Beyond Science and Decisions: From Problem Formulation to Dose-Response Assessment

Case Study: Cancer Risk Assessment for 1,3-Butadiene Based on New Data and Methods

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1. Provide a few sentences summarizing the method illustrated by the case study

An updated cancer dose-response assessment was conducted for 1,3-butadiene (BD) using the best available data and methods. This assessment incorporates the most recent animal cancer bioassay (**Tables A.2** and **A.3**) and epidemiology data available (**Table B.1**) for characterizing the relationship between exposure and cancer risk.

Rodent-Based Potency Estimates

- *New Data:* Cancer bioassay data for BD assessed in rats and mice remain the same as was relied upon in USEPA (2002) (**Tables A.2** and **A.3**). New biomarker data (hemoglobin adducts) are available to estimate internal doses of reactive metabolites to support interspecies extrapolation (**Table A.4**).
- *New Methods:* The methods of Fred et al. (2008)/Motwani and Tornqvist (2014) were extended and applied to BD using hemoglobin adduct data measured in mice, rats, and humans to quantify species differences in the internal doses for three reactive epoxide metabolites of BD (epoxybutane or EB, diepoxybutane or DEB, and epoxybutane diol or EBD; **Figure A.1**). The magnitude of the species differences in the metabolic activation of BD (mice>rats>humans) spans several orders of magnitude. Ratios of internal dose estimates as well as metabolite-specific relative genotoxic potency estimates were used to support the calculation of data-derived extrapolation factors (DDEFs) for interspecies extrapolation.

Epidemiology-Based Potency Estimates

- *New Data:* Exposure and mortality data for leukemia and bladder cancer from the Styrene-Butadiene Rubber (SBR) worker cohort were used. Data for this cohort have been updated multiple times since 2002, and now includes 18 additional years of follow-up, >5900 additional deaths, validation of exposure estimates, and observations for male and female workers (previously just males) (**Table B.1**).
- *New Methods:* Separate cancer potency estimates were derived for BD based on mortality data for: (1) leukemia, which has been used previously to estimate BD's cancer potency, and (2) bladder cancer, which is a new endpoint based upon a significant statistical association, but one that is uncertain due to potential confounders (e.g., smoking). In addition, to support a characterization of total risk a method for aggregating cancer endpoints (leukemia or bladder cancer) was evaluated within the context of the Cox proportional hazards modeling.

Resulting probability distributions for rodent-based and for epidemiology-based cancer potency estimates are compared in **Figure A.3B**.

2. Describe the problem formulation(s) the case study is designed to address. How is the method described in the case useful for addressing the problem formulation?

The existing cancer assessment for 1,3-butadiene on USEPA's IRIS database is nearly 20 years old. As such, the existing assessment does not reflect the current state of knowledge for this chemical, and no longer serves as an appropriate basis for quantifying risk in exposed human populations, as is needed for risk assessments conducted under TSCA. By incorporating the latest data and methods, the assessment presented in this case study is expected to provide a robust characterization of the potential risks associated with BD exposure.

- *Rodent-based potency estimates* – Large, quantitative differences in species metabolism of BD to three reactive epoxide metabolites are well-documented in *in vitro*, *in situ*, and *in vivo* studies for BD. To date, none of the PBPK models developed for BD are sufficient to quantify all of these differences to support interspecies extrapolation. Accounting for these differences represents a significant challenge for risk assessors, risk managers, and decision makers. For example, despite BD being a data-rich chemical, ATSDR (2012) opted to not derive minimal risk levels (MRLs) for BD "*due to the large species differences in the metabolism of 1,3-butadiene and the lack of chemical-specific data to adjust for these differences, which may result in the MRL overestimating the risk to humans.*"
- *Epidemiology-based potency estimates* – In their 2002 assessment for BD, USEPA applied an additional adjustment factor of 2 to the cancer potency estimate in part to account for potential multisite carcinogenicity, as observed in exposed rats and mice. For other chemicals, USEPA has more recently adopted an approach of summing separately modeled potency estimates for specific cancers (e.g., leukemia and breast cancer in workers exposed to ethylene oxide; USEPA, 2016) to address concerns for total risk. The former approach serves as an educated guess, and the latter approach invokes assumptions of normality that may not be valid, and ignores potential relationships (e.g., correlations) between endpoints. Neither of these approaches is ideal. The approach described here makes the best use of the dose-response data available for BD.

3. Comment on whether the method is general enough to be used directly, or if it can be extrapolated, for application to other chemicals and/or problem formulations. Please explain why or why not.

BD is a data-rich chemical, and as such the approaches described here can readily be applied to other chemicals with sufficient data as described below.

- *Rodent-Based Potency Estimates* - For chemicals that are reactive or are metabolized to reactive metabolites that form stable hemoglobin adducts, measured hemoglobin adduct data can be used to estimate internal doses in blood without the need for a PBPK model. Internal dose estimates in the test species and in humans can be used to

quantify species differences to inform extrapolations within the framework for data-derived extrapolation factors (DDEFs; USEPA, 2014).

- *Epidemiology-Based Potency Estimates* – For chemicals with epidemiology data that can support dose-response assessment for more than one cancer type, this approach can be used to characterize total risk without the need for using adjustment factors (as done for BD in USEPA 2002) or post-hoc methods (e.g., summation of Wald potency estimates) for combined separate potency estimates (as has been done for other chemicals such as ethylene oxide; USEPA, 2016).

4. Discuss the overall strengths and limitations of the methodology.

<u>Rodent-Based Potency Estimates</u>

- *Strengths:* Provides a robust characterization of the quantitative species differences in the metabolic activation of BD without the need for a PBPK model. Because stable hemoglobin adducts reflect circulating levels of reactive chemicals over the lifetime of erythrocytes, these biomarkers are good for estimating long-term exposures (i.e., over several months). Measured biomarker data are agnostic to metabolite formation, and therefore avoids the uncertainties in PBPK model in assigning metabolic activity to hepatic vs. extrahepatic tissues. The relative potency approach used to assess potential risk from exposure to BD metabolites is well-established risk assessment practice for chemical mixtures (e.g., PAHs, dioxin-like chemicals).
- *Limitations:* The approach is data intensive may require refined analytical methods. For example, the detection of low-level concentrations of hemoglobin adducts from DEB (pyr-Val) in exposed workers required substantial refinement to achieve the detection limits required (Swenberg et al., 2011; Boysen et al., 2012). The combined approach of using chemical-specific biomarkers and quantifying the relative potency increases the complexity of the assessment. There is uncertainty regarding which type of genotoxicity endpoint (e.g., DNA damage, mutations, clastogenicity) best reflects metabolite contribution to carcinogenic potency. There is uncertainty in the MOA associated with the contribution of other metabolites proposed for BD.

<u>Epidemiology-Based Potency Estimates</u>

- *Strengths:* The approach relies upon epidemiology data that was considered to be high quality by USEPA in 2002, and has greatly improved over time with multiple updates and validation of exposure estimates. As such, the epidemiology-based cancer potency estimates for BD based on leukemia continues to reflect the best available science for application to quantitative risk assessment. In addition, the use of an aggregate endpoint, which includes an endpoint (bladder cancer) that is uncertain due to potential confounders (smoking), within the Cox proportional hazards model provides a robust characterization of total risk that considered potential relationships/correlations between cancer types.
- *Limitations:* The appropriateness of combining different cancer endpoints requires both toxicological considerations (e.g., do the endpoints share a common mode of action?), statistical considerations (e.g., do they share common covariates and lag assumptions?),

and weight of evidence considerations (e.g., differing weight of evidence for each enpoint), and as such is likely to be controversial. Uncertainty remains as to whether or not a total risk characterization is needed, and if so, how and when should it be addressed.

5. Outline the minimum data requirements and describe the types of data needed.

Rodent-Based Potency Estimates

- Species differences in internal dose requires hemoglobin adduct data for measured exposures to chemical/metabolites in the test species and in humans. Relative potency estimates require genotoxicity data for each metabolite believed to contribute to a carcinogenic response.

Epidemiology-Based Potency Estimates

- Sufficient human data to support quantitative characterization of exposure-response relationship for more than one endpoint

6. Questions for the panel

Rodent-Based Potency Estimates

- For assessing relative potency estimates of BD metabolites, the average potency across all genotoxic endpoints (**Table A.5**). Should more weight be given to one or more genotoxic endpoints (e.g., DNA damage, mutations, micronuclei) as a predictor for carcinogenic potency?
- Is the use of hemoglobin adduct data to quantify species differences in the internal doses of epoxide metabolites appropriate?

Epidemiology-Based Potency Estimates

- Is a characterization of total or combined risk needed for BD, or should potency estimates be based on the leukemia endpoint alone (as has been done previously for BD)?
- Is the combining of cancer endpoints supportable for BD from a toxicological perspective? Is a common mode of action required for combination (i.e., if both endpoints are related to genotoxicity of BD metabolites)?
- From a statistical perspective, do potential confounders for bladder cancer (e.g., smoking) preclude its use in the quantitative assessment? How do differences in the underlying weight of evidence (e.g., statistical association for bladder cancer vs. causal association for leukemia) impact the decision to combine?
- What additional information should be included?
- Given the data on metabolic activation as a function of age and the acute cancer bioassay data in rodents, is an ADAF needed for BD?

Does your case study:

A. Describe the dose-response relationship in the dose range relevant to human exposure?
    a. *Rodent-Based Potency* – No, because rodent cancer bioassays utilize exposures to high concentrations of BD (i.e., well above expected human exposures), the rodent-based potency estimates require large extrapolations (i.e., linearity assumed) to human exposure levels.
    b. *Epidemiology-Based Potency* – Yes, because the points of departure for a $1x10^{-6}$ risk level fall within the range of observation defined by the SBR cohort.
B. Address human variability and sensitive populations?
    a. *Rodent-Based Potency* - No
    b. *Epidemiology-Based Potency* – Yes, in part. The SBR cohort includes men and women, and includes some minorities (black). The lack of data in female workers was once of the reasons USEPA included an additional factor of 2 to its cancer potency in 2002, and therefore is no longer applicable. Other minorities and people younger than 18 years of age are not represented in the cohort. Biomarker data for BD (Nieto et al., 2021; Fustinoni et al. 2002) suggest that human variation in toxicokinetic factors are generally consistent with historical default assumptions for intraspecies variation (i.e., ~3-fold). However, this variation is typically not considered in cancer risk assessment (reserved for noncancer assessment and definition of uncertainty factors).
C. Address background exposures or responses?
    a. *Rodent-Based Potency* – No.
    b. *Epidemiology-Based Potency* – No.
D. Address incorporation of existing biological understanding of the likely mode of action?
    a. *Rodent-Based Potency* – A genotoxic mode of action is assumed, and the relative potency of 3 BD metabolites was characterized using genotoxicity studies that studied all 3 metabolites in the same test system.
    b. *Epidemiology-Based Potency* – A genotoxic mode of action was assumed.
E. Address other extrapolations, if relevant – insufficient data, including duration extrapolations, interspecies extrapolation?
    a. *Rodent-Based Potency* – Biomarker data were used to support interspecies extrapolation based on toxicokinetic differences (primarily for metabolic activation). Potential toxicodynamic differences across species were ignored (i.e., it is assumed the rodents serve as relevant models for assessing human risk).
    b. *Epidemiology-Based Potency* – The need for including additional adjustments for potential early-life susceptibility (e.g., ADAF application) is discussed, and may not be required for BD based upon age differences in metabolic activation (i.e., infants & children < adults; see **Section B.5**), and the negative results obtained from an acute cancer bioassay for BD in male and female mice (**Table A.2**).
F. Address uncertainty?
    a. *Rodent-Based Potency* – This approach reduces the uncertainty associated with interspecies extrapolation when metabolism differences are known.

b. *Epidemiology-Based Potency* – This approach eliminates the need for an additional adjustment factor of 2 (used in USEPA's 2002 assessment) for characterizing total risk.

G. Allow the calculation of risk (probability of response for the endpoint of interest) in the exposed human population?
   a. *Rodent-Based Potency* – Yes, unit risk values were derived.
   b. *Epidemiology-Based Potency* - Yes, unit risk values were derived.

H. Work practically? If the method still requires development, how close is it to practical implementation?
   a. *Rodent-Based Potency* – With sufficient chemical-/metabolite-specific data, this approach can be implemented now.
   b. *Epidemiology-Based Potency* - With sufficient chemical-specific data, this approach can be implemented now.

Appendix A: Use of Biomarker Data and Relative Potencies of Mutagenic Metabolites to Support Derivation of Cancer Unit Risk Values for 1,3-Butadiene from Rodent Tumor Data

A.1. Background

This section provides background information on previous dose-response assessments for tumors in rodents, metabolism, and mode of action (MOA) studies for BD. These data are used here to support key decisions in the cancer dose-response assessments conducted for BD. In so doing, these data reduce the uncertainty associated with using rodent-derived data for BD human health risk assessment consistent with USEPA guidelines (USEPA, 2005, 2014).

A.1.1 Previous Rodent-Based Cancer Risk Assessments for BD

Previous rodent-based cancer risk assessments conducted by regulatory agencies and risk assessors are summarized in **Table A.1**. These potency estimates take into consideration tumor incidence at multiple tissue sites in mice (**Table A.2**) and rats (**Table A.3**) exposed to BD via inhalation. The rodent carcinogenicity database for BD is robust, and includes acute cancer bioassays in mice of both sexes (Bucher et al., 1993), a series of stop-exposure studies conducted in male mice (NTP, 1993), and lifetime cancer bioassays in mice and rats of both sexes (NTP, 1993; Owen et al., 1987). Upper bound cancer potency estimates based on mouse tumor data are significantly higher than corresponding values based on rat tumor data, which is generally attributed to underlying differences between species in metabolic activation and detoxification (see Section 2.2). More recent data and methods published since the time of these risk assessment now allow for these important differences to be addressed quantitatively in human health cancer risks assessments for BD (see **Section A.2**).

A.1.2 Metabolism Overview

BD is chemically inert, but is metabolized to several electrophilic epoxides that are capable of alkylating cellular macromolecules, to which the genotoxic and carcinogenic effects of BD are attributed (see **Section A.1.3** below). The metabolism of BD to reactive epoxide metabolites, including 2,3-epoxy-1-butene (EB), 1,2,3,4-diepoxybutane (DEB), and 3,4epoxybutane-1,2-diol (EBD), has been well studied in mice, rats, and humans (as reviewed in Himmelstein et al., 1997; Albertini et al., 2003; Kirman et al., 2010; Filser et al., 2010), which indicates that the metabolic pathways for BD are qualitatively similar, but exhibit large quantitative differences across species. Internal doses of these metabolites reflect pathways accounting for their formation (e.g., oxidation) as well as their clearance (e.g., hydrolysis, conjugation) as depicted in **Figure A.1**.

Large species differences in the metabolism of BD are consistently reported in *in vitro, in situ*, and *in vivo* studies. *In vitro* studies on Michaelis-Menten constants (Vmax and Km values) for activation and detoxification pathways of BD in microsomes indicate that mice have a significantly higher ratio of EB activation-to-detoxification than either rats or humans (Csanady et al., 1992; Schmidt and Loeser, 1985; Krause and Elfarra, 1997; Bond et al., 1993; Kreuzer et

al., 1991; Seaton et al., 1995; Motwani and Tornqvist, 2014). In the effluent of mouse livers perfused with BD, all three epoxides (EB, DEB and EBD) and BD-diol were observed, while in effluents from rat livers perfused with BD, only EB and BD-diol were detected. When the mouse and rat livers were perfused with EB, Filser et al., (2001, 2010) found that BD-diol, EBD, and DEB were formed, with BD-diol predominating in both species. DEB formation was greater in mouse than in rat livers (Filser et al., 2010). Following *in vivo* exposures of rats and mice to BD via inhalation, differences in circulating DEB levels have been reported to be over 100-fold greater in mice than in rats (Filser et al., 2007; Thornton-Manning et al., 1995a,b).

Quantitative differences in the *in vivo* production of BD metabolites are also reflected in the accumulations of metabolite-specific hemoglobin adducts. A DEB-specific hemoglobin adduct, N,N-(2,3-dihydroxy-1,4-butadiyl)-valine (pyr-Val), has been identified and measured, providing insights into species and exposure differences in BD metabolism (Boysen et al., 2004). The formation of pyr-Val hemoglobin adducts has been studied in male and female mice and rats exposed to 1.0 ppm by inhalation for 6 hours/day for four weeks (Swenberg et al., 2007), in which adduct burdens (i.e., concentrations in blood due to cumulative exposure) in rats were more than 30-fold lower than the corresponding values in mice. The formation of pyr-Val adducts in rats and mice of both sexes was assessed following 4-week inhalation exposures to either 1, 6.25, or 62.5 ppm BD for 6 hours/day (Georgieva et al., 2010). The difference in adduct levels between species was large (mice>rat by approximately an order of magnitude) and dose-dependent, with larger differences observed at higher concentration compared to low concentrations. Swenberg et al. (2007) compared results in occupationally-exposed workers in the Czech Republic to results in BD-exposed mice and rats for pyr-Val. Pyr-Val adducts were not detected (LOD of 0.3 pmol/g Hb) in occupationally exposed men and women with the mean exposures ranging from 0.18-0.8 ppm (Albertini et al., 2003, 2007). Using analytical methods with improved sensitivity, Swenberg et al. (2011)/Boysen et al. (2012) detected pyr-Val in humans. For a given exposure to BD, DEB blood levels in humans (estimated from measured pyr-Val adducts) were approximately 16-fold lower than the DEB blood levels in rats, which in turn are approximately 45-fold lower than the DEB blood levels in mice.

Motwani and Tornqvist (2014) estimated internal dose (i.e., blood AUCs per unit exposure) for BD metabolites in mice, rats, and humans using two approaches: (1) estimating blood dose from hemoglobin adduct data using second-order rate constants for adduct formation and erythrocyte half-lives; and (2) scaling up metabolite clearance rates from *in vitro* studies. For DEB, both approaches yielded consistent results in which large differences are estimated across species (mice>rats>humans). Of primary importance to human health risk assessment, relative species differences in DEB AUC between mice and humans are very large (approximately 2 to 3 orders of magnitude) (Motwani and Tornqvist, 2014). Based on hemoglobin adduct biomarkers (Motwani and Tornqvist, 2014) and urinary biomarker data (Kotapati et al., 2015), there is clear evidence that mice, rats, and humans are exposed internally to mixtures of BD metabolites that are qualitatively similar, but have important quantitative differences.

A.1.3 Mode of Action Summary

There is clear evidence from *in vivo* and *in vitro* studies that BD can produce genotoxicity through the formation of electrophilic metabolites (as reviewed in USEPA, 2002; Albertini et al., 2010). USEPA (2002) concluded *"...it is virtually certain that the carcinogenic effects are mediated by genotoxic metabolites of 1,3-butadiene*." Key events for a genotoxic mode of action, which can include point mutations and clastogenic events, were summarized in Kirman et al. (2010): (1) Exposure to BD; (2) Distribution of BD to metabolizing tissues (liver); (3) Metabolism of BD to electrophilic intermediates (epoxide metabolites); (4) Distribution of electrophilic intermediates to target tissues; (5) Formation of DNA adducts; (6) Error in DNA replication; (7) Viable cell with gene mutation; and (8) Tumor Progression. Because metabolic activation (Key event 3) is considered the molecular initiating event in the MOA, quantification of the large species differences in metabolism serves as an important challenge to quantitative risk assessment.

BD, through its metabolism, is both mutagenic and clastogenic. The types of genotoxic events (point mutations vs. chromosomal aberrations) may play differing roles in the various cancers associated with BD exposure in rodents and humans. Point mutations are generally assumed to play an initiating role in the carcinogenic process, and often serve as the basis for assumptions of low-dose linearity as a matter of risk assessment policy. However, specific chromosomal aberrations are known to play a key role in some human leukemias (e.g., Philadelphia chromosome and chronic myelogenous leukemia), but interestingly were not observed in human cells exposed to DEB *in vitro* despite increases in DNA double strand breaks (Walker et al., 2019). For cancer types requiring clastogenic events (e.g., reciprocal translocations/deletions), a nonlinear dose-response relationship may better reflect the underlying mode of action for specific structural chromosome alterations requirement of two-hits during a single round of DNA replication for their production (Preston, 1999). The aberrations will arise at a frequency proportional to the square of exposure concentration, and therefore cancers that are dependent upon reciprocal translocations or interstitial deletions are expected to exhibit a quadratic component to their dose-response relationship.

A.2 Methods

A.2.1 Unit Risk Derivation

Cancer URs for BD were calculated from rodent bioassay data in a manner consistent with USEPA methodology (USEPA, 2005, 2012) using the following equation:

$$UR = BMR / POD_{HEC} \qquad Eq.1$$

Where,

- *UR* = unit risk (per ppm, continuous exposure);
- *BMR* = Benchmark response rate (e.g., 10%); and
- POD_{HEC} = Point of departure (e.g., benchmark dose) expressed in terms human equivalent concentration (ppm, continuous exposure), after adjusting for discontinuous exposures and species differences in the toxicokinetics of BD.

UR derivation is a multistep process that includes key decisions for: (1) Human Equivalent Concentration Calculation; (2) Endpoint/Dataset Selection; (3) Dose-Response Modeling; (4) Point of Departure (POD) Selection; (5) Low-Dose Extrapolation; and (6) Additional adjustments.

A.2.2.1 Human Equivalent Concentration Calculation

As discussed in **Section A.1.2**, there are clear species differences in the metabolism of BD, which need to be accounted for when calculating human equivalent concentrations (HECs) from test concentrations of BD administered to rodents. Accordingly, HECs were calculated using the following equation:

$$HEC = (TC * AF) / EF_{AK} \qquad Eq.2$$

Where,

- *HEC* = human equivalent concentration (ppm, continuous exposure);
- *TC* = test concentration administered to mice or rats (ppm, discontinuous exposure);
- *AF* = adjustment factor to account for discontinuous exposure in toxicity studies (e.g., 6 hours/24 hours per day, 5 days/7 days per week); and
- EF_{AK} = data-derived extrapolation factor (DDEF) to account for species differences in the toxicokinetics of BD.

DDEF values were derived in a manner consistent with USEPA (2014) guidelines. Based upon consideration of the uncertainty in the mode of action for systemic tumors (see **Section A.1.3**), the reactive metabolites of BD (EB, DEB, EBD) are assumed to each contribute to carcinogenic effects of BD. Although DEB is considered to be the most potent metabolite of BD with respect to genotoxicity (see MOA discussion above), a potential role for other reactive metabolites cannot be ruled out. To estimate the combined contribution of BD metabolites in quantitative manner (dose additivity assumed), a genotoxicity index approach was applied using the following equation:

$$GI_S = \sum[(AUC_{EB} \times RP_{EB}) + (AUC_{DEB} \times RP_{DEB}) + (AUC_{EBD} \times RP_{EBD})] \quad Eq.3$$

Where,

- GI_S = Species-specific genotoxicity index, calculated separately for male and female mice, rats, and humans (nM*hr per ppm*hr BD)
- *AUC* = Species-specific unit AUCs for each metabolite, which reflects the internal dose of each metabolite in each species (nM*hr per ppm*hr BD; **Table A.4**)
- *RP* = Relative potency of each metabolite for producing genotoxicity in mammalian cells (unitless; summarized in **Table A.5**).

Accordingly, EF_{AK} values for interspecies extrapolations can be calculated using a ratio approach (USEPA, 2014) as defined by the following equation:

$$EF_{AK} = \frac{GI_A}{GI_H} \qquad Eq.4$$

Where,

- EF_{AK} = Data-derived extrapolation factor for interspecies extrapolation due to toxicokinetic differences (unitless); and
- *GI* = Genotoxicity Index for BD metabolites in laboratory animals (A) or humans (H) (**Table A.5**).

In the absence of data, no attempts were made to account for potential toxicodynamics differences across species with respect to the carcinogenic effects of BD.

A.2.2.2 Endpoint/Dataset Selection

Target tissues for cancer risk assessment were selected based upon a review of risk assessments by regulatory agencies and risk assessors available for 1,3-butadiene (**Table A.1**), and based on the review of the recently published literature. Data sets used to estimate the cancer potency of BD include the lifetime cancer bioassay incidence data for the following:

- *Female Mice* – lymphoma, histiocytic sarcoma, mammary gland, ovary, Harderian gland, liver, forestomach, lung, heart tumors (**Table A.2**);
- *Male Mice* – lymphoma, histiocytic sarcoma, preputial, kidney, Harderian gland, liver, forestomach, alveolar-bronchiolar, heart tumors (**Table A.2**);
- *Female Rats* – uterus, mammary gland, thyroid, and Zymbal's gland tumors (**Table A.3**); and
- *Male Rats* – pancreas, testes, and glial cell tumors (**Table A.3**).

Incidence data for acute exposures (all sexes and species) were not used to estimate cancer potency since no significant tumor incidences were reported. Similarly, incidence data from the stop-exposure study in male mice were not used to estimate cancer potency since sufficient data were available from lifetime studies.

A.2.2.3 Benchmark Dose Modeling

Each tumor data set was modeled separately using the multistage model (1st through 5th degree polynomial) (BMDS, version 3.2). The best fitting degree of the multistage model was selected based on a consideration of AIC, goodness of fit p-value, and visual inspection.
The multistage model was used to estimate the EC10 value, as well as its 95% lower confidence limit (LEC10), 95% upper confidence limit (UEC10), and the cumulative distribution function (CDF; 1st – 99th percentile values). Within each species, a distribution of endpoint-specific unit risk values was determined using the CDFs generated by BMDS and *Eq.1* (i.e., 10%/EC10). A distribution for the multi-site unit risk value was calculated for each species/sex by summing across cancer endpoints:

$$UR_{Combined} = \sum(UR_{Endpoint1} + UR_{Endpoint2} \ldots) \qquad Eq.5$$

Where,

- *$UR_{Combined}$ = combined unit risk across endpoint calculated for each sex and species (ppm^{-1}); and*
- *$UR_{Endpoint}$ = tumor endpoint specific unit risk within each sex/species (ppm^{-1}).*

A distribution for the combined UR values was generated using Monte Carlo methods (Crystal Ball for Excel; version 7.3) based on a simulation of 10,000 iterations. The 5th and 95th percentiles for the combined UR distributions were adopted as the lower and upper confidence limits, respectively, for each combined data set.

A.3 Results

A.3.1 Human Equivalent Concentrations

EF_{AK} values of 0.0300, 0.0228, 0.531, and 0.556 were calculated to extrapolate to humans from female mice, male mice, female rats, and male rats, respectively (**Table A.6**). These values account for species differences in the internal dose of reactive epoxide metabolites (**Table A.4**) as well as metabolite differences in genotoxic potency (**Table A.5**). For comparison purposes, EF_{AK} values of 0.000886, 0.000630, 0.0165, and 0.0175 were similarly calculated for DEB alone, if an alternative the hypothesis were adopted assuming the clastogenic effects of DEB are solely responsible for the observed tumor response (*i.e.*, assuming contributions of EB and EBD tumor response are negligible). The approach for BD used here is similar to that proposed by Fred et al. (2008) to address differences in the genotoxic potency of BD metabolites for cancer endpoints for tumors observed in mice and rats, but has been expanded to include humans as well as additional data sets for assessing relative genotoxic potency.

A.3.2 Unit Risk Values and Species Concordance

Central tendency (MLE) estimates for unit risk values based on combined target tissue cites were determined to be 0.00088, 0.00034, 0.000067, and 0.000014 (ppm^{-1}) based on data for female mice, male mice, female rats, and male rats, respectively (**Table A.7**). Corresponding lower bound values were determined to be 0.00057, 0.00028, 0.000042, and 0.0000075 (ppm^{-1}), respectively. Similarly, corresponding upper bound values were determined to be 0.0012, 0.00043, 0.000096, and 0.000021 (ppm^{-1}), respectively. A comparison of the species-specific distributions for BD unit risk values is provided in **Figure A.3**. This comparison shows that the data-derived extrapolation factor adjustments to account for species differences in metabolic activation BD improves the overall concordance across species as evidence by the reduced spread of the distributions in the adjusted unit risk values (**Figure A.3B**) as compared to unadjusted values (**Figure A.3A**). In addition, the range of adjusted unit risk values based on rodent tumor data compare reasonably well to the unit risk distribution derived from epidemiology data for exposed styrene-butadiene rubber (SBR) workers based on data for leukemia and bladder cancer (Valdez-Flores and Kirman, see **Appendix B**). This unit risk value is

based on a Cox proportional hazard regression for the two cancer endpoints, based on the most recent follow-up and exposure data (Sathiakumar et al., 2021a,b).

A.3 Consideration of Sensitive Subpopulations and Additional Adjustments

As a matter of policy, derivation of unit risk values should also consider potentially sensitive subpopulations (USEPA, 2005). Potential sensitivity to the carcinogenic effects of BD can be attributed to toxicokinetic and/or toxicodynamic factors as summarized below.

- *Toxicokinetic Factors* - With respect to toxicokinetics, the mode of action for BD's carcinogenic action involves metabolic activation to reactive epoxides (Albertini et al., 2010). Blood and urinary biomarker data for BD can be used to characterize human variation in metabolism due to: (1) gender differences; (2) ethnicity differences; and (3) genetic polymorphisms. Gender differences have been reported for occupationally exposed men and women to BD with respect to hemoglobin adducts (Vacek et al., 2010) and urinary biomarkers (Kotapati et al., 2015). When expressed on a per mg/m^3 BD exposure basis, these differences are approximately 2-fold (females < males). Ethnicity differences, generally less than a factor of 2, have been reported for urinary biomarkers for BD metabolites, including significantly higher concentrations of MHBMA in whites as compared to Japanese Americans and Native Hawaiians (Park et al., 2014), and significantly higher concentrations of DHBMA in African Americans compared to whites (Boldry et al., 2017). In addition, ethnic differences for urinary excretion of repaired DNA adducts (EB-GII) have been reported (Sangaraju et al., 2017; Jokipii Krueger et al., 2020). Differences across ethnic groups are generally up to 2- to 3-fold. Some of the ethnic differences in BD biomarkers may be related to known genetic polymorphisms across ethnic groups (Fernandez-Salguero et al., 1995; Wormhoudt et al., 1999; London et al., 2000; Yoshikawa et al., 2000), especially GSTT1 gene copy number (Boldry et al., 2017). *In vitro* studies have shown that human cell lines with differing status in glutathione-S-tranferase (GST-T1) differ in sensitivity to EB (GSTT1- cells exhibiting greater sensitivity than GSTT1+ cells; Degner et al., 2020). The effects of genetic polymorphisms for various enzyme systems (P450, GST, EH) alone and combined were assessed for the DEB-specific hemoglobin adduct levels (THBVal). Specific polymorphisms (particularly for GSTT1) showed significant effects on THBVal levels (Fustinoni et al., 2002). THBVal levels across different metabolism groups (i.e., combinations of genetic polymorphisms) were found to be generally within a factor of 2 of the overall mean. The weight of evidence from available biomarker studies for BD suggests that human variation based on toxicokinetic (TK) factors is likely near or below the default uncertainty factor for intraspecies variation (i.e., $UF_{tk} \lesssim 3$).
- *Toxicodynamic Factors* – Because BD is metabolized to reactive epoxides capable of producing genotoxic events, conditions and disease states associated with reduced repair of DNA damage are expected to be potentially sensitive to the carcinogenic effects of BD. For example, sensitivity to BD metabolite and clastogen, 1,2,3,4-diepoxybutane (DEB), is specifically used in the diagnosis of Fanconi's anemia

(Auerbach, 2015). However, quantification of potential risks to specific disease states exposed to BD is beyond the scope of this paper.

As a matter of policy, genotoxic chemicals such as BD are expected to pose an increased risk when exposures occur early in life, a time period that is not directly covered by data from animal cancer bioassays or epidemiology studies of occupational cohorts. Some evidence is available for BD that suggests early-life exposures are not associated with increased risk. For example, BD is metabolically activated to epoxide metabolites by cytochrome P450, principally isozyme CYP2E1. Based on the ontogenesis of CYP2E1 activity in humans (Hines, 2007; Johnsrud et al., 2003), metabolic activation of BD is expected to be much lower in neonates, infants, and children when compared to adults. In additional, acute cancer bioassays conducted for BD in mice indicate that single, high exposures relatively early in life do not initiate tumors over the course of their lifetime (Bucher et al., 1993). For these reasons, application of an ADAF may not be required to ensure the protection of human health.

A.4. Discussion and Conclusions

A series of analyses were conducted using the robust rodent tumor data available for BD to determine human relevance. From these analyses, several conclusions can be supported:

(1) *Risk from Acute Exposures* - Acute exposures, even those associated with extremely high levels of BD , do not appear to be associated with an increased risk of cancer. An apparent duration threshold, which falls between 1 day (no increase in tumors observed) and 91 days (the shortest duration with an observed increase in tumors) likely exists for BD tumors in mice. For this reason, quantitative cancer risk assessment may not be required for acute human exposures scenarios to BD.
(2) *Use of DDEF Adjustments for Interspecies Extrapolation* - By accounting for species differences in metabolic activation, concordance across species in the cancer potency estimates for BD is improved (**Figure A.3B**). Remaining differences across species in BD cancer potency may reflect toxicodynamic differences, for which no adjustments were made. To reconcile the differences in cancer potencies for BD in mice and rats in **Figure A.3B** (i.e., for the distributions to overlap) would require the existence of a toxicodynamic difference of approximately 13- to 25-fold between the two species (mouse>rat).

UR values were derived for BD based upon tumor incidences reported in laboratory mice and rats following lifetime exposures. The UR values derived here are considerably lower than those derived previously by regulatory agencies (see **Table A.1**), since they account for species differences in metabolic activation. Substantial species differences in the metabolism of BD result in humans and rodents experiencing internal doses of reactive metabolites that are qualitatively similar (i.e., all 3 reactive metabolites are formed in all species), but exhibit large quantitative differences. Accounting for these differences serves as an important challenge to human health risk assessment. The methods of Fred et al. (2008)/Motwani and Tornqvist (2014) were extended and applied to this assessment to account for species differences in

metabolism, as well as differences in metabolite mutagenic potency. This approach made use of biomarker data (metabolite-specific hemoglobin adducts) to quantify species differences in the internal doses of BD metabolites experienced in mice, rats and humans. The use of hemoglobin adducts for BD here is consistent with USEPA's practice in the assessment of other chemicals (e.g., acrylamide risk assessment; IRIS, 2010). The availability of biomonitoring data across species enables a data-driven approach to better place rodent tumor results into the context of human equivalent exposure. Although the use of a relative potency approach to address the contributions from mixtures of metabolites originating from a single chemical may be viewed as novel, this approach has been applied to risks assessments for chemical mixtures that are believed to act via a common mechanism, including polycyclic aromatic hydrocarbons (USEPA, 2010a) and dioxin-like chemicals (USEPA, 2010b), and are therefore justified here.

Sources of uncertainty are identified for this assessment below.

- *Mode of Action* – For this assessment, it was assumed that all three epoxide metabolites contribute to the tumor responses observed in rodents. Because DEB is a bi-functional genotoxic agent that is capable of producing clastogenic effects, an alternative hypothesis that DEB is solely responsible for the observed tumor responses can be supported. Based on the DDEF values derived for DEB alone compared to those derived for the combined action of all three metabolites (**Table A.5**), UR values based on DEB alone would be approximately 32- to 36-fold lower than derived here, which is consistent with the comparatively low levels of DEB produced by humans. For this reason, the assumption that all three BD epoxide metabolites contribute to cancer risk is considered to be a health-protective assumption.
- *Relative Potency Approach* – The relative genotoxic potency estimates calculated here (EB=1.0, DEB=85, EDB=1.5) are similar to the mutagenic potencies estimated by Fred et al. (2008) for these three metabolites (EB=1.0, DEB=32, EDB=0.21), with the exception of EDB. The genotoxic potency of EDB was higher in this assessment, largely based on the results of Meng et al. (2010) who reported considerable differences in potency across the four stereoisomers of EBD, for which some steroisomers (e.g., 2R,3S-EBD) were found to be more potent than EB. The approach used in this assessment could be expanded to address stereochemistry differences in metabolism, with the collection of data to characterize species differences in stereoisomer formation and their internal doses. However, some uncertainty remains regarding the potential contribution of other BD metabolites to adverse effects, including hydroxymethylvinyl ketone (HMVK), as well as proposed chlorohydroxy metabolites produced via myeloperoxidase (Elfarra and Zhang, 2012; Wang et al., 2018; Wu et al., 2019). The approach used in this assessment could be expanded to include additional BD metabolites if their importance is warranted from a mechanistic standpoint, and if the data needed to estimate internal doses (e.g., from hemoglobin adduct data) and relative potencies are generated.
- *Human Equivalent Concentration Calculation* – Uncertainty in the internal dose estimates calculated from hemoglobin adducts per Motwani and Tornqvist (2014) is considered low. Some uncertainty remains in the use of hemoglobin adduct data collected from male workers (**Table A.4**) to estimate internal doses in human

populations that include males and females. Small differences (<2-fold) in internal dose estimates are noted between male and female mice, and between male and female rats (Motwani and Tornqvist, 2014; Swenberg et al., 2011). However, these observations in rodents differ from findings in humans, which showed similar or lower formation of adducts in women compared to men (Swenberg et al., 2011).

- *Dose-Response Modeling* – Uncertainty in the dose-response modeling is considered to be relatively low. The BMD:BMDL ratios for the combined UR values calculated for each sex and species, which serve as overall indicators of the uncertainty associated with fitted model parameters, were found to be less than a factor of 1.5 (**Table A.7**) in this assessment.

Despite these sources of uncertainty, overall confidence in the UR values derived for BD here is high. The key data sets are defined by well-conducted studies that have been consistently selected by regulatory agencies to support cancer risk assessments for BD. Confidence in the dosimetry of the assessment is also considered high, since they are derived from excellent biomarker data that are metabolite-specific and have been quantified in all three species of interest (mice, rats, and humans). Confidence in the cancer database is considered high, since the carcinogenicity of BD has been well-studied in rodents and humans and the database is considered robust.

Charge Questions

1) "Is the use of hemoglobin adduct data to quantify species differences in the internal doses of epoxide metabolites appropriate?"
2) Given the data on metabolic activation as a function of age and the acute cancer bioassay data in rodents, and is an ADAF needed for BD?

Table A.1. Summary of Rodent-Based Cancer Risk Assessments for 1,3-Butadiene

Assessor (Year)	Endpoint	Data set	DR Model	POD Type	POD Value	Species Extrapolation Assumption	Low-Dose Extrapolation Assumption	Unit Risk (ppm^{-1})
USEPA (2002)	Leydig cell, pancreatic exocrine cell, Zymbal gland, Mammary gland, thyroid follicular cell	Male & Female Rats (Hazleton, 1981)	Multistage	LEC10	NS	Air concentration equivalence	Linear	0.0042-0.056
	Lymphocytic lymphomas, histiocytic sarcomas, heart hemangiosarcomas, lung, forestomach, Harderian gland, liver, preputial gland, ovary, mammary gland	Male & Female Mice (NTP, 1993)	Multistage-Weibull time-to-tumor	LEC10	0.7-13.3 ppm	Air concentration equivalence	Linear	0.0064-0.29
Health Canada (2000)	Multiple	Male & Female Rats (Hazleton, 1981)	Multistage	TC05	4.7-905 mg/m^3	Air concentration equivalence	NA	0.00012-0.024*
	Multiple	Male & Female Mice (NTP, 1993)	Multistage	TC05	1.4-23 mg/m^3	Air concentration equivalence	NA	0.0048-0.079*
OEHHA (2009, 2011)	Multiple	Female Mouse Male & Female Mice (NTP, 1993); Male & Female Rats (Hazleton, 1981)	Multistage	NS	NS	Surface area scaling	Linear	0.077 0.002-0.16

*Linear potency estimate calculated by dividing the benchmark response rate (5%) by the TC05 value

NS = not specified; NA = not applicable; LC10 = 95% lower confidence on the concentration producing a 10% increase in extra risk; TC05 = total concentration associated with a 5% increase in tumor incidence.

Table A.2. Mouse Tumor Data Following Inhalation Exposure to 1,3-Butadiene (NTP, 1993)

Gender	Duration (reference)	Exposure	Concentration (ppm)	HEC* (ppm, cont)	Lymphoma	Histiocytic sarcoma	Heart	Alveolar-bronchiolar	Forestomach	Mammary Gland	Liver	Harderian	Preputial, Ovary
Male	Acute (Bucher et al., 1993)	2 hours (1x)	0	0	7/59	NR	NR	8/59	0/59	0/59	17/59	NR	NR
			1000	3655	8/58	NR	NR	9/58	1/58	0/58	21/58	NR	NR
			5000	18275	8/58	NR	NR	12/57	1/58	0/58	21/58	NR	NR
			10000	36550	10/58	NR	NR	8/58	3/58	1/58	18/58	NR	NR
	Long-term (NTP, 1993)	6 hr/d, 5 d/wk, 40 wks	200	1566	8/50	5/50	15/50	36/50	3/50	NR	33/49	27/50	1/50
		6 hr/d, 5 d/wk, 52 wks	312	2444	8/50	7/50	33/50	32/50	9/50	NR	25/50	30/50	4/50
		6 hr/d, 5 d/wk, 13 wks	625	4895	22/50	2/50	7/50	28/50	7/50	NR	24/49	23/50	5/50
		6 hr/d, 5 d/wk, 26 wks	625	4895	33/50	2/50	13/50	17/50	10/50	NR	13/50	13/50	3/50
	Lifetime (NTP, 1993)	6 hr/d, 5 d/wk, 103 wks	0	0	4/50	0/50	0/50	21/50	1/50	NR	21/50	6/50	0/50
			6.25	49	2/50	0/50	0/49	23/50	0/50	NR	23/50	7/50	0/50
			20	157	4/50	4/50	1/50	19/50	0/50	NR	30/50	9/50	0/50
			62.5	490	6/50	5/50	5/48	31/49	1/50	NR	25/48	20/50	0/50
			200	1566	2/50	7/50	20/48	35/50	8/50	NR	33/48	31/50	5/50
			625	4895	51/73	$4/73^{+}$	$4/73^{+}$	$3/73^{+}$	$4/73^{+}$	NR	$5/72^{+}$	$6/73^{+}$	$0/73^{+}$
Female	Acute (Bucher et al., 1993)	2 hours (1x)	0	0	13/57	NR	NR	3/56	0/57	2/57	5/56	NR	0/53
			1000	2778	19/56	NR	NR	4/56	1/56	1/56	6/55	NR	0/52
			5000	13889	18/57	NR	NR	0/57	0/57	3/57	8/57	NR	1/53
			10000	27778	13/58	NR	NR	3/58	0/58	4/58	3/58	NR	0/56

	Lifetime (NTP, 1993)	6 hr/d, 5 d/wk, 103 wks	0	0	6/50	3/50	0/50	4/50	0/50	0/50	15/49	8/50	1/49
			6.25	52	12/50	2/50	0/50	15/50	0/50	2/50	14/49	10/50	0/49
			20	167	11/50	7/50	0/50	19/50	3/50	4/50	15/50	7/50	1/48
			62.5	521	7/50	4/50	1/49	24/50	2/50	12/50	19/50	15/50	9/50
			200	1667	9/50	7/50	21/50	25/50	4/50	15/50	16/50	20/50	8/50
			625	5208	32/80	4/80+	23/80+	22/78+	22/80+	16/80+	2/80+	9/80+	6/79+

*Calculated using EFAK values of 0.0300 and 0.0228 for female and male mice, respectively (**Table A.6**).

+Due to early deaths primarily attributed to lymphomas this dose group was excluded from dose-response modeling for other tumor types

NR = not reported

Table A.3. Rat Tumor Data Following Inhalation Exposure to 1,3-Butadiene (Owen and Glaister, 1990; Melnick and Huff, 1993)

					Target Tissues					
Gender	**Duration**	**Exposure**	**Concentration (ppm)**	**HEC (ppm, cont)**	**Pancreas**	**Zymbal**	**Mammary**	**Thyroid**	**Glial Cell**	**Testis, Uterus**
Male	Lifetime	6 hr/d, 5 d/wk, 103 wks	0	0	3/100	1/100	1/100	3/100	1/100	0/100
			1000	321	1/100	1/100	2/100	5/100	4/100	3/100
			8000	2569	11/100	2/100	0/100	1/100	5/100	8/100
Female	Lifetime	6 hr/d, 5 d/wk, 103 wks	0	0	2/100	0/100	50/100	0/100	NR	1/100
			1000	336	0/100	0/100	79/100	4/100	NR	4/100
			8000	2690	0/100	4/100	81/100	11/100	NR	5/100

*Calculated using EFAK values of 0.531 and 0.556 for female and male rats, respectively (**Table A.6**).

Table A.4. Use of Hemoglobin Adduct Data in Mice, Rats, and Humans to Quantify Species Differences in Internal Dose of BD Epoxide Metabolites (adapted from Motwani and Tornqvist, 2014)

	Metabolite-Specific Unit Internal Dose (nM*hr per ppm*hr BD)[1]				
	Mouse		Rat		Human[1]
Metabolite	**Female**	**Male**	**Female**	**Male**	**Male**
EB	13±2	15±2	0.77±0.1	0.72±0.1	0.11±0.076
DEB	27±7	38±8	1.45±0.2	1.37±0.3	0.024±0.020
EBD	266±71	210±30	19±0.9	19±2	52±36

[1]Calculated as the pooled arithmetic mean±SD using two data sets for exposed male workers (Motwani and Tornqvist, 2014)

Table A.5. Relative Genotoxic Potencies for BD Metabolites in Mammalian Cells[1]

	Metabolite				
Endpoint	**EB**	**DEB**	**EDB**	***In Vitro* Cell System**	**Reference**
DNA Damage	1.00	11.21	0.961	Human hepatocytes, pH 11.9	Wen et al. 2011; Zhang et al. 2012
	1.00	4.22	0.955	Human hepatocytes, pH 9	
DNA Damage Mean±SD	*1.00*	*7.72±4.94*	*0.96±0.004*		
Mutations	1.00	81.66	2.10	Human TK6 (HPRT)	Meng et al. 2010
	1.00	277.12	4.46	Human TK6 (TK)	
	1.00	58.10	0.45	Human TK6 (HPRT)	Cochrane and Skopec (1994)
	1.00	114.83	0.71	Human TK6 (TK)	
	1.00	49.08	0.35	BB Mouse Fibroblasts	Erexson and Tindall (2000)
	--[2]	--[2]	--[2]	BB Rat Fibroblasts	
	1.00	4.20	3.87	SA T100	Adler et al. (1997)
Mutations Mean±SD	*1.00*	*97.5±95.3*	*1.99±1.81*		
Micronuclei	1.00	128.28	0.58	BB Mouse Fibroblasts	Erexson and Tindall (2000)
	1.00	124.08	0.74	BB Rat Fibroblasts	
	--[2]	--[2]	--[2]	Rat spermatids	Sjoblom and Kahdetie, 1996
Micronuclei Mean±SD	*1.00*	*126.18±2.97*	*0.66±0.12*		
Overall Mean±SD[3]	***1.00***	***85.28±82.81***	***1.52±1.48***		

[1]Calculated based on the ratio of linear slopes for each metabolite relative to the slope for EB assessed in the same cell test system.

[2]Only DEB yielded a positive response, therefore relative potencies were not estimated for this data set.

[3]Values used to support calculation of data-derived extrapolation factors (**Table A.6**).

Table A.6. Data-Derived Extrapolation Factors to Quantify Species Differences in BD Toxicokinetics (EF_{AK})

		Individual Metabolites			
Parameter (units)	**Species/ Extrapolation**	**EB**	**DEB**	**EBD**	**Metabolites Combined**[3]
Genotoxicity Index (nM*hr per ppm*hr BD)[1]	Female Mouse	13.0	2303	404	2719
	Male Mouse	15.0	3241	319	3574
	Female Rat	0.77	124	28.8	153
	Male Rat	0.72	117	28.5	146
	Human	0.109	2.04	79.2	81.4
EF_{AK} (Unitless)[2]	Human:Female Mouse	0.00842	0.000886	0.196	0.0300[4]
	Human:Male Mouse	0.00730	0.000630	0.249	0.0228[4]
	Human:Female Rat	0.142	0.0165	2.75	0.531[4]
	Human:Male Rat	0.152	0.0175	2.75	0.556[4]

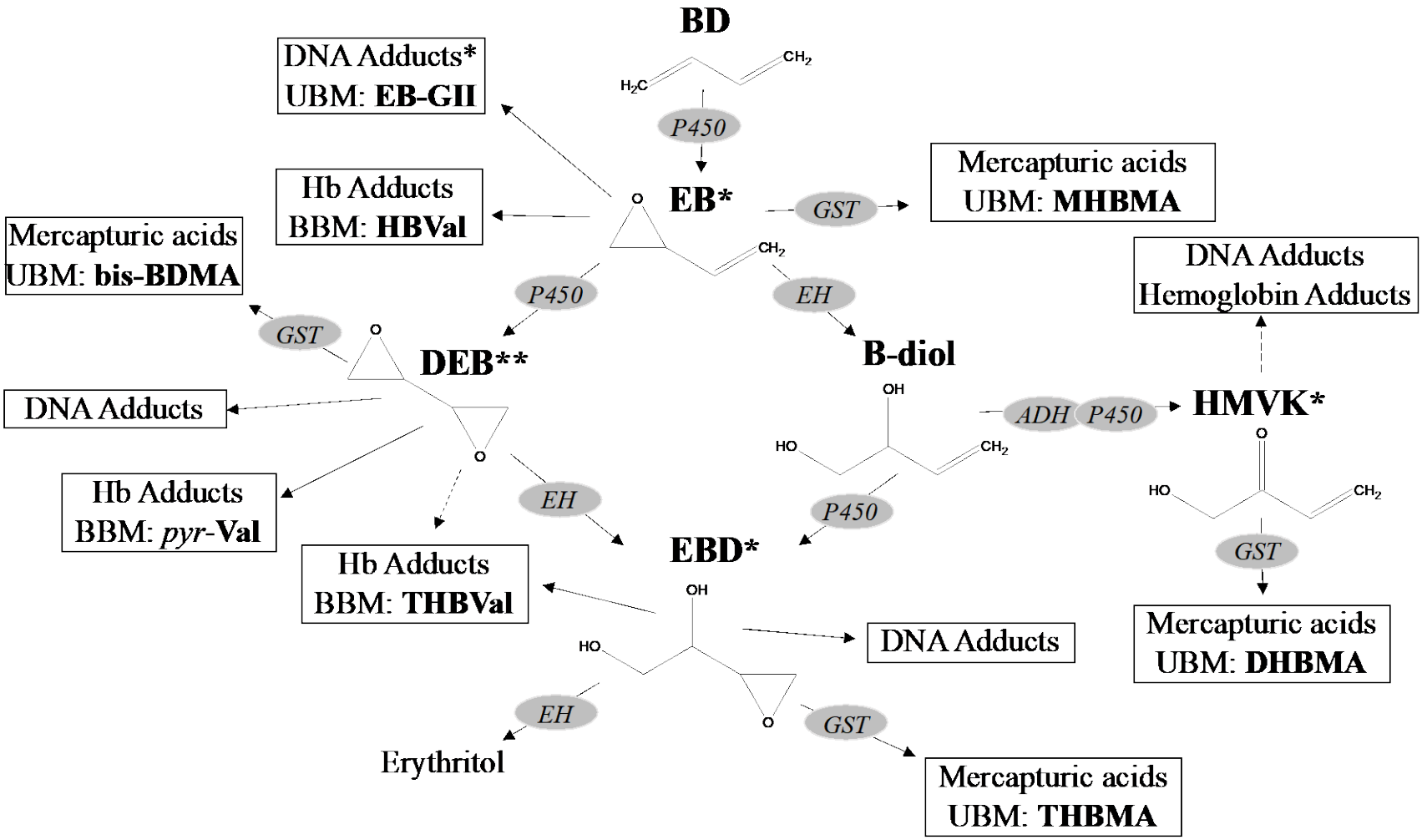
[1]Calculated as the product of unit internal dose value (**Table A.4**) and relative cytotoxic potency (**Table A.5**), units of nM*hr per ppm*hr BD.
[2]Calculated as the ratio of genotoxicity indices for each species, unitless.
[3]Calculated as the sum across metabolites.
[4]Values used to calculate human equivalent concentrations for tumor PODs attributed to all 3 epoxide metabolites combined.

Table A.7. Unit Risk Values Based on Tumors in Mice and Rats

Data Set			Range of Model Fit Statistics for Individual Tumor Types		Unit Risk for Combined Tumor Types (ppm^{-1} HEC)*
Data Set	**N**	**Range of Observation, (HEC, ppm continuous)**	**p-Values**	**AICs**	
Female Mouse (**Table A.2**)	558	52-27800	0.103-0.867	81.6-349.1	8.8E-04 (5.7E-04 – 1.2E-03)
Male Mouse (**Table A.2**)	756	49-36550	0.052-0.966	35.6-337.3	3.5E-04 (2.8E-04 – 4.3E-04)
Female Rat (**Table A.3**)	300	336-2690	0.00016-0.969	35.7-357	6.7E-05 (4.2E-05 – 9.6E-05)
Male Rat (**Table A.3**)	300	321-2570	0.131-0.163	88.7-109	1.4E-05 (7.5E-06 – 2.1E-05)

*HEC = Interspecies adjustments made assuming all 3 genotoxic epoxide metabolites contribute to the observed tumorigenic response in rodents.

Fig. A.1 Metabolism of BD



*monofunctional alkylating agent; **bifunctional alkylating agent

Fig. A.2 Species Differences in Relative Internal Dose of Genotoxic Equivalents (internal dose x genotoxic potency) from BD Metabolites (pie surface area proportionate to the magnitude of internal dose; dark shading = DEB, medium shading = EB, light shading = EBD).

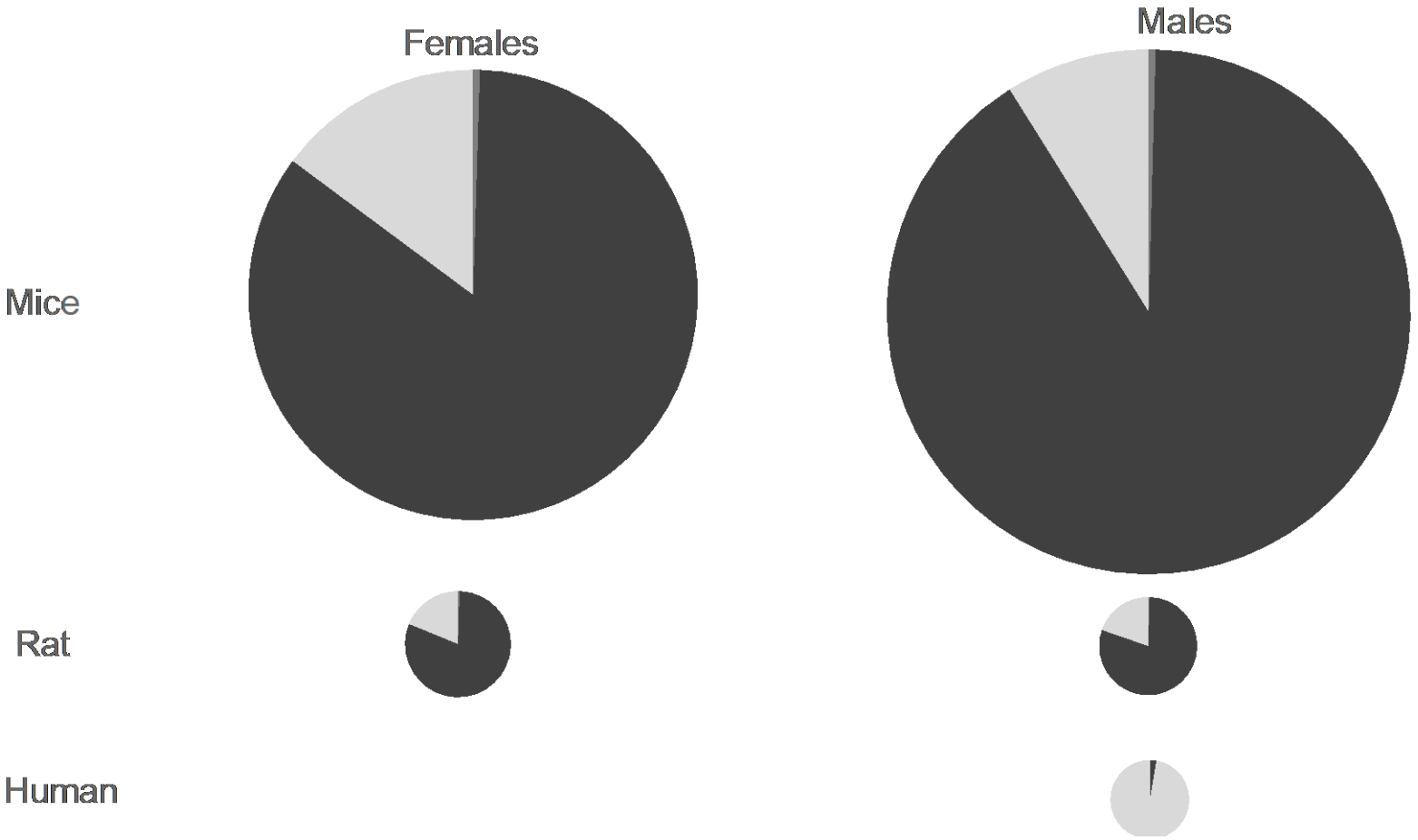
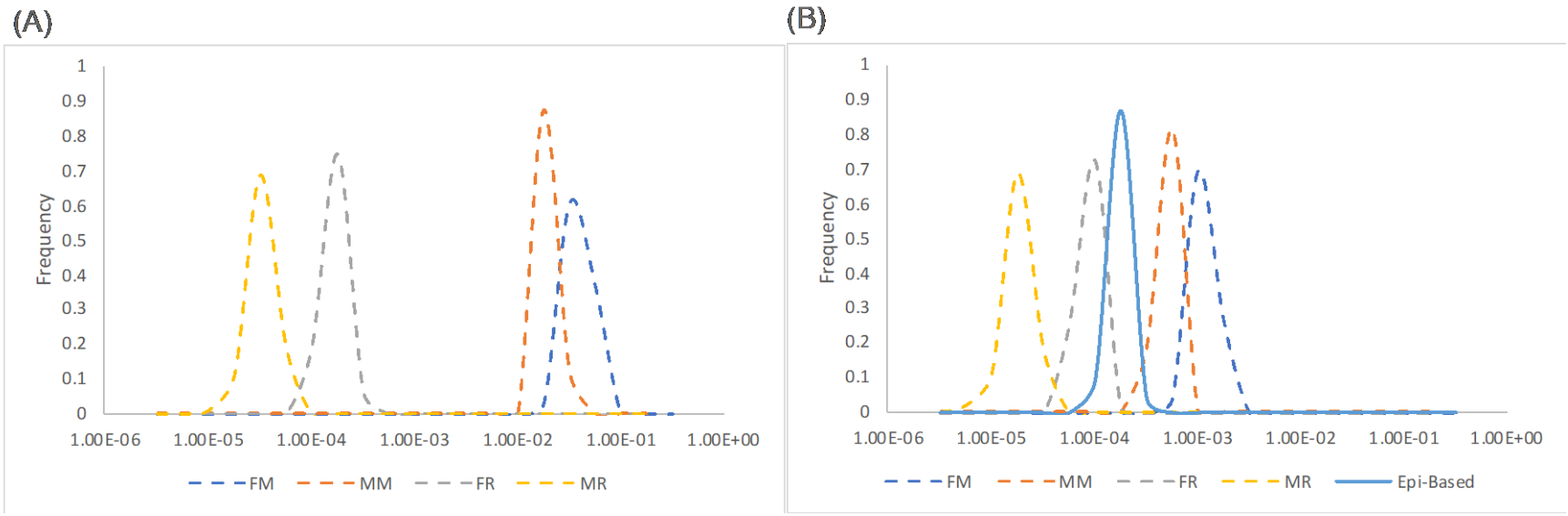


Fig. A.3. Concordance of Unit Risk Distributions: (A) Unadjusted Exposure; and (B) Adjusted for Species Differences in Internal Dose and Genotoxic Potency of BD Metabolites; FM=female mouse, MM=male mouse, FR=female rat, MR=male rat; Epi-based=based on epidemiology data (Valdez-Flores and Kirman, in prep)



Appendix B: An Updated Lymphohematopoietic and Bladder Cancers Risk Evaluation for Occupational and Environmental Exposures to 1,3-Butadiene

B.1 Introduction

Exposures to 1,3-butadiene (BD) have been the subject of regulatory interest for several decades in the USA and most developed countries (e.g., USEPA (1985), NIOSH (1991), OSHA (1996), Cagen et al. (1996), IARC (1986, 1992)). Exposure-response modeling and the evaluation of potential risks from exposures to BD were initially based on animal studies (NTP, 1993; Melnick and Huff, 1993). In recent years, however, advances in exposure-response modeling and the availability of good epidemiological data have spawned the development of risk characterizations that better reflect the risks of health effects in human populations exposed to BD. Since the early 1990s, the University of Alabama at Birmingham (UAB) developed epidemiological data of workers exposed to BD in the North American styrene-butadiene rubber (SBR) industry. Originally, the UAB epidemiological data (hereinafter referred to as the SBR study) included only male workers that were followed up from 1944 through 1991. The study was updated to add seven more years of follow up, through 1998, and then in 2002 women were also included. In 2009, the most recent update of the SBR study, the cohort includes male and female workers. Table B.1 lists the update history for the SBR epidemiological data.

Update	Exposure Characterization (years estimated)	Period of Follow up	Sex	Number of workers	Number of Deaths
Original[1]	Original (1944-1991)	1944-1991	Male	17,964	4,665
Update 1[2]	Refined[5] (1944-1991)	1944-1998	Male	17,924	6,237
Update 2[3]	Refined[5] (1944-1991)	1943-2002	Female	4,863	1,198
Update 3[4]	Refined[5] (1944-1991)	1943-2009	Male and Female	22,785	10,617

[1]Delzell, E., N. Sathiakumar, M. Macaluso, M. Hovinga, R. Larson, F. Barbone, C. Beall, P. Cole, J. Julina, and D.C.F. Muir. 1995. A Follow-Up Study of Synthetic Rubber Workers. Submitted to The International Institute of Synthetic Rubber Producers. October 2, 1995.

[2]Sathiakumar N., J. Graff, M. Macaluso, G. Maldonado, R. Matthews, and E. Delzell. 2005. An updated study of mortality among North American synthetic rubber industry workers. Occup Environ Med. 62:822-829.

[3]Sathiakumar N. and E. Delzell. 2009. A follow-up study of mortality among women in the North American synthetic rubber industry. J Occup Environ Med.51:1314-1325.

[4]Sathiakumar N., M. Tipre, M. Leader, I. Brill, and E. Delzell. 2019. Mortality among men and women in the North American synthetic rubber industry, 1943 to 2009. J Occup Environ Med. 61:887-897.

[5]Macaluso M, Larson R, Lynch J, Lipton S, Delzell E. Historical estimation of exposure to 1,3-BD, styrene, and dimethyldithiocarbamate among synthetic rubber workers. J Occup Environ Hyg 2004;1:371-390.

Table B.1. SBR epidemiological study updates, including workers with and without exposure estimates

USEPA (2002) used the original SBR epidemiological study (first row in Table B.1) and the Texas Commission for Environmental Quality (TCEQ, 2008) used the first update (second row in Table B.1) to develop their cancer potency assessments because it provided the best data available and included individual exposure metrics to evaluate risks to humans exposed to BD via the inhalation route. After extensive literature search, the TCEQ (2008) concluded that there were no other epidemiological studies with exposure history that could be used to develop exposure-response models for cancer risk assessment of BD.

USEPA (2002) evaluation was based on Poisson regression modeling published by Health Canada (2000) and did not explore co-exposure variables other than cumulative styrene (STY). TCEQ (2008) assessment, on the other hand, was based on Cox proportional hazards modeling (Cox, 1972). More recently (e.g., assessment for ethylene oxide, USEPA 2016), regulatory agencies have used Cox proportional hazards modeling over Poisson regression modeling because Cox proportional hazards models control better for the effect of age on cancer development. It is important to optimally control for age in cancer mortality or incidence studies because age is usually the most important factor associated increases in cancer mortality and incidence rates. We also analyzed the 1998 SBR study data and reported our results in Sielken et al. (2007). Follow up papers by Sielken and Valdez-Flores (2011, 2013, and 2015) analyzed the update of the SBR data for male workers with follow up through the end of 1998. The present paper uses the most recent update of male and female workers of the SBR study with 11 more years of follow up through the end of 2009. Table B.2 presents the breakdown of workers by gender, plant, race, and vital status that are included in our analyses because they have exposure estimates that can be used in exposure-response modeling.

Statistic	All Workers	Male	Female
Number of Employees	21,087	16,579	4,508
Plant, Location, Number			
1. Kentucky	1,563	1,391	172
2. Louisiana	2,463	1,988	475
3. Louisiana	2,849	2,084	765
4. Texas	2,929	2,328	601
5. Ontario	7,044	5,356	1,688
6. Texas	4,239	3,432	807
Race			
White/Unknown	18,674	14,486	4,188

Black/Other	2,413	2,093	320
Vital Status			
Alive	11,180	8,228	2,952
Deceased	9,665	8,214	1,451
Unknown	242	137	105

Table B.2. Summary statistics of workers with exposure estimates in the most recent SBR study update

USEPA designated BD as a high priority chemical under TSCA in December 2019, and the chemical is currently undergoing risk evaluation. The USEPA (2002) assessment for BD was conducted approximately 20 years ago and used exposures to cumulative BD (ppm-years) as the predictor variable of all leukemia mortality based on the original SBR study. Based upon the presence of multiple updates for this cohort (Table B.1), USEPA's assessment no longer reflects the best available data and science for BD, and therefore is not an appropriate basis for assessing and managing risks for BD under TSCA. Here, we analyze the most recent SBR data that now includes approximately 5,000 women in addition to the more than 17,000 workers included in the original SBR study. The updated SBR study also includes 18 more years of follow up and approximately 6,000 more deaths than the 4,665 deaths in the original SBR study (Table B.1). EPA's assessment relied on Health Canada's Poisson regression modeling (Health Canada, 2000) and did not explicitly adjust the model for other exposure metrics (e.g., cumulative number of high intensity tasks (HITs) that counted the number of tasks that involved exposures above 100 ppm for BD or 50 ppm for STY, etc.). In addition, as indicated by Sielken and Valdez-Flores (2015), the Poisson regression model developed by Health Canada excluded a large proportion of person-years due to an over-partition of age groups and other covariates. This over-partitioning in Poisson regression results in groups (partitions) with no cases at any exposure group but person-years of follow up. Person-years in groups with zero cases in every exposure group are implicitly left out of the likelihood and, therefore, do not affect the exposure-response relationship. This results in slope estimates that are biased high (TCEQ 2008 and Breslow and Day 1980, 1987). Cox proportional hazards models are used herein, and adjustment for other non-exposure and exposure covariates are included using non-parametric estimation.

Cumulative exposures weight concentration level and duration equally (i.e., cumulative [ppm × duration]). Though assigning different weights to exposure level and duration have been proposed (e.g., cumulative [$ppm^c \times duration^d$] for different values of c and d), particularly for lymphoma in mice exposed to BD (ten Berge, 1986, 1999; Kirman et al., in prep) none has resulted in improved model prediction for endpoints in humans. Similarly, cumulative exposures for different windows of time, whereby only exposures inside a period defined by the window of x and lag years (x>lag) from the observation time are considered relevant for the response, have been explored but

have not improved the model fit (Sielken and Valdez-Flores, 2015). Herein, these exposure metrics are considered again with the updated SBR study.

Because leukemia was the only endpoint that increased with cumulative exposure to BD among workers in the early SBR study updates, USEPA (2002) and TCEQ (2008) based their assessments primarily on all leukemia mortality. The most recent update of the SBR study found exposure response increases for all leukemia and bladder/urinary cancer mortality with cumulative exposure to BD (Sathiakumar et al. 2019, 2021). Although bladder/urinary cancer mortality increases with increasing BD exposures, it cannot be ascertained that the increase is attributed to BD exposures because workers were exposed to other chemicals (Sathiakumar et al., 2021). Sathiakumar et al. concludes that the association between BD and styrene exposures with increasing bladder/urinary cancer mortality "*could be due to uncontrolled confounding by smoking.*" All leukemia and bladder/urinary cancer mortality, along with some leukemia subtypes are considered in this paper (Table B.3).

Endpoint	All Workers	Male	Female
Leukemia	132 (29)	116 (16)	16 (13)
Lymphoid Leukemia	52 (13)	45 (7)	7 (6)
Myeloid Leukemia	67 (14)	61 (8)	6 (6)
Multiple Myeloma	60 (17)	52 (11)	8 (6)
NHL	110 (34)	93 (19)	17 (15)
Bladder/Urinary	95 (19)	85 (10)	10 (9)

[1]The International Agency for Research on Cancer (IARC) classified BD as a human carcinogen based on leukemia and lymphohematopoietic cancers. In a recent publication, Sathiakumar et al. (2019, 2021) found a positive relationship between leukemia and bladder cancer, respectively, exposures to BD
[2]Number in parentheses indicate the number of cancer deaths in groups with no occupational exposure to BD

Table B.3. Number of deaths n occupationally and non-occupationally exposed workers in 6 SBR plants (deaths with no exposure to BD) by cancer type[1,2]

B.2 Methods

B.2.1 Data

The analyses presented here are based on the most recent individual data of the SBR study that includes male and female workers that work or have worked in the SBR industry and have job history exposure estimates. Delzell et al. (1995) first published the results of the SBR data and included male workers followed up through the end of 1991. The SBR study was updated to include follow up through the end of 1998 and was

published by Sathiakumar et al. (2005). This update included substantially improved estimates of exposures to BD and STY (Macaluso et al., 2004). Complete individual exposure estimates, however, were available for workers in six of the eight plants in the study. Sathiakumar et al. (2019) published the most recent update of the SBR study that includes 11 years of additional follow up (through the end of 2009) and an increased number of mortalities. Table B.2 lists the number of male and female workers in each of the six plants with exposure estimates in the most recent update of the SBR study included in the analyses performed herein. In the six plants with exposure estimates, the data includes 116 leukemia deaths among male workers as compared to 81 in the 2000 update and 58 in the original 1995 study. The total number of decedents in all eight plants increased from 4,665 males in the 1995 update, to 6,237 males in the 2000 and to 10,617 males and females in the most recent update of the SBR data (note that the 1995 and 2000 studies included only male workers, the most recent study includes male and female workers). Table B.4 lists selected statistics of the distribution of cumulative BD ppm-years for workers in the six plants with exposure estimates in the most recent update of the SBR study and for decedents with selected cancer endpoints.

Characteristic	All Workers	Leukemia Decedents	Bladder/urinary Cancer Decedents
Number	21,087	132	95
Percent with no occupational exposure to BD	33.50%	21.97%	20.00%
50th percentile of BD ppm-years[1]	10.4 (0.049)[2] [0.231][3]	51 (0.240) [1.133]	50 (0.235) [1.111]
75th percentile of BD ppm-years	87 (0.409) [1.933]	263 (1.235) [5.844]	218 (1.024) [4.844]
95th percentile of BD ppm-years	561 (2.635) [12.467]	1,095 (5.143) [24.333]	1,256 (5.899) [29.911]
Maximum BD ppm-years	9,269 (43.533) [205.978]	7,743 (36.366) [172.067]	7,900 (37.104) [175.556]

[1]The percentile of BD ppm-years distribution is in terms of occupational exposures in the SBR study
[2]Equivalent environmental average concentration for 70 years. To convert to an environmental BD ppm concentration, multiply the cumulative occupational BD ppm-years by 10/20 (daily occupational inhalation rate in m^3/day / daily environmental inhalation rate in m^3/day) times 240/365 (number of occupational days in a year / number of calendar days in a year) divided by the duration of environmental exposure (e.g., 70 years).
[3]Equivalent occupational average concentration for 45 years (20 to 65 years of age). To convert to an occupational BD ppm concentration, divide the cumulative occupational BD ppm-years by 45 years.

Table B-4. Distributional characteristics of the cumulative BD ppm-years in the 2019 SBR study update for all workers included in the analyses, all leukemia decedents and

bladder/urinary cancer decedents and the equivalent average environmental and occupational BD ppm concentrations

USEPA (2002) and TCEQ (2008) assessments used leukemia mortality as the health endpoint for their risk evaluation because exposure-response models suggested that this endpoint had the highest risks with increasing exposures to cumulative BD exposures. Sielken and Valdez-Flores (2011, 2013, and 2015) developed exposure-response models based on leukemia and subtypes of leukemia using the 2000 update of the SBR study. In the 2019 update with follow up through the end of 2009, however, bladder/urinary cancers, in addition to leukemia, were found to be associated with increasing cumulative BD exposures (Sathiakumar et al. 2019 and 2021). However, the association between cumulative BD exposure and bladder/urinary cancer mortality cannot be ascertained as causal because workers were exposed to other chemicals and the apparent increase "*could be due to uncontrolled confounding by smoking*" (Sathiakumar et al., 2021).

The results presented here are based on mortality data for all leukemia, select subtypes of leukemia, and bladder/urinary cancer. Table B.3 lists the individual endpoints analyzed along with the number of deaths associated with each endpoint for male and female workers. Similar to Sielken and Valdez-Flores (2015), this paper considers seven cumulative exposure metrics in addition to cumulative BD ppm-years. Considered here are also cumulative BD ppm-years after adjusting for other exposures and exposure metrics. In the process of adjusting the exposure-response relationship between an endpoint and cumulative BD ppm-year, it is important to note that these adjustments do not necessarily result in smaller magnitude of the impact of BD ppm-years in the exposure-response model.

Life-table methodology is used to calculate excess risks based on the most 2017 U.S. survival probabilities and 2019 U.S. endpoint-specific mortality rates along with cumulative exposures to BD. Calculations of environmental risks assume continuous exposures 24 hours a day every day from birth to the lifetime of the individual. Occupational risks, on the other hand, are calculated assuming that workers are exposed 8 hours a day, 240 days per year each year during 45 years of occupational tenure starting at 20 years of age.

B.2.2 Analyses of Individual Endpoints

The six individual endpoints analyzed herein, were selected based on significant associations with cumulative BD exposures reported in Sathiakumar et al. (2021) and are listed in Table B.3 along with the number of male and female decedents with the response. No causal analysis or conclusions were attempted in this work. All endpoints analyzed had sufficient endpoint-specific decedents with non-zero cumulative exposure to BD to fit an exposure-response model. When the data were restricted to male workers only, there were still sufficient endpoint-specific decedents for all endpoints to fit an exposure-response model (tables for results restricted to male workers are

presented in the supplement to this paper). However, when the data were restricted to female workers, there were three or fewer endpoint-specific deaths with non-zero cumulative exposure. The number of endpoint-specific female decedent workers with non-zero exposures were insufficient to fit exposure-response models that includes only female workers.

Exposure-response models were fit to leukemia, lymphoid leukemia, myeloid leukemia, multiple myeloma, non-Hodgkin's lymphoma, and bladder/urinary cancer mortality data for male and female workers combined and for the set of male workers only. The most recent update of the SBR study includes information on six non-exposure variables (age, years since hire, calendar year, sex, race, and plant) and eight exposure variables (cumulative BD ppm-years, cumulative STY ppm-years, cumulative number of BD high intensity tasks (HITs), cumulative number of STY HITs, cumulative ppm-years with BD≤100 ppm, cumulative ppm-years with BD>100 ppm, cumulative ppm-years with STY ≤50 ppm, and cumulative ppm-years with STY>50 ppm) that may be related to the endpoints. These six non-exposure variables and eight exposure variables were available for every worker with time-dependent exposures estimates for the entire period of follow up. (Models did not adjust for smoking because no data were available for this covariate. Not adjusting for a covariate is tantamount to assuming that the effect of the covariate is homogeneous for all workers, and this assumption does not result in deflation or inflation of cancer risk estimates.) Using the individual information and exposure history of every worker, the Cox proportional hazards model was fit including one or more of the six non-exposure variables and the eight exposure variables for each of the endpoints. The SBR study includes individual information on other exposure metrics, besides BD ppm concentrations, that the UAB developed and validated (Macaluso et al., 2004).

The effective ppm concentration (EC) resulting in an excess risk of 1/1,000,000 was identified as an appropriate point of departure (POD) and was used to evaluate risks for cancer mortality. As discussed in the results section, the exposure concentration corresponding to an excess risk of 1/1,000,000 is in the heart of the exposure cumulative ppm-days exposures (more than the 40^{th} percentile) in the SBR study. The unit risk factor (URF_{mle}) corresponding to a linear extrapolation for exposures less than the EC(1/100,000) was also derived ($URF_{mle} = 1\times10^{-5}$/ EC(1/100,000)). The 95% lower confidence limit on the EC(1/100,000) (LEC(1/100,000)) was evaluated along the upper bound on the unit risk factor (URF_{ub}) corresponding to a linear extrapolation for exposures less than the LEC(1/100,000). URFs for a range of excess risks (1/1,000,000 to 1/1,000) for 70- and 80-year lifetimes were evaluated for the endpoints with and without adjustments for the most statistically significant covariates.

Risks (ECs and their 90% confidence intervals) were calculated using life-table methods. These stepwise methods use population-specific survival probabilities and endpoint-specific mortality rates at every age of the population. The methods go through time until a specified target age for risk evaluation. Exposure metrics and hazard rates are evaluated at every point of time through a target age (e.g., 0 to 70 years). The life-

table methods incorporate the survival probabilities, the mortality rates, the calculated exposure metrics and the Cox proportional hazards model fit to the observed data in the evaluation of the hazard rates. The cumulative hazard rates are then used to calculate the endpoint-specific excess risks by the target age.

The life-table method to evaluate ECs and LECs is adapted from the BEIR IV methodology (NRC, 1988) and is described in detail in Sielken and Valdez-Flores (2009a, 2009b). This method has been used by USEPA (2002), TCEQ (2008), and Sielken and Valdez-Flores (2011, 2013, and 2015) in addition to other risk assessments by EPA and other publications by the same authors. The 2017 survival probabilities for the U.S. population used to calculate ECs and LECs were taken from Arias and Xu (2019) while the 2019 cancer-specific mortality rates were extracted from the CDC WONDER http://wonder.cdc.gov on July 2, 2021.

No attempt was made here to estimate risks for cancer incidence rather than cancer mortality. The SBR data includes only cancer mortality information such that models for cancer incidence cannot be derived. Although exposure-response models fit to cancer mortality can be used in conjunction with background cancer incidence rates to purportedly estimate risks of cancer incidence, this practice is unfounded. Models for cancer mortality can be very different than models for cancer incidence. Using models fit to cancer mortality coupled with cancer incidence background hazard rates will certainly result in higher cancer risks, but not necessarily in the correct risk estimates of cancer incidence. If, however, an exposure-response model is used to predict cancer incidence from mortality data, then the correct life-table method adapted from the BEIR IV methodology (NRC, 1988) should be applied as discussed by Sielken and Valdez-Flores (2009b).

B.2.3 Assessment of Total Risk Based on an Aggregate Endpoint

In USEPA's previous assessment for BD (2002), an adjustment of 2 was included in their cancer potency estimate, based upon concerns for potentially underestimating total risk for multiple cancer types (based on observations in rodents) when relying on a single cancer type in humans (leukemia) to estimate cancer potency. As noted above, Sathiakuma et al. (2021a,b) reported associations for leukemia and bladder/urinary cancers with the latest SBR cohort data. One approach used by USEPA to estimate unit risk values for total risk from different cancer types by combining Wald-based estimates, as was done for the cancer potency estimate for ethylene oxide using leukemia and breast cancer risks (USEPA, 2016). However, this approach ignores potential correlations between the cancer endpoints, and requires assumptions of normality that may not be valid. As an alternative, an aggregate endpoint was considered here within the context of the Cox proportional hazards modeling.

To include the possibility of mortality due to leukemia and/or bladder/urinary cancer as a potential regulatory endpoint, an aggregated endpoint that includes leukemia or bladder/urinary cancer is also considered. Though combining leukemia and bladder/urinary cancers is uncertain due to potential differences in mechanism of action,

different weights of evidence (e.g., potential confounding by smoking for bladder cancer), both endpoints are expected to be related to a genotoxic mode of action for BD through the formation of reactive metabolites. The analyses of the aggregate endpoint presented here, which combines endpoints that may have different etiology, were performed as a resource for risk assessors interested in characterizing conservative estimates of total risk in human populations exposed to BD.

B.3 Exposure-Response Modeling

B.3.1 Model Form

The Cox proportional hazards model is used here to fit the most recent SBR study data. Sielken and Valdez-Flores (2011, 2013, and 2015) used the Cox proportional hazards model to estimate the rate ratio (RR) as a function of cumulative exposure to BD. The Cox model assumes that the baseline hazard rate ($\lambda_0(t)$) is a function of time (age) and that the RR, in addition to cumulative exposures, depends on $\lambda_0(t)$ and the effect of multiplicative covariates. The hazard rate ($\lambda(t)$) can then be written as:

$$\lambda(t) = \lambda_0(t) \times CV_{NE} \times CV_{OE} \times RR(BD\ ppm - years)$$

where, CV_{NE} = categorical effect of non-exposure covariates, CV_{OE} = categorical effect of other exposure covariates (i.e., exposures other than cumulative BD ppm-years), and RR(BD ppm-years) = the hazard rate ratio as a continuous function of cumulative BD ppm-years. The Cox proportional hazards model assumes that the hazard rate at any age ($\lambda(t)$) is the product of the baseline hazard rate ($\lambda_0(t)$), the effect on non-exposure covariates (CV_{NE}), the effect of other exposure covariates (CV_{OE}), and the effect of BD ppm-years.

The standard Cox proportional hazards model assumes that the RR is a log-linear function of the cumulative exposure to BD; that is,

$$\ln\big(RR(BD\ ppm - years)\big) = \beta \times (BD\ ppm - years)$$

This log-linear model is preferred because it is approximately linear for low values of cumulative BD ppm-years and is the standard and most widely accepted model for Cox proportional hazards analyses. In order to free the model fitting from parametric assumptions, non-exposure and exposure covariates were treated as categorical variables. Time-dependent covariates were categorized by splitting on the number of cause-specific deaths by cumulative exposure quintiles. Sielken et al. (2007) performed sensitivity analyses on the effect of number of categories and the use of continuous, rather than categorical covariates, using the SBR study.

Under a confidentiality agreement, the UAB provided the authors with anonymized, individual worker exposure histories for all workers with exposure estimates in the SBR study. The individual data were used to fit the Cox proportional hazards model for the endpoints listed in Table B.3. Table B.5 lists the six non-exposure variables and eight

exposure variables included with the SBR epidemiological data set and considered in the analyses presented here.

Six non-exposure variables
1. Age
2. Years Since Hire
3. Calendar Year
4. Sex
5. Race
6. Plant

Nine exposure variables
1. Cumulative BD ppm-years
2. Cumulative STY ppm-years
3. Cumulative number of BD high-intensity tasks (HITs); i.e., tasks with exposures ≥ 100 ppm BD
4. Cumulative number of STY HITs; i.e., tasks with exposures ≥ 50 ppm STY
5. Cumulative exposure to BD ppm-years concentrations ≤ 100 ppm (concentrations > 100 ppm set equal to 100 ppm)
6. Cumulative exposure to BD ppm-years concentrations > 100 ppm (concentrations ≤ 100 ppm set equal to 0 ppm)
7. Cumulative exposure to STY ppm-years concentrations ≤ 50 ppm (concentrations > 50 ppm set equal to 50 ppm)
8. Cumulative exposure to STY ppm-years concentrations > 50 ppm (concentrations ≤ 50 ppm set equal to 0 ppm)

Table B.5. The 2009 update of the SBR study includes individual worker information and exposure history to extract the following non-exposure and exposure variables

B.3.2. Assessment of Covariates

Initially, a mortality model for every endpoint was fit with the RR being a continuous function of the eight exposure variables using Cox proportional hazards, each assessed individually. Table B.6 shows the statistical significance (p-value) of including each of the cumulative exposures as the explanatory variable. The smaller the p-value, the more significant is the impact of the exposure variable in explaining the variability in the observed mortality. The main purpose of Table B.6 was to explore whether any of the eight exposure variables was a better predictor than cumulative BD ppm-years. The table shows that there was at least one exposure variable that predicts the observed data better (has smaller p-value) than cumulative BD ppm-years. The exposure variable that most consistently outperformed cumulative BD ppm-years is the cumulative BD ppm-years concentrations ≤ 100 ppm. However, this exposure metric caps BD exposure concentrations at 100 ppm and leaves out the part of the concentrations exceeding 100 ppm in the workplace. By truncating BD exposures at 100 ppm, this exposure metric does not quantify the magnitude of high exposures to BD, similar to cumulative number

of BD HITs. Thus, cumulative BD ppm-years concentrations ≤ 100 ppm is somewhat related to cumulative number of BD HITs in that both metrics incorporate the number of instances in which BD concentrations exceed 100 ppm, regardless of magnitude. Because cumulative BD ppm-years concentrations ≤ 100 ppm does not include only concentrations of BD below 100 ppm and the SBR study included concentrations above 100 ppm, the variable is not meaningful as a predictive exposure variable. Cumulative BD HITs outperformed cumulative BD ppm-years for leukemia, but cumulative BD ppm-years outperformed BD HITs for bladder/urinary cancers. Leukemia and bladder/urinary cancers had a statistically significant p-value at the 1% significance level for cumulative BD ppm-years while lymphoid leukemia was significant at the 5% significance level. Other endpoints (myeloid leukemia, multiple myeloma, and NHL) did not have statistically significant increase in the likelihood when cumulative BD ppm-years was the exposure variable.

Exposure Variable	Leukemia	Lymphoid Leukemia	Myeloid Leukemia	Multiple Myeloma	NHL	Bladder/ Urinary
BD (cumulative ppm-years)	0.0091**,c	0.0113*,b	0.4114	0.4480	0.7503	0.0035**,b
Cumulative # of BD HITs	0.0043**,b	0.0801	0.0960[a]	0.8612	0.0510[b]	0.2921
BD > 100 ppm (cumulative ppm-years)	0.0309*	0.0234*,c	0.5169	0.3983	0.7393	0.0085**
BD ≤ 100 ppm (cumulative ppm-years)	0.0002**,a	0.0050**,a	0.2141[b]	0.8642	0.0141*,a	0.0021**,a
STY (cumulative ppm-years)	0.0421*	0.0257*	0.8021	0.1219[b]	0.9719	0.0131*
Cumulative # of STY HITs	0.3604	0.4772	0.4449	0.7126	0.5412	0.8049
STY > 50 ppm (cumulative ppm-years)	0.1302	0.0427*	0.8618	0.1104[a]	0.3128	0.0355*
STY ≤ 50 ppm (cumulative ppm-years)	0.0106*	0.0342*	0.3802[c]	0.3301[c]	0.1530[c]	0.0073**,c

1Analyses are based on male and female workers combined and cumulative exposure includes all exposures

[a,b,c]Rank of the maximum log-likelihood for the specified endpoint ([a]Implies the best fitting model, [b]Implies second best, and [c]Implies third best).

Shaded cells have maximum log-likelihoods better than the maximum log-likelihood for cumulative BD ppm-years.

*p-Value for the slope of the log-linear model is less than 0.05 implying that the slope is statistically significantly different than zero at the 5% significance level.

**p-Value for the slope of the log-linear model is less than 0.01 implying that the slope is statistically significantly different than zero at the 1% significance level.

Table B-6. Statistical significance (p-value) of the effect of using one exposure variables in the Cox proportional hazards model as the only explanatory variable.[1]

Table B.7 lists the statistical significance in the improvement of the likelihood of the model after adding one of the non-exposure or one of the categorical exposure covariates to the Cox proportional hazards model for cumulative BD ppm-years. The p-values of the covariate effects that significantly improve the likelihood of the model for each of the endpoints is highlighted. The p-value reflects the change in the likelihood after incorporating the covariate to the model that included only cumulative BD ppm-years as the predictive variable. A smaller p-value implies that the improvement in the likelihood of the model is larger after adjusting of the covariate. Because age is the index variable used in the Cox proportional hazards model, it is implicitly incorporated and does not have to be included as an explicit covariate. Table B.7 shows that, except for sex, none of the other four non-exposure covariates (years since hire, calendar year, race, or plant) improves the likelihood over the model that includes only cumulative BD ppm-years significantly at the 5% significance level and have a positive slope for cumulative BD ppm-years for any of the endpoints studied. In contrast, only one of the exposure covariates (cumulative BD ≤ 100 ppm-years) did not improve the likelihood (at the 1% significance level) over the maximum likelihood of the model that uses cumulative BD ppm-years for any of the six endpoints analyzed. Herein, we used a significance level of 1% to determine whether a covariate significantly improved the likelihood of the model. For leukemia seven of the eight exposure covariates (i.e., all but cumulative exposure to BD ≤ 100 ppm) improved the likelihood with p-values less than 1%. BD Hits is the only exposure covariate that increased the model's likelihood at the 1% significance level for lymphoid leukemia. Although some of the exposure covariates significantly improved the likelihood at the 1% level for myeloid leukemia, multiple myeloma and NHL, these models resulted in an estimated negative relationship between the rate ratio of the endpoint and cumulative BD ppm-years (this is indicated by a negative sign in front of the p-values in Table B.7). For bladder/urinary cancers, none of the exposure covariates improved the likelihood of the model at the 1% significance level. The slope per cumulative BD ppm-year is statistically significantly greater than zero (at the 5% significance level) for leukemia, lymphoid leukemia, and bladder urinary cancer for the model not adjusted for any covariates. The slope is not statistically significant at the 5% significance level for leukemia after adjusting for the most significant covariate (BD HITs). For lymphoid leukemia and bladder/urinary the slope remains statistically significant at the 5% significance level for after adjusting for the most significant covariate (BD HITs and sex, respectively). The slope per cumulative BD ppm-year for myeloid leukemia, multiple myeloma and NHL were not statistically significantly greater than zero at the 5% significance level before or after adjusting for the most significant non-exposure or exposure covariate.

Covariate	Leukemia	Lymphoid Leukemia	Myeloid Leukemia	Multiple Myeloma	NHL	Bladder/ Urinary

Years Since Hire	0.7212	0.3290	0.4306	0.6198	0.9151	0.9161
Calendar Year	0.7787	0.5059	0.9925	0.2764	0.9883	0.9276
Sex	0.0019**	0.1051	0.0024**	0.0416*	0.0339*	0.0009**
Race	0.5446	0.2761	0.9643	-0.0039**	0.9643	0.0395*
Plant	0.4012	0.2064	0.7967	0.5794	0.4820	0.7362
STY (cumulative ppm-years)	0.0090**	0.4226	0.0600	-0.3412	-0.1707	0.1249
Cumulative # of BD HITs	6.0×10^{-7}**	0.0040**	-0.0005**	0.6506	-0.3018	0.0448*
Cumulative # of STY HITs	2.6×10^{-5}**	0.0442*	-0.0008**	0.0602	0.2402	0.0378*
BD ≤ 100 ppm (cumulative ppm-years)	0.0209*	0.0822	0.2945	0.5790	-0.5415	0.0185*
BD > 100 ppm (cumulative ppm-years)	0.0003**	0.0977	-0.0092**	-0.2327	-0.0049**	0.1744
STY ≤ 50 ppm (cumulative ppm-years)	0.0063**	0.2072	-0.1381	0.7996	-0.1980	0.1503
STY > 50 ppm (cumulative ppm-years)	0.0001**	0.0299*	-0.0329*	-0.2266	-0.5438	0.0584

*Statistically significant improvement in the likelihood at the 5% significance level.
**Statistically significant improvement in the likelihood at the 1% significance level.
[1]The lightly shaded cells indicate where significant improvements occur at the 5% significance level.
[2]The moderately shaded cells indicate where significant improvements occur at the 1% significance level.
[3]A negative sign preceding a p-value indicates that the estimated slope for cumulative BD ppm-years is negative.

Table B-7. Statistical significance (p-value) of the effect of adding one of the non-exposure or exposure covariates to the Cox proportional hazards model with the rate ratio being a log-linear function of cumulative BD ppm-years.[1,2,3]

Table B.8 lists the significance of the slope of cumulative BD ppm-years before and after incorporating each of the non-exposure and the categorical exposure covariates. This table highlights (shaded cells) the significance of the slope of cumulative BD ppm-years after incorporating the non-exposure or exposure covariate

that had the most impact (smallest p-value for each endpoint in Table B.7) in the model. Although the cumulative BD ppm-years slope for leukemia was significant (p-value=0.0091) when no covariates were included in the model, the slope is not significant (p-value=0.2794) after adjusting the model for BD HITs. For lymphoid leukemia and bladder/urinary cancers, the slope for cumulative BD ppm-years became less significant (larger p-values) after adjusting the model for the most significant covariate (smallest p-values in Table B.7). The slope for cumulative BD ppm-years for myeloid leukemia, multiple myeloma and NHL were not significantly positive before adjusting for any covariates (p-values=0.4113, 0.4479, and 0.7506, respectively) and the slopes became not significant negative (p-values shown in parentheses indicate the estimate of the slope is negative) after adjusting for the most significant covariate (smallest p-values in Table B.7).

Covariate	Leukemia	Lymphoid Leukemia	Myeloid Leukemia	Multiple Myeloma	NHL	Bladder/ Urinary
No Covariates	0.0091**	0.0113*	0.4113	0.4479	0.7506	0.0035**
Years Since Hire	0.0089**	0.0113*	0.4011	0.4292	0.7390	0.0036**
Calendar Year	0.0096**	0.0119*	0.4201	0.4670	0.7579	0.0036**
Sex	0.0234*	0.0188*	0.6264	0.6087	0.9643	0.0105*
Race	0.0212*	0.0402*	0.4363	(0.9496)	0.7506	0.0005**
Plant	0.0037**	0.0022**	0.3865	0.5564	0.5795	0.0019**
STY (cumulative ppm-years)	0.1223	0.0667	(0.9333)	(0.7733)	(0.3591)	0.0189*
Cumulative # of BD HITs	0.2794	0.0447*	(0.6539)	0.4463	(0.7870)	0.0419*
Cumulative # of STY HITs	0.2231	0.0239*	(0.5771)	0.5125	0.9643	0.0094**
BD ≤100 ppm (cumulative ppm-years)	0.3207	0.3774	0.9333	0.7604	(0.4233)	0.0694
BD > 100 ppm (cumulative ppm-years)	0.3014	0.1399	(0.9643)	(0.3118)	(0.4103)	0.1331

STY ≤ 50 ppm (cumulative ppm-years)	0.1187	0.0745	(0.9748)	0.8129	(0.3655)	0.0283*
STY > 50 ppm (cumulative ppm-years)	0.0989	0.0153*	(0.9436)	(0.6507)	(0.7323	0.0140*

[1]Shaded cells are the p-values of the slope after including the most statistically significant covariate
*Statistically significant improvement in the likelihood at the 5% significance level.
**Statistically significant improvement in the likelihood at the 1% significance level.

Table B-8. Statistical significance (Likelihood Ratio Test p-value) of the slope of cumulative BD ppm-years after adding one of the non-exposure or exposure covariates to the Cox proportional hazards model with the rate ratio being a log-linear function of cumulative BD ppm-years.[1]

The likelihood of the models fit with the most significant covariate did not improve significantly, at the 1% significance level, after adding another covariate for any of the endpoints analyzed. The models with the most significant covariate were investigated after adjusting them for sex, but similar results were obtained with smaller slopes for cumulative BD ppm-years.

B.3.3. Exposure Lag/Windows of Exposure

The best models (i.e., models that had cumulative BD ppm-years as the explanatory variable adjusted for the most significant non-exposure or exposure covariate) were evaluated using lagged cumulative exposures. In addition to excluding the most recent exposures to BD (lag), windows of exposure were considered in Sielken and Valdez-Flores (2015). Windows of exposure exclude not only recent exposures to BD but also exposures that occurred more than y years ago. Thus, a window of exposure with a 5-year lag and excluding exposures that occurred more than 30 years ago, includes only a maximum of 25 years of exposures (i.e., exposure that occurred between 30 years ago and 5 years ago). The EPA's Science Advisory Board (SAB) (1998) recommended consideration of windows of exposure for BD. The SAB suggested that in addition to considering the impact of BD high intensity tasks (HITs), the effect of dose-time relationship be explored. Here, in addition to windows of exposure, we explored cumulative exposures where the concentration and the time of the exposure had different weights, i.e., sum of C^n*t^k, for different values of n and k. These exposure metrics are consistent with SAB's suggestion of considering "a model that assumes a limited effect time (i.e., that leukemia risk during a given year of age is affected largely by the BD exposures received during the previous, say, 20 years, and only slightly or not at all by more distant ones)." The models that adjusted for the most significant covariates were not statistically significantly improved by lagging the cumulative exposure to BD for any of the six endpoints analyzed.

The model for each endpoint with positive exposure-response relationship and adjusted for the most significant covariate in addition to unlagged cumulative BD ppm-years was

evaluated assuming 0, 5, …, 30-year lagged cumulative BD ppm-years. None of the non-zero lagged cumulative BD ppm-years fit the models statistically significantly better than the model with unlagged cumulative BD ppm-years at the 5% significance level. In fact, the likelihood of the model with non-zero lags cumulative BD ppm-years was less than the likelihood of the model with unlagged cumulative BD ppm-years for leukemia, lymphoid leukemia and bladder/urinary cancers. The same models were also evaluated using the cumulative BD ppm-years restricted to the 60, 50, 40, 30, and 20 most recent years and compared with the likelihood of the model that included all the cumulative BD ppm-years. None of the models with restricted exposure duration had a statistically significant likelihood larger than the likelihood of the model that included all BD exposures for any of the six endpoints considered. Because excluding old BD concentrations and excluding recent BD concentrations from the cumulative BD ppm-years did not improve the likelihood of the model's fit to the observed data, no attempt was made to find a window of exposure that only includes BD concentrations that occurred within a window of time.

B.3.4. Shape of Exposure-Response Relationship

Although the standard Cox proportional hazards model assumes a log-linear model of the rate ratio as a function of cumulative exposure, this relationship can take on different forms when the ten Berge (1986) exposure metrics are considered. That is, when exposures of the form sum of $C^n * t^k$, for different values of n and k, the log-linear relationship of the rate ratio as a function of cumulative-exposure can take different shapes that depend on the values of n and k (i.e., potential sub-linear or supra-linear relationships). Using ten Berge (1986) exposure metrics the rate ratio as a function of the exposure metric is given by

$$\ln(RR) = \beta \times (BD\ ppm)^n \times (Duration\ days)^k$$

Figure B.1 shows shapes of the relationship between rate ratios and cumulative hypothetical exposures for different values of n and k of the ten Berge (1986) exposure metrics assuming a constant concentration of BD ppm.

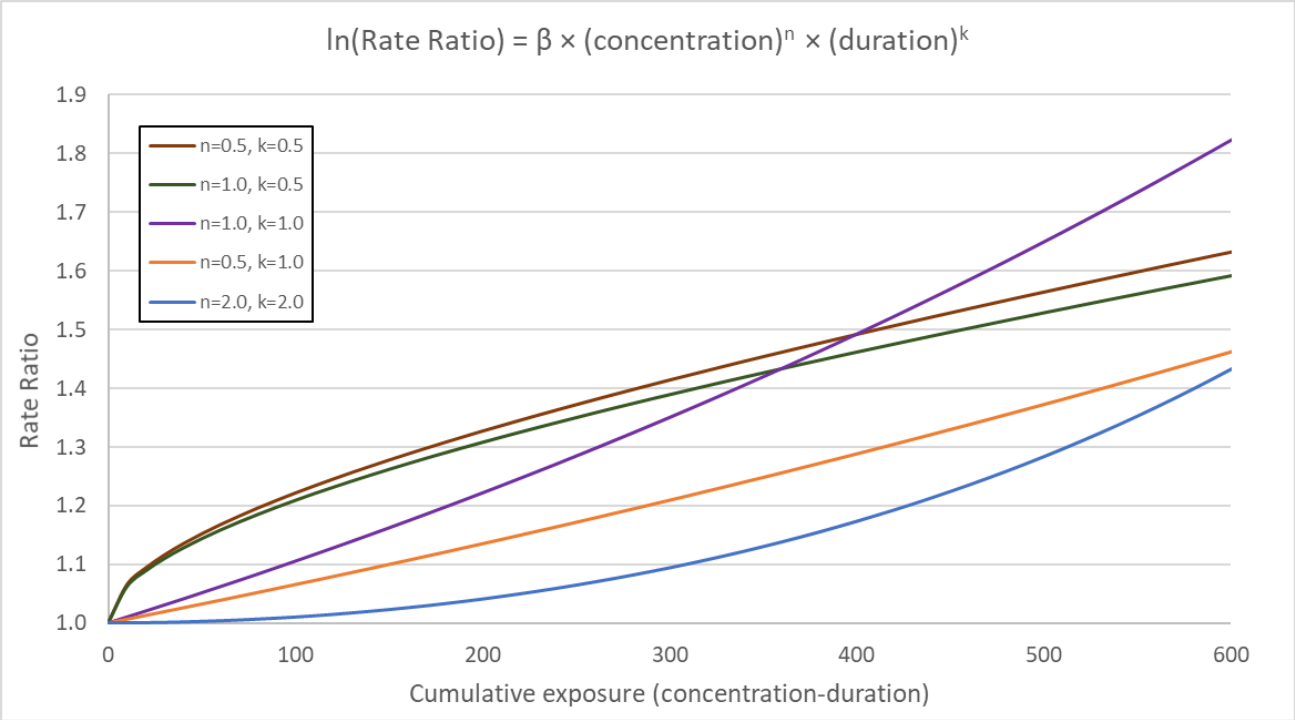


Figure B.1. Hypothetical example of exposure-response shapes of the rate ratio as a function of cumulative exposures for ten Berge (1986) exposure metrics for different values of n and k of the assuming a constant concentration

The Cox proportional hazards models for leukemia were also fit using the exposure metrics $C^n \times T^k$ for values of n and k = 0.1, 0.25, 0.5, 0.75, 1.0, and 2. The deviance of the models decrease with smaller values of n (the exponent of BD ppm concentration). As noted by Sielken and Valdez-Flores (2013) smaller values of n reduce the effect of larger values of BD TWA ppm in cumulative exposures. The impact of n on the deviance disappeared when the models were adjusted for the number of BD HITs. In addition, the parameter for the exposure metrics was statistically indistinguishable from zero when BD HITs was added as a covariate to the models. This lends support to the results discussed earlier that cumulative number of BD HITs is more predictive of leukemia mortality than cumulative BD ppm-years in the SBR study. Here, we present risk characterizations for leukemia of the Cox proportional hazards model with no covariate adjustment and with adjustment for BD HITs and other covariates that significantly improved the model fit.

B.4 Results

Table B.9 reports the maximum likelihood estimates of the excess cancer mortality risk for environmental exposures and occupational exposures to 1 ppm of BD for a lifetime of 70 years (i.e., unit risk values). Environmental exposures assume the individuals are exposed 24 hours a day every day of their lifetime while occupational exposures assume that workers are exposed to 1 ppm of BD for eight hours a day, 240

days a year from 20 to 65 years of age. The table shows the results for the unadjusted models for each endpoint adjustment and for each covariate that significantly (at the 1% significance level) improved the likelihood of the model with only cumulative BD ppm-years. The results in Table B.9 show that the excess risks by age 70 years for the six endpoints included here at an environmental exposure of 1 ppm for the entire lifetime range from 6.7×10^{-8} for NHL to 8.1×10^{-5} for all leukemia. If only the models that adjust for the most significant covariate are considered for each of the endpoints, the range is 0 for several endpoints to 4.9×10^{-5} for bladder/urinary cancer. In Table B.9, the range of excess cancer risks by age 70 years for an occupational exposure to 1 ppm from age 20 to age 65 is 1.4×10^{-8} for NHL to 1.7×10^{-5} for all leukemia. Considering only the models that adjust for the most significant covariate for each endpoint the range of excess cancer mortality risks is 0 for several endpoints to 1.0×10^{-5} for bladder/urinary cancer. Table 10 shows the best estimates of the average environmental BD concentration (EC in ppm) for a lifetime exposure corresponding to different excess risk and ages 70 and 85 years. Although risk estimates for a lifetime of 70 years are more robust and are more consistent with the data, USEPA (2002) estimated risks for an 85-year lifetime. The table shows the results for the models for each endpoint that have a positive slope per BD ppm-year and includes adjustment for each covariate that significantly (at the 1% significance level) improved the likelihood of the model with only cumulative BD ppm-years. The minimum average lifetime concentrations associated with an excess risk of one in a million using the model that adjusts for the most significant covariate is for bladder/urinary cancer with 0.021 and 0.004 ppm for 70 years and 85 years, respectively. The table lists the environmental EC's for excess risks ranging from 1 in 10,000 to 1 in 1,000,000. Table B-11 shows the best estimates of the average occupational BD concentration (EC in ppm) for an occupational exposure from 20 to 65 years of age corresponding to different excess risks and ages 70 and 85 years. The minimum average lifetime concentrations associated with an excess risk of one in ten thousand using the model that adjusts for the most significant covariate is for bladder/urinary cancer with 9.12 and 2.24 ppm for 70 years and 85 years, respectively. The table lists the occupational EC's for excess risks ranging from 1 in 1,000 to 1 in 100,000. Table B.12 lists U.S. age-dependent survival probabilities and age-dependent mortality rates used in the life-table calculations of excess risks and ECs for the six points evaluated.

Endpoint	Covariate[1]	Slope[2] (MLE)	Slope (Std Dev)	Stat. Sig.[3] of Slope	Lag[4]	Added Risk[5] (Unit Risk per ppm)	
						Environ.	Occup.
Leukemia	BD HITs	0.0001316	0.0001079	NS	0	3.7×10^{-5}	7.7×10^{-6}
	STY HITs	0.0001542	0.0001121	NS	0	4.4×10^{-5}	9.1×10^{-6}
	STY>50ppm	0.0002060	0.0001072	NS	0	5.9×10^{-5}	1.2×10^{-5}

	BD>100ppm	0.0001439	0.0001247	NS	0	4.1×10^{-5}	8.5×10^{-6}
	Sex	0.0002483	0.0000879	SS(5%)	0	7.1×10^{-5}	1.5×10^{-5}
	STY≤50ppm	0.0001970	0.0001089	NS	0	5.6×10^{-5}	1.2×10^{-5}
	STYppm-yrs	0.0001953	0.0001091	NS	0	5.6×10^{-5}	1.1×10^{-5}
	None	0.0002808	0.0000838	SS(1%)	0	8.1×10^{-5}	1.7×10^{-5}
Lymphoid Leukemia	BD HITs	0.0003250	0.0001273	SS(5%)	0	2.1×10^{-5}	4.2×10^{-6}
	None	0.0003540	0.0001023	SS(5%)	0	2.3×10^{-5}	4.6×10^{-6}
Myeloid Leukemia	BD HITs	-0.0001297	0.0003170	NS	0	0[6]	0
	STY HITs	-0.0001557	0.0003079	NS	0	0	0
	Sex	0.0001029	0.0001936	NS	0	1.9×10^{-5}	3.9×10^{-6}
	BD>100ppm	-0.0000115	0.0002857	NS	0	0	0
	None	0.0001656	0.0001746	NS	0	3.0×10^{-5}	6.3×10^{-6}
Multiple Myeloma	Race	-0.0000115	0.0002857	NS	0	0	0
	None	0.0001489	0.0001722	NS	0	2.5×10^{-5}	5.4×10^{-6}
NHL	BD>100ppm	-0.0002542	0.0003696	NS	0	0	0
	None	0.0000579	0.0001726	NS	0	6.7×10^{-8}	1.4×10^{-8}
Bladder/ Urinary	Sex	0.0002802	0.0000852	SS(5%)	0	4.9×10^{-5}	1.0×10^{-5}
	None	0.0003159	0.0000813	SS(1%)	0	5.5×10^{-5}	1.2×10^{-5}

[1]Covariate is a non-exposure or exposure covariate that results in a statistically significant (at the 1% significance level) increase in the maximum likelihood over the maximum likelihood for the model with only cumulative BD ppm-years. Covariates are listed in the order from most to least significant

improvement. (Adjusting for Sex as another covariate, resulted in smaller slope estimates for BD ppm-years: data not shown.)
[2]Slope is the coefficient of cumulative BD ppm-years in the Cox model.
[3]SS (1%) implies that the slope is statistically significantly different than zero (at the 1% significance level); SS (5%) implies that the slope is statistically significantly different than zero (at the 5% significance level); NS implies that the slope is not statistically significantly different than zero (at the 5% significance level). Based on likelihood ratio test.
[4]Lag in years. Statistically significant (at the 1% significance level) improvement in the maximum likelihood.
[5]Environmental exposure corresponds to the persons being exposed continuously from birth until the end of calculations (70 years). Occupational exposure corresponds to 45 potential years of work with the persons being exposed between 20 and 65 years of age. Added risks are calculated using life-table methodology with 2019 U.S. mortality rates and 2017 U.S. survival probabilities.
[6]An added risk equal to 0 indicates that the slope per cumulative occupational BD ppm-year was non-positive, resulting in an exposure–response relationship that would not estimate a positive added risk for any positive exposure to BD.

Table B.9. Added risk by age 70 years for an environmental and occupational BD exposure concentration of 1.0 ppm using the maximum likelihood estimate of the Cox proportional hazards log-linear models: Models with maximum log-likelihoods statistically significantly (at the 1% significance level) improved over the maximum log-likelihood for the model with exposure characterized only by cumulative BD ppm-years are shaded; Models for the same endpoint are ordered with the largest maximum log-likelihood first and the smallest maximum log-likelihood last. This table includes only models with positive slopes.

Environmental							
Response	Covariate	Average BD concentration (ppm)					
		by age 70 years			by age 85 years		
		Added risk			Added risk		
		1 in 1,000,000 or 0.000001	1 in 100,000 or 0.00001	1 in 10,000 or 0.0001	1 in 1,000,000 or 0.000001	1 in 100,000 or 0.00001	1 in 10,000 or 0.0001
All leukemia	Cumulative # of BD HITs	0.027	0.270	2.629	0.007	0.070	0.689
	Cumulative # of STY HITs	0.023	0.231	2.244	0.006	0.059	0.588
	STY > 50 ppm (cumulative ppm-years)	0.017	0.173	1.679	0.004	0.044	0.440
	BD > 100 ppm (cumulative ppm-years)	0.025	0.247	2.404	0.006	0.064	0.630
	Sex	0.014	0.143	1.393	0.004	0.037	0.365
	STY ≤ 50 ppm (cumulative ppm-years)	0.018	0.181	1.756	0.005	0.046	0.460
	STY (cumulative ppm-years)	0.018	0.182	1.771	0.005	0.047	0.464
	None	0.013	0.127	1.232	0.003	0.033	0.323
Lymphoid Leukemia	Cumulative # of BD HITs	0.048	0.477	4.261	0.011	0.114	1.100
	None	0.044	0.438	3.912	0.011	0.105	1.010
Myeloid Leukemia	Sex	0.055	0.543	5.191	0.015	0.149	1.464
	None	0.034	0.337	3.226	0.009	0.092	0.910
Multiple Myeloma	None	0.040	0.403	3.833	0.010	0.101	0.994
NHL	None	13.969	90.167	265.097	3.264	27.622	126.958
Bladder/Urinary	Sex	0.021	0.210	2.000	0.004	0.044	0.438
	None	0.019	0.186	1.774	0.004	0.039	0.389

[1]The results in the first row of each response are for the model after adjusting for the most statistically significant covariate

Table B-10. Estimates of the average environmental BD exposure concentrations (ppm) for a lifetime of exposure (starting at birth) corresponding to specified excess risks and specified ages (70 and 85 years)[1]

Occupational							
Response	Covariate	Average BD concentration (ppm)					
		by age 70 years			by age 85 years		
		Added risk			Added risk		
		1 in 100,000 or 0.00001	1 in 10,000 or 0.0001	1 in 1,000 or 0.001	1 in 100,000 or 0.00001	1 in 10,000 or 0.0001	1 in 1,000 or 0.001
All leukemia	Cumulative # of BD HITs	1.29	12.55	98.70	0.36	3.54	32.50
	Cumulative # of STY HITs	1.10	10.71	84.25	0.30	3.02	27.74
	STY > 50 ppm (cumulative ppm-years)	0.83	8.02	63.05	0.23	2.26	20.76
	BD > 100 ppm (cumulative ppm-years)	1.18	11.48	90.27	0.33	3.23	29.72
	Sex	0.69	6.65	52.31	0.19	1.87	17.22
	STY ≤ 50 ppm (cumulative ppm-years)	0.86	8.38	65.94	0.24	2.36	21.71
	STY (cumulative ppm-years)	0.87	8.46	66.51	0.24	2.38	21.90
	None	0.61	5.88	46.26	0.17	1.66	15.23
Lymphoid Leukemia	Cumulative # of BD HITs	2.35	20.78	107.90	0.59	5.72	43.20
	None	2.16	19.08	99.06	0.55	5.26	39.66
Myeloid Leukemia	Sex	2.57	24.51	175.09	0.76	7.46	65.34
	None	1.59	15.23	108.79	0.47	4.64	40.60
Multiple Myeloma	None	1.84	17.52	123.02	0.51	5.04	44.18
NHL	None	424.21	1,227.42	2,152.60	143.45	663.43	1,492.32
Bladder/Urinary	Sex	0.96	9.12	64.28	0.23	2.24	20.01
	None	0.85	8.09	57.02	0.20	1.99	17.75

Table B-11. Estimates of the average occupational BD exposure concentrations (ppm) for 45 years of exposure (starting at age 20 years) corresponding to specified excess risks and specified ages (70 and 85 years).

Age range (years)	Survival (probability survival past age range)	Mortality rate per 100,000 persons / ICD10 Codes					
		Leukemia C91-C95	Lymphoid Leukemia C91.0-C91.5, C91.7, C91.9	Myeloid Leukemia C92.0-C92.5, C92.7, C92.9	Multiple Myeloma C88.0-C88.3, C88.7, C88.9, C90.0-C90.2	NHL C96.1-C96.3, C96.7, C96.9	Bladder/ Urinary C66-C67, C68.0, C68.1, C68.8, C68.9
<1	1.00000	0.3436	0.1850	0.1057	0.0000	0.0000	0.0000
1 to 4	0.99422	0.5698	0.2279	0.2533	0.0000	0.0063	0.0000
5 to 9	0.99326	0.3961	0.2179	0.1436	0.0050	0.0000	0.0000
10 to 14	0.99268	0.5241	0.2789	0.1875	0.0000	0.0000	0.0048
15 to 19	0.99191	0.5367	0.2850	0.1472	0.0000	0.0000	0.0000
20 to 24	0.98937	0.8459	0.3652	0.3652	0.0046	0.0046	0.0046
25 to 29	0.98466	0.7997	0.2722	0.3913	0.0128	0.0043	0.0255
30 to 34	0.97872	0.7712	0.2719	0.4012	0.0134	0.0045	0.0401
35 to 39	0.97163	1.2099	0.3266	0.7361	0.1150	0.0000	0.1058
40 to 44	0.96321	1.3704	0.3162	0.8533	0.3614	0.0050	0.2811
45 to 49	0.95275	2.0051	0.4412	1.3237	0.7256	0.0147	0.6324
50 to 54	0.93797	2.8666	0.5470	1.9485	1.6311	0.0195	1.3185
55 to 59	0.91538	4.9595	1.1107	3.1448	3.0305	0.0229	2.9300
60 to 64	0.88226	7.8119	1.7209	4.9924	5.4931	0.0243	5.9890
65 to 69	0.83696	13.5262	2.9218	8.7711	9.1607	0.0516	9.5445
70 to 74	0.77697	22.6968	4.8188	14.4136	14.4706	0.0998	16.4309
75 to 79	0.69418	37.1504	9.0234	22.1079	22.6777	0.1347	28.1269
80 to 84	0.57839	55.4201	15.4657	30.2982	33.3692	0.3483	45.9064
85+	0.42382	55.4201	15.4657	30.2982	33.3692	0.3483	45.9064

[a]Survival probabilities: Arias, Elizabeth and Jiaquan Xu. National Vital Statistics Report. Volume 68, Number 7. Hyattsville, MD: National Center for Health Statistics, June 24, 2019. https://www.cdc.gov/nchs/data/nvsr/nvsr68/nvsr68_07-508.pdf

[b]Mortality rates: http://wonder.cdc.gov accessed on 7/02/21

Table B-12. U.S. total population (male, female, and all races combined) 2017 survival rates[a] and 2019 mortality rates[b]

Endpoint	Covariate[1]	Slope[2] (MLE)	Slope (Std Dev)	Stat. Sig.[3] of Slope	Lag[4]	Average Environmental Concentration (ppm)[5]
						EC (LEC, UEC)
Model not adjusted for the effect of covariates						
Leukemia	None	0.0002808	0.0000838	SS(1%)	0	0.01271 (0.00853, 0.02497)

Bladder/ Urinary	None	0.0003159	0.0000813	SS(1%)	0	0.01873 (0.01315, 0.03247)
Aggregate (Leukemia or Bladder/ Urinary)	None	0.0002991	0.0000583	SS(1%)	0	0.00745 (0.00564, 0.01096)
Model adjusted for statistically significant covariates						
Leukemia	BD HITs	0.0001316	0.0001079	NS	0	0.02712 (0.01155, n/a[6])
Bladder/ Urinary	Sex	0.0002802	0.0000852	SS(5%)	0	0.02111 (0.01407, 0.42236)
Aggregate (Leukemia or Bladder/ Urinary)	BD HITs and Sex	0.0001726	0.0000725	SS(5%)	0	0.01291 (0.00763, 0.04177)

[1]Covariate is a non-exposure or exposure covariate that results in a statistically significant (at the 1% significance level) increase in the maximum likelihood over the maximum likelihood for the model with only cumulative BD ppm-years. Covariates are listed in the order from most to least significant improvement. (Adjusting for Sex as another covariate, resulted in smaller slope estimates for BD ppm-years: data not shown.)
[2]Slope is the coefficient of cumulative BD ppm-years in the Cox model.
[3]SS (1%) implies that the slope is statistically significantly different than zero (at the 1% significance level); SS (5%) implies that the slope is statistically significantly different than zero (at the 5% significance level); NS implies that the slope is not statistically significantly different than zero (at the 5% significance level). Based on likelihood ratio test.
[4]Lag in years. Statistically significant (at the 1% significance level) improvement in the maximum likelihood.
[5]Environmental exposure corresponds to the persons being exposed continuously from birth until the end of calculations (70 years). Added risks are calculated using life-table methodology with 2019 U.S. mortality rates and 2017 U.S. survival probabilities.
[6]n/a means that the upper bound of the EC cannot be estimated because the lower bound on the slope for BD ppm-years is zero or negative

Table B.13. Estimates of the average environmental BD exposure concentrations (ppm) for a lifetime of exposure (starting at birth) corresponding to an excess risks of 1 in a million by age 70 years using the maximum likelihood estimate (EC) of the Cox proportional hazards log-linear models and its 95% lower and upper confidence limits (LEC, UEC): Model with BD ppm-years as the predictor variable with no covariates and with statistically significant covariates for leukemia, bladder/urinary and the aggregate (leukemia or bladder/urinary cancer).

Environmental concentrations of BD in air corresponding to an excess risk of 1 in a million are summarized in Table B.13. For comparison purposes, Sielken and Valdez-Flores (2015) reported excess risks and ECs for leukemia based on male workers of the 2000 SBR study. The environmental risks of leukemia for an exposure to 1 ppm BD concentration with the model that adjust for BD HITs was reported as 8.2×10^{-5}, which is

more than two-fold greater than the excess risk of 3.7×10^{-5} that we obtained with the most recent SBR data. Correspondingly the EC for 1 in a million reported by Sielken and Valdez-Flores (2015) is 0.012 ppm compared with 0.027 ppm for the same endpoint and model based on the most recent SBR data.

The ECs and LECs were calculated for an excess risk of one in a million. Oftentimes, the ECs or LECs are defined as a point of departure for linear extrapolation from the concentration corresponding to the EC or LEC down to zero concentration. In animal studies, this POD is usually defined as the dose where the models predict approximately a 10% increase in the response rate and such that the dose for the POD is within the experimental doses (USEPA, 2005). Epidemiological studies, however, would hardly have exposure-response data where a 10% increase in the response rate is observed. So, a POD is better defined, in these cases, by a dose that is within the range of the dose metric used in the exposure-response model fitted to the observed data. Here cumulative exposure BD ppm-years was used as the dose metric for exposure-response modeling. The distribution of cumulative BD ppm-years at the end of the follow up period is given in Figure B.2. The figure indicates that the cumulative occupational BD ppm-years for 34% of the workers was zero. Table B.4 lists selected statistics of the distribution of cumulative BD ppm-years for workers in the most recent update of the SBR study. The 75-th and 95-th percentile of the cumulative BD ppm-years in the most recent update of the SBR study are 87 and 561 ppm-years and it never exceeds 9,269 ppm-years. The cumulative BD ppm-years is occupational; that means it is accumulated only over working hours and days. In other words, an environmental exposure of 0.013 ppm ($EC_{0.000001}$) over a period of 70 years is equivalent to approximately 2.75 cumulative BD ppm-years ($0.013\times(20/10)\times(365/240)\times70$) of occupational exposure, which corresponds to the 41.4-th percentile of the distribution of cumulative BD ppm-years in the most recent update of the SBR study. In any case, we recommend that the best estimate for a POD for environmental BD concentration (i.e., estimated using the maximum likelihood estimate or EC_{bmr}) would not exceed 2.6 ppm, which is equivalent to 95-th percentile (561 cumulative BD ppm-years) of occupational BD ppm-years exposures in the SBR study. In contrast, any BD concentration above zero guarantees that more than one third of the workers in the SBR study had exposures below the POD. In summary, we recommend that, for models fit to the most recent update of the SBR study, the benchmark risk (BMR) be the point of departure (EC_{BMR}) such that the resulting EC_{BMR} is less than 2.6 ppm.

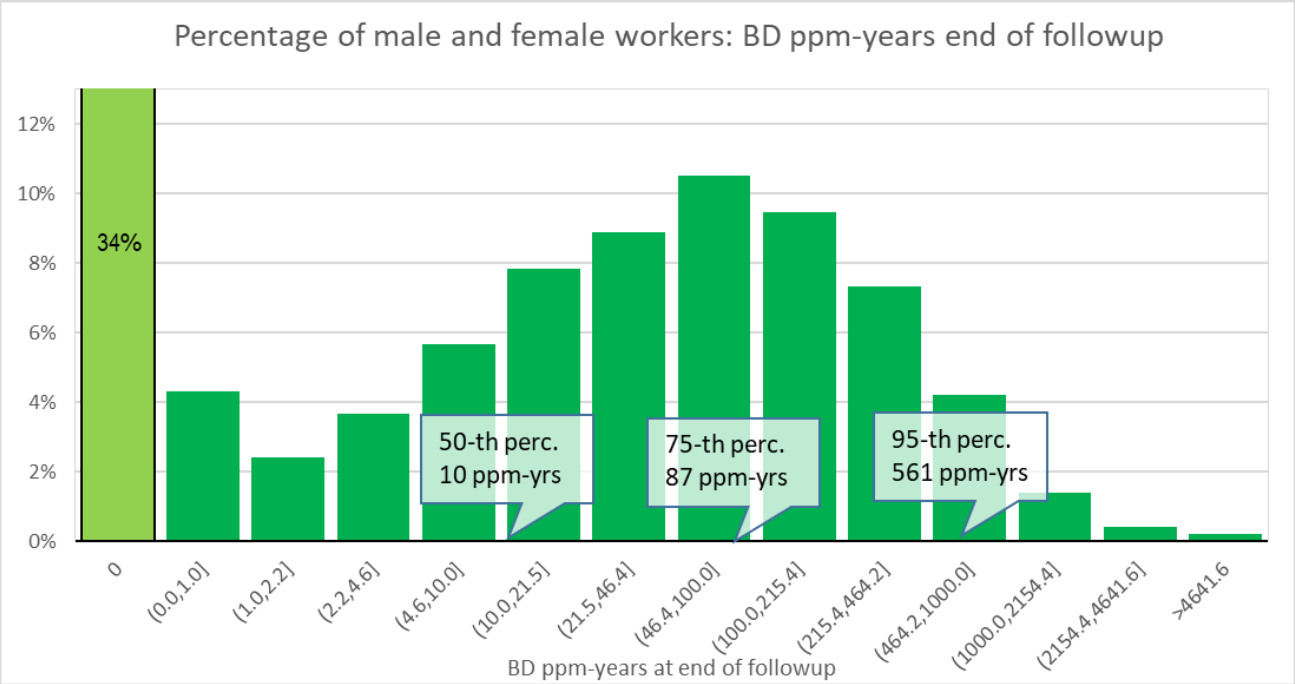


Figure B.2. Distribution of the cumulative BD ppm-years at the end of follow up for all workers included in the model fitting of the most recent update of the SBR study

In order to present a range of alternative risk estimates, the URFs for the two endpoints with the highest risks (leukemia and bladder/urinary) are presented in Table B.14. The URFs in Table B.14 are based on the 95% lower confidence limits of ECs (LECs) for excess risks ranging from 1 in a million to 1 in a thousand and for 70- and 85-year lifetimes. The ratio of the URFs based on an excess risk of 1/1,000 to those based on an excess risk of 1/1,000,000 are less than 1.5 for a 70-year lifetime and less than 1.2 for an 85-year lifetime. This small ratio of URFs at low and higher excess risks is an indication that the models are essentially linear at low cumulative exposures. It is also noteworthy that all, but one of the ECs for an excess risk of 1/1,000, exceeds the 95th percentile of the distribution of occupational BD ppm-years in the SBR study and for some endpoints listed in Table B.14 exceed the maximum occupational exposure (BD ppm-years) in the SBR study. The one instance where the EC for 1/1,000 excess risk does not exceed the 95th percentile of BD ppm-years it exceeds the 75-th percentile. Although URFs based on excess risks 1/1,000 are presented in Table B.14 for the sake of completeness, URFs based on excess risks greater than or equal to 1/1,000 are unreliable because they result in ECs larger than cumulative exposure concentrations observed in the SBR study (i.e., require extrapolating upwards from the range of observation).

Endpoint	Covariate	70-year lifetime				85-year lifetime			
		0.000001	0.00001	0.0001	0.001	0.000001	0.00001	0.0001	0.001
Model not adjusted for the effect of covariates									
Leukemia	None	1.17×10^{-4}	1.18×10^{-4}	1.21×10^{-4}	1.52×10^{-4}	4.57×10^{-4}	4.58×10^{-4}	4.62×10^{-4}	5.03×10^{-4}
Bladder/ Urinary	None	7.60×10^{-5}	7.64×10^{-5}	8.02×10^{-5}	1.14×10^{-4}	3.61×10^{-4}	3.62×10^{-4}	3.66×10^{-4}	4.11×10^{-4}
Aggregate (Leukemia or Bladder/Urinary)	None	1.77×10^{-4}	1.78×10^{-4}	1.81×10^{-4}	2.12×10^{-4}	7.47×10^{-4}	7.47×10^{-4}	7.51×10^{-4}	7.90×10^{-4}
Model adjusted for statistically significant covariates									
Leukemia	BD HITs	8.66×10^{-5}	8.68×10^{-5}	8.93×10^{-5}	1.12×10^{-4}	3.38×10^{-4}	3.38×10^{-4}	3.41×10^{-4}	3.71×10^{-4}
Bladder/ Urinary	Sex	7.11×10^{-5}	7.14×10^{-5}	7.50×10^{-5}	1.06×10^{-4}	3.38×10^{-4}	3.38×10^{-4}	3.42×10^{-4}	3.84×10^{-4}
Aggregate (Leukemia or Bladder/ Urinary)	BD HITs and Sex	1.31×10^{-4}	1.31×10^{-4}	1.34×10^{-4}	1.57×10^{-4}	5.52×10^{-4}	5.52×10^{-4}	5.55×10^{-4}	5.84×10^{-4}

Table B-14. Unit risk estimates per unit of environmental BD exposure concentrations (ppm) for a lifetime of exposure (starting at birth) based on a range of excess risks (1 in 1,000 to 1 in a million) by age 70 and 85 years using the 95% lower confidence limit on the EC (LEC), Model with BD ppm-years as the predictor variable with no covariates and with statistically significant covariates for leukemia, bladder/urinary and the aggregate leukemia or bladder/urinary cancer

Total Risk Based on an Aggregate Endpoint

The aggregate endpoint of leukemia or bladder/urinary cancer was also evaluated, resulting in estimations for the environmental concentration of BD in air corresponding to an excess risk of 1 in a million (Table B.13) and corresponding URF values (Table B.14). These two endpoints were considered because they, individually, result in the strongest exposure response relationship with cumulative BD ppm-years. The models fit to each single endpoint (unadjusted or adjusted for covariates) had slopes that were not statistically significantly different at the 5% significance level. That is, the exposure-response relationships between leukemia and BD ppm-years and between bladder/urine cancer and BD ppm-years were not statistically significantly different and could be combined in a model that considered the aggregate response of leukemia and/or bladder/urinary cancer. Inspection of the unit risk values for the aggregate endpoint compared to leukemia alone suggests that USEPA's application of an adjustment factor of 2 in its previous assessment for BD was slightly too large. For example, for a 70-year lifetime and a BMR of 0.000001, the ratio of URF values for the aggregate endpoint to leukemia alone is approximately 1.5 (1.77×10^{-4} / 1.17×10^{-4}). Fitting a single model to an aggregated endpoint, in addition to increasing the statistical power of the model, may be useful for risk assessors seeking a conservative characterization of total risk. Use of the URF based on the aggregate endpoint may be viewed as conservative since it includes an assumption that the statistical association between cumulative BD exposure and bladder/urinary cancer is causal, and ignores the contribution of potential confounding factors (e.g., smoking) for this endpoint in SBR workers.

B.5 Discussion and Conclusion

Epidemiological data and exposure-response modeling have been greatly updated and improved since the last EPA's assessments (1998, 2002), which were based on Poisson regression and an outdated version of the SBR study with considerably fewer workers, fewer years of follow up, fewer cancer deaths, fewer overall deaths, and less precise exposure estimates. Sielken and Valdez-Flores (2011, 2013, and 2015) and TCEQ (2008) used an updated SBR study with more years of follow up and the Cox proportional hazards model that controls for the effect of age better than the Poisson regression model.

While TCEQ (2008) did not control for the effect of covariates in the exposure-response model and assumed that BD ppm-years was the only effect on leukemia, Sielken and Valdez-Flores (2011, 2013, 2015) considered models that adjusted for statistically significant covariates, in addition to BD ppm-years. Sielken and Valdez-Flores used the likelihood ratio test to determine whether a non-exposure or exposure covariate made a statistically significant improvement in the model fit to the epidemiological data. Here, this latter approach has been followed, so that the most parsimonious model that explains the variability observed in the SBR study is considered as the most parsimonious model that should be used for risk assessment. The analyses in this paper, however, use the most recent SBR study data which is considerably richer than the SBR study data used by Sielken and Valdez-Flores (2011,

2013, and 2015). In addition, this paper confirmed what Sathiakumar et al. (2021) had reported; namely, in addition to leukemia mortality, bladder/urinary cancer was statistically significantly related to BD ppm-years. Although, like Sathiakumar et al. (2021), no additional analyses were conducted to ascertain the impact of uncontrolled confounding by smoking on the association of BD exposure and bladder/urinary cancer. As such, no analysis of causality is provided or implied

A novel approach was taken in considering an aggregate cancer response (leukemia or bladder cancer) within the context of Cox proportional hazards model, for the purpose of providing a characterization of total risk in human populations exposed to BD. This approach is offered as an alternative to the inclusion of an adjustment factor (as done USEPA's 2002 assessment for BD) or post-hoc combination of individual cancer types (as done in USEPA's 2016 assessment for ethylene oxide). Use of BD cancer potency estimates based on this aggregate endpoint may be viewed as conservative since includes an assumption that the statistical association between cumulative BD exposure and bladder/urinary cancer is causal, and ignores the contribution of potential confounding factors (e.g., smoking) for this endpoint in SBR workers.

Subpopulations of people may be considered sensitive to the potential carcinogenic effects of BD due to toxicokinetic and/or toxicodynamic factors.

- Toxicokinetic Factors - With respect to toxicokinetics, the mode of action for BD's carcinogenic action involves metabolic activation to reactive epoxides (Albertini et al., 2010). Blood and urinary biomarker data for BD, most of which has been collected since USEPA's 2002 assessment, can be used to characterize human variation in metabolism due to: (1) gender differences; (2) ethnicity differences; and (3) genetic polymorphisms. Gender differences have been reported for exposed men and women with respect to hemoglobin adducts (Vacek et al., 2010) and urinary biomarkers (Kotapati et al., 2015). When expressed on a per mg/m^3 BD exposure basis, these differences are approximately 2-fold (females < males). Ethnicity differences have been reported for urinary biomarkers for BD metabolites (Park et al., 2014; Bouldry et al., 2017). In addition, ethnic differences for urinary excretion of repaired DNA adducts (EB-GII) have been reported (Sangaraju et al., 2017; Jokipii Krueger et al., 2020). Differences across ethnic groups are generally up to 2- to 3-fold. Some of the ethnicity differences in BD biomarkers may be related to known genetic polymorphisms across ethnic groups. In vitro studies have shown that human cell lines with differing status in glutathione-S-transferase (GST-T1) differ in sensitivity to EB (GSTT1- cells exhibiting greater sensitivity than GSTT1+ cells; Degner et al., 2020). The effects of genetic polymorphisms for various enzyme systems (P450, GST, EH) alone and combined were assessed for THBVal levels. Specific polymorphisms (particularly for GSTT1) showed significant effects on THBVal levels (Fustinoni et al., 2002). THBVal levels across different metabolism groups (i.e., combinations of genetic polymorphisms) were found to be generally within a factor of 2 of the overall mean. The weight of evidence from available biomarker studies for BD

suggests that human variation based on toxicokinetic (TK) factors is likely near or below the default uncertainty factor for intraspecies variation (i.e., UFtk≲3). Because the SBR cohort is large, and includes female workers, sensitive subpopulations due to toxicokinetic factors are likely to be represented in the worker population.

- Toxicodynamic Factors – Because BD is metabolized to reactive epoxides capable of producing genotoxic events, conditions and disease states associated reduced repair of DNA damage are expected to be potentially sensitive to the carcinogenic effects of BD. For example, sensitivity to BD metabolite and clastogen, 1,2,3,4-diepoxybutane (DEB), is specifically used in the diagnosis of Fanconi's anemia (Auerbach, 2015). Such conditions are relatively rare and are accompanied by health conditions (bone marrow failure, cancer, early mortality) such that they are not expected to be represented in a healthy worker population.

As a matter of policy, genotoxic chemicals such as BD are expected to pose an increased risk when exposures occur early in life, a time period that is not directly covered by data from occupational cohorts (USEPA, 2005). Some evidence is available for BD that suggests early-life exposures are not associated with increased risk. For example, BD is metabolically activated to epoxide metabolites by cytochrome P450, principally CYP2E1. Based on the ontogenesis of CYP2E1 activity (Hines, 2007; Johnsrud et al., 2003), metabolic activation of BD is expected to be much lower in neonates, infants, and children when compared to adults. In additional, acute cancer bioassays conducted for BD in mice indicate that single, high exposures relatively early in life do not initiate tumors over the course of their lifetime (Bucher et al., 1993). For these reasons, application of age-dependent adjustment factors (ADAF) may not be required to ensure the protection of human health. TCEQ (2008) also discusses why the ADAF are unnecessary because "children are not more susceptible to chemical leukemogenesis than adults" (Levine and Bloomfield, 1992; Pyatt et al., 2005, 2007). In addition, TCEQ (2008) showed that when the ADAF's are appropriately incorporated in the life-table calculations of BD risks according to USEPA (2002) guidelines the BD cancer risk estimates with ADAFs are only about 1% larger. TCEQ (2008) implementation of ADAFs is different to what USEPA (2002) used in their evaluation for BD, which generally results in increased potency by a factor of approximately 1.7-fold for lifetime exposures (USEPA, 2005).

URF values presented herein are not adjusted for early-life exposures to BD as suggested in USEPA (2005) report. EPA's evaluations usually apply those adjustments by multiplying risks by a factor that is calculated based on the age of the risk calculation. In contrast, Sielken and Valdez-Flores (2009a) suggest a modified life-table analysis to incorporate EPA's proposed ADAFs according to the interpretation in USEPA (2005) guidance. Because applying ADAF factors is a policy decision and regulatory agencies usually do this adjustment a-posteriori, they were not included in the results presented here.

For the general population, exposures to BD are expected to be low. However, some subpopulations are expected to have higher exposures to BD than the general population. In addition, users of tobacco products may experience higher exposures to BD. For example, Nieto et al. (2021) reported that the median urinary excretion of a BD biomarker (N-acetyl-S-(4-hydroxy-2-buten-1-yl)-L-cysteine) was approximately 8-fold higher in smokers compared to non-smokers (31.5 vs. 4.11 ug/g creatinine) and was highly correlated with the number of cigarettes smoked per day.

Models fit to the SBR study did not adjust for smoking because these data were not available. Not adjusting for smoking is equivalent to assuming that the probability and intensity of smoking was the same for all workers in the SBR study and that BD was the causative agent. However, a recent study by Hsu et al. (2019) indicates that hourly workers tend to smoke more than salaried workers. In addition, hourly workers usually perform jobs where exposures are higher. If Hsu et al. (2019) findings hold for the SBR study cohort, the results presented here are biased high; that is, slopes of models unadjusted for smoking are steeper than what they would be if the models adjusted for smoking. In other words, the unadjusted models explain the increases in cancer mortality without accounting for the effect that smoking is having in the increased cancer mortalities in highly exposed workers.

The SBR study is the best available epidemiological data for exposure-response modeling. This study is backed by more than 17 papers published on this data by the UAB scientists that maintain this cohort. The USEPA (2002) and Health Canada (2000) used the first SBR study in evaluating BD. IARC (2008 and 2012) relied on the SBR study for evaluation of BD risks. The Health Effects Institute (HEI, Delzell et al. 2006) performed an audit of the study. HEI's main concern with the SBR study data was that the uncertainty associated with the estimation of worker's exposures. Sathiakumar et al. (2007) published an assessment of the exposure concentrations estimates and measurements of BD exposures for the SBR cohort. The findings indicated that estimated BD concentrations between 1970 and 1984 were lower than measured BD concentrations by a factor of about two and estimated BD concentrations after 1984 were about three times larger than measured BD concentrations. TCEQ (2008) reported the findings of the effect on the uncertainty of exposure estimates. TCEQ found that applying a correction factor to the exposures in the SBR study to align with Sathiakumar et al. (2007) validation findings, resulted in less conservative estimates of risk (Section 4.2.5.3 in TCEQ 2008). TCEQ furthered their sensitivity analyses by using the 5^{th} and 95^{th} percentile of the BD exposure estimates that Macaluso et al. (2004) had developed. The findings of this latest analyses (Appendix 7 in TCEQ 2008) indicate that using the 5^{th} percentile of the BD concentration distribution as opposed to the mean, results in approximately a two-fold increase in the risk estimates. In contrast, using the 95^{th} percentile of the BD concentration distribution as opposed to the mean, results in approximately a two-fold decrease in the risk estimates.

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