	0.80	105%	0.17
200	18.00	21	3.987	0.87	93%	0.22
1000	16.80	20	2.996	0.67	87%	0.18
Extragestational weight (gm)						
0	35.50	18	2.036	0.48	100%	0.06
40	35.10	19	1.918	0.44	99%	0.05
200	34.50	21	2.108	0.46	97%	0.06
1000	34.10	20	1.610	0.36	96%	0.05

* Hackett et al. (1987b)
** Standard deviation = standard error x square root of number of litters

Table 2B. Dose-Response Data Fetal Toxicity Endpoints

Dose (ppm)	Mean	Number of Litters	Calculated Standard Deviation **	Standard Error	% Control response	Coefficient of Variation (CV)
Mean placental weight (mg) males and females (mean per litter)						
0	86.80	18	12.685	2.99	100%	0.15
40	85.40	19	9.982	2.29	98%	0.12
200	78.60	21	14.848	3.24	91%	0.19
1000	72.60	20	8.408	1.88	84%	0.12
Mean fetal weight (gm) males and females (mean per litter)						
0	1.34	18	0.127	0.03	100%	0.09
40	1.28	19	0.044	0.01	96%	0.03
200	1.13	21	0.092	0.02	84%	0.08
1000	1.04	20	0.134	0.03	78%	0.13
Abnormal sternebrae (mean percent per litter)						
0	0.60	18	0.900		100%	1.50
40	0.40	19	0.700		67%	1.75
200	0.40	21	0.800		67%	2.00
1000	0.80	20	1.300		133%	1.63
Supernumerary ribs (mean percent per litter)						
0	1.70	18	2.300		100%	1.35
40	1.60	19	2.100		94%	1.31
200	6.00	21	3.600		353%	0.60
1000	9.90	20	3.000		582%	0.30
Reduced ossification (all sites combined) (mean percent per litter)						
0	1.70	18	1.700		100%	1.00
40	1.20	19	1.500		71%	1.25
200	2.70	21	2.700		159%	1.00
1000	3.90	20	2.600		229%	0.67

* Hackett et al. (1987b)
** Standard deviation = standard error x square root of number of litters

Table 2C. Summary of BMC Modeling Results for Maternal Effects

Linear Model 4 doses		Linear Model 3 doses		Unrestricted Power Model 4 doses	
Whole-body weight (gm) Day 18					
homogeneous variance		homogeneous variance		homogeneous variance	
Test 1 0.02995		Test 1 0.3509	**X**	Test 1 0.02995	
Test 2 0.6543		Test 2 0.9013		Test 2 0.6543	
Test 3 0.6543		Test 3 0.9013		Test 3 0.6543	
Test 4 0.2575		Test 4 0.4884		Test 4 0.1941	
AIC	320.569569			AIC	321.54225
Scaled residual	< abs value of 2			Scaled residual	< abs value of 2
BMC10 =	1343.75			BMC10 =	1403.64
BMCL10 =	895.747			BMCL10 =	598.977
BMC 1 SD =	1120.78			BMC 1 SD =	962.47
BMCL 1SD =	732.341			BMCL 1SD =	304.564
Extragestational weight gain (gm)					
homogeneous variance		homogeneous variance		homogeneous variance	
Test 1 0.01364		Test 1 0.1505	**X**	Test 1 0.01364	
Test 2 0.2158		Test 2 0.6481		Test 2 0.2158	
Test 3 0.2158		Test 3 0.6481		Test 3 0.2158	
Test 4 0.0927	**X**	Test 4 0.5343		Test 4 0.4245	
				AIC	164.106882
				Scaled residual	< abs value of 2
				BMC10 =	31.362
				BMCL10 =	3.46E-05
				BMC 1 SD =	722.796
				BMCL 1SD =	51.3032
Body weight gain (gm) Days 11-16					
homogeneous variance		homogeneous variance		homogeneous variance	
Test 1 0.001187		Test 1 0.03683		Test 1 0.001187	
Test 2 0.2651		Test 2 0.2566		Test 2 0.2651	
Test 3 0.2651		Test 3 0.2566		Test 3 0.2651	
Test 4 0.07957	**X**	Test 4 0.7342		Test 4 0.339	
		AIC	153.301194	AIC	199.608973
		Scaled residual	< abs value of 2	Scaled residual	< abs value of 2
		BMC10 =	145.382	BMC10 =	108.232
		BMCL10 =	94.2853	BMCL10 =	5.96473
		BMC 1 SD =	237.988	BMC 1 SD =	392.348
		BMCL 1SD =	148.203	BMCL 1SD =	63.495
Gravid uterine weight (gm)					
homogeneous variance		homogeneous variance		homogeneous variance	
Test 1 0.05228	**X**	Test 1 0.369	**X**	Test 1 0.05228	**X**
Test 2 0.4485		Test 2 0.6955		Test 2 0.4485	
Test 3 0.4485		Test 3 0.6955		Test 3 0.4485	
Test 4 0.2653		Test 4 0.2733		Test 4 0.1333	
Extragestational weight (gm)					
homogeneous variance		homogeneous variance		homogeneous variance	
Test 1 0.263	**X**	Test 1 0.6113	**X**	Test 1 0.263	**X**
Test 2 0.6542		Test 2 0.9104		Test 2 0.6542	
Test 3 0.6542		Test 3 0.9104		Test 3 0.6542	
Test 4 0.4253		Test 4 0.7356		Test 4 0.6608	

X =Test 1-4 results unacceptable **Test 1** p values > 0.05; **Test 2** determines whether a homogeneous or nonhomogeneous variance applies (p > 0.1 = homogeneous variance, p < 0.1 = nonhomogeneous variance); **Test 3** p value < 0.1; **Test 4** goodness of fit p value < 0.1

Table 2D. Summary of BMC Modeling Results for Fetal Effects

Linear Model 4 doses		Linear Model 3 doses		Unrestricted Power Model 4 doses	
Mean placental weight per litter (mg) males and females					
nonhomogeneous variance *		homogeneous variance		nonhomogeneous variance *	
Test 1 0.0004354		Test 1 0.09712	**X**	Test 1 0.0004354	
Test 2 0.05768		Test 2 0.215		Test 2 0.05768	
Test 3 0.04312	**X**	Test 3 0.215		Test 3 0.04312	**X**
Test 4 0.7669		Test 4 0.9487		Test 4 0.9837	
AIC	466.096041			AIC	467.565607
Scaled residual	< abs value of 2			Scaled residual	< abs value of 2
BMC05 =	344.446			BMC05 =	123.276
BMCL05 =	255.57			BMCL05 =	4.16675
BMC 1 SD =	1063.26			BMC 1 SD =	874.047
BMCL 1SD =	733.771			BMCL 1SD =	233.341
Mean fetal weight per litter (gm) males and females					
nonhomogeneous variance		nonhomogeneous variance *		nonhomogeneous variance	
Test 1 <.0001		Test 1 <.0001		Test 1 <.0001	
Test 2 <.0001		Test 2 0.0001236		Test 2 <.0001	
Test 3 <.0001	**X**	Test 3 <.0001	**X**	Test 3 <.0001	**X**
Test 4 <.0001	**X**	Test 4 0.3503		Test 4 0.01814	**X**
		AIC	-212.273267		
		Scaled residual	< abs value of 2		
		BMC05 =	65.7926		
		BMCL05 =	54.7521		
		BMC 1 SD =	94.7601		
		BMCL 1SD =	71.78		
Abnormal sternebrae (Mean percent per litter)					
nonhomogeneous variance		homogeneous variance		nonhomogeneous variance	
Test 1 0.07281	**X**	Test 1 0.7441	**X**	Test 1 0.07281	**X**
Test 2 0.02859		Test 2 0.5637		Test 2 0.02859	
Test 3 0.9033		Test 3 0.5637		Test 3 0.9033	
Test 4 0.2526		Test 4 0.4958		Test 4 0.1857	
Supernumerary ribs (Mean percent per litter)					
nonhomogeneous variance		nonhomogeneous variance		nonhomogeneous variance	
Test 1 <.0001		Test 1 <.0001		Test 1 <.0001	
Test 2 0.06411		Test 2 0.02879		Test 2 0.06411	
Test 3 0.364		Test 3 0.8166		Test 3 0.364	
Test 4 <.0001	**X**	Test 4 0.07209	**X**	Test 4 0.001961	**X**
Reduced ossification (all sites combined) (Mean percent per litter)					
nonhomogeneous variance		nonhomogeneous variance		nonhomogeneous variance	
Test 1 0.0002402		Test 1 0.008605		Test 1 0.0002402	
Test 2 0.02047		Test 2 0.01733		Test 2 0.02047	
Test 3 0.6049		Test 3 0.6737		Test 3 0.6049	
Test 4 0.01897	**X**	Test 4 0.08082	**X**	Test 4 0.01417	**X**

X =Test 1-4 results unacceptable **Test 1** p values > 0.05; **Test 2** determines whether a homogeneous or nonhomogeneous variance applies (p > 0.1 = homogeneous variance, p < 0.1 = nonhomogeneous variance); **Test 3** p value < 0.1; **Test 4** goodness of fit p value < 0.1

* Both a nonhomogeneous and homogeneous variance were used to model the data. The scaled residuals for a nonhomogeneous variance produced slightly smaller scaled residuals in the low-dose region of the dose response curve, so the results from a nonhomogeneous variance are reported.

Table 2E. Table of Data and Estimated Values of Interest

Whole-body weight Day 18 Linear Model 4 doses

Table of Data and Estimated Values of Interest

Dose	N	Obs Mean	Est Mean	Obs Std Dev	Est Std Dev	Scaled Res.
0	18	54.9	54.6	5.13	4.56	0.256
40	19	55.4	54.5	4.75	4.56	0.897
200	21	52.5	53.8	4.63	4.56	-1.32
1000	20	50.8	50.6	3.85	4.56	0.236

Mean placental weight per litter Linear Model 4 doses

Table of Data and Estimated Values of Interest

Dose	N	Obs Mean	Est Mean	Obs Std Dev	Est Std Dev	Scaled Res.
0	18	86.8	84.5	12.7	13	0.746
40	19	85.4	84	9.98	12.8	0.469
200	21	78.6	82.1	14.9	12.1	-1.31
1000	20	72.6	72.2	8.41	8.6	0.187

Whole-body weight Day 18 Unrestricted Power Model 4 doses

Table of Data and Estimated Values of Interest

Dose	N	Obs Mean	Est Mean	Obs Std Dev	Est Std Dev	Scaled Res.
0	18	54.9	55.2	5.13	4.53	-0.324
40	19	55.4	54.4	4.75	4.53	0.961
200	21	52.5	53.3	4.63	4.53	-0.781
1000	20	50.8	50.6	3.85	4.53	0.171

Mean placental weight per litter Unrestricted Power Model 4 Doses

Table of Data and Estimated Values of Interest

Dose	N	Obs Mean	Est Mean	Obs Std Dev	Est Std Dev	Scaled Res.
0	18	86.8	86.3	12.7	13.1	0.156
40	19	85.4	84	9.98	12.4	0.476
200	21	78.6	80.6	14.9	11.5	-0.814
1000	20	72.6	72.1	8.41	9.17	0.224

Extragestational weight gain Unrestricted Power Model 4 doses

Table of Data and Estimated Values of Interest

Dose	N	Obs Mean	Est Mean	Obs Std Dev	Est Std Dev	Scaled Res.
0	18	7.6	7.62	2.04	1.65	-0.052
40	19	6.99	6.81	1.66	1.65	0.473
200	21	6.2	6.42	1.74	1.65	-0.605
1000	20	5.91	5.83	1.25	1.65	0.209

Mean fetal weight per litter Linear Model - 3 doses

Table of Data and Estimated Values of Interest

Dose	N	Obs Mean	Est Mean	Obs Std Dev	Est Std Dev	Scaled Res.
0	18	1.34	1.33	0.127	0.0958	0.408
40	19	1.28	1.29	0.044	0.0936	-0.481
200	21	1.13	1.13	0.092	0.0844	0.0803

Body weight gain (GD11-16) Linear Model 3 doses

Table of Data and Estimated Values of Interest

Dose	N	Obs Mean	Est Mean	Obs Std Dev	Est Std Dev	Scaled Res.
0	18	13.3	13.2	2.55	2.16	0.213
40	19	12.7	12.8	1.74	2.16	-0.259
200	21	11.4	11.4	2.29	2.16	0.0494

Body weight gain (GD11-16) Unrestricted Power Model 4 doses

Table of Data and Estimated Values of Interest

Dose	N	Obs Mean	Est Mean	Obs Std Dev	Est Std Dev	Scaled Res.
0	18	13.3	13.4	2.55	2.07	-0.114
40	19	12.7	12.4	1.74	2.07	0.622
200	21	11.4	11.7	2.29	2.07	-0.684
1000	20	10.6	10.5	1.79	2.07	0.202

Benchmark Modeling Results Using the Power Model (11/19/07 Email from Bruce Allen)

From: "Bruce Allen" <bruce_allen@verizon.net>
To:"'Roberta Grant'" <RGrant@tceq.state.tx.us>
Date: 11/19/2007 8:10:50 AM
Subject: RE: Benchmark modeling results using the power model

Dr. Grant,

Sorry to take so long in getting back to you. Your table is correct on the estimates from the unrestricted power model. Note that the AIC values are all the same, because you are fitting the same model to the same data set each time; changing the definition of the BMR does not change any of that. And, as the full output shows, the fit to the data points is quite good.

As to the differences in the BMC and BMCL - that is totally a product of the curve shape and it can become pronounced when an unrestricted model is used. In such a model, you can get very steep initial (low-dose) slopes and in the search for lower bounds, such a model allows for even greater initial slopes among the candidates that might give the BMCL. So, many people have avoided such models because they can indeed give bigger differences between the BMC and BMCL.

The reason that the restricted model does not give such big differences is the fact the restriction essentially makes the linear fit the worst case (low-dose slope does not get progressively larger). So, as in this case, when the best fit is linear, then the search for lower bounds cannot include anything more extreme than a linear fit and the class of possible model parameter values that gives a good enough likelihood (in the BMCL determination) only includes some slightly steeper (but still linear) dose response shapes.

This, to me, is an arbitrary constraint, especially when the fit to the data is so bad with a restricted model. The results indicate a highly nonlinear dose-response, so why not let the model capture that behavior? That is what the unrestricted model does. Or (probably not an option here) find a better model that does not allow for extreme low-dose shapes but still does capture the observed dose-response pattern.

So, I am left with the impression that the unrestricted model fit to all the data points is still the best - but I would stick with a BMR defined in terms of 1 sd change. As we discussed on the phone, that allows for consistency across endpoints and assessments. And, in this particular case, it does not get you into the region where the low-dose shape is too dominant in determining what your BMCL is. That may not always be the case, but it does help you here. Just my 2 cents.

Bruce

———

From: Roberta Grant [mailto:RGrant@tceq.state.tx.us]
Sent: Wednesday, October 31, 2007 10:22 AM

To: bruce_allen@verizon.net
Cc: Joan Strawson; Angela Curry; Joseph Haney; Michael Honeycutt
Subject: Benchmark modeling results using the power model

Dr. Allen, it was a pleasure to participate in the teleconference with you yesterday. Your comments and suggestions were very helpful. There was a question about the output from the unrestricted power model using a BMR of 1 x SD, 0.77 x SD, and a 10% reduction. I've attached the BMCL modeling results using these different BMR rates. Using four doses and an unrestricted power model, these are the values I get:

Decrease in Extragestational Weight Gain 4 Doses

Bmc bmcl AIC
Unrestricted Power 1 x SD
722.8 51.3 164.1
Unrestricted Power 0.77 x SD
250.2 1.89 164.1
Unrestricted Power 10% reduction
31.36 3.45E-05 164.1

As discussed yesterday during the teleconference, I get exactly the same results as you did when using the BMR of 1 x SD, but very different results when using 0.77 x SD. I notice that there are big differences between the BMC and BMCL values. Is that normal or acceptable? I notice that for the restricted power model, the differences between the BMC and BMCL are generally less than two (see attached "extragestational review table").

Again, thanks for your comments. Roberta

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CC:"'Joan Strawson'" <jstrawson@nc.rr.com>, "'Angela Curry'" <ACurry@tceq.state.tx.us>, "'Joseph Haney'" <JHaney@tceq.state.tx.us>, "'Michael Honeycutt'" MHoneycu@tceq.state.tx.us

Appendix 3. Statistical Analyses of Reproductive Endpoints

Robert L. Sielken Jr., Ph.D., and Ciriaco Valdez Flores, Ph.D., P.E.

Sielken & Associates Consulting Inc.

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August 6, 2007

TCEQ Contract 582-7-81521

EPA's 2002 final risk assessment for BD (USEPA. 2002. Health Assessment of 1,3-Butadiene. EPA/600/P-98/001F) derived a reference concentration using the ovarian atrophy in female mice exposed to butadiene via inhalation. This animal study was conducted by the NTP in 1993 (NTP. 1993. Toxicology and carcinogenesis studies of 1,3-butadiene (CAS No. 106-99-0) in B6C3F1 mice (inhalation studies). Research Triangle Park, NC: National Toxicology Program, U.S. Public Health Service, U.S. Department of Health and Human Services. TR 434). EPA used a Weibull time-to-tumor dose-response model to fit the time-to-ovarian atrophy data and excluded the highest dose group because of excessive early mortality. The ECs and LECs for ovarian atrophy were calculated at an equivalent human age of 50 years "to reflect only the time before average age at menopause when follicles are no longer present and available for ovulation, because in the mouse studies of ovarian atrophy, the atrophy occurs as a result of follicular failure."

In the NTP 1993 critical study, female mice were exposed to 0, 6.25, 20, 62.5, 200, or 625 ppm BD for 6 hours/day, 5 days/week for two years (i.e., equivalent to 0, 1.12, 3.57, 11.2, 35.7, and 111.6 ppm BD of continuous exposure – for example, $6.25 \times (5/7) \times (6/24) = 1.12$). The air concentration 6.25 ppm was identified as a LOAEL for ovarian atrophy. The final 2002 EPA's risk assessment for BD reports several analyses of these data, including application of a log-logistic model, a quantal Weibull model, and a Weibull time-to-response model.

The final Weibull time-to-response model that EPA used is linear in dose with time raised to a power. EPA used TOX_RISK version 3.5 (Crump *et al.*, ICF Kaiser International, Ruston, LA) for the model fitting and the estimation of the ECs and LECs. In February 2006, the Olefins Panel of the American Chemistry Council asked the Sapphire Group, Inc. to recalculate EPA's ECs and LECs for ovarian atrophy (Kirman, C. R. and M. L. Gargas. 2006. Benchmark Dose Analyses for Reproductive and Developmental Endpoints for 1,3-Butadiene, Submitted to Olefins Panel, American Chemistry Council, Arlington, VA, February 2006). The Sapphire Group, Inc.'s report included the time-to-response data for ovarian atrophy of the NTP 1993 study, and those data are reproduced here in Attachment A.

Sielken & Associates Consulting, Inc. reanalyzed the ovarian atrophy data using the Weibull time-to-response model and the data presented in Attachment A. The linear Weibull time-to-response model had the following form:

Probability of a response (ovarian atrophy) by week T at dose d =

$$1 - \exp \{ - [Q_0 + Q_1 \times d] \times T^Z \}.$$

Tables 1 and 2 list the results of the analyses when the highest exposure group is not included in the estimation of the model and when all exposure groups are included, respectively. The results labeled SA# were calculated using Sielken & Associates, Inc.'s GEN.T software package – however, Sielken & Associates verified that the parameter estimates are identical to those estimated with TOX_RISK version 3.5. The LEC_{10} values for the SA# analyses in the table were estimated using 99 simulated bootstrap data sets. The two analyses in addition to EPA's analyses included in Tables 1 and 2 are:

1) Analysis SA1 parallels the analysis performed by EPA. The small discrepancies between the SA1 and EPA analyses may be due to assumptions that EPA may have made and did not describe in their report.
2) Analysis SA2 uses a modified data set in which all animals that lived beyond age 521 days (74.3 weeks – which is equivalent to 50 years in a 70-year human lifetime -- (50/70) × 104 weeks) were excluded from the parameter estimation.

In Tables 1 and 2, the range of EC_{10} values derived by EPA, SA1, and SA2 analyses is 1.05 to 1.25 ppm whereas the range of the LEC_{10} values derived by EPA, SA1, and SA2 analyses is 0.768 to 0.958 ppm.

Table 1 and 2 also show the results for concentrations corresponding to an extra risk of 0.05. Because the Weibull time-to-tumor model in these analyses is linear in dose, the EC_{05} and LEC_{05} values are approximately half the corresponding EC_{10} and LEC_{10} values.

Table 1. Parameters (Q_0, Q_1, and Z) for Weibull time-to-response model for ovarian atrophy and corresponding human benchmark 1,3-butadiene exposure concentrations for extra risks of 0.1 and 0.05 at 50 years of age using different methods of calculation – **excluding** the highest dose group

Analysis	Q_0	Q_1	Z	EC_{10}	LEC_{10}	EC_{05}	LEC_{05}
EPA	4.86×10^{-6}	7.06×10^{-6}	2.21	1.05	0.878	n/a	n/a
SA1	6.96×10^{-6}	8.62×10^{-6}	2.15	1.15	0.881	0.560	0.429
SA2	6.76×10^{-23}	6.90×10^{-5}	1.66	1.18	0.768	0.573	0.374

Table 2. Parameters for Weibull time-to-response model for ovarian atrophy and corresponding human benchmark 1,3-butadiene exposure concentrations for extra risks of 0.1 and 0.05 at 50 years of age using different methods of calculation – **including** the highest dose group

Analysis	Q_0	Q_1	Z	EC_{10}	LEC_{10}	EC_{05}	LEC_{05}
EPA	9.01×10^{-6}	1.32×10^{-6}	2.58	1.13	0.958	n/a	n/a
SA1	1.68×10^{-6}	2.04×10^{-6}	2.47	1.25	0.949	0.607	0.462
SA2	3.61×10^{-25}	1.95×10^{-6}	2.49	1.17	0.812	0.569	0.396

The estimated values of EC_{10} and LEC_{10} are close to the lowest experimental dose (1.12 ppm) while the values of EC_{05} and LEC_{05} are approximately half way between the lowest experimental dose and zero. The values of EC_{05} and LEC_{05} can be used if the dose-response relationship below the lowest experimental dose is believed to be the linear Weibull time-to-response model fit to the data. The assumption of linearity below the lowest experimental dose is usually conservative and, therefore, health protective. However, the motivation behind the benchmark dose methodology is to identify the point of departure (EC or LEC) to be within the range of the experimental data (the range of the non-zero doses in the experimental data) and to be a dose whose risk can be reasonably reliably estimated without undue sensitivity to the dose-response model selected or the model estimation. Here, the EC_{05} and LEC_{05} in the SA1 and SA2 analyses are below the range of the experimental data and, hence, introduce an additional element of uncertainty into the point of departure.

The EPA and SA1 analyses include ovarian atrophy responses beyond the equivalent of age 50 years in humans. These older-age responses in mice may not be relevant to humans and may inappropriately impact the fitted dose-response model used to estimate the risk at age 50. SA2 eliminates all animals that lived beyond the equivalent of age 50. However, it is known that some of these animals did not have an observed response (ovarian atrophy) and this information is

ignored/lost and not incorporated into the dose-response modeling as it should be. The fitted models for all the mice (analyses SA1) are very similar to the fitted models for only mice that died on or before week 74.3 (analyses SA2). This suggests that the older-age animals in the SA1 analyses are not distorting those analyses. Therefore, the results for analyses SA1 are preferable to the SA2 analyses because the SA1 analyses include more data (i.e., mice that lived past 74.3 weeks) and the inclusion of mice older than 74.3 weeks does not distort the fit of the model. In other words, the models fit to either all the mice (analyses SA1) or only to mice that died on or before week 74.3 are (analyses SA2) very similar but the confidence limits for analyses SA1 are more reliable because they are based on more animals.

The ovarian atrophy data were analyzed excluding the highest dose group (Table 1) and also including all the data (Table 2). The analyses that exclude the high dose were performed to parallel those analyses used by EPA. Traditionally, EPA drops the highest dose group when the model does not fit the data well due to some biological phenomenon or when quantal data are fit with a quantal model and there is high mortality in the highest dose group. The ovarian atrophy data, however, were modeled with a time-to-response model (i.e., a model that accounts for the time of death) as opposed to a quantal model which do not account for time of death. Furthermore, the model fit to the data that excluded the highest dose group was not better than the model fit to the data that included the highest dose group. Figure 1 shows the fit of analysis SA1 to the lower four dose groups and the control group while Figure 2 shows the fit of analysis SA1 to all dose groups and the control group.

In summary, the SA1 analysis in Table 2 that includes all the exposure groups and all animals in each exposure group is the most statistically sound analysis of the ovarian atrophy study because: 1) the model fit using all animals is similar to the model fit using only animals that died on or before 74.3 weeks of age, 2) the model fit using all dose groups is similar to the model fit to only the four lowest dose groups, and 3) using all the data results in more reliable maximum likelihood estimates and corresponding confidence limits.

Figure 1. Observed versus multistage-Weibull model predicted proportions of mice with ovarian atrophy when only the four lowest dose groups and the control group are used to fit the model

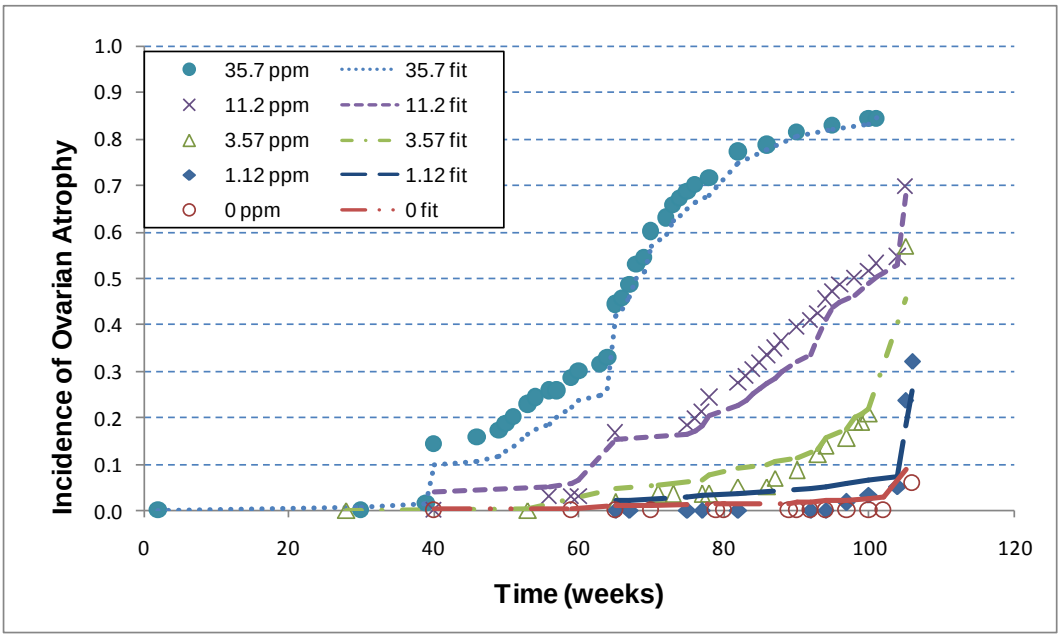
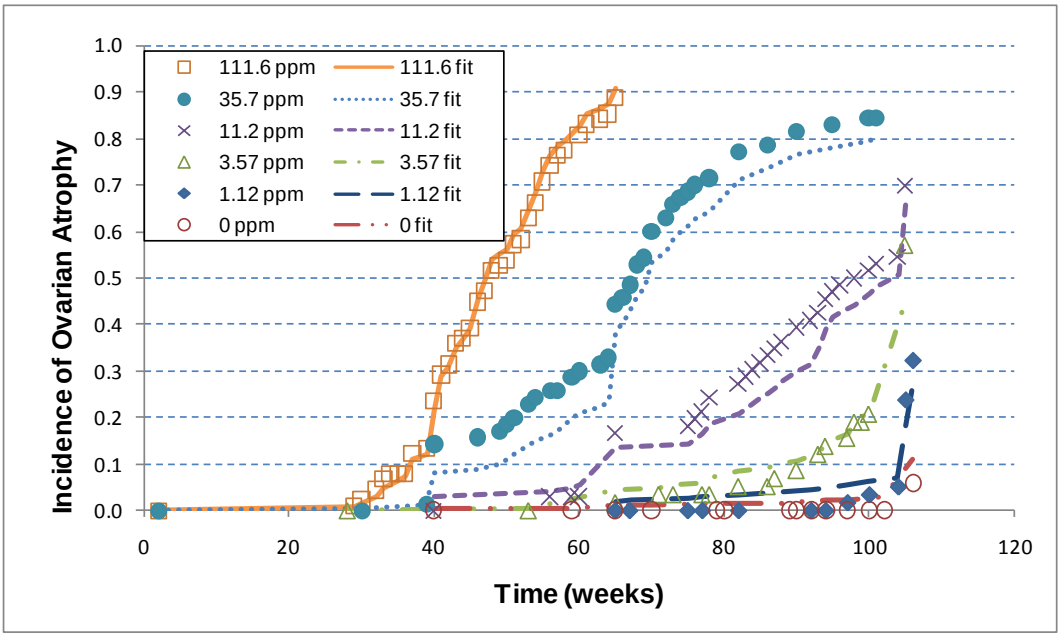


Figure 2. Observed versus multistage-Weibull model predicted proportions of mice with ovarian atrophy when all five dose groups and the control group are used to fit the model.



Attachment A

Time-to-response for ovarian atrophy as reported by the Sapphire Group, Inc. of the NTP 1993 study (NTP. 1993. Toxicology and carcinogenesis studies of 1,3-butadiene (CAS No. 106-99-0) in B6C3F1 mice (inhalation studies). Research Triangle Park, NC: National Toxicology Program, U.S. Public Health Service, U.S. Department of Health and Human Services. TR 434).

Concentration (ppm)	Responders	Non-Responders	n	Day	Week
0	0	10	10	280	40
0	0	1	1	413	59
0	0	10	10	455	65
0	0	1	1	490	70
0	0	1	1	553	79
0	0	1	1	560	80
0	0	1	1	623	89
0	0	1	1	630	90
0	0	1	1	644	92
0	0	1	1	658	94
0	0	1	1	679	97
0	0	1	1	700	100
0	0	3	3	714	102
0	4	32	36	742	106
6.25	0	10	10	455	65
6.25	0	1	1	469	67
6.25	0	2	2	525	75
6.25	0	1	1	539	77
6.25	0	1	1	574	82
6.25	0	3	3	644	92
6.25	0	1	1	658	94
6.25	1	0	1	679	97
6.25	1	1	2	700	100
6.25	1	0	1	728	104
6.25	11	10	21	735	105
6.25	5	10	15	742	106
20	0	1	1	196	28
20	0	1	1	371	53
20	1	9	10	455	65
20	1	0	1	497	71
20	0	1	1	511	73
20	0	1	1	539	77
20	0	2	2	546	78
20	1	1	2	574	82

20	0	1	1	602	86
20	1	0	1	609	87
20	1	0	1	630	90
20	2	0	2	651	93
20	1	2	3	658	94
20	1	1	2	679	97
20	2	1	3	686	98
20	0	1	1	693	99
20	1	0	1	700	100
20	21	3	24	735	105
62.5	0	10	10	280	40
62.5	2	0	2	392	56
62.5	0	1	1	413	59
62.5	0	1	1	420	60
62.5	9	1	10	455	65
62.5	1	0	1	525	75
62.5	1	0	1	532	76
62.5	1	0	1	539	77
62.5	2	0	2	546	78
62.5	2	0	2	574	82
62.5	1	0	1	581	83
62.5	1	0	1	588	84
62.5	1	0	1	595	85
62.5	1	0	1	602	86
62.5	1	0	1	609	87
62.5	1	0	1	616	88
62.5	2	0	2	630	90
62.5	1	0	1	644	92
62.5	1	2	3	651	93
62.5	2	1	3	658	94
62.5	1	1	2	665	95
62.5	1	0	1	672	96
62.5	1	0	1	686	98
62.5	1	1	2	700	100
62.5	1	0	1	707	101
62.5	1	1	2	728	104
62.5	10	1	11	735	105
200	0	1	1	14	2
200	0	1	1	210	30
200	1	0	1	2733*	390.4286
200	9	1	10	280	40
200	1	0	1	322	46

200	1	0	1	343	49
200	1	0	1	350	50
200	1	0	1	357	51
200	2	0	2	371	53
200	1	0	1	378	54
200	1	0	1	392	56
200	0	1	1	399	57
200	2	0	2	413	59
200	1	0	1	420	60
200	1	0	1	441	63
200	1	0	1	448	64
200	8	4	12	455	65
200	1	0	1	462	66
200	2	0	2	469	67
200	3	0	3	476	68
200	1	0	1	483	69
200	4	0	4	490	70
200	2	0	2	504	72
200	2	0	2	511	73
200	1	0	1	518	74
200	1	0	1	525	75
200	1	0	1	532	76
200	1	0	1	546	78
200	4	1	5	574	82
200	1	1	2	602	86
200	2	0	2	630	90
200	1	0	1	665	95
200	1	0	1	700	100
200	0	1	1	707	101
625	0	1	1	14	2
625	1	0	1	203	29
625	1	0	1	210	30
625	2	0	2	224	32
625	2	0	2	231	33
625	1	0	1	238	34
625	0	1	1	245	35
625	0	1	1	252	36
625	4	0	4	259	37
625	1	0	1	273	39
625	9	1	10	280	40
625	5	2	7	287	41
625	2	0	2	294	42

625	4	0	4	301	43
625	1	1	2	308	44
625	2	0	2	315	45
625	5	1	6	322	46
625	2	2	4	329	47
625	4	0	4	336	48
625	1	0	1	343	49
625	1	0	1	350	50
625	3	0	3	357	51
625	1	0	1	364	52
625	4	0	4	371	53
625	3	0	3	378	54
625	4	0	4	385	55
625	3	0	3	392	56
625	2	0	2	399	57
625	1	0	1	406	58
625	3	0	3	420	60
625	2	0	2	427	61
625	1	0	1	441	63
625	1	0	1	448	64
625	3	0	3	455	65

***2733 was replaced by 273 in our analyses**

Appendix 4. Leukemia Mortality/Incidence Rates and Survival Rates

US Total Population 2000-2003		Texas Statewide 1999-2003	Texas Statewide 1999-2003
Total Leukemia Mortality Rates per 100,000 [1]		Total Leukemia Mortality Rates per 100,000 [2]	Total Leukemia Incidence Rates per 100,000 [2]
	Rate	Rate	Rate
00 years	0.7	0.9	5.1
01-04 years	0.9	0.9	8.7
05-09 years	0.7	0.6	3.8
10-14 years	0.8	0.9	3.5
15-19 years	1.1	1.3	3.1
20-24 years	1.2	1.5	2.6
25-29 years	1.1	1.1	2.8
30-34 years	1.3	1.4	2.9
35-39 years	1.6	1.5	3.5
40-44 years	2.0	1.8	4.4
45-49 years	2.9	3.4	6.8
50-54 years	4.4	4.2	10.5
55-59 years	7.5	8.4	16.8
60-64 years	12.9	13.2	24.6
65-69 years	20.8	21.3	35.7
70-74 years	33.0	31.8	48.8
75-79 years	47.0	43.4	62.6
80-84 years	63.2	65.5	82.7
85+ years	81.5	81.3	91.3

[1] Table XIII-8, Seer Cancer Statistics Review 2000-2003Surveillance, Epidemiology, and End Results database (SEER 2006))

[2] Texas-specific mortality and incidence rates for 1999-2003 for all leukemia and Texas-specific survival rates for 2003 were kindly provided by the Texas Department of State Health Services, Cancer Epidemiology and Surveillance Branch, Texas Cancer Registry.

2000 US All [1]		Total Texas Population 2003 [2]	
Age	**Survival**	**Life Tables**	
0	1	0	1
1	0.99307	1	0.99342
5	0.99177	5	0.99191
10	0.99095	10	0.99105
15	0.98992	15	0.99005
20	0.98654	20	0.98659
25	0.98181	25	0.9818
30	0.97696	30	0.9772
35	0.97132	35	0.97192
40	0.96349	40	0.9641
45	0.9521	45	0.95248
50	0.93522	50	0.93546
55	0.91113	55	0.91092
60	0.87498	60	0.87584
65	0.82131	65	0.82385
70	0.74561	70	0.75079
75	0.64244	75+	0.65073
80	0.51037		
85	0.34959		

[1] US survival rates for 2000 (Arias 2002)

[2] Texas-specific survival rates for 2003 were kindly provided by the Texas Department of State Health Services, Cancer Epidemiology and Surveillance Branch, Texas Cancer Registry.

Appendix 5. Calculating Excess Risk with Age-Dependent Adjustment Factors

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March 12, 2007

TCEQ Contract 582-7-81521

In order to obtain a copy of Appendix 5, please send an email to the Toxicology Division providing the name of the DSD and the requested appendices to the following email address: tox@tceq.texas.gov.

Appendix 6. Cox Proportional Hazards Models Not Included in Cheng *et al.* (2007)

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Cheng *et al.* presented several analyses with the objective of showing different alternatives they thought could be relevant. For example, they restricted the analyses to include only cumulative ppm-years, average intensity or lagged cumulative ppm-years as the relevant doses. There is no evidence that any of these measures of dose is the relevant dose. They also fit models that adjusted for race, year of birth, race, years since hire, plant and number of high intensity tasks (HITs) and exposures to DMDTC. Cheng *et al.* did not give any biological reasons to include or exclude from the model. Ideally, the final model should adjust for effects that are biologically relevant to the outcome of study. However, there is not enough scientific knowledge to indicate what, if any, covariate effects should be included in a model of leukemia mortality with cumulative exposure to butadiene. The research closest to shedding some light on which covariates to include in the model is that published by Albertini *et al.* (2007), which seems to indicate that leukemia does not occur at low exposure to butadiene.

Although the decision of whether or not to adjust for a confounder should ideally be based on biological knowledge, Sielken *et al.* (2007) adjustment for confounders was determined using a statistically-based approach. The use of statistical methodology instead of biological arguments serves for the purpose of corroborating new biological evidence about possible confounders – specifically the role of the number of high intensity tasks in leukemia rate ratios. That is, the inclusion of the number of HITs as a covariate, although based on statistical arguments, was consistent with the biological findings of Albertini *et al.* (2007). In other words, not only was the number of HITs a plausible explanation of the increase in the number of leukemia deaths from a biological and mechanistic standpoint but also the statistical analysis of the data reached the same conclusion. Other attributes to see in model selection are issues like: consistency with biological expectations (i.e., the model should make biological sense), model parsimony (i.e., include as few variables as necessary to explain the relationship when there is no sufficient biological knowledge to justify the inclusion or exclusion of a variable), etc.

Cheng *et al.* (2007) presented a model that adjusts for age and the number of HITs (BD peaks). That is, $\beta = 2.5\times10{-}4$, $p = 0.03$ presented in Section 3.5 of the Cheng *et al.* (2007) paper. This results in a S.E. of $1.2\times10{-}4$. This model is close to the Poisson regression model in the Sielken

et al. (2007) paper with the exceptions that: 1) Sielken *et al.* adjusted for the number of HITs using a nonparametric relation based on quintiles whereas Cheng *et al.* adjusted for the number of HITs using a parametric linear relationship, 2) Cheng *et al.* models assume an log-linear relationship between rate ratios and cumulative BD ppm-years whereas Sielken *et al.* uses a linear relationship, 3) Cheng *et al.* use Cox proportional hazards model and Sielken *et al.* use Poisson regression model, and 4) Cheng *et al.* use continuous cumulative BD ppm-years and Sielken *et al.* uses BD ppm-years mean-scored deciles.

Model	**Covariates**	**Parameter Estimate**		**URF[a] (ppm^{-1}) Air Concentration for an excess risk of 1 in 100,000 (ppb)**	
		β (S.E.)	95% UCL	URF (MLE)	URF(95% UCL); 95% LCL on Conc.
Cox regression Cheng *et al.* (2007) ppm-years continuous[b], # of HITS continuous[c]	Age number of HITs > 100 ppm	2.5E-04 (1.2E-04)	4.474E-04	1.284E-04 77.88	2.298E-04 43.52
Cox regression ppm-years continuous[b], # of HITS categorical[d]	Age number of HITs > 100 ppm	2.0E-04 (1.3E-04)	4.138E-04	1.027E-04 97.35	2.125E-04 47.05
Cox regression ppm-years mean-scored deciles[e], # of HITS categorical[d]	Age number of HITs > 100 ppm	2.8E-04 (2.4E-04)	6.748E-04	1.438E-04 69.53	3.466E-04 28.85
Poisson regression (Sielken *et al.* (2007) ppm-years mean-scored deciles[e], # of HITS categorical[d]	Age number of HITs > 100 ppm	1.89E-04 (3.6E-04)	7.812E-04	8.083E-05 123.7	3.314E-04 29.93

[a] URF(MLE) = 0.001 / EC_{001} and URF(95% UCL) = 0.001 / LEC_{001}

[b]ppm-years is included as a continuous variable (untransformed) in a parametric model of the effect of ppm-years

[c] number of HITS > 100 ppm is included as a continuous variable (untransformed) in a parametric model of the effect of the number of HITS > 100 ppm

[d] number of HITS > 100 ppm is included as a categorical variable (based on quintiles) in a nonparametric model of the effect of the number of HITS > 100 ppm

[e] ppm-years is included as a categorical variable (based on mean-scored deciles, untransformed) in a parametric model of the effect of ppm-years

Despite all these differences, the models are close and converge to very similar results if some of the discrepancies are resolved. For example, if the Cox proportional hazards log-linear model presented by Cheng *et al.* were non-parametrically adjusted for BD peaks, then the estimate of the coefficient for cumulative BD ppm-years would be $\beta = 0.00020$ (S.E.=0.00013), which is close to the parameter estimates reported in Sielken *et al.* (i.e., $\beta = 0.000189$, S.E.=0.00036) for the Poisson linear model. If, in addition to adjusting for the number of HITs nonparametrically, the Cox proportional hazards log-linear model used BD ppm-years mean-scored deciles instead of continuous exposures, then the coefficient for cumulative BD ppm-years would be $\beta = 0.00028$ (S.E.=0.00024). This last model differs from Sielken *et al.* model only in that Sielken *et al.* used a Poisson regression model and a linear relationship as opposed to the Cox proportional hazards model and a log-linear relationship. The following table summarizes the results of the Cox proportional hazards model and the Sielken *et al.* Poisson regression model when adjusting for the number of HITs.

In the above discussion, a parametric model is a model that assumes a specified functional form (e.g., linear or log-linear), and a nonparametric model is a model that does not assume a specified functional form. This is analogous to the difference between regression which assumes a specified functional form (e.g., linear or polynomial) and hence is parametric and analysis of variance (ANOVA or AOV) which is nonparametric. Continuing with the analogy, if a treatment can be characterized by a number (e.g., concentration or amount), then in a regression analysis (say, a linear regression) the magnitudes of the different treatment values are important and a treatment with twice the magnitude has twice the effect. On the other hand, in an analysis of variance the different treatments are dealt with nonparametrically (say, as treatments A, B, C, etc.) and the magnitudes (numerical values) are ignored. Therefore, in an analysis of variance there is no functional relationship specified between the effects of the different treatments.

If a variable is said to be treated continuously, then each individual value of that variable is used – the values are not grouped and no representative values for the groups are used. On the other hand, if a variable is treated categorically, then the individual values of that variable are grouped and representative values for the groups replace the individual values in the analysis. Cumulative butadiene ppm-years and cumulative number of HITS > 100 ppm can both be treated either as continuous or categorical variables. Since the categorical (group) values for these variables are numerical, a categorical variable could be included in both parametric and nonparametric models.

In the table above, both the Cox and Poisson regressions assume a parametric model for the effect of cumulative butadiene ppm-years. The model for the effect of ppm-years is log-linear in Cox regression and is linear in Poisson regression. In Cox regression, ppm-years is treated as a

continuous variable in the first two models and treated as a categorical variable in the third model. In the Poisson regression, ppm-years is treated as a categorical variable.

In the first model in the table above, the cumulative number of HITS > 100 ppm is treated as a continuous variable and treated parametrically. In the other three models, the cumulative number of HITS > 100 ppm is treated as a categorical variable and treated nonparametrically.

Albertini, R., Sram, R. J., Vacek, P. M., Lynch, J., Rossner, P., Nicklas, J. A., McDonald, J. D., Boysen, G., Georgieva, N., and Swenberg, J. A. (2007). Molecular epidemiological studies in 1,3-butadiene exposed Czech workers: Female-male comparisons. Chemico-Biological Interactions, Volume 166, Issues 1-3, 20 March 2007, Pages 63-77.

Appendix 7. Sensitivity Analysis: Exposure Estimation Errors

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Appendix 8. Calculating Excess Risk When Specified Response Is Mortality Versus Incidence

Issues in Quantitative Epidemiology

Calculating Excess Risk When Specified Response is Mortality

Vs When the Specified Response is Incidence

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Appendix 9. 24-Hour Reference Value (TCEQ 2015)

For chemicals detected in the ambient air monitoring network, short-term AMCVs have generally been derived by the TCEQ to evaluate 1-h reported concentrations and long-term AMCVs were derived to evaluate annual averages. Since a significant amount of ambient air data is collected over a 24-h duration, the derivation of chemical-specific 24-h AMCV values is needed to better evaluate ambient 24-h data. TCEQ believes using a short-term, 1-h AMCV or long-term AMCV to evaluate a 24-h ambient air sample is not appropriate because toxic effects induced by 24-h exposure may be governed by modes of action somewhat different than those influencing toxicity due to 1-h or chronic exposure. A 24-h Reference Value (ReV) is derived for human health hazards associated with threshold dose-response relationships (typically effects other than cancer) and is defined as an estimate of an inhalation exposure concentration that is likely to be without an appreciable risk of adverse effects to the human population (including susceptible subgroups) for a 24-h exposure. The ReV is used as the AMCV (TCEQ 2015).

The critical step in deciding whether or not to derive a 24-h AMCV is the availability of appropriate toxicity studies that provide meaningful information to evaluate a 24-h exposure duration. An evaluation of the mode of action, dose metric, and the toxicokinetics and toxicodynamics of the chemical of concern as well as exposure duration adjustments that are unique for the derivation of a 24-h AMCV is conducted. The same analytical steps used to derive acute 1-h AMCVs and chronic AMCVs (TCEQ 2015) are used to derive a 24-h AMCV. OECD (2010) also provides guidance applicable to the development of acute reference concentrations.

The purpose of this document is to summarize the main steps involved in the development of the 24-h AMCV for BD. General steps discussed below for developing a 24-h value include:

- availability of appropriate toxicity studies that provide meaningful information to evaluate a 24-h exposure duration;
- identification of a point of departure (POD) for the critical effect(s) based on review of dose-response data for relevant toxicity endpoints;
- consideration of an exposure duration adjustment;
- animal-to-human inhalation dosimetric adjustment;
- selection and application of applicable uncertainty factors; and
- derivation of the 24-h AMCV.

Please refer to the 1,3-Butadiene Development Support Document (TCEQ 2008) for detailed information on human and animal studies, mode of action information, etc.

Acute 24-H AMCV

Key Studies

BD has very low acute toxicity (TCEQ 2008). The toxicity of BD is shown in Figure 1 as an exposure response array for acute (less than 24 h) and subacute studies which were considered for the development of a 24-h AMCV. Effects in humans (slight smarting of the eyes and difficulty in focusing) occurred at 2000 ppm after a 7-h BD exposure (Carpenter et al. 1944). Animal studies show BD is a potential reproductive/developmental hazard to humans. The following studies were considered for the development of a 24-h AMCV:

- Developmental toxicity (decrease in maternal body weight gain and fetal body weight) occurs in mice, the most sensitive species, after BD exposure (6 h/day, gestational day 6-15) with a lowest observed adverse effect level (LOAEL) of 200 ppm and a no-observed adverse effect level (NOAEL) of 40 ppm (Hackett *et al.* (1987b).

- In three different developmental studies in rats, the lowest critical effect is a decrease in body weight parameters. Toxicity occurs in rats at much higher concentrations than in mice. The LOAELs in rat studies ranged from 1000-1500 ppm and the NOAELs ranged from 200-300 ppm (IISRP 1982; Hackett *et al.* 1987a; ACC 2003).

- After a 6-h/day, 5-day exposure in male mice, decreased testes weight was observed with a LOAEL = 500 ppm and a NOAEL = 130 ppm (Pacchierotti et al. 1998). There was a concentration/duration effect on male reproductive effects. After 4 weeks of exposure in male mice, followed by mating, there was an increase in early fetal deaths with a LOAEL of 65 ppm (Anderson et al. (1998). After 10 weeks exposure, a LOAEL of 12.5 ppm was observed for fetal deaths and sperm abnormalities (Anderson et al. 1996).

Critical Effect

The TCEQ developed a 1-h AMCV in 2008 (TCEQ 2008) based on developmental toxicity in mice, the most sensitive species, after BD exposure (Hackett *et al.* 1987b). Reproductive/ developmental effects may have been caused by only a single day's exposure that occurred at a critical time during gestation. Therefore, this developmental study is relevant for derivation of a 24-h AMCV. This study has the lowest LOAEL and NOAEL (Figure 8) relevant for an acute exposure and was also selected for development of the 24-h AMCV, based on the following toxicokinetic and mode of action analysis.

Toxicokinetics and Mode of Action

BD is a highly volatile, colorless gas with a mildly aromatic odor, and is only slightly soluble in water. Absorption through the lung is limited by blood flow to the lung. After absorption, BD is distributed throughout the body. For both rats and mice after exposure to ^{14}C-butadiene, Bond et al. (1987) reported the following:

- Within 1 h after the end of exposure, respiratory tissue, gastrointestinal tract, liver, kidneys, urinary bladder and pancreas contained higher concentrations of radioactivity than other tissues

- Tissues of mice attained significantly greater concentrations than did rats per μmole of BD inhaled, although there were no apparent differences between rats and mice in tissue depots of BD

- Elimination of BD from tissues and blood was rapid, with 77% to 99% of the initial tissue burden being eliminated with half-times of 2 to 10 h.

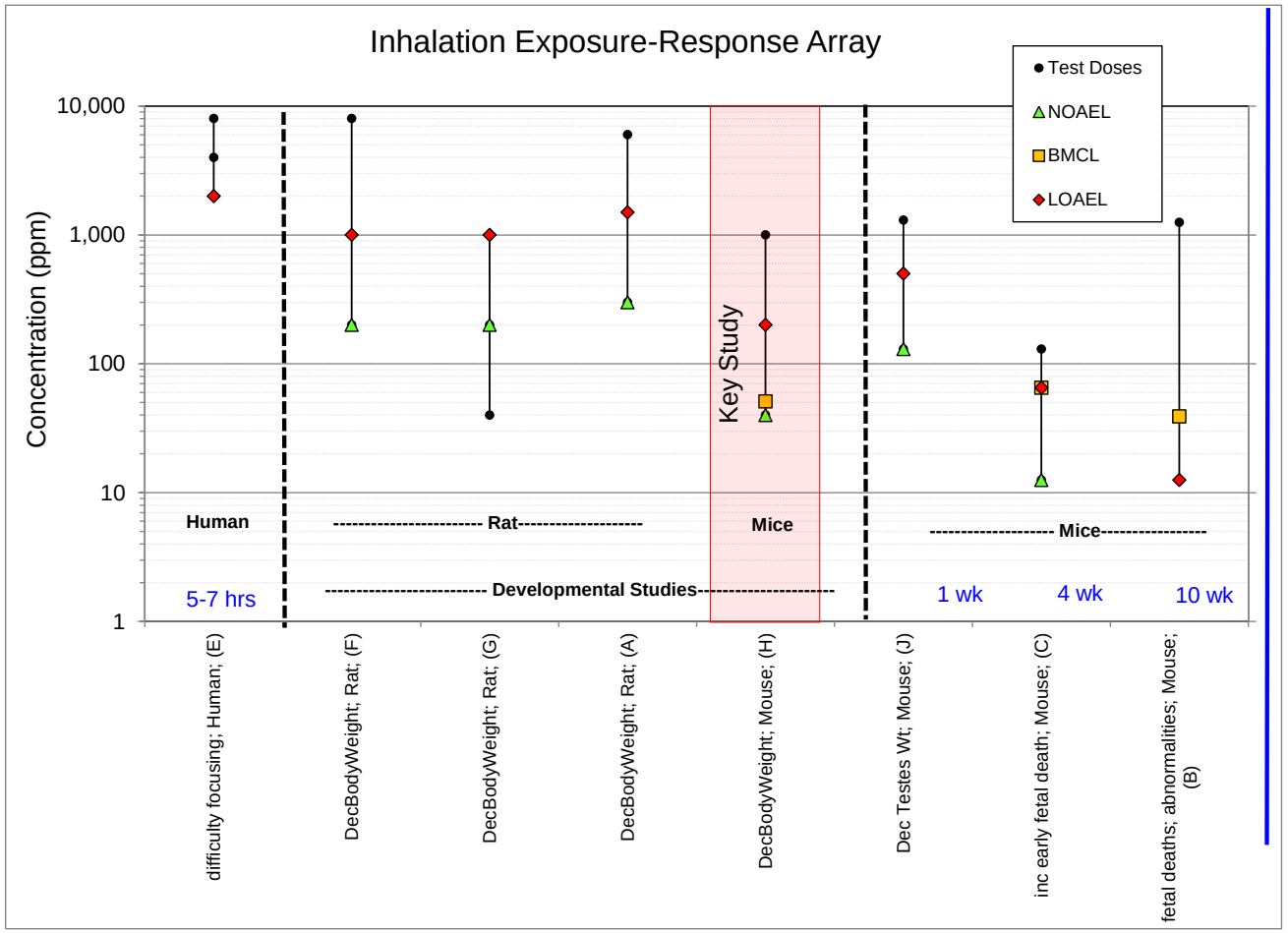

Figure 8 BD Exposure response array for acute (less than 24 h) and subacute studies

Citation	Key	Citation	Key
ACC (2003)	A	IISRP (1982)	F
Anderson et al. (1996)	B	Hackett et al. (1987a)	G
Anderson et al. (1998)	C	Hackett et al. (1987b)	H
Carpenter et al. (1944)	E	Pacchierotti et al. (1998)	J

Research has shown that BD produces toxicity when metabolized to reactive metabolites. After exposure to BD, the most reactive metabolites are 1,2-epoxy-3-butene (EB) and 1,2:3,4-diepoxybutane (DEB). There is a difference in the metabolism between mice and rats. Therefore, the basis of the species differences may be related to the greater production of toxic intermediates and a lower capacity for detoxification of these intermediates in mice compared to rats (USEPA 2002). Moreover, humans are more similar to rats in the metabolism of BD. Humans produce much lower levels of DEB than mice as demonstrated by experimental data on DEB-specific pyr-Val Hb adducts (Swenberg *et al.* (2007); Georgieva *et al.* (2007; 2008) and urinary metabolites (Sabourin *et al.* 1992).

The specific mechanism of action for the maternal reproductive/developmental effects produced by BD is unknown after acute exposure, although the mode of action (MOA) may involve DEB-induced ovarian atrophy and a decrease in serum progesterone levels, as shown by Spencer *et al.* (2001) and Chi *et al.* (2002). Although the amount of DEB produced by humans is much lower than mice, reproductive/developmental effects were assumed to be relevant to humans (Kirman and Grant 2012). However, using a study in mice to predict toxicity in humans is conservative. Refer to TCEQ (2008) for a detailed discussion of the metabolism and mode of action of BD.

Based on toxicokinetic and MOA information, the reproductive/developmental effects in mice are considered to have a threshold and to be concentration and duration dependent.

Dose Metric

For the reproductive/developmental key study (Hackett *et al.* 1987b), the most appropriate dose metric for a 24-h exposure is likely area under blood concentration curve of DEB or DEB concentration in target tissue; however, this data was not available. Therefore, data on the exposure concentration of the parent chemical was used as the default dose metric.

Dose-Response Modeling and Points of Departure (PODs)

The TCEQ (2008) performed benchmark concentration (BMC) modeling for numerous endpoints from Hackett et al. (1987b). The endpoint with the lowest relevant BMCL was decrease in maternal extragestational weight gain ($BMCL_{1\ SD}$ = 51.3 ppm with a $BMC_{1\ SD}$ of 723 ppm), followed by decrease in fetal body weight ($BMCL_{05}$ = 54.7 ppm BMC_{05} of 65.8 ppm). The POD for development of the 24-h AMCV is the $BMCL_{1\ SD}$ of 51.3 ppm.

Duration and Default Animal-to-Human Dosimetry Adjustments

Duration adjustments from a 6-h exposure to a 24-h exposure were conducted using Haber's Rule as modified by ten Berge (1986) with "n" = 1. The adjusted POD applicable for a 24-h exposure (POD_{ADJ}) is 12.8 ppm.

Default animal-to-human dosimetry adjustments were based on methods for Category 3 gases producing systemic effects (USEPA 1994; TCEQ 2015). For BD, the animal to human blood gas ratio [$(H_{b/g})_A$ / $(H_{b/g})_H$] is greater than 1 (TCEQ 2008). When $(H_{b/g})_A$ / $(H_{b/g})_H$, > 1, a default value of 1 is used for the regional gas dose ratio (RGDR) (USEPA 1994). The human equivalent POD (POD_{HEC}) is 12.8 ppm.

Uncertainty Factors and Derivation of the 24-H ReV

The default procedure for deriving health-protective concentrations for noncarcinogenic effects is to determine a POD and apply appropriate uncertainty factors (UFs) (i.e., assume a threshold/nonlinear MOA) (TCEQ 2015). The POD_{HEC} of 12.8 ppm was used and divided by the following UFs:

- Intraspecies human UF (UF_H) of 10 for intraspecies variability;
- Interspecies animal UF (UF_A) of 3 for extrapolation from animals to humans; and
- Database UF (UF_D) of 1 for database uncertainty.

A full UF_H of 10 was used to account for intraspecies variability. There is experimental evidence that indicates BD-sensitive human subpopulations may exist due to metabolic genetic polymorphisms (USEPA 2002), although recent studies indicate that variability due to genetic polymorphisms is less than 10 based on metabolism of BD in humans with different genotypes. (Albertini *et al.* 2001, 2003).

A UF_A of 3 was used for extrapolation from animals to humans because default dosimetric adjustments from animal-to-human exposure were conducted, which account for toxicokinetic differences but not toxicodynamic differences. This approach is likely conservative, since existing studies indicate that mice are relatively sensitive laboratory animals in regards to the reproductive effects of BD.

A database UF_D of 1 was used because the overall acute toxicological database for BD includes acute inhalation studies in humans; two inhalation bioassays in different species investigating a wide range of endpoints; and several prenatal developmental toxicity studies in different species (USEPA 2002; TCEQ 2008). Both the quality of the studies and the confidence in the acute database is high.

Thus, the 24-h ReV = 24-h AMCV =

$POD_{HEC} / (UF_H \times UF_A \times UF_D) = 12.8 \text{ ppm} / (10 \times 3 \times 1)$

$= 0.426.7 \text{ ppm}$

$= 430 \text{ ppb}$ (rounded to two significant figures)

A summary of the derivation of the 24-h AMCV is found in the following table.

Derivation of the Acute 24-H AMCV

Parameter	**Summary**
Study	Hackett *et al.* 1987b
Study population	CD-1 mice (18-21 pregnant mice per dose group)
Study quality	High
Exposure Methods	0, 40, 200, and 1,000 ppm BD on gestation days (GD) 6-15 for 6 h/day
Critical Effects	Reduction in extragestational weight gain and fetal body weight; developmental toxicity
POD	51.3 ppm ($BMCL_{1\ SD}$)
Duration	6 h
Extrapolation to 24-h	51.3 ppm ($BMCL_{1\ SD}$) x 6/24 = 12.8 ppm
24-h POD_{HEC}	12.8 ppm (gas with systemic effects, based on default RGDR = 1.0)
Total UFs	30
Interspecies UF	3
Intraspecies UF	10
LOAEL UF	Not applicable
Database UF	1
Database Quality	High
Acute 24- h ReV (HQ = 1)	**950 µg/m^3 (430 ppb)**
Acute 24-h AMCV	

Values for Air Monitoring Evaluation

The acute evaluation resulted in the derivation of the following values:

- 6-h acute health-based AMCV = 3,700 µg/m^3 (1,700 ppb) (TCEQ 2008)
- 24-h acute health-based AMCV = 950 µg/m^3 (430 ppb) (TCEQ 2015)
- 1-h acute odor-based AMCV = 510 µg/m^3 (230 ppb)) (TCEQ 2008)

For the evaluation of 24-h ambient air monitoring data, the 1-h acute odor-based AMCV of 510 µg/m^3 (230 ppb) is lower than the 24-h AMCV of 950 µg/m^3 (430 ppb), although both values may be used for the evaluation of 24-h ambient air monitoring data (Table 1). If the 24-h ambient air monitoring data is below the odor-based AMCV, it does not mean that odor-potential did not occur, since peak concentrations may exceed the odor-based AMCV.

The health-based 24-h AMCV of 430 ppb (950 µg/m^3) falls between the health-based TCEQ acute 1-h AMCV of 1,700 ppb (3,700 µg/m^3) and the chronic noncarcinogenic AMCV of 15 ppb (33 µg/m^3) and the carcinogenic value of 9.1 ppb (20 µg/m^3) (TCEQ 2008). It is sufficiently conservative for the adequate protection of public health for the exposure duration and adverse effects considered and would significantly complement TCEQ health effect evaluations of ambient air data, which currently utilize 1-h and chronic (i.e., lifetime) health-protective and welfare-based (i.e., odor, vegetation) AMCVs.

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