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Provisional Peer-Reviewed Toxicity Values for

The Aromatic Medium Carbon Range Total Petroleum Hydrocarbon (TPH) Fraction (various CASRNs)

U.S. EPA Office of Research and Development
Center for Public Health and Environmental Assessment

United States Environmental Protection Agency

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https://www.epa.gov/pprtv

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Center for Public Health and Environmental Assessment
Office of Research and Development
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Questions regarding the content of this PPRTV assessment should be directed to the U.S. EPA Office of Research and Development (ORD) Center for Public Health and Environmental Assessment (CPHEA) website at https://ecomments.epa.gov/pprtv.

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COMMONLY USED ABBREVIATIONS AND ACRONYMS

α2u-g	alpha 2u-globulin
ACGIH	American Conference of Governmental Industrial Hygienists
AIC	Akaike's information criterion
ALD	approximate lethal dosage
ALT	alanine aminotransferase
AR	androgen receptor
AST	aspartate aminotransferase
atm	atmosphere
ATSDR	Agency for Toxic Substances and Disease Registry
BMC	benchmark concentration
BMCL	benchmark concentration lower confidence limit
BMD	benchmark dose
BMDL	benchmark dose lower confidence limit
BMDS	Benchmark Dose Software
BMR	benchmark response
BUN	blood urea nitrogen
BW	body weight
C#	number of carbon atoms contained in a molecule
CA	chromosomal aberration
CAS	Chemical Abstracts Service
CASRN	Chemical Abstracts Service registry number
CBI	covalent binding index
CHO	Chinese hamster ovary (cell line cells)
CL	confidence limit
CNS	central nervous system
CPHEA	Center for Public Health and Environmental Assessment
CPN	chronic progressive nephropathy
CYP450	cytochrome P450
DAF	dosimetric adjustment factor
DEN	diethylnitrosamine
DMSO	dimethylsulfoxide
DNA	deoxyribonucleic acid
EC	equivalent carbonEPA Environmental Protection Agency
ER	estrogen receptor
FDA	Food and Drug Administration
FEV_1	forced expiratory volume of 1 second
GD	gestation day
GDH	glutamate dehydrogenase
GGT	γ-glutamyl transferase
GSH	glutathione
GST	glutathione-*S*-transferase
Hb/g-A	animal blood-gas partition coefficient
Hb/g-H	human blood-gas partition coefficient
HEC	human equivalent concentration
HED	human equivalent dose
i.p.	intraperitoneal
IRIS	Integrated Risk Information System
IVF	in vitro fertilization
LC_{50}	median lethal concentration
LD_{50}	median lethal dose
LOAEL	lowest-observed-adverse-effect level
MN	micronuclei
MNPCE	micronucleated polychromatic erythrocyte
MOA	mode of action
MTD	maximum tolerated dose
NAG	*N*-acetyl-β-D-glucosaminidase
NCI	National Cancer Institute
NOAEL	no-observed-adverse-effect level
NTP	National Toxicology Program
NZW	New Zealand White (rabbit breed)
OCT	ornithine carbamoyl transferase
ORD	Office of Research and Development
PBPK	physiologically based pharmacokinetic
PCNA	proliferating cell nuclear antigen
PND	postnatal day
POD	point of departure
POD_{ADJ}	duration-adjusted POD
QSAR	quantitative structure-activity relationship
RBC	red blood cell
RDS	replicative DNA synthesis
RfC	inhalation reference concentration
RfD	oral reference dose
RGDR	regional gas dose ratio
RNA	ribonucleic acid
SAR	structure-activity relationship
SCE	sister chromatid exchange
SD	standard deviation
SDH	sorbitol dehydrogenase
SE	standard error
SGOT	serum glutamic oxaloacetic transaminase, also known as AST
SGPT	serum glutamic pyruvic transaminase, also known as ALT
SSD	systemic scleroderma
TCA	trichloroacetic acid
TCE	trichloroethylene
TWA	time-weighted average
UF	uncertainty factor
UF_A	interspecies uncertainty factor
UF_C	composite uncertainty factor
UF_D	database uncertainty factor
UF_H	intraspecies uncertainty factor
UF_L	LOAEL-to-NOAEL uncertainty factor
UF_S	subchronic-to-chronic uncertainty factor
U.S.	United States of America
WBC	white blood cell

Abbreviations and acronyms not listed on this page are defined upon first use in the PPRTV document.

PROVISIONAL PEER-REVIEWED TOXICITY VALUES FOR THE AROMATIC MEDIUM CARBON RANGE TOTAL PETROLEUM HYDROCARBON (TPH) FRACTION

BACKGROUND

A Provisional Peer-Reviewed Toxicity Value (PPRTV) is defined as a toxicity value derived for use in the Superfund Program. PPRTVs are derived after a review of the relevant scientific literature using established U.S. Environmental Protection Agency (U.S. EPA) guidance on human health toxicity value derivations.

The purpose of this document is to provide support for the hazard and dose-response assessment pertaining to chronic and subchronic exposures to substances of concern, to present the major conclusions reached in the hazard identification and derivation of the PPRTVs, and to characterize the overall confidence in these conclusions and toxicity values. It is not intended to be a comprehensive treatise on the chemical or toxicological nature of this substance.

Currently available PPRTV assessments can be accessed on the U.S. EPA's PPRTV website at https://www.epa.gov/pprtv. PPRTV assessments are eligible to be updated on a 5-year cycle and revised as appropriate to incorporate new data or methodologies that might impact the toxicity values or affect the characterization of the chemical's potential for causing adverse human-health effects. Questions regarding nomination of chemicals for update can be sent to the appropriate U.S. EPA's eComments Chemical Safety web page (https://ecomments.epa.gov/chemicalsafety/).

QUALITY ASSURANCE

This work was conducted under the U.S. EPA Quality Assurance (QA) program to ensure data are of known and acceptable quality to support their intended use. Surveillance of the work by the assessment managers and programmatic scientific leads ensured adherence to QA processes and criteria, as well as quick and effective resolution of any problems. The QA manager, assessment managers, and programmatic scientific leads have determined under the QA program that this work meets all U.S. EPA quality requirements. This PPRTV was written with guidance from the CPHEA Program Quality Assurance Project Plan (PQAPP), the QAPP titled *Program Quality Assurance Project Plan (PQAPP) for the Provisional Peer-Reviewed Toxicity Values (PPRTVs) and Related Assessments/Documents (L-CPAD-0032718-QP),* and the PPRTV development contractor QAPP titled *Quality Assurance Project Plan—Preparation of Provisional Toxicity Value (PTV) Documents (L-CPAD-0031971-QP)*. As part of the QA system, a quality product review is done prior to management clearance. A Technical Systems Audit may be performed at the discretion of the QA staff.

All PPRTV assessments receive internal peer review by at least two CPHEA scientists and an independent external peer review by at least three scientific experts. The reviews focus on whether all studies have been correctly selected, interpreted, and adequately described for the purposes of deriving a provisional reference value. The reviews also cover quantitative and qualitative aspects of the provisional value development and address whether uncertainties associated with the assessment have been adequately characterized.

DISCLAIMERS

The PPRTV document provides toxicity values and information about the adverse effects of the chemical and the evidence on which the value is based, including the strengths and limitations of the data. All users are advised to review the information provided in this document to ensure that the PPRTV used is appropriate for the types of exposures and circumstances at the site in question and the risk management decision that would be supported by the risk assessment.

Other U.S. EPA programs or external parties who may choose to use PPRTVs are advised that Superfund resources will not generally be used to respond to challenges, if any, of PPRTVs used in a context outside of the Superfund program.

This document has been reviewed in accordance with U.S. EPA policy and approved for publication. Mention of trade names or commercial products does not constitute endorsement or recommendation for use.

QUESTIONS REGARDING PPRTVS

Questions regarding the content of this PPRTV assessment should be directed to the U.S. EPA ORD CPHEA website at https://ecomments.epa.gov/pprtv.

1. INTRODUCTION

This Provisional Peer-Reviewed Toxicity Value (PPRTV) assessment supports a fraction-based approach to risk assessment for mixtures of petroleum hydrocarbons U.S. EPA (2022). In this approach, total petroleum hydrocarbon (TPH) fractions are defined based on expected transport in the environment and analytical methods used to quantify environmental contamination by TPH mixtures. TPH components were first classified into aromatics and aliphatics, and each of these two major fractions were further separated into low, medium, and high carbon range fractions. This PPRTV assessment describes the derivation of both noncancer and cancer toxicity values for the aromatic medium carbon range fraction of TPH. The toxicity values described herein are used in the assessment of Complex Mixtures of Petroleum Hydrocarbons that is intended to replace current approaches used at TPH-contaminated sites U.S. EPA (2022).

In general, fraction-based approaches involve: (1) dividing a complex mixture into groups based on similarities in their chemical structures or chemical properties; (2) measuring the concentrations of these groups (or the components within the group) in environmental media or estimating the rates of human exposure (e.g., mg/kg-day) to these groups; (3) selecting an approach to characterize a dose-response relationship for the group; (4) combining the dose-response approach and the exposure estimates for all members of the group to estimate health risks from the group; and (5) estimating risks or hazards posed by exposure to the complex mixture using the risk characterization information from the individual groups [adapted from Atsdr (2018)].

1.1. DEFINITION OF THE AROMATIC MEDIUM CARBON RANGE FRACTION

The aromatic medium carbon range fraction includes aromatic hydrocarbons with a carbon (C) range of C9−C10 (contains between 9 and 10 carbons, inclusive) and an equivalent carbon (EC) number[1] index range of EC9−EC < 11[2] that occur in, or co-occur with, petroleum contamination. It should be noted that the aromatic high carbon range fraction also includes C10 compounds, but unlike the aromatic high carbon range fraction, the aromatic medium carbon range fraction is restricted to those with EC9−EC < 11; the aromatic high carbon range fraction includes those compounds with an EC11–EC35. The EC index is equivalent to the retention time of the compound on a boiling-point gas chromatography (GC) column (nonpolar capillary column), normalized to the *n*-alkanes NJ DEP (2010). EC numbers are the physical characteristic that underpin analytical separation of petroleum components. EC numbers are useful because they are more closely related to environmental mobility than carbon number. For instance, two chemicals with similar carbon numbers but different structures (e.g., aliphatic vs. aromatic) could partition differently into environmental media and, ultimately, have different environmental fates. Grouping based on EC numbers provides a consistent basis for logically placing petroleum hydrocarbon compounds into fractions because EC measures correlate with physicochemical properties such as water solubility, vapor pressure, Henry's law constant, and soil adsorption coefficient (log K_{oc}).

[1]Based on an empirical relationship, the EC value can be estimated from a compound's boiling point (BP; °C) using the following equation: EC = 4.12 + 0.02 (BP) + 6.5 × 10^{-5} $(BP)^2$; see Gustafson et al. (1997a).
[2]This range reflects EC values rounded to the nearest whole number. For instance, isopropylbenzene (EC = 8.66) is included in this fraction because its EC value rounds to 9.

Toxicological considerations also contributed to the definition of the aromatic medium carbon range fraction. Substituted benzenes (C9−C10; contained within the aromatic medium carbon fraction) were grouped separately from PAHs, naphthalenes, and 1,1-biphenyl (contained within the aromatic high carbon fraction), which generally exhibit greater carcinogenicity and noncancer toxicity. Example compounds in the aromatic medium carbon range fraction include isopropylbenzene (a C9 aromatic compound with an EC of 8.66) and *n*-butylbenzene (a C10 aromatic compound with an EC of 9.96). The selection of relevant compounds and mixtures is described in Section 2 and Appendix A.

1.2. OVERVIEW OF PHYSICOCHEMICAL PROPERTIES AND ENVIRONMENTAL FATE

The systematic chemical names, synonyms [following guidance in Nist (2020b)], CASRNs, chemical abbreviations, chemical structures, and molecular weights for chemicals in this document are listed in Table 1 and in Appendix B of U.S. EPA (2022). The physicochemical properties for chemicals of the aromatic medium carbon range fraction that have toxicity values compiled from the CompTox Chemicals Dashboard U.S. EPA (2021) are provided in Table 2. Section 2 details how fraction members with toxicity values were identified. As Table 2 shows, the eight chemicals with toxicity values include both C9 and C10 compounds. The eight chemicals selected to represent the components of the aromatic medium carbon range fraction are all liquids at room temperature with moderate water solubility and high vapor pressure. All fraction members contain an alkyl substituted aromatic ring; three of the members contain three methyl groups on an aromatic ring (CASRNs 108-67-8, 95-63-6, and 526-73-8), three have one branched alkyl group on an aromatic ring (CASRNs 98-82-8, 98-06-6, and 135-98-8), and two have one linear alkyl group on an aromatic ring (CASRNs 103-65-1 and 104-51-8). Members of this fraction are expected to have negligible to slow mobility in soil. Volatilization may occur from water and moist soil (based upon measured and estimated Henry's law constant values) and from dry soil surfaces (based upon measured vapor pressure data); however, adsorption to soil is expected to attenuate volatilization of the fraction members from soil. Measured aerobic and anaerobic biodegradation data are available for the representative compounds. Aromatic hydrocarbons typically have slow biodegradation rates under aerobic conditions and slow to no biodegradation under anaerobic conditions. However, more rapid biodegradation has been reported for some of the members of this fraction. For example, in activated sludge from a predominantly domestic wastewater treatment plant, *sec*-butylbenzene and *n*-butylbenzene had 56−67 and 72−80% removal, respectively, in 5 days at 25°C at test substance concentrations of 100 mg/L NCBI (2022a, b). Members of the aromatic medium carbon range fraction do not contain hydrolysable functional groups; therefore, the rate of hydrolysis is expected to be negligible for all fraction members. In the atmosphere, the rate of photooxidation is expected to range from slow (*tert*-butylbenzene) to rapid for the vapor-phase forms of the fraction members. The fraction members do not contain chromophores that absorb at wavelengths >290 nm and are therefore not expected to be susceptible to direct photolysis by sunlight Nlm (2013, 2008a, b, c, 2005, 2004a, b, 2003).

Table 1. Synonyms and Abbreviations for Chemicals in This Document[a]

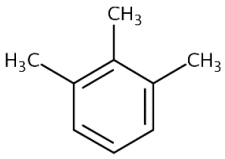
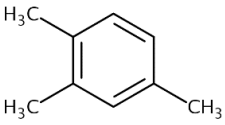
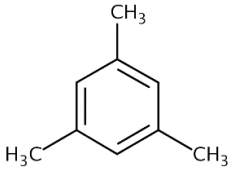
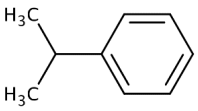
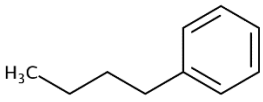
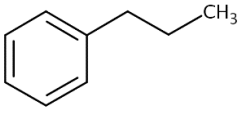
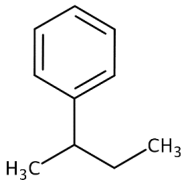
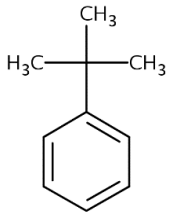
Chemical Name (common synonyms)[b]	CASRN	Abbreviation	Structure	Molecular Weight (g/mol)
1,2,3-Trimethylbenzene (benzene, 1,2,3-trimethyl-)	526-73-8	1,2,3-TMB		120.195
1,2,4-Trimethylbenzene (benzene, 1,2,4-trimethyl-)	95-63-6	1,2,4-TMB		120.195
1,3,5-Trimethylbenzene (benzene, 1,3,5-trimethyl-)	108-67-8	1,3,5-TMB		120.195
Isopropylbenzene (cumene; [propan-2-yl]benzene; benzene, [1-methylethyl]-)	98-82-8			120.195
HFAN (light aromatic solvent naphtha [petroleum]; solvent naphtha, petroleum, light aromatic; super high flash naphtha; aromatic solvent; solvent, aromatic petroleum; solvent naphtha; light aromatic solvent naphtha; low boiling point naphtha–unspecified; solvent naphtha [petroleum], light aromatic)	64742-95-6		Various	Various
***n*-Butylbenzene** (benzene, butyl-)	104-51-8			134.222
***n*-Propylbenzene** (propylbenzene; benzene, propyl-)	103-65-1			120.195

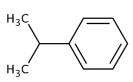
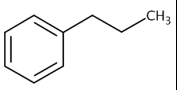
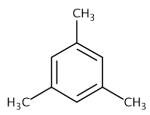
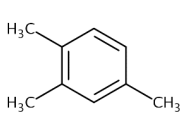
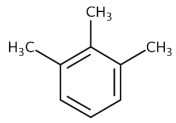
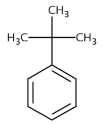
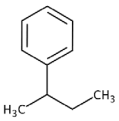
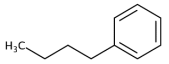
Table 1. Synonyms and Abbreviations for Chemicals in This Document[a]

Chemical Name (common synonyms)[b]	CASRN	Abbreviation	Structure	Molecular Weight (g/mol)
***sec*-Butylbenzene** ([butan-2-yl]benzene; benzene, [1-methylpropyl]-)	135-98-8			134.222
***tert*-Butylbenzene** (benzene, [1,1-dimethylethyl]-)	98-06-6			134.222

[a]Only chemicals with toxicity values are listed.
[b]Synonyms are listed according to National Institute of Standards and Technology Nist (2020b) and include valid synonyms from the U.S. EPA CompTox Chemicals Dashboard; https://comptox.epa.gov/dashboard; accessed 03-30-2020.

U.S. EPA = U.S. Environmental Protection Agency.

Table 2. Physicochemical Properties of Aromatic Medium Carbon Range Chemicals with Toxicity Values[a]

Chemical	Isopropyl-benzene	*n*-Propyl-benzene	1,3,5-TMB	1,2,4-TMB	1,2,3-TMB	*tert*-Butyl-benzene	*sec*-Butyl-benzene	*n*-Butyl-benzene
Structure								
CASRN	98-82-8	103-65-1	108-67-8	95-63-6	526-73-8	98-06-6	135-98-8	104-51-8
Molecular formula	C_9H_{12}	C_9H_{12}	C_9H_{12}	C_9H_{12}	C_9H_{12}	$C_{10}H_{14}$	$C_{10}H_{14}$	$C_{10}H_{14}$
EC number[b]	8.66	8.94	9.15	9.36	9.65	9.36	9.57	9.96
Molecular weight (g/mol)	120.195	120.195	120.195	120.195	120.195	134.222	134.222	134.222
Melting point (°C)	−96	−99.6	−44.9	−46.0	−25.4	−58.3	−73.6	−88.0
Boiling point (°C)	152	159	164	169	176	169	174	183
Vapor pressure (mm Hg at 25°C)	4.50	3.42	2.48	2.10	1.69	2.20	1.75	1.06
Henry's law constant (atm-m^3/mole at 25°C)	1.15×10^{-2}	1.05×10^{-2}	8.77×10^{-3}	6.16×10^{-3}	4.36×10^{-3}	7.89×10^{-3}	8.03×10^{-3}	8.05×10^{-3}
Water solubility (mol/L at 25°C)	5.24×10^{-4}	4.65×10^{-4}	3.99×10^{-4}	4.78×10^{-4}	6.30×10^{-4}	2.19×10^{-4}	1.30×10^{-4}	8.73×10^{-5}
Log K_{ow}	3.66	3.71	3.50	3.63	3.63	4.11	4.57	4.38
Log K_{oa}	3.98	4.09	4.54*	4.54*	4.54*	4.46*	4.31*	4.60*
Log K_{oc}	3.00*	2.87	2.82	3.05*	2.80	3.34*	3.76*	3.39

[a]Data are presented as experimental averages from the U.S. EPA CompTox Chemicals Dashboard unless otherwise stated; https://comptox.epa.gov/dashboard; updated 02-03-2021.
[b]EC number was developed by the TPHCWG and is proportional to the BP of a chemical. EC number is analogous to an *n*-paraffin retention time index and can be estimated using: EC = 4.12 + 0.02 (BP) + 6.5×10^{-5} $(BP)^2$ NIST (2020a; Edwards et al. (1997; Gustafson et al. (1997b).
*Predicted value.

BP = boiling point; EC = equivalent carbon; K_{oa} = octanol-air partition coefficient; K_{oc} = soil adsorption coefficient; K_{ow} = octanol-water partition coefficient; TMB = trimethylbenzene; TPHCWG = Total Petroleum Hydrocarbon Criteria Working Group.

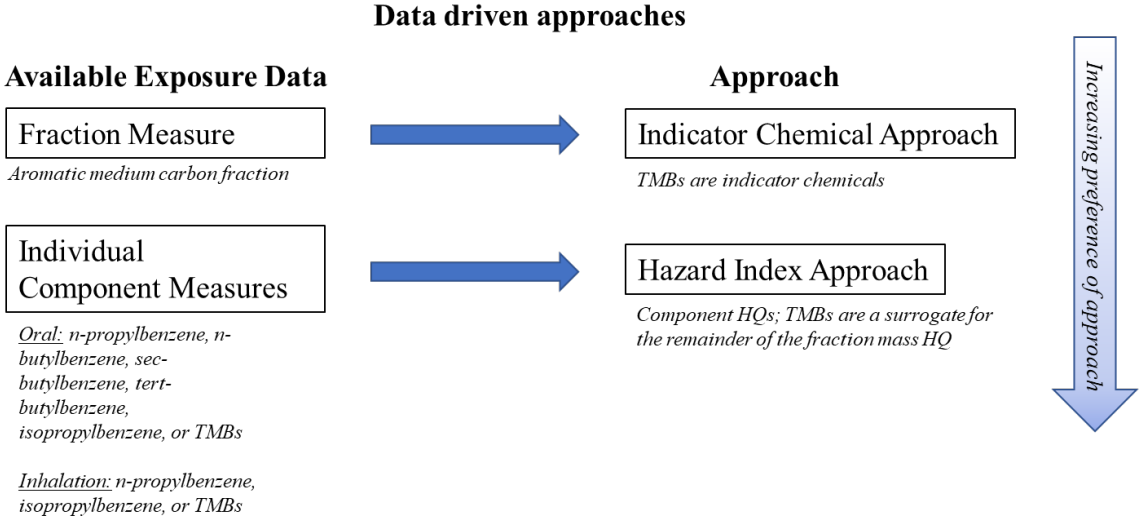
1.3. OVERVIEW OF MIXTURE ASSESSMENT METHODS

A number of different approaches have been developed and used to estimate risks and hazards posed by exposures to chemical mixtures encountered in the environment. Among the simplest of these approaches to implement is the indicator chemical approach Atsdr (2018). The indicator chemical approach estimates the risk or hazard of a mixture by evaluating the dose-response assessment developed for a component of the mixture to the exposure rate of the entire mixture. The indicator chemical approach is used when there are only measures of the concentrations of this fraction (i.e., no information is available on the concentrations of individual chemicals in this fraction) (see Section 1.3.1). The hazard index (HI) approach (the other approach that will be addressed in this PPRTV assessment) can be used when there are measured concentrations of specific chemical compounds. In the HI approach, the individual chemical intake rates (or concentrations in the air) are divided by the reference dose (RfD) (or reference concentration [RfC]) for the chemical to estimate a hazard quotient (HQ). The HQs are summed to estimate the HI (see Section 1.3.2). The indicator chemical approach has greater uncertainty than the HI approach (see Figure 1).

The U.S. Environmental Protection Agency's (U.S. EPA) *Supplementary Guidance for Conducting Health Risk Assessment of Chemical Mixtures* U.S. EPA (2000, 1986) describes the following two broad categories of approaches for assessing human health risks and health hazards associated with environmental exposures to chemical mixtures: component methods and whole mixture methods. Component-based approaches, which involve analyzing the toxicity of a mixture's individual components, have more uncertainty and are recommended when toxicity data on a complex mixture of concern, or on a sufficiently similar mixture (discussed below), are unavailable U.S. EPA (2000, 1986). In this PPRTV, a component approach, the HI approach, is described for assessing noncancer hazards posed by exposures to the aromatic medium carbon range fraction.

Chemical mixture assessments are conducted most appropriately with quantitative dose-response information resulting from comparable exposures to the mixture of concern. If the dose-response data are insufficient to develop a health reference value for the specific mixture of concern in the environment, the second option that the U.S. EPA's *Supplementary Guidance for Conducting Health Risk Assessment of Chemical Mixtures* U.S. EPA (2000, 1986) recommended is a "sufficient similarity" approach that uses a health reference value from a characterized surrogate mixture to estimate the hazard or risk associated with exposures to the mixture of concern. This method requires chemistry and toxicity data on both the potential surrogate mixture and the mixture of concern (e.g., a key event that is related to the apical endpoint observed in an epidemiological study or whole animal study), and a health reference value (e.g., from an in vivo study) on the surrogate mixture. If the chemistry and toxicity data indicate that the mixtures are "sufficiently similar" to one another, then the health reference value for the surrogate mixture can be used as a proxy for the mixture of concern. No data were identified that were suitable to implement a whole mixture approach.

The choice of a chemical mixtures risk assessment method is driven by the available data. Starting with the method requiring the least information and then discussing the method requiring more information, the following subsections summarize the indicator chemical approach and the HI approach. Figure 1 summarizes the two approaches and the preference for using each approach.



Two approaches are available to estimate the noncancer hazards associated with exposure to the aromatic medium range fraction. Approach selection should be driven by the available exposure data. Increased analytical characterization of fraction components allows for more refined health hazard estimates with less inherent uncertainty. Approach preference is inversely correlated with approach uncertainty.

HQ = hazard quotient; TMB = trimethylbenzene.

Figure 1. Provisional Peer-Reviewed Toxicity Approaches for the Aromatic Medium Carbon Range TPH Fraction Noncancer Assessment

1.3.1. Indicator Chemical Approach

When the chemical composition of a mixture or a mixture fraction is not known, or toxicity measures are not available for individual chemicals in a mixture, the toxicity of an individual chemical can be used as an indicator for the toxicity of a mixture or a mixture fraction Atsdr (2018). Atsdr (2018) describes an indicator chemical as "a chemical . . . selected to represent the toxicity of a mixture because it is characteristic of other components in the mixture and has adequate dose-response data." Indicator chemical approaches are typically implemented to assess health risks in a health-protective manner; the chemical chosen as an indicator is among the best characterized toxicologically and likely among the most potent components of the mixture. The indicator chemical needs to have adequate dose-response data to indicate hazard potential or a dose-response relationship for noncancer outcomes, depending on the purpose of the assessment. The health risk value of the indicator chemical is integrated with exposure estimates for the mixture or mixture fraction to estimate health hazard associated with the fraction (i.e., calculate fraction-specific HI for a specific exposure pathway). This approach does not scale for potency of individual constituents; instead, it assumes that the toxicity of all measured members of the fraction can be adequately estimated, given the purpose of the risk assessment, by the indicator chemical.

1.3.2. Hazard Index Approach

The HI approach combines estimated population exposures with toxicity information to characterize the potential for adverse effects. The HI is not a risk estimate, in that it is not expressed as a probability, nor is it an estimate of a toxicity measure (e.g., percentage decrement

in enzyme activity). Instead, the HI is an indicator of potential hazard. In the HI approach, a HQ is calculated as the ratio of human exposure (*E*) to a health hazard reference value (RfV) for each mixture component chemical (*i*) U.S. EPA (1986). These HQs are summed to yield the HI for the mixture. In health risk assessments, the U.S. EPA's preferred RfVs are the RfD for the oral exposure route and the RfC for the inhalation exposure route.

$$HI = \sum_{i=1}^{n} HQ_i = \sum_{i=1}^{n} \frac{E_i}{RfV_i}$$

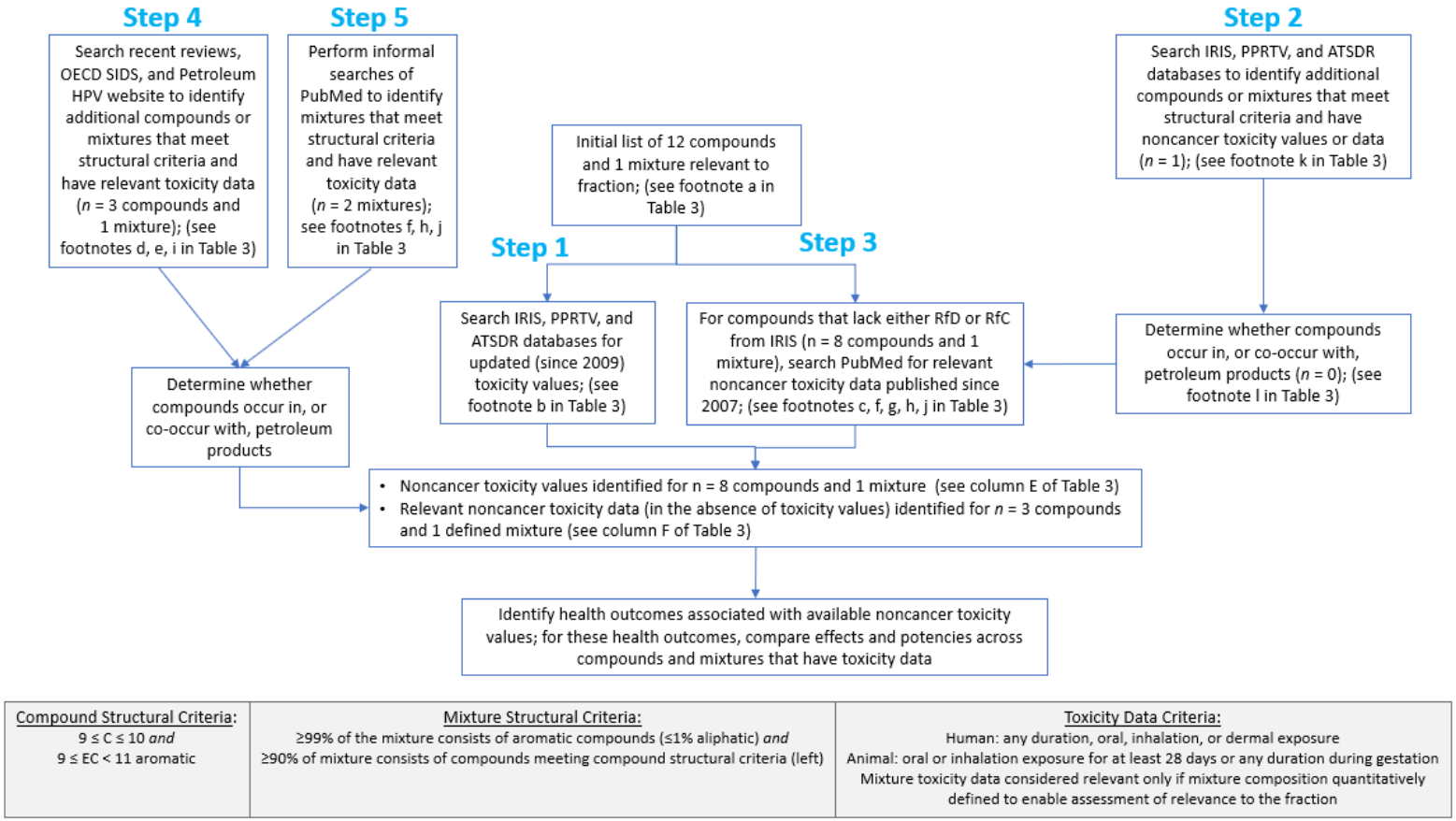
The HI is based on dose addition U.S. EPA (2000); Svendsgaard and Hertzberg (1994); the hazard is evaluated as the potency-weighted sum of the component exposures. The HI is dimensionless, so *E* and the RfV must be in the same units.

2. SUMMARY OF TOXICITY AND DOSE-RESPONSE ASSESSMENT APPROACH

Toxicity and dose-response assessment for the aromatic medium carbon range fraction entailed five basic steps, outlined here and described in more detail below. Mixtures and compounds that met structural criteria (see Section 1.1) and had available toxicity values from designated sources were identified. The dose-response assessment for the fraction can include selection of a health reference value from an indicator chemical. Alternatively, if exposure measurement data are available for component chemicals, those health reference values from component chemicals with existing toxicity values can be used to apply the HI approach.

In Step 1 and Step 2 of the assessment, literature searches were performed for the mixtures and compounds with toxicity values and for other mixtures and compounds that are relevant to the fraction. Searches date-limited to assessments published from 2009 forward (e.g., Agency for Toxic Substances and Disease Registry [ATSDR], Integrated Risk Information System [IRIS]) were conducted in February 2018, and updated most recently in August 2021. The start date of 2009 was selected to determine whether new information suggested that toxicity values for mixtures or compounds relevant to the fraction should be updated from those identified in the U.S. EPA (2009b) PPRTV for complex mixtures of aliphatic and aromatic hydrocarbons. Step 3 in the assessment involved searching PubMed for new noncancer toxicity data on compounds and mixtures lacking either Integrated Risk Information System (IRIS) oral or inhalation toxicity values. These literature searches were conducted in February 2018, updated in August 2021, and were date-limited to studies published from 2007 forward, in order to capture studies that were published since the searches performed in U.S. EPA (2009b). Step 4 in the assessment involved searching recent comprehensive reviews on the toxicity of petroleum components or classes of compounds relevant to the fraction, as well as Organisation for Economic Co-operation and Development (OECD) Screening Information Data Set (SIDS) assessments[3] and the Petroleum High Production Volume (HPV) Testing Group website, to identify other mixtures or compounds within this carbon range with existing toxicity data that may inform hazard identification for the fraction. Step 5 of the assessment involved informal searches of PubMed to identify mixtures that met structural criteria of the fraction and had toxicity data available. Toxicity data criteria included human studies of any duration by oral, inhalation, and dermal exposure, and animal studies of oral or inhalation exposure lasting at least 28 days (or any duration of gestational exposure). Mixture toxicity data were considered relevant only if the mixture composition under study was quantitatively defined to enable assessment of relevance to the fraction. Figure 2 shows a schematic depiction of the process, and further detail is provided below.

[3]The OECD Existing Chemicals Database (https://hpvchemicals.oecd.org) was reviewed for relevant categories, and dossiers for the following categories were screened: diethylbenzenes (DEBs), C9 aromatic solvents, and C10−C13 aromatic solvents. In addition, the Oecd (1994) hazard characterization for 1,4-DEB was reviewed for relevant toxicity data.



Compounds and mixtures relevant to the aromatic medium carbon range fraction with available toxicity values or data were identified in a five-step process. Table 3 lists individual compounds and mixtures and links them to their corresponding identification source during the literature search process. Table 3 also indicates compounds with available toxicity values, or in the absence of toxicity values, available toxicity data.

ATSDR = Agency for Toxic Substances and Disease Registry; C = carbon; EC = equivalent carbon; HPV = High Production Volume; IRIS = Integrated Risk Information System; OECD = Organisation for Economic Co-operation and Development; PPRTV = Provisional Peer-Reviewed Toxicity Value; RfC = reference concentration; RfD = reference dose; SIDS = Screening Information Data Set.

Figure 2. Selection of Compounds and Mixtures for Aromatic Medium Carbon Range Fraction Hazard Identification and Dose-Response Assessment

2.1. IDENTIFICATION OF RELEVANT MIXTURES AND COMPOUNDS WITH TOXICITY VALUES

Step 1 (see Figure 2) in the assessment of the toxicity for the aromatic medium carbon range fraction was to identify constituents of the fraction that have existing toxicity values from any of the sources considered for the U.S. EPA (2009b) PPRTV assessment for complex mixtures of aliphatic and aromatic hydrocarbons (these included IRIS, PPRTVs, ATSDR Minimal Risk Levels [MRLs], Massachusetts Department of Environmental Protection [MassDEP], Total Petroleum Hydrocarbon Criteria Working Group [TPHCWG], and Health Effects Assessment Summary Tables [HEAST]). Of these sources, only IRIS, PPRTVs, and ATSDR MRLs have been updated since 2009, so only these sources were consulted for toxicity values. Based on the U.S. EPA's previous assessments and assessment activities as well as those relevant chemicals reviewed by the MassDEP MassDep (2003) or TPHCWG Edwards et al. (1997), the U.S. EPA compiled an initial list of 12 chemicals and 1 mixture (i.e., high flash aromatic naphtha [HFAN]). [See full list in Appendix A and description of approach and results in Wang et al. (2012).] Table 3 lists the chemicals and mixtures for which PubMed searches were performed, published toxicity data were identified, and toxicity values were identified. At least one updated subchronic or chronic oral or inhalation reference value was available for eight chemicals (isopropylbenzene, *n*-propylbenzene, 1,3,5-trimethylbenzene [TMB], 1,2,4-TMB, 1,2,3-TMB, *tert*-butylbenzene, *sec*-butylbenzene, and *n*-butylbenzene), and one mixture (HFAN).

Table 3. Chemicals and Mixtures Identified in Literature Searches

CASRN	Chemical Name	Literature Search Identification Source	PubMed Searches Performed	Toxicity Values Identified	Toxicity Data Identified
526-73-8	1,2,3-Trimethyl-benzene	Initial list of 12 compounds + 1 mixture[a, b, c]		X	
95-63-6	1,2,4-Trimethyl-benzene	Initial list of 12 compounds + 1 mixture[a, b, c]		X	
135-01-3	1,2-Diethylbenzene	Recent reviews of petroleum toxicity[d, e]			X
108-67-8	1,3,5-Trimethyl-benzene	Initial list of 12 compounds + 1 mixture[a, b, c]		X	
141-93-5	1,3-Diethylbenzene	Recent reviews of petroleum toxicity[d, e]			X
105-05-5	1,4-Diethylbenzene	Recent reviews of petroleum toxicity[d, e]			X
620-14-4	1-Methyl-3-ethyl-benzene	Initial list of 12 compounds + 1 mixture[a]	X		
622-96-8	1-Methyl-4-ethyl-benzene	Initial list of 12 compounds + 1 mixture[a, f, g]	X		X
535-77-3	1-Methyl-3-iso-propylbenzene	Initial list of 12 compounds + 1 mixture[a]	X		
Various	Alkylbenzenes (various)	PubMed searches[f, h]	X		X
25340-17-4	Diethylbenzene mixture	OECD SIDS[i]			X
64742-95-6, 88845-25-4, and 64742-94-5	High flash aromatic naphtha	Initial list of 12 compounds + 1 mixture[a, b]	X	X	
538-93-2	Isobutylbenzene	Initial list of 12 compounds + 1 mixture[a]	X		
98-82-8	Isopropylbenzene	Initial list of 12 compounds + 1 mixture[a, b, c]		X	
104-51-8	*n*-Butylbenzene	Initial list of 12 compounds + 1 mixture[a, b]	X	X	
103-65-1	*n*-Propylbenzene	Initial list of 12 compounds + 1 mixture[a, b]	X	X	
Various	Naphtha solvent	PubMed searches[f, j]	X		X
99-87-6	*p*-Isopropyltoluene	Updated PPRTV, IRIS, and ATSDR MRL databases[k]			X[l]
135-98-8	*sec*-Butylbenzene	Initial list of 12 compounds + 1 mixture[a, b]	X	X	

Table 3. Chemicals and Mixtures Identified in Literature Searches

CASRN	Chemical Name	Literature Search Identification Source	PubMed Searches Performed	Toxicity Values Identified	Toxicity Data Identified
98-06-6	*tert*-Butylbenzene	Initial list of 12 compounds + 1 mixture[a, b]	X	X	

[a]U.S. EPA developed the initial list of 12 chemicals and 1 mixture relevant to the aromatic medium carbon range. The list included all individual hydrocarbons considered previously by the U.S. EPA's STSC in the evaluation of hydrocarbons, as well as all those with toxicity data reviewed by the MassDEP MassDep (2003) or TPHCWG Edwards et al. (1997).
[b]At least one updated (since 2009) subchronic oral or inhalation reference value was available for these compounds following searches of the IRIS, PPRTV, and ATSDR databases.
[c]Because these compounds had IRIS, PPRTV, or ATSDR toxicity values for both oral and inhalation routes, no additional literature searches were performed.
[d]These compounds/mixtures were identified in McKee et al. (2015), a recent review of petroleum toxicity.
[e]Toxicity data for these compounds were found in Gagnaire et al. (1990).
[f]These compounds were identified in PubMed searches date-limited to studies published from 2007 forward, conducted in February 2018.
[g]Toxicity data for this compound were found in Swiercz et al. (2000).
[h]Human olfaction data for these compounds were found in Cometto-Muniz and Abraham (2009).
[i]Toxicity data for this mixture were found in Oecd (2007).
[j]A human case report for this mixture was found in Magdalan et al. (2009).
[k]During review of the updated IRIS, PPRTV, and ATSDR MRL databases, these compounds were identified in the PPRTV database as meeting structural criteria for inclusion and having toxicity assessments.
[l]This compound was not considered relevant to this assessment because this compound does not typically occur or co-occur with petroleum contamination. The U.S. EPA (2011) PPRTV for isopropyltoluene identified no studies investigating health effects following oral exposures for short-term, subchronic, or chronic durations; further, no developmental or reproductive oral exposure studies were identified. The PPRTV reported data on *p*-isopropyltoluene-induced toxicity in animals exposed orally limited to a single-dose, acute toxicity study that reported depression, coma, bloody lacrimation, diarrhea, irritability, and scrawny appearance in treated Osbourne-Mendel rats. The PPRTV identified one subchronic inhalation study and two acute inhalation studies; these reported mortality in mice, but not in rats or guinea pigs. A necropsy in the mice revealed hyperemic lungs, mottled liver, and pale kidney.

ATSDR = Agency for Toxic Substances and Disease Registry; IRIS = Integrated Risk Information System; MassDEP = Massachusetts Department of Environmental Protection; MRL = minimal risk level; OECD = Organisation for Economic Co-operation and Development; PPRTV = provisional peer-reviewed toxicity value; SIDS = Screening Information Data Set; STSC = Superfund Technical Support Center; TPHCWG = Total Petroleum Hydrocarbon Criteria Working Group; U.S. EPA = U.S. Environmental Protection Agency.

In Step 2 (see Figure 2), all existing chemicals in the IRIS, PPRTV, and ATSDR MRL databases were searched to determine whether they included any other compounds or mixtures (not on the initial list) meeting the structural criteria for inclusion (C9−C10 and EC9−EC < 11 aromatics). Searches of the IRIS and ATSDR databases did not identify any additional compounds, but review of the PPRTV database identified one additional compound with a toxicity assessment (but no health risk values), *p*-isopropyltoluene (*p*-cymene). Review of the Petroleum Hydrocarbon Criteria Working Group's (1998) *Selection of Representative TPH Fractions Based on Fate and Transport Considerations* Gustafson et al. (1997b) indicated that this compound typically does not occur or co-occur with petroleum contamination, a finding further confirmed by *Ullmann's Encyclopedia of Industrial Chemistry* Eggersdorfer (2012).

Therefore, *p*-isopropyltoluene was not considered relevant to the assessment of the aromatic medium carbon range fraction and is not discussed further. Table 4 shows the health risk values available for the eight relevant compounds and the HFAN mixture.

Table 4. Summary of Available Toxicity Values for Mixtures and Constituents of Aromatic Medium Carbon Range (C9−C10, EC9−EC < 11) Fraction[a]

CASRN	Name	C	EC	Oral Reference Dose (mg/kg-d)		Inhalation Reference Concentration (mg/m^3)	
				Subchronic	Chronic	Subchronic	Chronic
98-82-8	Isopropylbenzene	9	8.66	–	0.1 (IRIS)	–	0.4 (IRIS)
103-65-1	*n*-Propylbenzene	9	8.94	0.1[b] (PPRTV*)	0.1[b] (PPRTV*)	1[b] (PPRTV*)	1[b] (PPRTV*)
108-67-8	1,3,5-TMB	9	9.15	0.04 (IRIS)	0.01 (IRIS)	0.2 (IRIS)	0.06 (IRIS)
95-63-6	1,2,4-TMB	9	9.36	0.04 (IRIS)	0.01 (IRIS)	0.2 (IRIS)	0.06 (IRIS)
98-06-6	*tert*-Butylbenzene	10	9.36	0.1[c] (PPRTV*)	0.1[c] (PPRTV*)	–	–
135-98-8	*sec*-Butylbenzene	10	9.57	0.1[c] (PPRTV*)	0.1[c] (PPRTV*)	–	–
526-73-8	1,2,3-TMB	9	9.65	0.04 (IRIS)	0.01 (IRIS)	0.2 (IRIS)	0.06 (IRIS)
104-51-8	*n*-Butylbenzene	10	9.96	0.1 (PPRTV)	0.05 (PPRTV)	–	–
64742-95-6	HFAN	9−10	NA	0.3 (PPRTV*)	0.03 (PPRTV*)	1 (PPRTV)	0.1 (PPRTV)

[a]Toxicity values shown were selected from the following sources in order of preference: IRIS, PPRTV, ATSDR, HEAST, MassDEP, or TPHCWG. None of the mixtures or constituents in this fraction has an OSF or IUR.
[b]Based on the identification of ethylbenzene (CASRN 100-41-4) as an appropriate analogue chemical.
[c]Based on the identification of isopropylbenzene (CASRN 98-82-8) as an appropriate analogue chemical.
*Screening provisional toxicity value. Screening values are not assigned confidence statements; however, confidence in these values is presumed to be low. Screening values are derived when the data do not meet all requirements for deriving a provisional toxicity value. Screening values are derived using the same methodologies and undergo the same development and review processes (i.e., internal and external peer review, etc.) as provisional values; however, there is generally more uncertainty associated with these values.

ATSDR = Agency for Toxic Substances and Disease Registry; C = carbon; EC = equivalent carbon; HEAST = Health Effects Assessment Summary Tables; HFAN = high flash aromatic naphtha; IRIS = Integrated Risk Information System; IUR = inhalation unit risk; MassDEP = Massachusetts Department of Environmental Protection; NA = not applicable; OSF = oral slope factor; PPRTV = provisional peer-reviewed toxicity value; TMB = trimethylbenzene; TPHCWG = Total Petroleum Hydrocarbon Criteria Working Group.

2.2. IDENTIFICATION OF OTHER RELEVANT TOXICITY DATA

Among the 12 compounds and 1 mixture on the initial list determined to be relevant to the fraction, both oral and inhalation IRIS toxicity values were available for four compounds (isopropylbenzene and the three TMB isomers). Therefore, these compounds were not included in the comprehensive literature searches. In Step 3 (see Figure 2), literature searches were conducted in PubMed to identify any new studies that could fill data gaps for the remaining eight compounds and one mixture. The literature searches were conducted in February 2018, were most recently updated in August 2021, and were date-limited to studies published from 2007 forward, in order to capture studies that were published since the searches performed for the 2009 PPRTV assessment for complex TPH mixtures. A summary of the literature search strategy

is provided in Appendix A. As detailed in the appendix, studies considered relevant to hazard identification included animal studies using inhalation or oral exposure routes, in which exposures continued for at least 28 days (or any duration of gestational exposure), at least one health outcome was assessed, and an untreated or vehicle control group was included. Human studies of any duration in which exposure was known or presumed to be through oral, inhalation, or dermal routes and at least one health outcome was assessed were considered relevant.

The updated literature search identified two human studies: an acute human olfaction study of alkylbenzenes Cometto-Muniz and Abraham (2009) and a case report of exposure to naphtha solvent Magdalan et al. (2009). The only relevant animal study that was identified in the updated literature search was a 4-week study of 1-methyl-4-ethylbenzene Swiercz et al. (2000). No relevant reviews or secondary sources were identified.

In Step 4 (see Figure 2), to determine whether additional relevant compounds or mixtures had been tested for repeat-dose and/or reproductive/developmental toxicity since 2007, recent reviews of petroleum toxicity McKee et al. (2015; Infante and Bingham (2012; OECD (2012a, b, 2007) and the Petroleum HPV Testing Group website were searched. Mixtures considered relevant to the fraction met the following criteria (see Figure 2):

1. at least 90% of the mixture consisted of identified compounds within the C9−C10 and EC9−EC < 11 ranges.
2. 99% of the mixture consisted of aromatic compounds (≤1% aliphatic).
3. the mixture had been tested in animals in at least one repeat-dose (≥28 days) or reproductive/developmental toxicity study using inhalation or oral exposure routes and including an untreated or vehicle control group.
4. human mixture studies of any duration by oral, inhalation, and dermal exposure, and animal studies of oral or inhalation exposure lasting at least 28 days (or any duration of gestational exposure).

Using the same criteria, in Step 5, PubMed searches were conducted to identify mixtures with relevant toxicity data.

None of the mixtures described on the Petroleum HPV Testing Group website met these criteria. Oecd (2007) described toxicity data on a commercial diethylbenzene (DEB) mixture that met these criteria. Thus, including HFAN, toxicity data for two mixtures were considered potentially relevant to the assessment of the aromatic medium carbon fraction. HFAN consists of TMB and ethyltoluene isomers; by definition, it must contain a combined total of 75% TMB and ethyltoluene isomers (of which at least 22% is ethyltoluene and at least 15% is TMB) U.S. EPA (2009c). Commercial DEB typically contains 60−65% 1,3-DEB, 27−30% 1,4-DEB, and 4−5% 1,2-DEB Oecd (2007). No further detail on the chemical compositions of these mixtures was available in the sources reviewed.

Searching the review by McKee et al. (2015) resulted in the identification of three additional compounds that met structural criteria and had toxicity data that met inclusion criteria (see Figure 2): 1,2-, 1,3-, and 1,4-DEB. The three DEB isomers were tested for neurotoxicity in a study of rats exposed orally for 10 weeks Gagnaire et al. (1990). No other mixtures or compounds with toxicity data were identified. Human and animal studies that met criteria

outlined above were reviewed to support selection of surrogates for the aromatic medium carbon range fraction toxicity values.

2.3. METHODS FOR INDICATOR CHEMICAL SELECTION

Only compounds or mixtures with at least one U.S. EPA or ATSDR toxicity value (see Table 4) were to be considered for use as potential indicator chemicals (or indicator mixtures) for derivation of the fraction-specific toxicity values; however, no ATSDR toxicity values were identified. Toxicity data for other compounds that did not have toxicity values were used for hazard identification and to assess consistency in effects and potency across the components of the fraction. The method for selecting indicator chemicals was adapted from the 2009 complex TPH mixtures document U.S. EPA (2009b). First, mixtures consisting of fraction component chemicals were preferred over individual compounds, provided that the mixture study was adequate and the mixture exhibited in vivo toxic effects similar to those exhibited by the individual fraction components. If suitable mixture data were lacking, a representative compound exhibiting in vivo toxic effects and potency similar to those exhibited by other compounds in the fraction was chosen. If the components of the fraction varied widely in toxic effects or potency, the toxicity value for the most potent component (i.e., the component with lowest reference value) was selected as an indicator chemical for the fraction. Finally, if toxicity values were available for many or most of the individual compounds in a fraction, and these compounds are typically monitored at sites of hydrocarbon contamination, then a component approach would be considered.

2.4. DEVELOPMENT OF EXPOSURE-RESPONSE ARRAYS

In order to assess consistency in effects and potency across the components of the fraction, experimental data from compound-specific IRIS and PPRTV documents and primary data sources (identified from literature searches) were used to create exposure-response arrays provided in Appendix B. Data were extracted only from certain studies (i.e., studies that provided dose-response data enabling the identification of no-observed-adverse-effect levels [NOAELs] and lowest-observed-adverse-effect levels [LOAELs]). Target-organ-specific NOAELs and LOAELs were determined using the following methodology.

1. Whenever possible, NOAELs and LOAELs were identified from existing IRIS or PPRTV assessments. For chemicals in which both types of assessments were available, preference was given to IRIS (in accordance with U.S. EPA Office of Superfund Remediation and Technology Innovation [OSRTI] hierarchy of human health toxicity values for Superfund assessments). In general, these assessments explicitly identified NOAEL and LOAEL values only for the most sensitive target of toxicity, so characterization of additional adverse effect levels allowed for a comprehensive comparison of toxic effects across additional endpoints and tissues.

2. All other target-organ-specific effect levels (i.e., for targets other than the most sensitive target identified in IRIS or PPRTV assessments, and all targets evaluated in newly identified studies) were determined using scientific judgment, taking into consideration factors such as statistical significance (at a p-value < 0.05), biological significance (e.g., a greater than or equal to 10% increase in liver weight), magnitude and direction of change, and study quality. In the case of chemicals with existing IRIS or PPRTV assessments, NOAELs and LOAELs could often be identified from existing study summaries.

Dose-response data were presented in exposure-response arrays by health outcome and exposure route (see Appendix B). From left to right, compounds exhibiting an effect are shown before those not exhibiting an effect, to facilitate identification of patterns. Within the group exhibiting an effect, compounds are ordered from lowest LOAEL to highest. For compounds that do not exhibit an effect, NOAELs in the arrays are ordered by EC number (low to high from left to right), with mixtures shown last. Both administered doses and exposure concentrations reported in the arrays and in text reflect time-weighted average (TWA) exposures, to facilitate comparisons across studies and compounds. Consistency across the fraction was evaluated by assessing if comparable outcomes were observed for members of the fraction, and if these effects were observed at similar dose levels.

3. REVIEW OF POTENTIALLY RELEVANT DATA

3.1. NONCANCER EVIDENCE

Compound-specific IRIS and PPRTV documents, supplemented by the literature search findings and the review articles described above [particularly McKee et al. (2015)] were assessed to evaluate available noncancer data for the aromatic medium carbon range fraction compounds. Critical effects identified with existing toxicity values include neurological, hepatic, renal, body-weight, hematological, endocrine, respiratory, and developmental effects. Appendix B summarizes the evidence provided by human and experimental animal studies of noncancer health outcomes. Table 5 presents an overview of the human and animal data available to evaluate the primary toxicological endpoints identified for the fraction (neurological, hepatic, renal, body weight, hematological, endocrine, respiratory, and developmental). As Table 5 shows, both oral and inhalation data available to assess consistency in effects across members of the fraction are limited. There are no dependable human or animal data for at least three members of the fraction (*n*-propylbenzene, and *tert*- and *sec*-butylbenzene). There are oral or inhalation body-weight data for 11 members, and there are neurotoxicity endpoint data available for 9 members. For all other primary toxicological endpoints, there are oral or inhalation data for 5−7 members of the fraction. Most of the animal data are from inhalation toxicity studies. Comprehensive systemic toxicity was evaluated in rats and mice in subchronic and chronic inhalation studies for one member of the fraction (isopropylbenzene). In general, studies ranged in duration from 4 to 18 weeks; several of these studies (e.g., DEBs and TMBs) evaluated only neurological endpoints. Developmental inhalation toxicity studies were available for four members of the fraction (isopropylbenzene, 1,3,5- and 1,2,4-TMB, and HFAN). Finally, unless otherwise specified, the term “significant,” used throughout the document, refers to statistical significance at a p-value of < 0.05.

Table 5. Overview of Human and Animal Data Availability for Evidence Integration[a, b]

CASRN	Name	C	EC	Neurological	Hepatic	Renal	Body Weight	Hemato-logical	Endocrine	Respiratory	Develop-mental
98-82-8	**Isopropylbenzene**	9	8.66	I	I	I, O	I	I	I	I	I
103-65-1	***n*-Propylbenzene[c]**	9	8.94								
622-96-8	1-Methyl-4-ethylbenzene	9	9.07				I			I	
108-67-8	**1,3,5-Trimethylbenzene**	9	9.15	H, I	O	O	O, I	H, O		H	I
95-63-6	**1,2,4-Trimethylbenzene**	9	9.36	H, I	I	I	I	H, I	I	H, I	I
98-06-6	***tert*-Butylbenzene[c]**	10	9.36								
135-98-8	***sec*-Butylbenzene[c]**	10	9.57								
526-73-8	**1,2,3-Trimethylbenzene**	9	9.65	H, I	I	I	I	H, I	I	H, I	
141-93-5	1,3-Diethylbenzene	10	9.91	O			O				
105-05-5	1,4-Diethylbenzene	10	9.96	O	O	O	O	O			O
104-51-8	***n*-Butylbenzene**	10	9.96		O	O	O		O		O
135-01-3	1,2-Diethylbenzene	10	9.96	O			O				
64742-95-6	**HFAN**	9−10	NA	I	O, I	O, I	O, I	O, I	O		O, I
NA	Diethylbenzenes (mixture)	10	NA	O, I			O, I				

[a]Includes human and animal studies meeting inclusion criteria. **Bolded** compounds and mixtures have at least one oral or inhalation toxicity value available (see Table 4).
[b]Compounds are arranged by increasing EC number.
[c]In the absence of human or animal data, screening toxicity values were derived using appropriate analogue chemicals (ethylbenzene and isopropylbenzene) in the PPRTV assessments of these compounds.

C = carbon; EC = equivalent carbon; H = human data; HFAN = high flash aromatic naphtha; I = animal inhalation studies; NA = not applicable; O = animal oral studies.

Critical effects used to derive oral or inhalation toxicity values for the aromatic medium carbon range fraction compounds and mixtures include neurological effects (decreased pain sensitivity), hepatic toxicity (hepatocellular hypertrophy), renal toxicity (increased kidney weight and histopathology), decreased body weight, hematological toxicity, endocrine system toxicity, and developmental toxicity (decreased fetal/pup body weights and delayed skeletal ossification). The available data for most of the aromatic medium carbon range compounds and mixtures are limited for endpoints other than body weight and are altogether absent for three members of the fraction (*n*-propylbenzene, and *tert*- and *sec*-butylbenzene). A majority of toxicity data are from subchronic studies of the inhalation route of exposure, and few compounds have been tested for toxicity following chronic oral or inhalation exposure.

Based on review of the available oral and inhalation toxicity data, there is evidence that several members of the fraction, and especially TMBs and DEBs, can induce neurological effects; however, most of the compounds in the fraction have not been evaluated for sensitive measures of neurological function. Information across half of the compounds (oral and inhalation exposure) composing the fraction suggests that aromatic medium carbon range fraction compounds and mixtures can induce hepatic effects in the form of increased liver weight, often accompanied by histological effects (most frequently hepatocellular hypertrophy via oral exposure). Similarly, data show that several members of the aromatic medium carbon range fraction induce significant increases in relative kidney weight after oral or inhalation exposure; this effect was seen (infrequently) in conjunction with serum chemistry changes (i.e., increased blood urea nitrogen [BUN] after oral exposure to 1,4-DEB) and in the absence of corresponding histological changes (other than rat-specific male nephropathy). Data on body-weight effects after oral and inhalation exposure to a variety of aromatic medium carbon range fraction compounds and mixtures indicate that members of the fraction can be expected to induce body-weight reductions generally at high doses.

The available data are not considered adequate to evaluate consistency in effects or potencies across fraction members for hematological or endocrine effects. Because there are data for only five compounds, data are insufficient to determine if respiratory effects are consistently associated with inhalation exposure to members of the aromatic medium carbon range fraction (there are no oral exposure data on respiratory effects). Finally, data from oral and inhalation developmental toxicity studies consistently identify decreased fetal body weights and delays in skeletal development for several members of the aromatic medium carbon range fraction.

3.2. CANCER EVIDENCE

3.2.1. Human Studies

No human studies were available to address the carcinogenic potential of the TMB isomers or other members of the aromatic medium carbon range fraction by any route of exposure.

3.2.2. Animal Studies—Oral

A single carcinogenicity study in rats orally exposed to 1,2,4-TMB for 104 weeks was identified Clark et al., 1989 as cited in U.S. EPA (2009c). The only noteworthy finding was a nonsignificant increase in the incidence of neuroesthesioepitheliomas (3/100 treated animals based on the combined sexes compared to 0/100 controls). Several study limitations were apparent, including use of one rodent species, treatment at a single dose level (800 mg/kg-day), and lack of quantitative mortality data ("slight" or "intermediate" reductions in survival were

reported). Therefore, the available oral data are not sufficient to adequately assess the carcinogenic potential of 1,2,4-TMB (or other members of the aromatic medium carbon range fraction).

3.2.3. Animal Studies—Inhalation

No neoplasms were reported in rats treated with HFAN for 12 months Clark et al., 1989 as cited in U.S. EPA (2009c).

New studies identified in the PubMed searches included a 105-week chronic toxicity/ carcinogenicity study of isopropylbenzene in rats and mice by the National Toxicology Program Ntp (2009). Statistically significantly increased incidences of respiratory epithelial adenomas of the nose in both sexes and renal adenoma or carcinoma (combined) in males were observed in rats. Increased interstitial cell adenomas were also reported in the male testis; however, the NTP report stated that these were possibly related to isopropylbenzene exposure. While the incidence of interstitial cell adenomas reported in the highest dose group in the male rats was statistically significantly increased compared to the control group and there was a positive trend in the incidences reported among all exposed groups, the incidence in the high-dose group was within the range for historical chamber controls when studies with all exposure routes were considered. Interstitial cell hyperplasia and adenoma are common proliferative lesions in F344/N rats (i.e., the test species) and reportedly will develop in nearly all male rats of this strain that are allowed to complete their natural life span Ntp (2009). In mice, the incidences of alveolar/ bronchiolar adenomas were significantly increased in both sexes; increased incidences of hemangiosarcomas and follicular cell adenomas in males[4] (possibly related to exposure) and hepatocellular adenomas or carcinomas in females also were reported. Based on these data, the NTP concluded that there was clear evidence of carcinogenicity in male rats and male and female mice, and some evidence of carcinogenic activity in female rats.

No studies evaluating carcinogenicity were available for other members of the aromatic medium carbon range fraction.

3.2.4. Summary of Cancer Evidence

Few data are available to assess the carcinogenic potential of compounds and mixtures in the aromatic medium carbon range fraction. No human data were identified. Animal data are limited to 1,2,4-TMB and isopropylbenzene. Several limitations were identified in the carcinogenicity study of 1,2,4-TMB. Only data from a newly identified study for isopropylbenzene Ntp (2009) are considered adequate to assess carcinogenic potential of individual fraction members. At this time, U.S. EPA has not formally evaluated the Ntp (2009) study and has not estimated the cancer potency associated with the study results.

[4]NTP described the increased incidence of hemangiosarcomas in males as "equivocal;" these tumors were observed only in the highest dose group. The incidences of follicular cell adenoma increased with a statistically significant positive trend in male mice; however, the incidence in the highest dose group was at the upper end of the historical ranges for chamber controls in inhalation studies and for historical controls (all exposure routes). NTP described these increases to be "possibly related to cumene exposure."

4. TOXICOKINETIC CONSIDERATIONS

Reviews of toxicokinetic information on aromatic medium carbon chain length hydrocarbons have been performed by McKee et al. (2015) and Infante and Bingham (2012). In general, these chemicals are well absorbed by oral and inhalation exposure and distributed widely throughout the body, with initial concentration in adipose tissue. Metabolism is efficient and occurs primarily via oxidation and conjugation of the alkyl side chains off the aromatic ring. These water-soluble metabolites are rapidly eliminated in the urine.

Compounds in the aromatic medium carbon range fraction are readily absorbed following oral exposure. F344 rats exposed to radiolabeled isopropylbenzene (cumene) by gavage showed maximum blood concentration levels 4 hours after dosing (the earliest time point sampled) at 33 mg/kg and 8−16 hours after dosing at 1,350 mg/kg U.S. EPA (1997). Based on recovery of urinary metabolites (time point not specified), absorption exceeded 70%. Absorption of *p*-isopropyltoluene (cymene) was at least 60−80% in rats and guinea pigs treated at 100 mg/kg and 52−74% in rats and various marsupial species treated at 50 or 200 mg/kg, based on urinary metabolites excreted within 48 hours U.S. EPA (2011). Absorption of *n*- and *tert*-butylbenzene was similarly found to exceed 66−81% in rabbits U.S. EPA (2012b). More than 99% of an orally administered dose of 1,2,4-TMB was absorbed (and subsequently eliminated) within 24 hours in rats McKee et al. (2015).

Absorption following inhalation exposure is rapid and extensive. Isopropylbenzene was detected in the blood of F344 rats within 5 minutes of the start of inhalation exposure U.S. EPA (1997). Based on recovery of urinary metabolites (time point not specified), absorption exceeded 70% in rats exposed to 100, 500, or 1,500 ppm isopropylbenzene U.S. EPA (1997). Mean respiratory tract retention was reported to be 50% (45−64%) in volunteers exposed to isopropylbenzene at 240, 480, or 720 mg/m^3 for 8-hour periods U.S. EPA (1997). Respiratory retentions for the three TMB isomers were approximately 70% in human subjects exposed at 5−150 mg/m^3 for 8 hours and 60% in subjects exposed to 2−25 ppm (10−125 mg/m^3) for 2 hours while performing light activity McKee et al. (2015); Infante and Bingham (2012). Consistent with these results, measured blood-air partition coefficients for these compounds are high in humans and laboratory animals. Human blood-air partition coefficients are 47 for *n*-propylbenzene and 37 for isopropylbenzene U.S. EPA (2009d). Blood-air partition coefficients for TMB isomers (1,2,4-TMB, 1,2,3-TMB, and 1,3,5-TMB, respectively) are 59.1, 66.5, and 43.0 in humans and 57.7, 62.6, and 55.7 in rats U.S. EPA (2016b).

Compounds in the aromatic medium carbon range fraction are widely distributed in the body after inhalation or oral exposure. Studies with isopropylbenzene, TMB isomers, and *tert*-butylbenzene all showed elevated concentrations of the administered chemical in fat (also stomach by oral route) >> kidney > liver > brain >blood in rats after exposure, regardless of route U.S. EPA (2016a, 2012b, 1997). Levels in fat were 10- to 100-fold higher than in other tissues. Modeling of tissue-air partition coefficients suggests that tissue distribution in humans would be similar to that seen in rats U.S. EPA (2016a; McKee et al. (2015). Due to the high lipophilicity of these chemicals, it has been estimated that 85% of alkylbenzene in the blood is bound to red blood cells (RBCs) U.S. EPA (2012b). TMBs and *tert*-butylbenzene have been shown to cross the placenta U.S. EPA (2016a; Infante and Bingham (2012).

Similar to absorption, metabolism of aromatic medium carbon range compounds is rapid and extensive. Metabolism proceeds primarily by oxidation of side chains on the aromatic ring to form the corresponding alcohols and carboxylic acids, followed by conjugation with glycine (hippuric acid), cysteine (mercapturic acid), glucuronic acid, or sulfate U.S. EPA (2016a); McKee et al. (2015); U.S. EPA (2012b, 2011, 2009d, 1997). Example chemicals (and principal metabolites) are isopropylbenzene (2-phenyl-2-propanol and its glucuronide and sulfate conjugates) U.S. EPA (1997), *n*-propylbenzene (glucuronides of ethylphenyl carbinol and benzyl methyl carbinol) U.S. EPA (2009d), 1,2,4-TMB (3,4-dimethylhippuric acid) U.S. EPA (2016a), 1,3,5-TMB (3,5-dimethylhippuric acid) McKee et al. (2015), *tert*-butylbenzene (2,2-dimethyl-2-phenylethyl glucuronide) U.S. EPA (2012b), and isopropyltoluene (2-[4-methylphenyl]propan-1-ol, 2-[4-methylphenyl]propan-2-ol) U.S. EPA (2011). Oxidation of the aromatic ring to form the corresponding phenol is a minor metabolic pathway for at least some of these chemicals U.S. EPA (2016a); Infante and Bingham (2012; U.S. EPA (2011). Where data in multiple species are available, metabolic profiles are similar in rats, rabbits, and humans U.S. EPA (2016a); McKee et al. (2015). Metabolism of aromatic medium carbon range compounds occurs in the liver, lung, and other tissues [e.g., kidney, adrenal, brain, and bone marrow U.S. EPA (2012b, 2009d, 1997)]. Several of these compounds have been shown to induce metabolic enzymes, and therefore, their own metabolism McKee et al. (2015; U.S. EPA (2012b, 2009d). There is some experimental evidence for saturation of metabolism at high exposure levels: blood concentrations of 4-ethyltoluene in rats were 10-fold higher after a single 6-hour exposure at 1,000 mg/m^3 than at 250 mg/m^3 [fourfold difference in exposure concentration McKee et al. (2015)].

Metabolites of aromatic medium carbon range compounds are water soluble and rapidly excreted in the urine McKee et al. (2015; U.S. EPA (2012b). Less than 1% of the absorbed fraction remained in the body 72 hours after inhalation exposure of isopropylbenzene at 1,200 ppm to rats U.S. EPA (1997). Similarly, >99% of an oral dose of 1,2,4-TMB was eliminated as metabolites in the urine within 24 hours after dosing in rats McKee et al. (2015). In studies of *p*-isopropyltoluene, at least 60−80% of an oral dose in rats and guinea pigs and 52−74% of an oral dose in rats and various marsupial species was excreted as metabolites in the urine within 48 hours U.S. EPA (2011). Small amounts of unchanged parent compound may also be found in the urine U.S. EPA (2016a, 2012b). Following inhalation exposure, unchanged parent compound may be exhaled via the lungs. Human subjects who retained 60% of inhaled TMB in the lung subsequently exhaled approximately 30% of the retained material Infante and Bingham (2012). In one human study, elimination of 1,3,5-TMB via breath was biphasic, with an initial half-life of 60 minutes and a terminal half-life of 600 minutes U.S. EPA (2016a; McKee et al. (2015). Breath concentrations of 1,3,5-TMB in this study returned to pre-exposure levels within 24 hours McKee et al. (2015).

5. MECHANISTIC CONSIDERATIONS AND GENOTOXICITY

Mechanistic information for health effects associated with exposure to compounds in the aromatic medium carbon range is limited. There is evidence that renal histopathology induced by isopropylbenzene in male rats reflects an alpha 2u-globulin (α2u-g)-specific nephropathy that is specific to male rats, and is therefore not an appropriate endpoint for human health risk assessment U.S. EPA (1997). Similar changes were noted in one study of an HFAN mixture U.S. EPA (2009c). Renal effects in studies of other fraction members were limited primarily to increases in kidney weight (see discussion of renal effects in Section 3.1). For isopropylbenzene, increases in kidney weight were unrelated to α2u-g-specific nephropathy, as they occurred in females as well as males [increased kidney weight in females was the critical effect for both the RfD and RfC for isopropylbenzene U.S. EPA (1997)]. Therefore, increases in renal weight associated with other fraction members does not necessarily result from α2u-g-specific nephropathy. In fact, there is no evidence for α2u-g-specific nephropathy among specific fraction member compounds other than isopropylbenzene (HFAN is a mixture of C8−C10 aromatics that can include isopropylbenzene).

Genotoxicity data for aromatic medium carbon range compounds primarily indicate little to no genotoxic potential. Almost all relevant mixtures or compounds were negative with or without metabolic activation in in vitro tests for point mutations in bacteria or mammalian cells U.S. EPA (2016a; OECD (2012a, b; U.S. EPA (2012b, 2011, 2009c, d; Oecd (2007; U.S. EPA (1997; Oecd (1994). The only exception was 1,2,3-TMB, which produced reverse mutations in *Salmonella* without, but not with, metabolic activation U.S. EPA (2016a). All three TMB isomers were at least weakly positive for sister chromatid exchange (SCE) in mice tested in vivo U.S. EPA (2016a). However, the larger C9 fraction to which TMB belongs was negative for SCE in Chinese hamster ovary (CHO) cells in vitro Oecd (2012b). Other tests of fraction members (including the TMB isomers) for clastogenicity (chromosomal aberrations [CAs] or micronucleus [MN] formation) in rodents in vivo or in rodent cells in vitro were negative or equivocal U.S. EPA (2016a; OECD (2012a, b; U.S. EPA (2012b; Oecd (2007; U.S. EPA (1997; Oecd (1994). By contrast, clastogenicity was reported for a commercial HFAN mixture in human lymphocytes with activation U.S. EPA (2009c). This HFAN mixture also was reported to produce deoxyribonucleic acid (DNA) damage in *Escherichia coli* without activation U.S. EPA (2009c). The only other finding relevant to DNA damage was equivocal evidence for unscheduled DNA synthesis (UDS) in rat hepatocytes by isopropylbenzene U.S. EPA (1997). There was also equivocal evidence for BALB/3T3 cell transformation by isopropylbenzene U.S. EPA (1997).

6. DERIVATION OF PROVISIONAL VALUES

6.1. DERIVATION OF ORAL REFERENCE DOSES

Subchronic provisional reference doses (p-RfDs) or RfDs are available for eight constituents of the fraction. The critical effects for these subchronic toxicity values are liver and kidney toxicity (*n*-propylbenzene, based on analogy to ethylbenzene), decreased pain sensitivity (1,3,5-, 1,2,4-, and 1,2,3-TMB), increased kidney weight (*tert*- and *sec*-butylbenzene, based on analogy to the chronic RfD for isopropylbenzene), liver histology (*n*-butylbenzene), and anemia (HFAN). There are nine available chronic RfDs for constituent compounds (isopropylbenzene, in addition to the compounds identified above). The chronic RfDs are based on the same studies and the same points of departure (PODs) as the corresponding subchronic RfDs. The chronic RfD for isopropylbenzene is based on increased kidney weights. Table 6 summarizes the subchronic and chronic RfDs for constituent compounds and mixtures, with PODs, uncertainty factors, critical effects, and confidence descriptors. As shown in Table 6 and discussed in Appendix B, the data available to assess consistency in critical effects across members of the fraction are limited for effects on endpoints other than body weight. The potencies with RfDs being within one order of magnitude of one another are comparable.

Table 6. Available RfD Values for Aromatic Medium Carbon Range Fraction (C9−C10, EC9−EC < 11)[a]

Indicator Chemical or Components	POD (mg/kg-d)	POD Type	UF_C	UF Components	p-RfD or RfD (mg/kg-d)	Confidence in p-RfD or RfD[a]	Critical Effect(s)	Species, Mode, and Duration	Primary Reference (source)
Subchronic									
n-Propylbenzene (C9, EC8.94)	97.1	$NOEL_{ADJ}$	1,000	UF_A, UF_H, UF_S	*0.1*[b]	Low	Based on ethylbenzene as an analogue; increased liver and kidney weights (hepatic, urinary); histopathologic changes in kidney	Rat, gavage, 5 d/wk for 182 d	Wolf (1956) as cited in U.S. EPA (2009d)
1,3,5-Trimethylbenzene (C9, EC9.15)	**3.5**	**BMDL (HED)**	**100**	**UF_A, UF_D, UF_H**	**0.04**	**Low**	**Decreased pain sensitivity in male Wistar rats[c] (nervous)**	**Rat, 6 h/d, 5 d/wk for 13 wk**	**Korsak and Rydzynski (1996) as cited in U.S. EPA (2016b)**
1,2,4-Trimethylbenzene (C9, EC9.36)	**3.5**	**BMDL (HED)**	**100**	**UF_A, UF_D, UF_H**	**0.04**	**Low**	**Decreased pain sensitivity in male Wistar rats[c] (nervous)**	**Rat, 6 h/d, 5 d/wk for 13 wk**	**Korsak and Rydzynski (1996) as cited in U.S. EPA (2016b)**
tert-Butylbenzene (C10, EC9.36)	110	$NOAEL_{ADJ}$	1,000	UF_A, UF_D, UF_H, UF_S	*0.1*[b]	Low	Based on isopropylbenzene as an analogue; increased kidney weight (urinary)	Rat, 5 d/wk for 194 d	Wolf (1956) as cited U.S. EPA (2012b)
sec-Butylbenzene (C10, EC9.57)	110	$NOAEL_{ADJ}$	1,000	UF_A, UF_D, UF_H, UF_S	*0.1*[b]	Low	Based on isopropylbenzene as an analogue; increased kidney weight (urinary)	Rat, 5 d/wk for 194 d	Wolf (1956) as cited in U.S. EPA (2012a)
1,2,3-Trimethylbenzene (C9, EC9.65)	**3.5**	**BMDL (HED)**	**100**	**UF_A, UF_D, UF_H**	**0.04**	**Low**	**Decreased pain sensitivity in male Wistar rats[c] (nervous)**	**Rat, 6 h/d, 5 d/wk for 13 wk**	**Korsak and Rydzynski (1996) as cited in U.S. EPA (2016b)**

Table 6. Available RfD Values for Aromatic Medium Carbon Range Fraction (C9−C10, EC9−EC < 11)[a]

Indicator Chemical or Components	POD (mg/kg-d)	POD Type	UF_C	UF Components	p-RfD or RfD (mg/kg-d)	Confidence in p-RfD or RfD[a]	Critical Effect(s)	Species, Mode, and Duration	Primary Reference (source)
n-Butylbenzene (C10, EC9.96)	137	BMDL	1,000	UF_A, UF_D, UF_H	0.1[b]	Low	Increased incidence of hepatocellular hypertrophy in F_0 and F_1 parent male rats (hepatic)	Rat, gavage, two-generation	Izumi et al. (2005) as cited in U.S. EPA (2010)
HFAN (C9−C10)	85	BMDL	300	UF_A, UF_D, UF_H	*0.3*[b]	Low	Mild anemia, evidenced by a decrease in RBC count (hematological)	Dog, gelatin capsules, 13 wk	Biodynamics (1990b) as cited in U.S. EPA (2009c)
Chronic									
Isopropylbenzene (C9, EC8.66)	110	$NOAEL_{ADJ}$	1,000	UF_A, UF_D, UF_H, UF_S	0.1	Low−medium	Increased average kidney weight in female Wistar rats (urinary)	Rat, 5 d/wk for 194 d	Wolf (1956) as cited in U.S. EPA (1997)
n-Propylbenzene (C9, EC8.94)	97.1	$NOEL_{ADJ}$	1,000	UF_A, UF_H, UF_S	*0.1*[b]	Low	Based on ethylbenzene as an analogue; increased liver and kidney weights (hepatic, urinary)	Rat; gavage; 5 d/wk for 182 d	Wolf (1956) as cited in U.S. EPA (2009a)
1,3,5-Trimethylbenzene (C9, EC9.15)	**3.5**	**BMDL (HED)**	**300**	**UF_A, UF_D, UF_H, UF_S**	**0.01**	**Low**	**Decreased pain sensitivity in male Wistar rats[c] (nervous)**	**Rat, 6 h/d, 5 d/wk for 13 wk**	**Korsak and Rydzynski (1996) as cited in U.S. EPA (2016b)**
1,2,4-Trimethylbenzene (C9, EC9.36)	**3.5**	**BMDL (HED)**	**300**	**UF_A, UF_D, UF_H, UF_S**	**0.01**	**Low**	**Decreased pain sensitivity in male Wistar rats[c] (nervous)**	**Rat, 6 h/d, 5 d/wk for 13 wk**	**Korsak and Rydzynski (1996) as cited in U.S. EPA (2016b)**

Table 6. Available RfD Values for Aromatic Medium Carbon Range Fraction (C9−C10, EC9−EC < 11)[a]

Indicator Chemical or Components	POD (mg/kg-d)	POD Type	UF_C	UF Components	p-RfD or RfD (mg/kg-d)	Confidence in p-RfD or RfD[a]	Critical Effect(s)	Species, Mode, and Duration	Primary Reference (source)
tert-Butylbenzene (C10, EC9.36)	110	$NOAEL_{ADJ}$	1,000	UF_A, UF_D, UF_H, UF_S	*0.1*[b]	Low	Based on isopropylbenzene as an analogue; increased kidney weight (urinary)	Rat, 5 d/wk for 194 d	Wolf (1956) as cited U.S. EPA (2012b)
sec-Butylbenzene (C10, EC9.57)	110	$NOAEL_{ADJ}$	1,000	UF_A, UF_D, UF_H, UF_S	*0.1*[b]	Low	Based on isopropylbenzene as an analogue; increased kidney weight (urinary)	Rat, 5 d/wk for 194 d	Wolf (1956) as cited in U.S. EPA (2012a)
1,2,3-Trimethylbenzene (C9, EC9.65)	**3.5**	**BMDL (HED)**	**300**	**UF_A, UF_D, UF_H, UF_S**	**0.01**	**Low**	**Decreased pain sensitivity in male Wistar rats[c] (nervous)**	**Rat, 6 h/d, 5 d/wk for 13 wk**	**Korsak and Rydzynski (1996) as cited in U.S. EPA (2016b)**
n-Butylbenzene (C10, EC9.96)	137	BMDL	3,000	UF_A, UF_D, UF_H, UF_S	0.05[b]	Low	Increased incidence of hepatocellular hypertrophy in F_0 and F_1 parent male Crj:CD (SD) IGS rats (hepatic)	Rat, gavage, two-generation	Izumi et al. (2005) as cited in U.S. EPA (2010)

Table 6. Available RfD Values for Aromatic Medium Carbon Range Fraction (C9−C10, EC9−EC < 11)[a]

Indicator Chemical or Components	POD (mg/kg-d)	POD Type	UF_C	UF Components	p-RfD or RfD (mg/kg-d)	Confidence in p-RfD or RfD[a]	Critical Effect(s)	Species, Mode, and Duration	Primary Reference (source)
HFAN (C9−C10)	85	BMDL	3,000	UF_A, UF_D, UF_H, UF_S	*0.03*[b]	Low	Mild anemia, evidenced by a decrease in RBC count (hematological)	Dog, gelatin capsules, 13 wk	Biodynamics (1990b) as cited in U.S. EPA (2009c)

[a]**Bolded** row shows the compound and toxicity value selected as the indicator chemical for the fraction if analytical chemistry data do not identify concentrations of individual chemicals composing this fraction.
[b]Toxicity values are provisional values obtained from an existing PPRTV assessment. Values in *italics* are screening provisional values obtained from an existing PPRTV assessment. Screening provisional values are not assigned confidence statements; however, confidence in these values is presumed to be low. Screening provisional values are derived when the available data do not meet the requirements for deriving a provisional toxicity value.
[c]Toxicity values based on route-to-route extrapolation (inhalation to oral) using a modified PBPK model.

ADJ = adjusted; BMDL = benchmark dose lower confidence limit; C = carbon; EC = equivalent carbon; HED = human equivalent dose; HFAN = high flash aromatic naphtha; NOAEL = no-observed-adverse-effect level; NOEL = no-observed-effect level; PBPK = physiologically based pharmacokinetic; POD = point of departure; PPRTV = provisional peer-reviewed toxicity value; RBC = red blood cell; p-RfD = provisional reference dose; RfD = reference dose; SD = standard deviation; UF = uncertainty factor; UF_A = interspecies uncertainty factor; UF_C = composite uncertainty factor; UF_D = database uncertainty factor; UF_H = intraspecies uncertainty factor; UF_S = subchronic-to-chronic uncertainty factor.

6.1.1. Oral Noncancer Assessment Using the Indicator Chemical Method for the Aromatic Medium Carbon Range Fraction

If available analytical chemistry data do not identify concentrations of individual chemicals composing this fraction, the subchronic and chronic p-RfDs (0.04 and 0.01 mg/kg-day, respectively) for TMBs are recommended as the indicator chemical for the aromatic medium carbon range fraction U.S. EPA (2016a). Given the limited data, the compounds that resulted in the lowest RfDs for these effects and target tissues were considered as the basis for indicator chemical selection. The RfDs for the TMBs, derived by route-to-route extrapolation (from inhalation to oral) using a modified physiologically based pharmacokinetic (PBPK) model, are based on neurological effects (decreased pain sensitivity; see Section 2.3). Available data generally support the nervous system as a target of the aromatic medium carbon compounds. Evaluation of available data as discussed in Appendix B suggests that use of the TMBs' RfD values is reasonably anticipated to be protective for effects associated with exposures to other constituents of the fraction. Users of the indicator chemical method should understand that there could be more uncertainty associated with the application of this toxicity value to the aromatic medium carbon range fraction than for its derivation in U.S. EPA (2016b).

The IRIS review of TMBs cited Korsak and Rydzynski (1996) as cited in U.S. EPA (2016b) as the principal study for the subchronic and chronic RfDs. A study summary was not provided in the IRIS assessment; however, the U.S. EPA (2007) PPRTV for 1,2,4-TMB (the compound that served as the driver for the toxicity value) provided the following summary:

> *In the subchronic experiment, rats were exposed to 1,2,4-trimethylbenzene at concentrations of 0, 25, 100 or 250 ppm (0, 123, 491 or 1,227 mg/m^3), 6 hours/day, 5 days/week for 3 months and observed for exposure-related clinical signs and body weight effects (Korsak and Rydzyński, 1996). Rotarod performance and hot-plate behavior were measured as indices of the neurotoxicity of trimethylbenzene isomers. Rotarod performance was tested prior to start of the study, weekly during exposure, and 2 weeks after the termination of the exposure. Hot-plate behavior was tested immediately after termination of the exposure. Fisher's exact test was used for analysis of rotarod performance and the Kruskall-Wallis test used for changes in pain sensitivity (hot plate behavior). Exposures to 1,2,4-trimethylbenzene did not result in any apparent body weight effects or clinical signs of toxicity. However, exposure-related indicators of neurotoxicity were noted. Rotarod performance failure increased in a concentration-related manner in the groups exposed to 1,2,4-trimethylbenzene, but reached the level of statistical significance (40% failure; $p < 0.05$) only in the highest (1,227 mg/m^3) exposure group following 8 or 13 weeks of exposure. The incidence of rotarod performance failure in control rats was 0% throughout the study period. Although the mean rotarod performance failure rate in the highest exposure group remained at 30% after a 2-week recovery period, the rate was not significantly different from controls. Pain-sensitivity was also decreased in a concentration dependent manner (evidenced by increased latency of the paw-lick response). As shown in Table 2, the increased latency reached the level of statistical significance in the 491- and 1,227-mg/m^3 groups. After a 2-week recovery period, the highest (1,227 mg/m^3) exposure group no longer exhibited a significant difference in pain sensitivity, relative to controls. This study identified*

a NOAEL of 123 mg/m^3 and a LOAEL of 491 mg/m^3 (6 hours/day, 5 days/week) for significantly decreased pain sensitivity.

U.S. EPA (2016a) used PBPK model estimates of internal blood dose metrics for 1,2,4-TMB coupled with benchmark dose (BMD) modeling to generate a POD. First, BMD modeling of the data (for decreased pain sensitivity following 1,2,4-TMB exposure) identified a benchmark concentration lower confidence limit with one standard deviation ($BMCL_{1SD}$) of 140.54 mg/m^3 (based on external air concentrations and subsequently adjusted for continuous exposure). Using the available PBPK model, the $BMCL_{1SD}$ was converted to a duration-adjusted POD (POD_{ADJ}) of 0.099 mg/L. The POD_{ADJ} value represents the internal blood dose metric of average weekly venous blood concentration of 1,2,4-TMB, which is considered by the U.S. EPA to be the most relevant internal dose metric. To derive an oral toxicity value based on these (inhalation) data, an oral exposure component was added to the PBPK model by the U.S. EPA. The model assumed 100% absorption of ingested 1,2,4-TMB. The human PBPK model was run to estimate a human BMDL (HED) that would result from the same weekly average venous blood concentration observed in the POD_{ADJ} in animals (0.099 mg/L). The resultant BMDL (HED) of 3.5 mg/kg-day was used to derive the subchronic and chronic RfDs for 1,2,4-TMB, which was applied to all TMBs (see Table 6).

Confidence in the principal study was low to medium. Although the study was well-conducted, peer-reviewed, and amenable to dose-response analyses (i.e., used an appropriate number of exposure groups), there was uncertainty with respect to the actual concentrations achieved (only target concentrations were reported), and reported measures of variance (type of measures [e.g., standard deviation] not explicitly specified). Confidence in the oral database (for TMBs) was low, because only acute neurotoxicity data and one subchronic toxicity study (for 1,3,5-TMB) were available. Owing to low confidence in the oral database, low to medium confidence in the principal study, and uncertainty associated with the applicability of the PBPK model for route-to-route extrapolation, confidence in the subchronic and chronic RfDs was low. While toxicological data from mixtures such as HFAN might be preferred in some cases, the p-RfD for HFAN is based on a screening value, and the Agency has more confidence in EPA's IRIS TMB oral assessments as the indicator chemical.

6.1.2. Alternative Oral Noncancer Assessment Using the Hazard Index Method for the Aromatic Medium Carbon Range Fraction

If the available analytical chemistry data quantify the concentrations of TMBs, *n*-propylbenzene, *n*-butylbenzene, *sec*-butylbenzene, *tert*-butylbenzene, or isopropylbenzene separately from the remainder of the aromatic medium carbon range fraction, it is recommended that HQs for the individual chemicals with analytical data be calculated and an HI for the mixture be developed using the calculated HQs.

For subchronic oral exposures, the following subchronic RfDs or p-RfDs can be used as the denominator in the HQ equations: TMBs (0.04 mg/kg-day), *n*-propylbenzene (0.1 mg/kg-day), *n*-butylbenzene (0.1 mg/kg-day), *sec*-butylbenzene (0.1 mg/kg-day), and *tert*-butylbenzene (0.1 mg/kg-day). In this alternative approach, the subchronic RfD for TMBs (0.04 mg/kg-day) is recommended for use with the remainder of the fraction, including any other fraction members analyzed individually (see Table 6).

For chronic oral exposures, the following chronic RfDs or p-RfDs can be used as the denominator in the HQ equations: TMBs (0.01 mg/kg-day), isopropylbenzene (0.1 mg/kg-day), *n*-propylbenzene (0.1 mg/kg-day), *n*-butylbenzene (0.05 mg/kg-day), *sec*-butylbenzene (0.1 mg/kg-day), and *tert*-butylbenzene (0.1 mg/kg-day). In this alternative approach, the chronic RfD for TMBs (0.01 mg/kg-day) is recommended for use with the remainder of the fraction, including any other fraction members analyzed individually (see Table 6).

In some cases, toxicological data from mixtures such as HFAN might be preferred; however, the p-RfD for HFAN is based on a screening value. The Agency has more confidence in an HI approach as an alternative to the indicator chemical approach than for the surrogate mixture approach for this fraction.

6.2. DERIVATION OF INHALATION REFERENCE CONCENTRATIONS

The available subchronic and chronic RfC values, with PODs, uncertainty factors, critical effects, and confidence ratings are presented in Table 7. As shown in the table, there are subchronic and chronic RfCs or provisional reference concentrations (p-RfCs) for one mixture (HFAN) and four individual compounds (*n*-propylbenzene and 1,3,5-, 1,2,4-, and 1,2,3-TMB) relevant to the aromatic medium carbon range fraction. In addition, there is a chronic RfC for isopropylbenzene. Critical effects for the RfCs included maternal toxicity (decreased body weight), developmental toxicity, increased adrenal and kidney weights, and decreased pain sensitivity.

Table 7. Available RfC Values for Aromatic Medium Carbon Range Fraction (C9−C10, EC9−EC < 11)[a]

Indicator Chemical or Components	POD	POD Type (all are HECs)	UF_C	UF Components	RfC or p-RfC (mg/m^3)	Confidence in p-RfC or RfC[a]	Critical Effect(s)	Species, Mode, and Duration	Primary Reference (source)
Subchronic									
n-Propylbenzene (C9, EC8.94)	434	NOAEL	300	UF_A, UF_H, UF_D	*I*[b]	Low	Based on ethylbenzene as an analogue; developmental toxicity (developmental)	Rat, 6−7 h/d, 7 d/wk for 3 wk prior to mating and GDs 1−19; rabbits, 6−7 h/d, 7 d/wk on GDs 1−24	Andrews (1981) and Hardin (1981) as cited in U.S. EPA (2009a)
1,3,5-Trimethylbenzene (C9, EC9.15)	**18.15**	**BMCL**	**100**	**UF_A, UF_H, UF_D**	**0.2**	**Low−medium**	**Decreased pain sensitivity in male Wistar rats (nervous)**	**Rat, 6 h/d, 5 d/wk for 13 wk**	**Korsak and Rydzynski (1996) as cited in U.S. EPA (2016b)**
1,2,4-Trimethylbenzene (C9, EC9.36)	**18.15**	**BMCL**	**100**	**UF_A, UF_H, UF_D**	**0.2**	**Low−medium**	**Decreased pain sensitivity in male Wistar rats (nervous)**	**Rat, 6 h/d, 5 d/wk for 13 wk**	**Korsak and Rydzynski (1996) as cited in U.S. EPA (2016b)**
1,2,3-Trimethylbenzene (C9, EC9.65)	**18.15**	**BMCL**	**100**	**UF_A, UF_H, UF_D**	**0.2**	**Low−medium**	**Decreased pain sensitivity in male Wistar rats (nervous)**	**Rat, 6 h/d, 5 d/wk for 13 wk**	**Korsak and Rydzynski (1996) as cited in U.S. EPA (2016b)**
HFAN (C9−C10)	125	LOAEL	100	UF_A, UF_H, UF_L	1[b]	Moderate	Decreased maternal body weight vs. controls (reproductive) in CD-1 mice	Mouse, 6 h/d, 7 d/wk on GDs 6−15	McKee et al. (1990) as cited in U.S. EPA (2009c)

Table 7. Available RfC Values for Aromatic Medium Carbon Range Fraction (C9−C10, EC9−EC < 11)[a]

Indicator Chemical or Components	POD	POD Type (all are HECs)	UF_C	UF Components	RfC or p-RfC (mg/m^3)	Confidence in p-RfC or RfC[a]	Critical Effect(s)	Species, Mode, and Duration	Primary Reference (source)
Chronic									
Isopropylbenzene (C9, EC8.66)	435	NOAEL	1,000	UF_A, UF_d, UF_H, UF_S	0.4	Medium	Increased kidney weights in female rats and adrenal weights in male and female F344 rats (endocrine, urinary)	Rat, 6 h/d, 5 d/wk for 13 wk	Cushman (1995) as cited in U.S. EPA (1997)
n-Propylbenzene (C9, EC8.94)	434	NOAEL	300	UF_A, UF_D, UF_H	*I*[b]	Low	Based on ethylbenzene as an analogue; developmental toxicity (developmental)	Rat, 6−7 h/d, 7 d/wk for 3 wk prior to mating and GDs 1−19; rabbit, 6−7 h/d, 7 d/wk on GDs 1−24	Andrews (1981) and Hardin (1981) as cited in U.S. EPA (2009a)
1,3,5-Trimethylbenzene (C9, EC9.15)	**18.15**	**BMCL**	**300**	**UF_A, UF_D, UF_H, UF_S**	**0.06**	**Low−medium**	**Decreased pain sensitivity in male Wistar rats (nervous)**	**Rat, 6 h/d, 5 d/wk for 13 wk**	**Korsak and Rydzynski (1996) as cited in U.S. EPA (2016b)**
1,2,4-Trimethylbenzene (C9, EC9.36)	**18.15**	**BMCL**	**300**	**UF_A, UF_D, UF_H, UF_S**	**0.06**	**Low−medium**	**Decreased pain sensitivity in male Wistar rats (nervous)**	**Rat, 6 h/d, 5 d/wk for 13 wk**	**Korsak and Rydzynski (1996) as cited in U.S. EPA (2016b)**
1,2,3-Trimethylbenzene (C9, EC9.65)	**18.15**	**BMCL**	**300**	**UF_A, UF_D, UF_H, UF_S**	**0.06**	**Low−medium**	**Decreased pain sensitivity in male Wistar rats (nervous)**	**Rat, 6 h/d, 5 d/wk for 13 wk**	**Korsak and Rydzynski (1996) as cited in U.S. EPA (2016b)**

Table 7. Available RfC Values for Aromatic Medium Carbon Range Fraction (C9−C10, EC9−EC < 11)[a]

Indicator Chemical or Components	POD	POD Type (all are HECs)	UF_C	UF Components	RfC or p-RfC (mg/m^3)	Confidence in p-RfC or RfC[a]	Critical Effect(s)	Species, Mode, and Duration	Primary Reference (source)
HFAN (C9−C10)	125	LOAEL	1,000	UF_A, UF_H, UF_L, UF_S	0.1[b]	Moderate	Decreased maternal body weight vs. controls (reproductive) on GD 15 in CD-1 mice	Mouse, 6 h/d, 7 d/wk on GDs 6−15	McKee et al. (1990) as cited in U.S. EPA (2009c)

[a]**Bolded** row shows the compounds and toxicity value selected as the indicator chemical for the fraction if analytical chemistry data do not identify concentrations of individual chemicals composing this fraction.
[b]Toxicity values are provisional values obtained from an existing PPRTV assessment. Values in *italics* are screening provisional values obtained from an existing PPRTV assessment. Screening provisional values are not assigned confidence statements; however, confidence in these values is presumed to be low. Screening provisional values are derived when the available data do not meet the requirements for deriving a provisional toxicity value.

BMCL = benchmark concentration lower confidence limit; C = carbon; EC = equivalent carbon; GD = gestation day; HEC = human equivalent concentration; HFAN = high flash aromatic naphtha; LOAEL = lowest-observed-adverse-effect level; NOAEL = no-observed-adverse-effect level; POD = point of departure; PPRTV = Provisional Peer-Reviewed Toxicity Value; p-RfC = provisional reference concentration; RfC = reference concentration; UF = uncertainty factor; UF_A = interspecies uncertainty factor; UF_C = composite uncertainty factor; UF_D = database uncertainty factor UF_H = intraspecies uncertainty factor; UF_S = subchronic-to-chronic uncertainty factor.

As shown in Table 7, the values of RfCs for fraction members show consistency across the fraction with respect to the toxicological effects exerted (most frequently neurological and developmental effects). The data show that an indicator chemical identifying effects on these targets would be reasonably anticipated to be representative of the effects of the fraction as a whole. Therefore, the compounds that resulted in the lowest RfCs for these effects were considered as the basis for surrogate selection (see Section 2.3).

6.2.1. Inhalation Noncancer Assessment Using the Indicator Chemical Method for the Aromatic Medium Carbon Range Fraction

If available analytical chemistry data do not identify concentrations of individual chemicals in this fraction, the lowest subchronic and chronic p-RfCs (0.2 mg/m^3 and 0.06 mg/m^3, respectively) for TMBs (0.2 and 0.06 mg/m^3, respectively; see Table 7); these values are recommended as indicator chemicals for the aromatic medium carbon range fraction U.S. EPA (2016a). The RfCs for TMBs are based on neurological effects (decreased pain sensitivity), and available data generally support the nervous system as a target of the aromatic medium carbon compounds. Use of these values is anticipated to be protective for exposure to other constituents based on the available toxicological information (see Appendix B). However, users of the indicator chemical method should understand that there could be more uncertainty associated with the application of this toxicity value to the aromatic medium carbon range fraction than for its derivation in the IRIS assessment U.S. EPA (2016b) and application to individual TMBs in the environment.

The IRIS review of TMBs cited Korsak and Rydzynski (1996) as cited in U.S. EPA (2016b) as the principal study for the subchronic and chronic RfCs. A summary of this study was provided in the preceding section (see Section 6.1). The POD_{ADJ} of 0.099 mg/L (described in the preceding section) was converted to a BMCL (HEC) of 18.15 mg/m^3 based on the available PBPK model; this was used to derive subchronic and chronic RfCs for 1,2,4-TMB, which were applied to all TMBs (see Table 7). As indicated in the preceding section, confidence in the principal study was low to medium. Confidence in the inhalation database was also low to medium. Although acute, short-term, subchronic, and developmental inhalation toxicity studies in rats and mice are available, there were no chronic or developmental neurotoxicity studies. In addition, supporting studies (with respect to the critical effect) were primarily from the same research group. Taken together, confidence in the subchronic and chronic RfCs was also low to medium.

Previously, in the PPRTV TPH Mixtures document U.S. EPA (2009b), the HFAN subchronic and chronic p-RfCs were recommended for assessing noncancer hazards associated with inhalation route exposures to this fraction, based on a 2009 PPRTV assessment U.S. EPA (2009c). In 2016, the U.S. EPA IRIS Program published TMB subchronic and chronic p-RfCs of 0.2 and 0.06 mg/m^3, respectively U.S. EPA (2016a), which are lower than the respective HFAN values of 1 and 0.1 mg/m^3, respectively U.S. EPA (2009c) (see Table 7). Because these are IRIS values rather than PPRTVs, these IRIS single chemical values should be used in the indicator chemical approach rather than HFAN-based surrogate mixture approach. The 2009 TPH mixture assessment indicates that HFAN toxicity values are similar to values for other individual compounds in the fraction, which supports using HFAN as a surrogate for the fraction; however, the 2016 TMB values are much lower than the HFAN values and that logic is not applicable.

6.2.2. Alternative Inhalation Noncancer Assessment Using the Hazard Index Method for the Aromatic Medium Carbon Range Fraction

If the available analytical chemistry data quantify the concentrations of TMBs, *n*-propylbenzene, or isopropylbenzene separately from the remainder of the aromatic medium carbon range fraction, it is recommended that HQs for the individual chemicals with analytical data be calculated and an HI for the mixture be developed using the calculated HQs.

For subchronic inhalation exposures, the subchronic RfCs or p-RfCs for TMBs (0.2 mg/m^3) or *n*-propylbenzene (1.0 mg/m^3) can be used as the denominator in the HQ equations (see Table 7). In this alternative approach, the subchronic RfC for TMBs (0.2 mg/m^3) is recommended for use with the remainder of the fraction, including any other fraction members analyzed individually.

For chronic inhalation exposures, the following chronic RfCs or p-RfCs can be used in the denominator of the HQ equations: TMBs (0.06 mg/m^3), isopropylbenzene (0.4 mg/m^3), and *n*-propylbenzene (1.0 mg/m^3) (see Table 7). In this alternative approach, the chronic RfC for TMBs (0.06 mg/m^3) is recommended for use with the remainder of the fraction, including any other fraction members analyzed individually. As stated in Section 6.2.1, in the U.S. EPA's PPRTV TPH Mixtures document U.S. EPA (2009b), the HFAN subchronic and chronic p-RfCs were recommended for assessing noncancer hazards associated with inhalation route exposures to this fraction, based on a 2009 PPRTV assessment U.S. EPA (2009c). In 2016, the U.S. EPA IRIS Program published TMB subchronic and chronic p-RfCs of 0.2 and 0.06 mg/m^3, respectively (U.S. EPA, 2016a), which are lower than the HFAN values of 1 and 0.1 mg/m^3 U.S. EPA (2009c) (see Table 7). Because these are IRIS values rather than PPRTVs, the U.S. EPA has more confidence in using these IRIS single chemical values in an HI approach rather than the HFAN values in surrogate mixture approach.

6.3. SUMMARY OF NONCANCER PROVISIONAL REFERENCE VALUES

Table 8 summarizes the noncancer health reference values for indicator chemicals used when available analytical data and exposure estimates are limited to either air concentrations of or oral exposure rates associated with the whole fraction. When analytical results, air concentrations, or exposure rate measures for individual compounds with reference values are available, then the hazards associated with these compounds can be assessed separately, using the HI approach and reference values reported in Tables 6 and 7.

Table 8. Summary of Noncancer Reference Estimates for Indicator Chemicals for Aromatic Medium Carbon Range (C9−C10; EC9−EC < 11) Fraction of Total Petroleum Hydrocarbons

Toxicity Type (units); Indicator Chemical	Species/ Sex	Critical Effect	p-Reference Value	POD Method	POD (HED/HEC)	UF_C	Reference
Subchronic p-RfD (mg/kg-d); trimethylbenzenes	Rat/M	Neurotoxicity (decreased pain sensitivity)	0.04 mg/kg-d	BMDL (HED)[a]	3.5	100	Korsak and Rydzynski (1996) as cited in U.S. EPA (2016b)
Chronic p-RfD (mg/kg-d); trimethylbenzenes	Rat/M	Neurotoxicity (decreased pain sensitivity)	0.01 mg/kg-d	BMDL (HED)[a]	3.5	300	Korsak and Rydzynski (1996) as cited in U.S. EPA (2016b)
Subchronic p-RfC (mg/m^3); trimethylbenzenes	Rat/M	Neurotoxicity (decreased pain sensitivity)	0.2 mg/m^3	BMDL (HEC)	18.15	100	Korsak and Rydzynski (1996) as cited in U.S. EPA (2016b)
Chronic p-RfC (mg/m^3); trimethylbenzenes	Rat/M	Neurotoxicity (decreased pain sensitivity)	0.06 mg/m^3	BMDL (HEC)	18.15	300	Korsak and Rydzynski (1996) as cited in U.S. EPA (2016b)

[a]Based on route-to-route extrapolation (inhalation to oral) using a modified PBPK model.

BMDL = benchmark dose lower confidence limit; HEC = human equivalent concentration; HED = human equivalent dose; M = male(s); PBPK = physiologically based pharmacokinetic; POD = point of departure; p-RfC = provisional reference concentration; p-RfD = provisional reference dose; UF_C = composite uncertainty factor.

6.4. CANCER WEIGHT-OF-EVIDENCE DESCRIPTOR

Carcinogenicity assessments for mixtures and individual components of the aromatic medium carbon range fraction that have assessments are shown below in Table 9. For all components of the fraction, there are either inadequate data to assess carcinogenic potential (via the oral or inhalation routes of exposure), or the available studies have not been formally evaluated by the U.S. EPA, and the U.S. EPA has not estimated the cancer potency associated with the study results.

Table 9. Available Cancer Weight-of-Evidence Evaluations for Aromatic Medium Carbon Range Fraction (C9−C10, EC9−EC < 11)

Compound or Mixture	Cancer WOE	Source
Isopropylbenzene (C9, EC8.66)	*"Inadequate Information to Assess Carcinogenic Potential"*	U.S. EPA (1997)
n-Propylbenzene (C9, EC8.94)	*"Inadequate Information to Assess Carcinogenic Potential"*	U.S. EPA (2009d)
1,3,5-Trimethylbenzene (C9, EC9.15)	*"Inadequate Information to Assess Carcinogenic Potential"*	U.S. EPA (2016a)
1,2,4-Trimethylbenzene (C9, EC9.36)	*"Inadequate Information to Assess Carcinogenic Potential"*	U.S. EPA (2016a)
tert-Butylbenzene (C10, EC9.36)	*"Inadequate Information to Assess Carcinogenic Potential"*	U.S. EPA (2012b)
sec-Butylbenzene (C10, EC9.57)	*"Inadequate Information to Assess Carcinogenic Potential"*	U.S. EPA (2012a)
1,2,3-Trimethylbenzene (C9, EC9.65)	*"Inadequate Information to Assess Carcinogenic Potential"*	U.S. EPA (2016a)
n-Butylbenzene (C10, EC9.96)	*"Inadequate Information to Assess Carcinogenic Potential"*	U.S. EPA (2010)
High flash aromatic naphtha (C9−C10)	*"Inadequate Information to Assess Carcinogenic Potential"*	U.S. EPA (2009c)

C = carbon; EC = equivalent carbon; WOE = weight of evidence.

While data on genotoxicity testing of compounds and mixtures in the aromatic medium carbon range fraction are limited, available information suggests little to no genotoxic potential (see Section 5).

6.5. DERIVATION OF PROVISIONAL CANCER RISK ESTIMATES

None of the mixtures or constituents in this fraction had an oral slope factor (OSF) or inhalation unit risk (IUR) from IRIS, PPRTVs, HEAST, MassDEP, or TPHCWG. Thus, a provisional oral slope factor (p-OSF) or provisional inhalation unit risk (p-IUR) was not derived for the fraction (see Table 10).

Table 10. Summary of Cancer Risk Estimates for Aromatic Medium Carbon Range (C9−C10; EC9−EC < 11) Fraction of TPHs

Toxicity Type (units)	Species/Sex	Tumor Type	Cancer Risk Estimate	Principal Study
p-OSF $(mg/kg\text{-}d)^{-1}$	NDr			
p-IUR $(mg/m^3)^{-1}$	NDr			

C = carbon; EC = equivalent carbon; NDr = not determined; p-IUR = provisional inhalation unit risk; p-OSF = provisional oral slope factor; TPH = total petroleum hydrocarbon.

APPENDIX A. LITERATURE SEARCH AND SCREENING

Literature searches were conducted in February 2018 and updated in August 2021 for studies relevant to the derivation of provisional toxicity values for the aromatic medium carbon range fraction of total petroleum hydrocarbons (TPHs). The following substances (CASRNs), 12 chemicals and 1 mixture, were included in the initial list for the revised aromatic medium carbon range fraction: *n*-butylbenzene (104-51-8), isobutylbenzene (538-93-2), *tert*-butylbenzene (98-06-6), *sec*-butylbenzene (135-98-8), isopropylbenzene (535-77-3), *n*-propylbenzene (103-65-1), 1-methyl-4-ethylbenzene (622-96-8), 1-methyl-3-isopropylbenzene (535-77-3), 1-methyl-3-ethylbenzene (620-14-4), 1,2,3-trimethylbenzene (TMB; 526-73-8), 1,2,4-TMB (95-63-6), 1,3,5-TMB (108-67-8), and high flash aromatic naphtha (HFAN; 64742-95-6, 88845-25-4, and 64742-94-5). Because Integrated Risk Information System (IRIS) assessments were available for isopropylbenzene and the three TMB isomers, these compounds were not included in the literature searches. Literature searches were conducted for studies relevant to the derivation of provisional toxicity values for the remaining eight chemicals and one mixture listed above. Initial searches were date limited from 2007 to 2018 and were conducted using the U.S. Environmental Protection Agency (U.S. EPA) Health and Environmental Research Online (HERO) database of scientific literature. The PubMed database was searched using the HERO interface. The updated search was conducted similarly using the same search strings in PubMed and Web of Science from February 2018 through August 2021.

The results of the PubMed searches (title and abstract) were screened for relevance using the Population, Exposure, Comparator, and Outcome (PECO) criteria outline in Table A-1. Full-text screening for relevance to hazard identification was performed using the refined PECO criteria shown in Table A-2.

Table A-1. PECO Criteria for Screening of Total Petroleum Hydrocarbon Constituent Literature Search Results

PECO Element	Inclusion Criteria
Population	Humans (any population) or laboratory mammals (any life stage).
Exposure	Human: Exposure to the subject material alone or as the primary component of a mixture, known or presumed to occur by oral, inhalation, and/or dermal routes. Animal: In vivo, exposure to the subject material alone, by oral or inhalation (including instillation) routes, for all durations of exposures (durations <28 d will be captured as supporting information), including any duration during gestation. Other routes of exposure will be captured as supporting information.
Comparator	Human: Includes any comparison/referent group (no exposure, lower exposure). Animal: Includes concurrent negative (untreated, sham-treated, or vehicle) control.
Outcome	Assesses any cancer or noncancer endpoint in any tissue, organ, or physiological system.

PECO = Population, Exposure, Comparator, and Outcome.

Table A-2. PECO Criteria for Relevance to Hazard Identification

PECO Element	Inclusion Criteria
Population	Humans (any population) or laboratory mammals (any life stage).
Exposure	Human: Exposure to the subject material alone or as the primary component of a mixture, known or presumed to occur by oral or inhalation routes. Animal: In vivo, exposure to the subject material alone, by oral or inhalation routes, for durations ≥28 d or any duration during gestation.
Comparator	Human: Includes any comparison/referent group (no exposure, lower exposure). Animal: Includes concurrent negative (untreated, sham-treated, or vehicle) control.
Outcome	Assesses any cancer or noncancer health outcome in any tissue, organ, or physiological system.

PECO = Population, Exposure, Comparator, and Outcome.

APPENDIX B. POTENTIALLY RELEVANT NONCANCER EVIDENCE

DEVELOPMENT OF EXPOSURE-RESPONSE ARRAYS

As described in the main document, dose-response data were presented in exposure-response arrays by health outcome and exposure route. In order to assess consistency in effects and potency across the components of the fraction, experimental data from compound specific Integrated Risk Information System (IRIS) and Provisional Peer Reviewed Toxicity Value (PPRTV) documents and primary data sources (identified from literature searches) were used to create exposure-response arrays. Exposure-response arrays present dose-response data by health outcome and exposure route. From left to right, compounds exhibiting an effect are shown before those not exhibiting an effect, to enable identification of patterns. Within the group exhibiting an effect, compounds are ordered from lowest lowest-observed-adverse-effect level (LOAEL) to highest. For compounds that do not exhibit an effect, no-observed-adverse-effect levels (NOAELs) in the arrays are ordered by equivalent carbon (EC) number index (low to high from left to right), with mixtures shown last. Both administered doses and exposure concentrations reported in the arrays and in text reflect time-weighted average (TWA) exposures, to facilitate comparisons across studies and compounds. Consistency across the fraction was evaluated by assessing if comparable outcomes were observed for members of the fraction, and if these effects were observed at similar dose levels. Unless otherwise specified, the term "significant," used throughout this appendix, refers to statistical significance at a p-value < 0.05.

NEUROLOGICAL EFFECTS

A nervous system endpoint (decreased pain sensitivity) is the critical effect for the subchronic and chronic reference concentration (RfC) values for trimethylbenzene (TMB) isomers, with data for 1,2,4-TMB being the driver for these values U.S. EPA (2016b). Oral toxicity values (i.e., subchronic and chronic reference doses [RfDs]) for TMBs were derived based on route-to-route extrapolation (using a modified physiologically based pharmacokinetic [PBPK] model) from the inhalation values. Neurological effects (including effects on motor coordination, cognitive function, vision, and the inner ear) have been reported in humans occupationally exposed to solvents including TMBs; however, the effects cannot be attributed to specific compounds. Neurotoxicity data in humans are limited to TMBs, and there are no human data for other members of the aromatic medium carbon range fraction. Animal studies examining neurological endpoints are available for most of the compounds or mixtures with toxicity data; however, the studies varied widely with respect to the nature of the neurological endpoints evaluated.

Human Studies

U.S. EPA (2016b) reviewed the evidence for neurotoxicity in humans exposed to TMBs alone or in complex mixtures. Much of the epidemiological evidence is from occupational exposures to complex mixtures including TMBs, and the relative contribution of TMBs compared with other constituents is not known. Associations between exposure of dockyard and shipyard painters to solvent mixtures possibly containing TMBs and impaired performance in a battery of neurological tests, including short-term memory (symbol digit substitution), motor speed/coordination (finger tapping), and peripheral nerve function tests, were reported in several studies U.S. EPA (2016b). Other neuropsychological symptoms (mood changes, equilibrium complaints, and sleep disturbances) were also observed among shipyard painters. Similarly, paint factory workers exposed to multiple unspecified solvents had detrimental neuropsychological

effects (memory problems, dizziness, hand tremble), and construction workers exposed to solvent mixtures had impaired performance in memory tasks U.S. EPA (2016b).

Other researchers reported damage or dysfunction of the inner ear and increased incidence of vertigo following workplace exposure (in paint and varnish factories and histology laboratories) to TMBs and other organic solvents U.S. EPA (2016b). Suggestive evidence of visual impairment (altered color vision and contrast) among furniture factory workers was reported following exposure to complex solvent mixtures. Increased latencies for visual evoked potentials (VEPs) were found in gasoline-exposed workers. There is suggestive evidence from multiple studies of human exposure to solvent mixtures containing TMB isomers that exposure results in toxicological effects on neuromuscular function and balance in humans, including increased reaction time, increased hand tremble, decreased hand-eye coordination, and vertigo U.S. EPA (2016b). Finally, symptoms associated with central nervous system (CNS) depression (e.g., lightheadedness, fatigue) have been reported in workers occupationally exposed to solvent mixtures containing TMBs U.S. EPA (2016b).

A significant, positive association between exposure and neurological symptoms (such as abnormal fatigue) was reported among asphalt workers exposed to 1,2,4-TMB; however, the association was not evident among asphalt workers with exposure to lower levels of 1,2,3-TMB or 1,3,5-TMB U.S. EPA (2016b). Paint shop workers exposed to 49−295 mg/m^3 of a solvent mixture containing 50% 1,2,4-TMB, 30% 1,3,5-TMB, and unspecified amounts of 1,2,3-TMB (listed as possibly present) exhibited a variety of neurological effects including nervousness, tension, headaches, vertigo, and anxiety U.S. EPA (2016b).

Studies of adult volunteers who were acutely exposed to mixtures containing 1,2,4-TMB reported significant and consistent increases in reaction time, although it is unclear whether 1,2,4-TMB or other constituents in the mixtures were responsible for the observed effects. Neurobehavioral impairment was either weakly or inconsistently associated with exposure in a volunteer study in which participants were exposed to aromatic or dearomatized white spirit (white spirit contains a mixture of 1,2,4- and 1,3,5-TMB) for 4 hours U.S. EPA (2016b).

The few available controlled human exposure studies of TMBs alone have not shown neurological effects U.S. EPA (2016b). No neurological abnormalities were reported in routine clinical examinations in two studies investigating the toxicokinetics of TMBs following controlled human exposures to 5−150 mg/m^3 of 1,2,3-, 1,2,4-, or 1,3,5-TMB; however, neither results data nor details regarding the specific neurological tests performed were provided. In another controlled toxicokinetics study, no overt CNS depression (measured as heart rate and respiration) or increase in subjective CNS symptoms (headache, fatigue, nausea, dizziness, intoxication) was observed in volunteers exposed to concentrations ≤123 mg/m^3 of TMB isomers U.S. EPA (2016b).

Animal Studies

Animals exposed orally to diethylbenzenes (DEBs) (individual isomers, as well as a mixture containing 7% 1,2-DEB, 58% 1,3-DEB, and 35% 1,4-DEB) have been evaluated for peripheral nervous system and CNS effects. No oral data on the neurotoxicity of the other members of the aromatic medium carbon range fraction were located. Figure B-1 is an exposure-response array containing studies for which neurotoxicity effects levels could be reliably determined. Reductions in sensory conduction velocity (SCV), motor nerve conduction velocity (MCV), and amplitude of the sensory action potential (SAP) (of the tail nerve) occurred after exposure to 57 mg/kg-day 1,2-DEB and ≥357 mg/kg-day of the DEB mixture (lowest doses tested) Gagnaire et al. (1990). The same parameters were unaffected in rats treated with 1,3- or 1,4-DEB at 357 mg/kg-day. In a follow-up study, rats administered 1,2-DEB at ≥43 mg/kg-day also showed significantly increased latencies with respect to parameters of brainstem auditory evoked potentials (BAEPs) Gagnaire et al. (1992a).

Neurological effects evaluated after inhalation exposure to aromatic medium carbon range compounds include clinical signs of neurotoxicity, neurobehavioral changes, peripheral nervous system and CNS function, and brain histopathology. Figure B-2 is an exposure-response array containing studies for which neurotoxicity effect levels could be reliably determined. Data were available for six members of the fraction. Clinical signs of neurotoxicity (side-to-side movement and head tilt) were observed in rats treated with isopropylbenzene at ≥92 mg/m^3 for 4 weeks Monsanto Company, 1986 as cited in U.S. EPA (1997). Significant neurobehavioral changes (impairments in active and passive avoidance, increased motor activity, and/or reduced pain sensitivity) were observed in rats treated with 1,2,4-, 1,3,5-, or 1,2,3-TMB at ≥88 mg/m^3 Wiaderna et al., 2002, Gralewicz and Wiaderna, 2001, Wiaderna et al., 1998, Gralewicz et al., 1997a, and Korsak and Rydzynski, 1996, Lutz, 2010, all as cited in U.S. EPA (2016b). Decreased pain sensitivity in rats treated with 1,2,4-TMB at ≥88 mg/m^3 serves as the basis for oral and inhalation subchronic and chronic RfDs and RfCs for all three TMB isomers Korsak and Rydzynski, 1996 as cited in U.S. EPA (2016b). Pain sensitivity was not significantly affected in rats exposed to HFAN at up to 1,157 mg/m^3 for 90 days Douglas et al., 1993 as cited in U.S. EPA (2009c). There was no effect on nervous system histology in this study or on brain weight or histology in rats or mice following exposure to isopropylbenzene for 14 or 105 weeks at concentrations up to 907 mg/m^3 Ntp (2009). Male rats exposed to a DEB mixture for 18 weeks showed decreased SCV, MCV, and amplitude of SAP (≥486 mg/m^3) and increased latency of BAEP parameters (at ≥633 mg/m^3) Gagnaire et al. (1992b). In a developmental study, no neurobehavioral effects were reported in rats exposed to HFAN at up to 500 mg/m^3 on gestation days (GDs) 7−15; however, it is not clear if only pups or pups and dams were evaluated for effects Lehotsky, 1989 as cited in U.S. EPA (2009c).

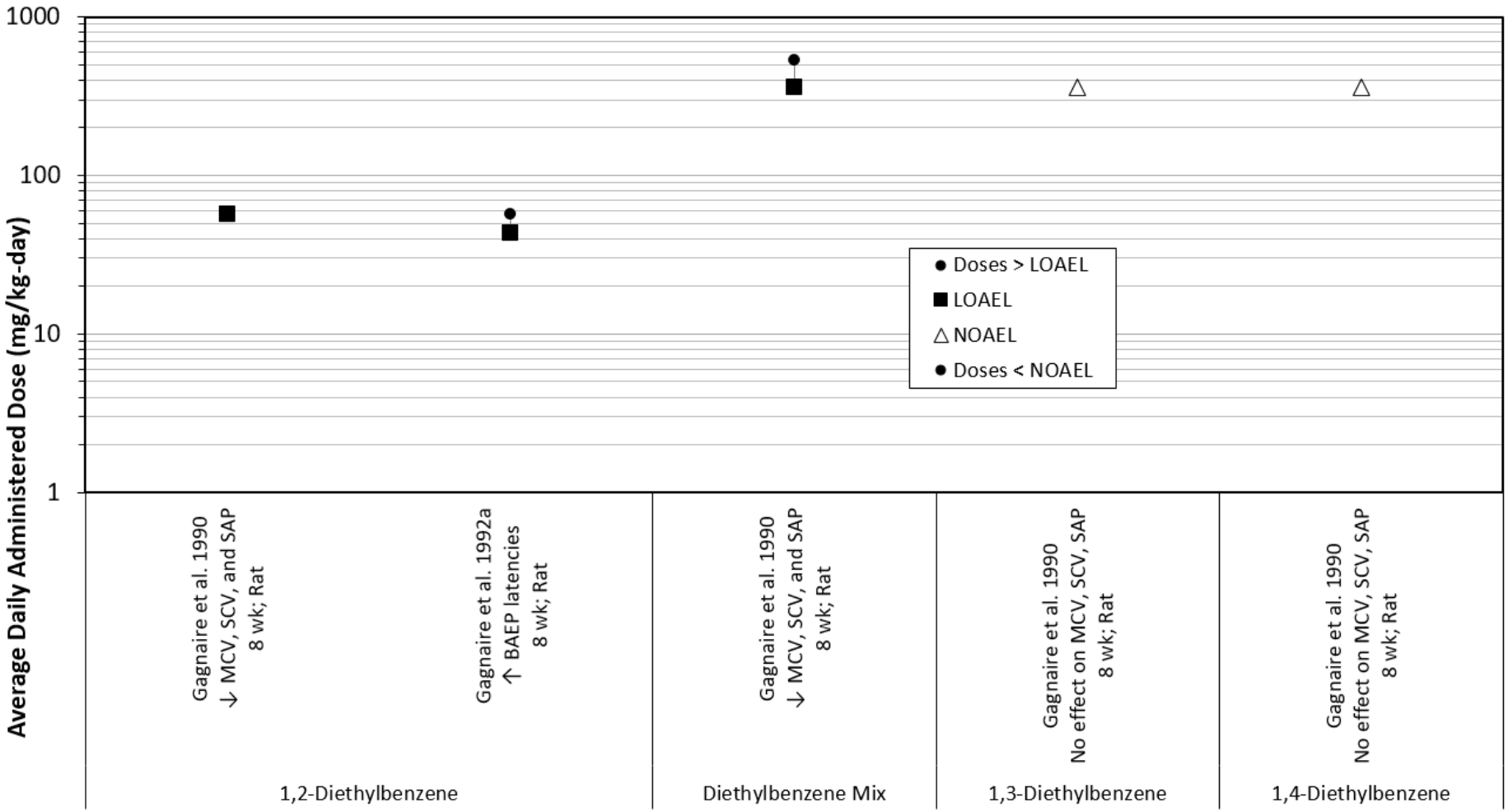


Figure B-1. Neurological Effects in Animals after Oral Exposure to Aromatics Medium Carbon Range Compounds

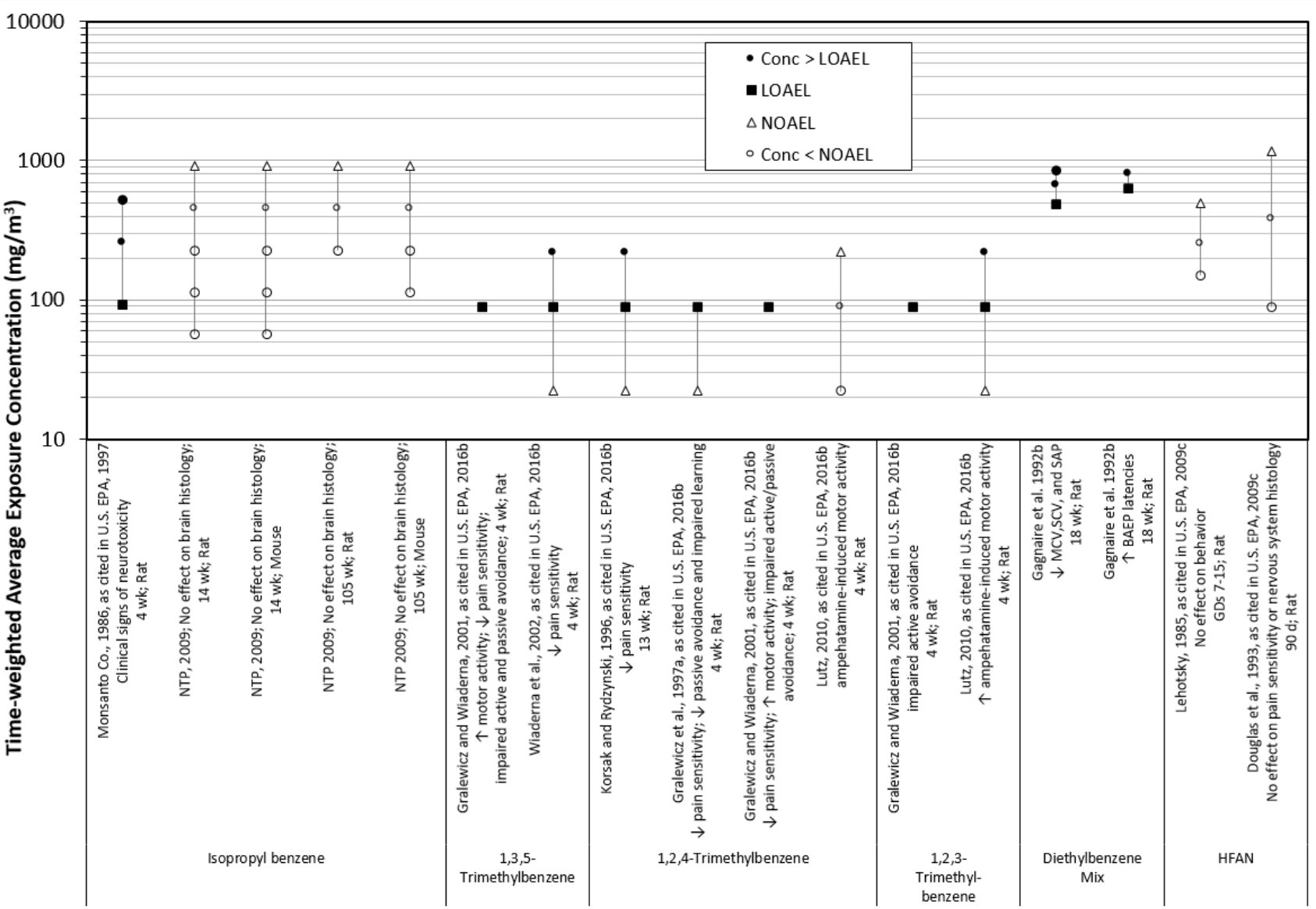


Figure B-2. Neurological Effects in Animals after Inhalation Exposure to Aromatic Medium Carbon Range Compounds

Summary of Potentially Relevant Evidence

Available data indicate that neurological effects are associated with oral or inhalation exposure to some fraction members. In oral and inhalation toxicity studies using individual DEB isomers or a mixture of DEB isomers, effects on peripheral nervous system and/or CNS (SCV, MCV, amplitude of SAP, and BAEPs) were consistently observed for 1,2-DEB and the DEB mixture (but not 1,3-DEB or 1,4-DEB). Studies in animals show that exposure to TMBs via inhalation induces neurobehavioral effects (specifically, decreased pain sensitivity and increased motor activity and passive and active avoidance); data from human studies are available but are insufficient to establish causal relationships (owing to coexposures with other compounds). Limited data are available for the other members of the fraction. CNS effects (consisting of clinical signs of neurotoxicity) were observed in at least one other inhalation study (in rats exposed to isopropylbenzene). However, additional studies using this compound were not dedicated to neurotoxicity studies; only brain histopathology was evaluated (no effects were observed in subchronic and chronic studies in rats and mice). The most sensitive chemical- or mixture-specific LOAELs for neurological endpoints ranged between 43 and 357 mg/kg-day in subchronic oral toxicity studies in rats (see Figure B-1) and between 88 and 633 mg/m^3 in subchronic inhalation toxicity studies in rats (see Figure B-2).

Taken together, the available data indicate that some members of the aromatic medium carbon range fraction can induce neurological effects. However, there are a number of compounds comprising the aromatic medium carbon range fraction that have not been evaluated for sensitive measures of neurological function.

HEPATIC EFFECTS

A hepatic effect (hepatocellular hypertrophy) is the critical effect for the subchronic and chronic provisional reference doses (p-RfDs) for *n*-butylbenzene U.S. EPA (2010). In addition, effects on liver histopathology serve as a cocritical effects (with effects on kidney histopathology) for the screening-level subchronic and chronic p-RfDs for *n*-propylbenzene [based on the use of ethylbenzene (CASRN 100-41-4) as an analogue chemical U.S. EPA (2009a). No human data pertaining to the hepatotoxicity of aromatic medium carbon range fraction members were identified. As shown in Table 5, oral and/or inhalation data on hepatic effects in animals were located for seven members of the fraction. In general, the hepatic endpoints evaluated in the studies were clinical chemistry parameters, liver weight, and histology.

Human Studies

No human studies were available to address the potential for hepatic toxicity of the TMB isomers or other members of the aromatic medium carbon range fraction by any route of exposure.

Animal Studies

Animals orally administered four materials (three individual compounds and HFAN) in the aromatic medium carbon range have been evaluated for hepatotoxicity. Figure B-3 is an exposure-response array containing studies for which hepatic effects levels could be reliably determined. Increased absolute and/or liver weight was observed in rats treated with 1,3,5-TMB at 428 mg/kg-day for 90 days Adenuga et al., 2014 as cited in U.S. EPA (2016b), 1,4-DEB at 750 mg/kg-day for 6 weeks MHW, 1993b as cited in Oecd (1994), *n*-butylbenzene at 300 mg/kg-day for two generations Izumi et al., 2005 as cited in U.S. EPA (2010), and HFAN at ≥357 mg/kg-day for 13 weeks Biodynamics, 1990a and Mobil Oil Company, 1994 as cited in U.S. EPA (2009c). Relative liver weight was likewise increased in HFAN-treated dogs at 500 mg/kg-day; however, this effect may have been influenced by decreased terminal body weights (20% lower than controls) Biodynamics, 1990b as cited in U.S. EPA (2009c). With the exception of rats treated with 1,3,5-TMB, rats that exhibited increased liver weights also showed increased incidences of hepatocellular hypertrophy. A significantly increased incidence of hepatocellular hypertrophy (in F_0 and F_1 parental males) was the basis for subchronic and chronic RfDs for *n*-butylbenzene Izumi et al., 2005 as cited in U.S. EPA (2010). In addition, liver and kidney toxicity were designated as cocritical effects for screening-level subchronic and chronic RfDs for *n*-propylbenzene, based on analogy to ethylbenzene U.S. EPA (2009d).

Data evaluating hepatotoxicity via inhalation exposure were available for four members of the aromatic medium carbon range fraction. Figure B-4 is an exposure-response array containing studies for which hepatic effects levels could be reliably determined. Relative liver weight was biologically and statistically significantly increased at concentrations of ≥227 mg/m^3 in rats and ≥454 mg/m^3 in mice treated with isopropylbenzene for 14 weeks [NTP (2009); Cushman et al., 1995 as cited in U.S. EPA (1997)] and at 2,823 mg/m^3 in rabbits treated with isopropylbenzene on GDs 6−18 Darmer et al. (1997). Increased organ weight was accompanied by histopathological evidence of liver damage (e.g., eosinophilic foci and necrosis) after 105 weeks in mice Ntp (2009), but not rats Ntp (2009). No significant, treatment-related effects on liver weights and/or histopathology were observed in rats exposed to 1,2,4- or 1,2,3-TMB at concentrations up to 220 mg/m^3 for 3 months Korsak et al. 2000a, b as cited in U.S. EPA (2016b), in rats exposed to HFAN at up to 327 mg/m^3 for 3 months Clark et al., 1985 as cited in U.S. EPA (2009c), or in rabbits exposed to HFAN at up to 1,000 mg/m^3 Clark et al., 1989 and Ungvary and Tartal, 1985 as cited in U.S. EPA (2009c).

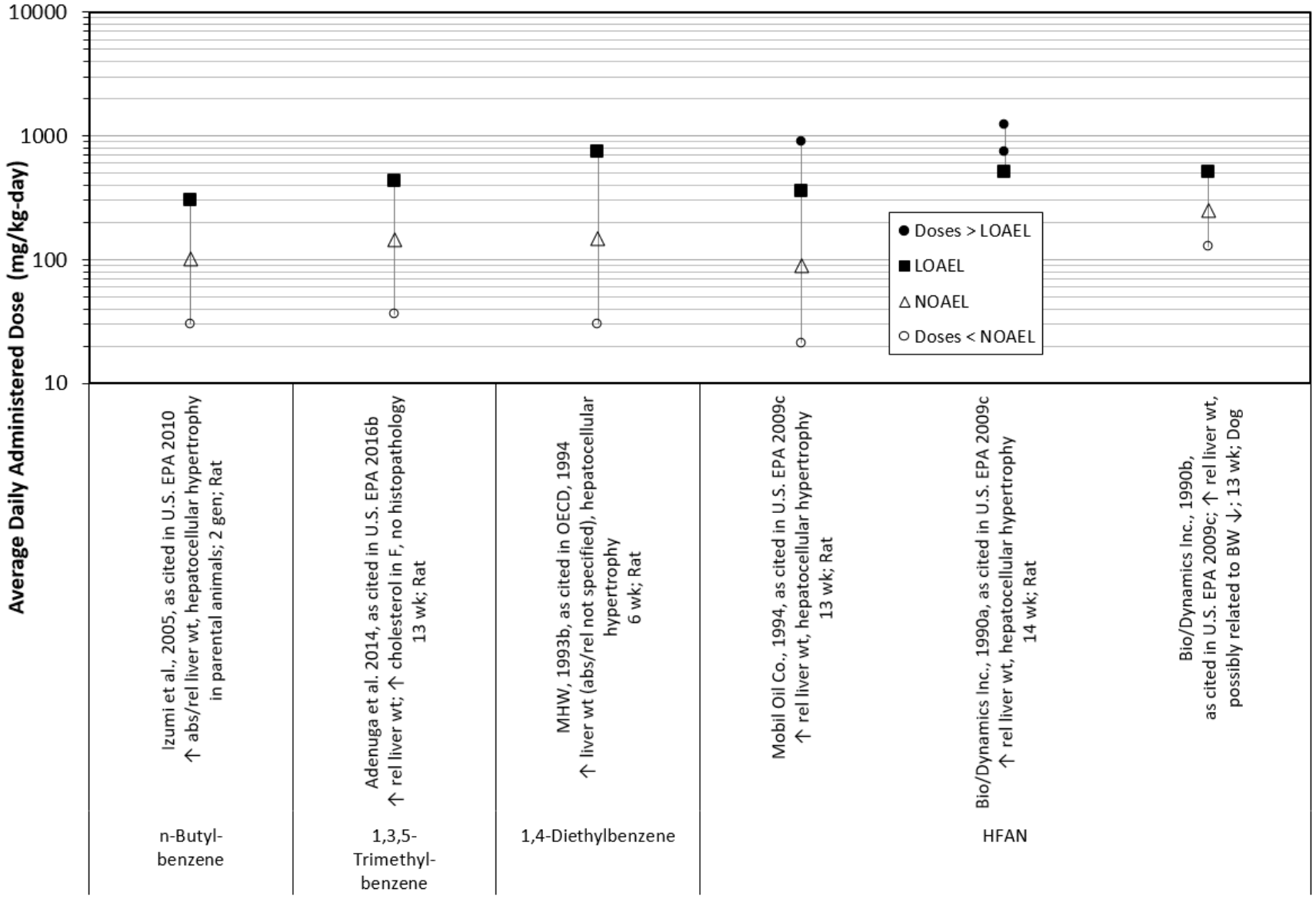


Figure B-3. Hepatic Effects in Animals after Oral Exposure to Aromatic Medium Carbon Range Compounds

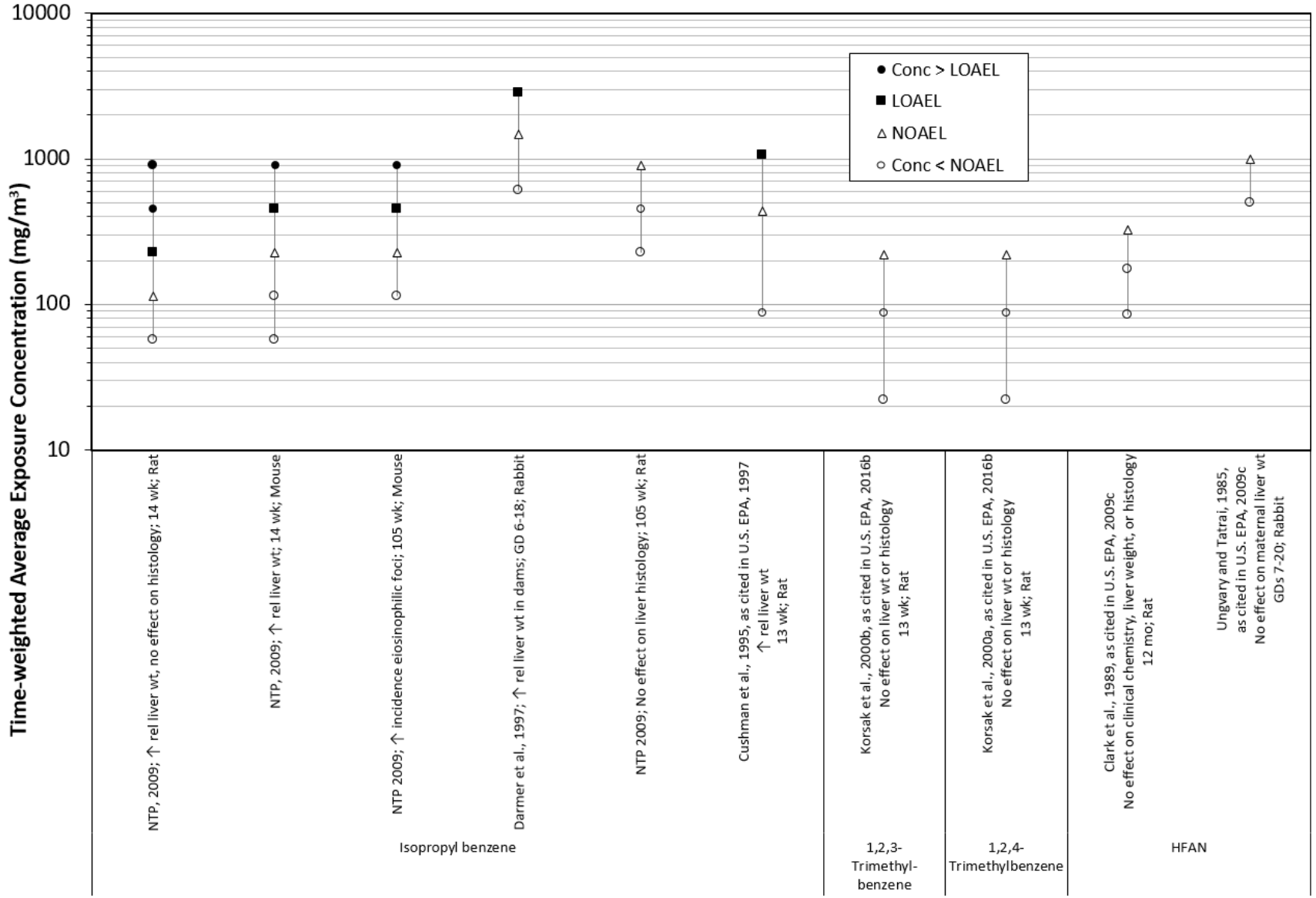


Figure B-4. Hepatic Effects in Animals after Inhalation Exposure to Aromatic Medium Carbon Range Compounds

Summary of Potentially Relevant Evidence

Oral studies examining liver effects were limited to three compounds and one mixture (HFAN) in studies of 6 weeks to about 3 months in duration. All oral studies showed increases in liver weight; this effect was typically accompanied by changes in liver histopathology (namely, hepatocellular hypertrophy). Hepatic effects, predominantly consisting of increased relative weights, were also seen in inhalation studies in laboratory animals exposed to members of the aromatic medium carbon range fraction. Histological changes observed in the livers of animals exposed to members of the fraction included hepatocellular hypertrophy in multiple subchronic oral toxicity studies and eosinophilic foci in singular chronic inhalation toxicity studies (with several studies reporting no histological effects). Lowest LOAELs for hepatic endpoints ranged from 300 to 750 mg/kg-day in oral studies in rats (see Figure B-3; a LOAEL of 500 mg/kg-day was also identified in dogs), and from 227 to 454 mg/m^3 in subchronic inhalation studies in rats and mice (see Figure B-4; excluding a LOAEL of 2,823 mg/m^3 for increased liver weight in a developmental toxicity study in rabbits). In aggregate, the data suggest that many aromatic medium carbon range fraction compounds and mixtures can promote increases in rodent liver weight, sometimes accompanied by histological changes.

RENAL EFFECTS

A renal endpoint (increased kidney weights in female F344 rats) is the critical effect for the chronic RfD and the cocritical effect (with increased adrenal weights) for the chronic RfC for isopropylbenzene U.S. EPA (1997). In addition, increased kidney weight serves as the critical effects for the screening-level subchronic and chronic p-RfDs for *tert*- and *sec*-butylbenzene U.S. EPA (2012a, b); these toxicity values are based on the use of isopropylbenzene as an analogue chemical. Effects on kidney histopathology were also identified as the critical effect in the study used to derive screening-level subchronic and chronic p-RfDs for *n*-propylbenzene U.S. EPA (2009d), based on the use of ethylbenzene as an analogue chemical. No human data pertaining to the renal toxicity of aromatic medium carbon range fraction members were identified. As shown in Table 5, data on renal effects in animals were located for seven members of the fraction. In general, the renal endpoints evaluated in the studies were kidney weight and histology; many also measured clinical chemistry parameters.

Human Studies

No human studies were available to address the potential renal effects of the TMB isomers or other members of the aromatic medium carbon range fraction by any route of exposure.

Animal Studies

Reliable data evaluating renal toxicity in animals after oral exposure to aromatic medium carbon range compounds were available for five members of the fraction. Figure B-5 is an exposure-response array containing studies for which renal effects levels could be reliably determined. These studies identified increased relative kidney weight as the most sensitive renal effect. Increased organ weight was observed in rats treated at ≥331 mg/kg-day with isopropylbenzene over 192 days (females only evaluated) Wolf, 1956 as cited in U.S. EPA (1997), 428 mg/kg-day with 1,3,5-TMB for 90 days Adenuga et al., 2014 as cited in U.S. EPA (2016b), 150 mg/kg-day with 1,4-DEB for about 6 weeks MHW, 1993b as cited in Oecd (1994), 300 mg/kg-day with *n*-butylbenzene for two generations (both generations) Izumi et al., 2005 as cited in U.S. EPA (2010), and ≥357 mg/kg-day with HFAN for about 13 weeks Biodynamics, 1990a and Mobil Oil Company, 1994 as cited in U.S. EPA (2009c). Kidney weight was likewise

increased in HFAN-treated dogs at 500 mg/kg-day; however, this effect may have been influenced by decreased terminal body weights (20% lower than controls) Biodynamics, 1990b as cited in U.S. EPA (2009c). Increased serum blood urea nitrogen (BUN) was observed at the same dose as increased kidney weights in rats treated with 1,4-DEB MHW, 1993b as cited in Oecd (1994). Although changes in kidney histopathology were noted in at least one study of HFAN Mobil Oil Company, 1994 as cited in U.S. EPA (2009c), effects were seen in males only and the "male kidney sections had changes that may be consistent with nephropathy typical of male rats (dose-related hyaline droplet deposition, and nondose-related cortical tubular degeneration, consisting primarily of epithelial swelling)" U.S. EPA (2009c). These toxicities were consistent with rat-specific nephropathy, and U.S. EPA (1991) reported that these pathologies may not be considered relevant to humans. As such, the results of this are not presented in Figure B-5.

Renal effects seen after inhalation exposure to aromatic medium carbon range compounds are almost strictly limited to significantly increased relative kidney weight. Figure B-6 is an exposure-response array containing studies for which renal effects levels could be reliably determined. Organ weight was increased in rats exposed to isopropylbenzene at ≥454 mg/m^3 for 14 weeks Ntp (2009), 1,055 mg/m^3 for 13 weeks Cushman et al., 1995 as cited in U.S. EPA (1997), and 526 mg/m^3 for 4 weeks Monsanto Company, 1986 as cited in U.S. EPA (1997); kidney weights were unaffected in mice treated at up to 907 mg/m^3 for 14 or 105 weeks Ntp (2009). Although histopathological effects (e.g., granular casts, mineralization of renal papilla) were noted in isopropylbenzene-exposed rats, effects were seen in males only and were considered in the assessment to be consistent with rat-specific nephropathy (i.e., their relevance to humans is considered questionable) U.S. EPA (1997). In rats exposed to 1,2,4- or 1,2,3-TMB at up to 220 mg/m^3 for 3 months, there was no evidence of renal toxicity based on kidney weights or histopathology Korsak et al. 2000a, b as cited in U.S. EPA (2016b). Similarly, rats exposed to HFAN at up to 327 mg/m^3 for 12 months showed no consistent signs of kidney damage based on evaluations of clinical chemistry parameters and kidney histopathology Clark et al., 1989 as cited in U.S. EPA (2009c).

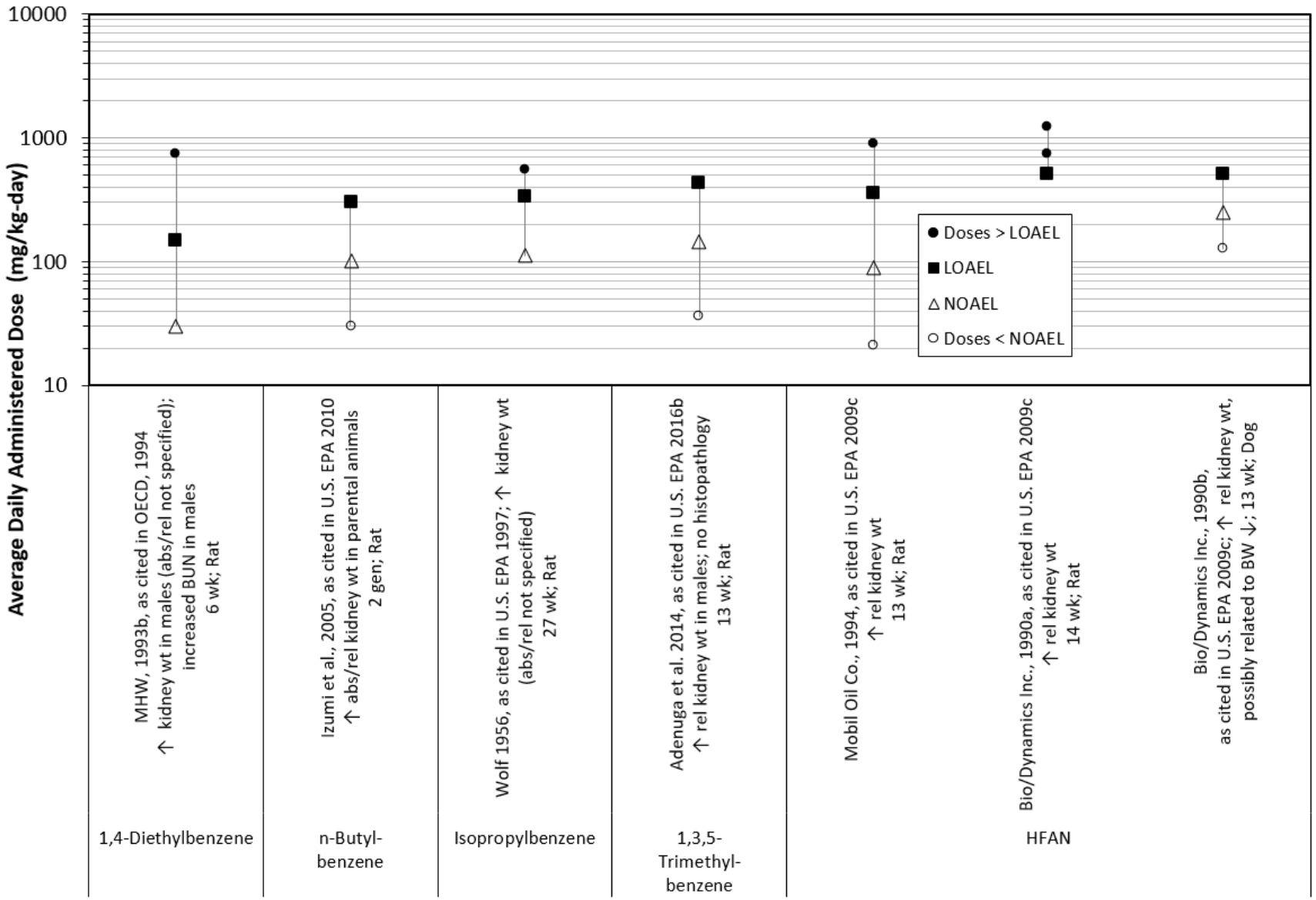


Figure B-5. Renal Effects in Animals after Oral Exposure to Aromatic Medium Carbon Range

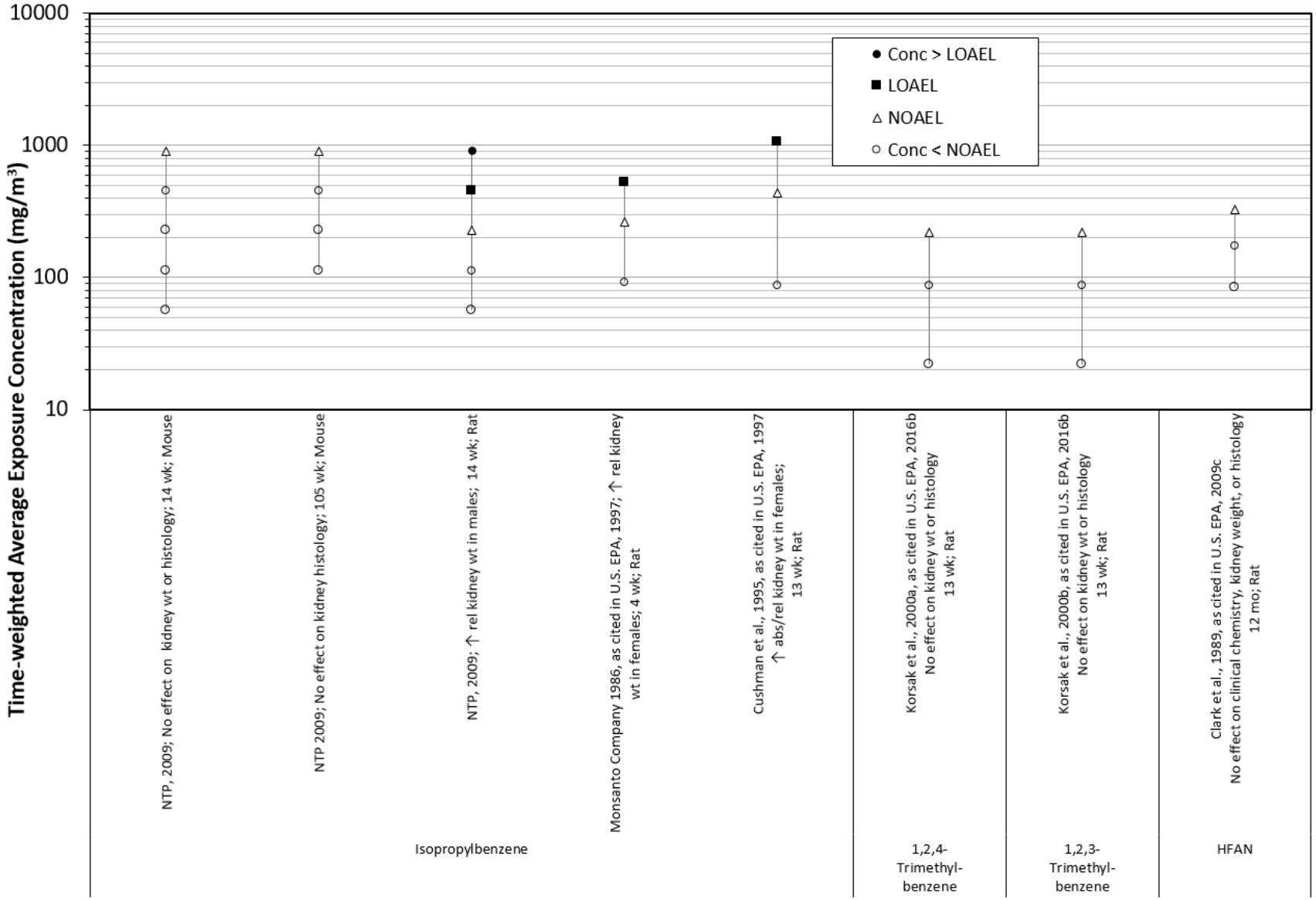


Figure B-6. Renal Effects in Animals after Inhalation Exposure to Aromatic Medium Carbon Range Compounds

Summary of Potentially Relevant Evidence

Oral studies examining kidney effects were limited to four compounds and one mixture (HFAN) in studies of 4−27 weeks in duration. All studies showed increases in kidney weight; one study also reported a change in serum chemistry consistent with kidney damage (i.e., increased BUN). Changes in kidney histology were reported only in male rats for the four compounds and one mixture; these effects were consistent with rat-specific nephropathy. Kidney effects (increased relative kidney weights) were also seen in inhalation studies of animals exposed to one member of the aromatic medium carbon range fraction (isopropylbenzene). The lowest LOAELs (by compound or mixture) for increased kidney weights ranged from 150 to 500 mg/kg-day in oral studies in rats and dogs (see Figure B-5). For inhalation exposure, the lowest LOAEL was 454 mg/m^3 for subchronic exposure to isopropylbenzene in rats (see Figure B-6). Inhalation studies of other compounds comprising the medium carbon fraction (including TMB isomers) did not indicate significant, treatment-related changes in kidney weights or histology. Taken together, the data indicate that several members of the aromatic medium carbon range fraction compounds and mixtures can produce increases in rodent kidney weight, sometimes accompanied by serum chemistry and histological changes.

BODY-WEIGHT EFFECTS

Decreased maternal body weight on GD 15 is the critical effect in the study used to derive the subchronic and chronic provisional reference concentrations (p-RfCs) for HFAN U.S. EPA (2009c). No human studies examining body-weight effects of aromatic medium carbon range compounds were identified in the sources reviewed. As Table 5 shows, animal studies (oral or inhalation) that examined body weight as an endpoint are available for nearly all of the compounds and mixtures with toxicity data. Exceptions are *n*-propylbenzene and *tert*- and *sec*-butylbenzene. In this section, body-weight changes of at least 10% relative to controls in adult animals are considered LOAELs, and smaller changes are not. For studies that reported body-weight gain but did not report absolute body weights, and for studies of maternal weight gain during gestation, statistically significant changes from control are described.

Human Studies

No human studies were available to address the potential for impacts on body weight of the TMB isomers or other members of the aromatic medium carbon range fraction by any route of exposure.

Animal Studies

Figure B-7 shows the effects of orally-administered aromatic medium carbon range compounds and mixtures on body weight; data are available for seven materials. In studies of 1,3,5-TMB, 1,3-DEB, and *n*-butylbenzene, no treatment-related effects on body weight or body-weight gain were observed Adenuga et al., 2014 as cited in U.S. EPA (2016b); Izumi, 2005 as cited in U.S. EPA (2010; Gagnaire et al. (1990). However, decreased body weight/body-weight gain was seen after exposure to rats to 1,4-DEB at 750 mg/kg-day for 6 weeks MHW, 1993b as cited in Oecd (1994), 1,2-DEB at ≥43 mg/kg-day for 8 weeks Gagnaire et al. (1992a; Gagnaire et al. (1990), HFAN at ≥625 mg/kg-day on GDs 6−15 Biodynamics, 1990c as cited in U.S. EPA (2009c), and a mixture of DEB isomers at ≥357 mg/kg-day for 10 weeks Gagnaire et al. (1990). Dogs administered HFAN at 500 mg/kg-day (highest dose tested) for up to 90 days showed a 20% reduction in terminal body weights [albeit not statistically significant Biodynamics, 1990c as cited in U.S. EPA (2009c)].

Body-weight effects evaluated in subchronic and chronic inhalation toxicity studies are shown in Figure B-8; effects evaluated in developmental inhalation toxicity studies are shown in Figure B-9. Data via the inhalation route were available for six members of the aromatic medium carbon range fraction and a mixture of members of this fraction. As shown in Figure B-8, significant effects in subchronic or chronic studies are limited to rats exposed to mixtures comprising the aromatic medium range carbon fraction. HFAN-treated rats showed decreased body weights after treatment at 1,157 mg/m^3 for 90 days Douglas et al., 1993 as cited in U.S. EPA (2009c). In addition, body-weight gain was significantly reduced in rats exposed to mixed DEB isomers at ≥486 mg/m^3 for 18 weeks Gagnaire et al. (1992b). In contrast, no significant effects on body weight were observed following exposure of rats and mice to isopropylbenzene at up to 907 mg/m^3 for 14 or 105 weeks Ntp (2009) or in rats exposed to isopropylbenzene at up to 1,055 mg/m^3 for 13 weeks Cushman et al., 1995 as cited in U.S. EPA (1997). In addition, rats exposed to 1-methyl-4-ethylbenzene at up to 417 mg/m^3 for 4 weeks Swiercz et al. (2000), or any of the three individual TMB isomers at up to 220 mg/m^3 for 3 months showed no significant, treatment-related effects on body weight or body-weight gain Wiaderna et al., 2002, Korsak et al., 2000a, b, Gralewicz and Wiaderna, 2000, Wiaderna et al., 1998, Gralewicz et al., 1997a, and Korsak and Rydzynski, 1996, 1997, all as cited in U.S. EPA (2016b).

Significant effects on body weight/body-weight gain were observed in dams exposed to members of the aromatic medium carbon range fraction during the gestational period (see Figure B-9). Rats and rabbits exposed to high concentrations of isopropylbenzene during gestation (at ≥1,488 and 2,823 mg/m^3, respectively) showed significantly reduced body-weight gains Darmer et al. (1997). After gestational exposure to 1,3,5-TMB (at ≥369 mg/m^3) or 1,2,4-TMB (at ≥738 mg/m^3), body-weight gain was likewise significantly decreased Saillenfait et al., 2014 as cited in U.S. EPA (2016b). Rabbits exposed to HFAN at up to 1,000 mg/m^3 on GDs 7−20 showed no significant, treatment-related effects on body weight Ungvary and Tatrai, 1985 as cited in U.S. EPA (2009c), but mice exposed during gestation were affected at concentrations as low as 125 mg/m^3 McKee et al., 1990 as cited in U.S. EPA (2009c).

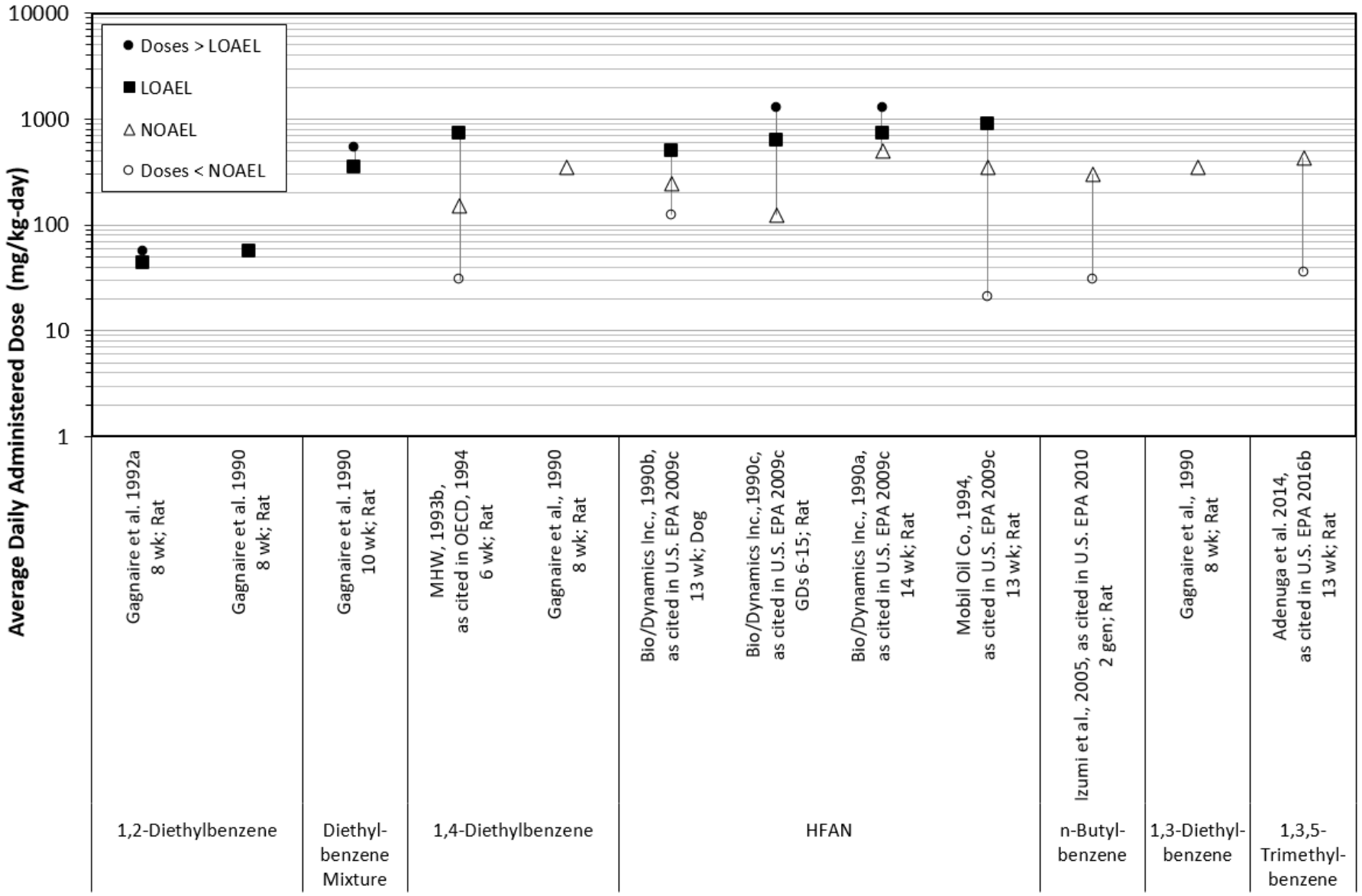


Figure B-7. Body-Weight Effects in Animals after Oral Exposure to Aromatic Medium Carbon Range Fraction

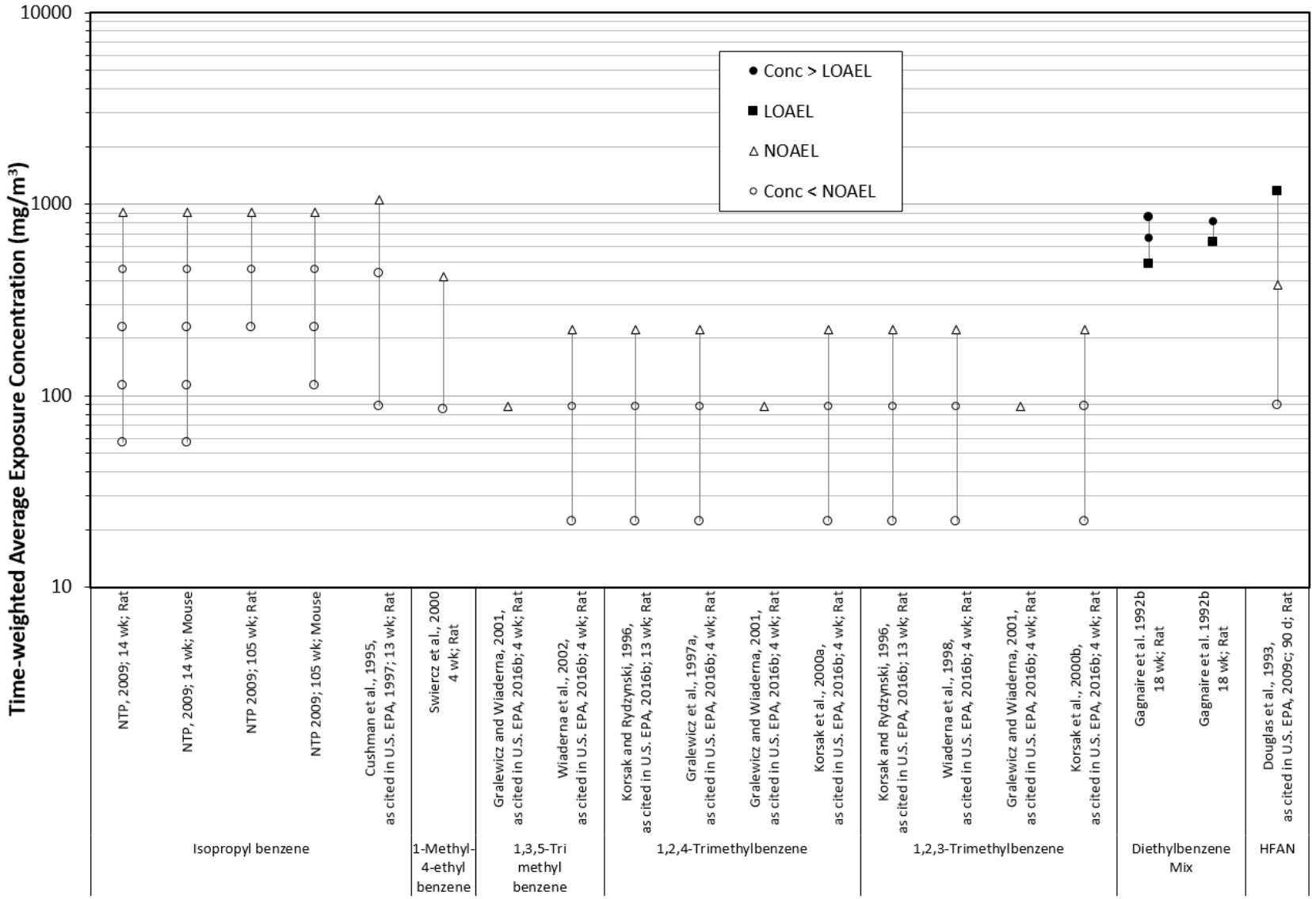


Figure B-8. Body-Weight Effects in Animals after Subchronic and Chronic Inhalation Exposure to Aromatic Medium Carbon Range Compounds

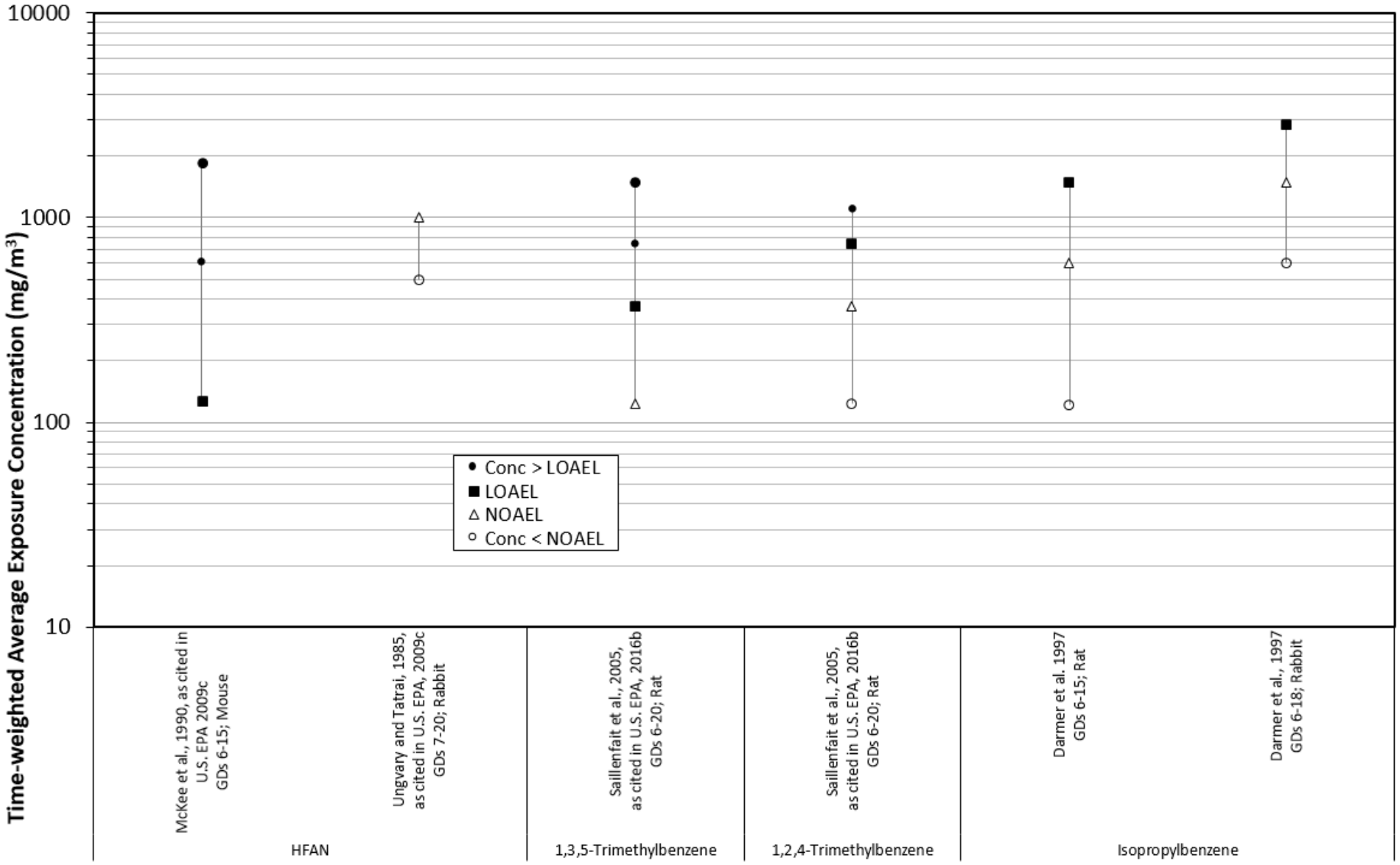


Figure B-9. Body-Weight Effects in Animals after Gestational Inhalation Exposure to Aromatic Medium Carbon Range Compounds

Summary of Potentially Relevant Evidence

Compounds and mixtures in the aromatic medium carbon range fraction have been shown to reduce body weights of rats, mice, rabbits, and dogs after oral and inhalation exposure. Members that induced body-weight changes in oral studies in rats included HFAN and DEBs (both individual isomers and mixed isomers); LOAELs for these effects ranged from 43 mg/kg-day (for 1,2-DEB) to 893 mg/kg-day (for HFAN) (see Figure B-7). In subchronic inhalation toxicity studies, rats exposed to HFAN and DEB mixtures (at 1,157 and ≥486 mg/m^3, respectively) showed reductions in body weight/body-weight gain, whereas rats and mice exposed to individual compounds from the fraction (in the case of many compounds, at lower concentrations and/or for shorter time periods) did not (see Figure B-8). In general, rats, mice, and rabbits exposed to aromatic medium carbon range fraction compounds or mixtures during gestation showed treatment-related body-weight deficits; LOAELs for this effect were 125 mg/m^3 for mice exposed to HFAN, between 369 and 1,488 mg/m^3 for rats exposed to 1,3,5-TMB, 1,2,4-TMB, or isopropylbenzene, and 2,823 mg/m^3 in rabbits exposed to isopropylbenzene (see Figure B-9). Taken together, the inhalation and oral animal data indicate that compounds in the aromatic medium carbon range fraction can be expected to induce body-weight reductions at sufficiently high doses (generally ≥500 mg/kg-day or duration-adjusted concentrations ≥300 mg/m^3 for most compounds or mixtures).

HEMATOLOGICAL EFFECTS

A hematological endpoint (anemia in dogs, with males being affected more than females) is the critical effect for the screening-level subchronic and chronic p-RfDs for HFAN U.S. EPA (2009c). Hematological effects (including anemia and effects on blood clotting) have been reported for humans occupationally exposed to solvent mixtures including TMBs; hematotoxicity cannot be attributed to specific isomers. Hematological data in humans are limited to TMBs; there are animal data (inhalation or oral) for six members of the aromatic medium carbon range fraction.

Human Studies

Hematological effects have been reported in workers exposed by inhalation to mixtures containing TMB isomers. Workers exposed to paint solvent containing 50% 1,2,4-TMB, 30% 1,3,5-TMB, and unspecified amounts of 1,2,3-TMB (listed as possibly present) exhibited alterations in blood clotting and anemia at 295 mg/m^3 U.S. EPA (2016b). Because occupational exposure studies involve solvent mixtures, hematological and clinical chemistry effects cannot be attributed to TMB isomers and may be due to other agents in the mixture. For example, the hematological effects may be attributed to trace amounts of benzene present in the solvent mixture U.S. EPA (2016b).

Animal Studies

Reliable data regarding the hematotoxicity of aromatic medium carbon range compounds following oral exposure are limited to three members of the fraction. Figure B-10 is an exposure-response array containing studies for which hematological effects levels could be reliably determined. Increased monocytes were observed in rats treated with 1,3,5-TMB at ≥143 mg/kg-day for 90 days Adenuga et al., 2014 as cited in U.S. EPA (2016b), but no significant effects were reported after treatment with 1,4-DEB at up to 750 mg/kg-day for 6 weeks MHW, 1993b as cited in Oecd (1994). Although one 13-week study reported no significant, treatment-related effects on hematological parameters in rats treated with HFAN at up to 893 mg/kg-day Mobil Oil Company, 1994 as cited in U.S. EPA (2009c), two other studies of similar duration identified effects consistent with anemia (i.e., reductions in red blood cells [RBCs], hematocrit [Hct], and/or hemoglobin [Hb] levels) in rats (females only) treated at 1,250 mg/kg-day for up to 96 days Biodynamics, 1990a as cited in U.S. EPA (2009c) and in dogs treated at ≥250 mg/kg-day for 90 days Biodynamics, 1990b as cited in U.S. EPA (2009c).

There are limited data for hematological effects following inhalation exposure to members of the aromatic medium carbon range fraction. Figure B-11 is an exposure-response array containing studies for which hematological effects levels could be reliably determined. No significant, treatment-related hematological effects were observed in rats or mice exposed to isopropylbenzene at up to 907 mg/m^3 for 14 weeks Ntp (2009). Rats exposed to TMB isomers (1,2,4- and 1,2,3-TMB) at ≥88 mg/m^3 showed decreased clotting time, decreased segmented neutrophils, decreased RBCs, and/or changes in differential white blood cell (WBC) counts Korsak et al. 2000a, b as cited in U.S. EPA (2016b). Effects consistent with anemia (decreased Hct and mean corpuscular volume [MCV]) were reported in mice exposed to HFAN at 1,858 mg/m^3 (highest concentration tested) on GDs 6−15 McKee et al., 1990 as cited in U.S. EPA (2009c); in contrast, a different study in rats reported no hematological effects following 12 months of exposure to HFAN at concentrations up to approximately 300 mg/m^3 Clark et al., 1989 as cited in U.S. EPA (2009c).

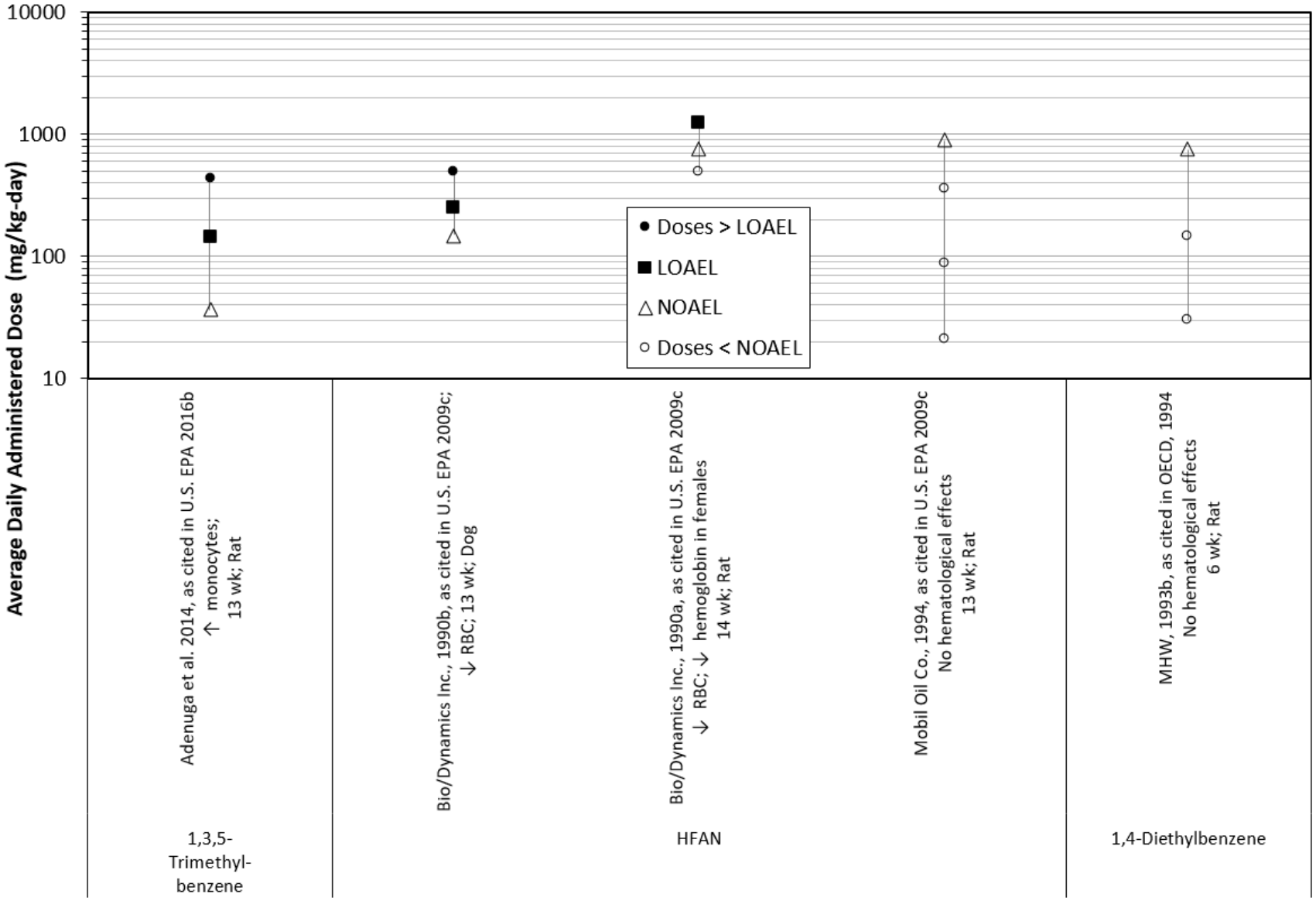


Figure B-10. Hematological Effects in Animals after Oral Exposure to Aromatic Medium Carbon Range Compounds

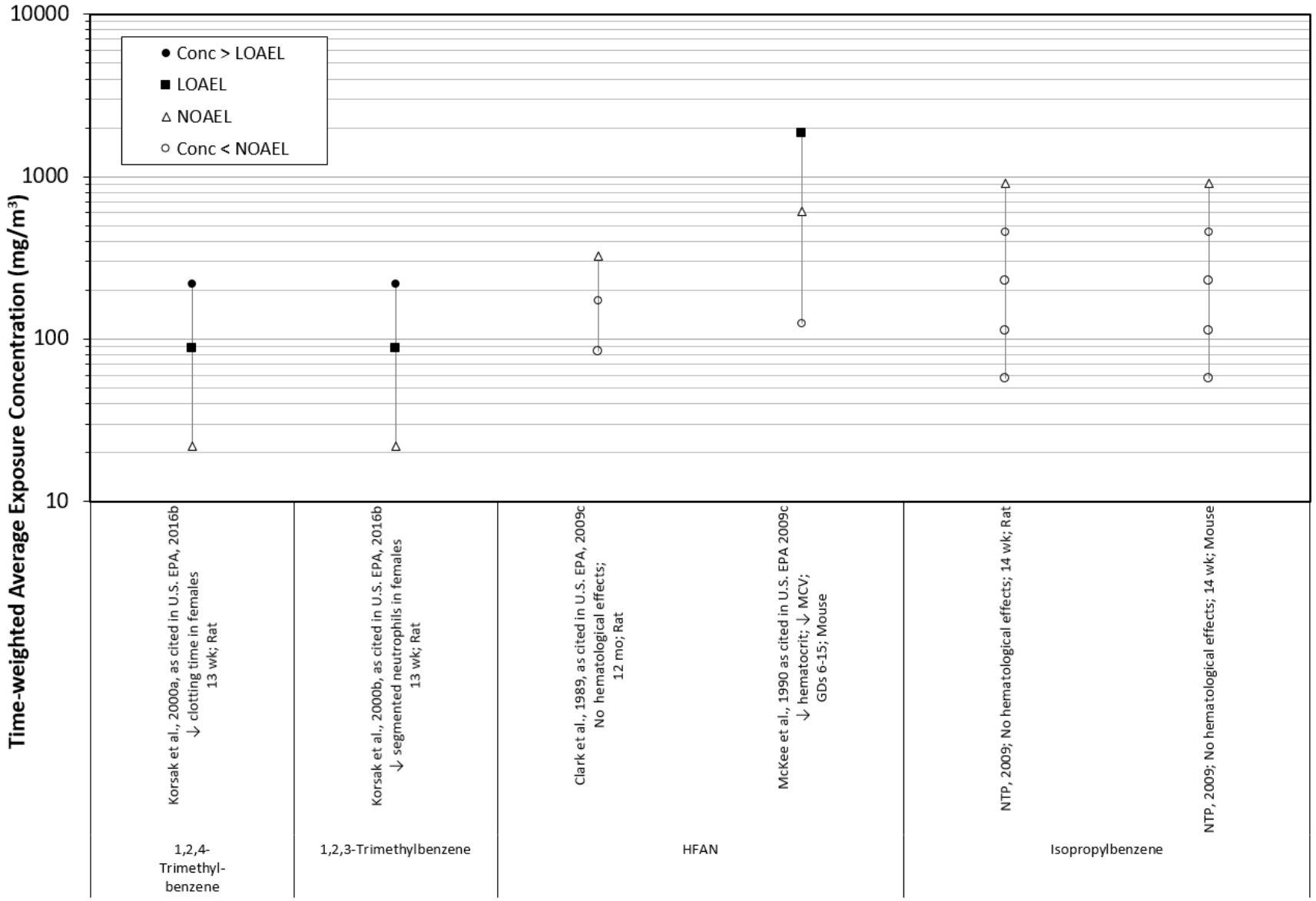


Figure B-11. Hematological Effects in Animals after Inhalation Exposure to Aromatic Medium Carbon Range Compounds

Summary of Potentially Relevant Evidence

Available data indicate that hematological effects associated with oral or inhalation exposure to TMBs (and HFAN) include changes in blood clotting parameters and other effects consistent with the development of anemia. Limited data are available for the other members of the fraction. Although available human data suggest that exposure to TMBs induces these effects, human data are insufficient to establish causal relationships (owing to coexposures with other compounds). In oral toxicity studies, rats treated with 1,3,5-TMB (at ≥143 mg/kg-day) and rats and dogs treated with HFAN (at 1,250 and ≥250 mg/kg-day, respectively) for 90−96 days showed increased monocytes and decreased Hct, Hb, and/or RBC counts (but no effects were reported in rats treated with HFAN or 1,4-DEB at lower doses). Rats exposed via inhalation to 1,2,4- or 1,2,3-TMB (at adjusted concentrations of ≥88 mg/m^3) and mice exposed to HFAN (at 1,858 mg/m^3 during gestation) showed similar types of effects (including decreased RBC counts, Hct, and MCV; and effects on differential WBC counts and clotting parameters), but no significant effects were reported after exposures of similar duration to isopropylbenzene. Taken together, the data suggest that at least some members of the aromatic medium carbon range fraction have the potential to induce anemia-like hematological effects. However, owing to the scarcity of data, it is not possible to evaluate the consistency in these endpoints and their potencies across members of the fraction.

ENDOCRINE EFFECTS

An endocrine endpoint (increased adrenal weight) is the cocritical effect (with increased kidney weight) for the chronic p-RfC for isopropylbenzene U.S. EPA (1997). No human data pertaining to the endocrine effects of aromatic medium carbon range fraction members were identified. As shown in Table 5, data (oral or inhalation) on endocrine effects in animals were located for five members of the fraction. The endpoints evaluated in the studies were organ weight and histology (adrenals, thyroid, parathyroid, and/or pituitary).

Human Studies

No human studies were available to address the potential for endocrine effects for any members of the aromatic medium carbon range fraction by any route of exposure.

Animal Studies

Studies that identify reliable effect levels for endocrine effects following oral exposure are limited to two members of the aromatic medium carbon range fraction. Significantly increased absolute and/or relative adrenal gland weights were reported in F_1 female rats treated with *n*-butylbenzene at 300 mg/kg-day for two generations Izumi, 2005 as cited in U.S. EPA (2010) and in female rats treated with HFAN at 893 mg/kg-day for 13 weeks Mobil Oil Company, 1994 as cited in U.S. EPA (2009c). In the former study, changes in adrenal gland weights occurred in the absence of histopathological changes; in the latter study, the adrenal glands were not examined microscopically, although the study authors performed gross necropsies on all animals and weighed major organs including the adrenal glands.

Data regarding endocrine effects from inhalation exposure to the aromatic medium carbon range compounds were identified for three members of the fraction. Based on organ weights and/or histopathology, there were no significant, treatment-related endocrine effects reported in rats or mice exposed to isopropylbenzene at up to 907 mg/m^3 for 14 or 105 weeks (including microscopic evaluations of the adrenals, pituitary, thyroid, and parathyroid) Ntp (2009) or rats exposed to 1,2,4- or 1,2,3-TMB at up to 220 mg/m^3 for 3 months (including

adrenal weights and histopathology) Korsak et al., 2000a, b as cited in U.S. EPA (2016b). However, absolute and relative adrenal gland weights were significantly increased in male and female rats following exposure to isopropylbenzene at 1,055 mg/m^3 for 13 weeks Cushman et al., 1995 as cited in U.S. EPA (1997).

Summary of Potentially Relevant Evidence

Data from animal toxicity studies suggest that members of the aromatic medium carbon range fraction induce effects on the endocrine system (specifically, increased adrenal gland weights). In oral toxicity studies, rats treated with *n*-butylbenzene at 300 mg/kg-day showed increased adrenal weights (in the absence of significant histological effects); rats treated with HFAN at 893 mg/kg-day also showed increased adrenal weights (histology was not evaluated). Oral data were not available for other members of the fraction. Data for endocrine effects via the inhalation route of exposure were limited to studies of isopropylbenzene and two isomers of TMB. 1,2,4- and 1,2,3-TMB induced no significant changes in adrenal gland weights or histopathology in rats. Comprehensive subchronic and chronic studies of isopropylbenzene in rats and mice (at exposures up to 907 mg/m^3) also failed to identify significant endocrine effects (including histological examinations of multiple endocrine organs). However, significantly increased adrenal gland weights were reported in rats exposed to a higher concentration of isopropylbenzene (1,055 mg/m^3) for 13 weeks. Taken together, the data suggest that at least some members of the aromatic medium carbon range fraction have the potential to induce endocrine effects (increased adrenal weights). However, owing to the scarcity of data, it is not possible to evaluate the consistency in these endpoints and their potencies across members of the fraction.

RESPIRATORY EFFECTS

None of the toxicity values for members of the aromatic medium carbon range fraction identify respiratory effects as the critical effect. Respiratory effects (mainly irritative effects) have been reported for humans occupationally exposed to solvents including TMBs; respiratory toxicity cannot be attributed to specific isomers. Respiratory toxicity data in humans are limited to TMBs; there are no data for most members of the aromatic medium carbon range fraction. Animal studies examining respiratory tract endpoints are available for five compounds (see Table 5); the preponderance of the animal data is from subchronic or chronic inhalation studies.

Human Studies

There is evidence for respiratory toxicity in humans from occupational and residential studies involving inhalation exposure to complex volatile organic compound (VOC) mixtures that include TMBs, but the effects cannot be attributed to any individual compound. Occupational exposure to complex VOC mixtures containing TMB isomers has been associated with respiratory irritation, including laryngeal and/or pharyngeal irritation and asthmatic bronchitis U.S. EPA (2016b). Residential exposure to mixtures containing 1,2,4-TMB were observed to be associated with an increase in asthma; however, as these studies involved exposures to mixtures containing multiple TMB isomers and other VOCs, it is difficult to ascertain the specific contribution of each TMB isomer to the specific health effects reported. U.S. EPA (2016b). Multiple studies in volunteers involving controlled acute (<4 hours) inhalation exposures to TMB isomers up to 25 ppm (123 mg/m^3) have not reported substantial irritation of the respiratory tract U.S. EPA (2016b). Studies of occupational and residential exposure to complex VOC mixtures suggest an association with asthmatic symptoms and

sensory irritation; however, because these exposures are to complex mixtures, the effects cannot be attributed to any specific constituent U.S. EPA (2016b).

Animal Studies

None of the available oral studies of compounds or mixtures relevant to the aromatic medium carbon range fraction examined respiratory tract effects in animals.

Data describing respiratory effects following inhalation exposure in animals for four members of the aromatic medium carbon range fraction were identified. Changes in the composition of the bronchoalveolar lavage (BAL) fluid were observed following inhalation exposure of rats to 1-methyl-4-ethylbenzene at 417 mg/m^3 for 4 weeks; this effect was accompanied by changes in lung histopathology (i.e., bronchitis, bronchopneumonia, and perivascular lymphocyte infiltration) Swiercz et al. (2000). Histopathological lesions were also observed in animals exposed to isopropylbenzene, 1,2,4-TMB, and 1,2,3-TMB. Increased incidences of hyperplasia of the nose (olfactory and respiratory epithelia) were reported in rats exposed to isopropylbenzene at ≥227 mg/m^3 for 105 weeks. Mice similarly exposed to isopropylbenzene showed lesions (metaplasia of the alveolar epithelium and bronchiole in the lung, accompanied by atrophy and hyperplasia in the nasal passages at higher concentrations) at ≥227 mg/m^3 (males) or ≥113 mg/m^3 (females) Ntp (2009). Significantly increased incidences of lesions of the lower respiratory tract were also noted in rats exposed to TMBs at ≥88 mg/m^3 for 3 months Korsak et al., 2000a, 2000b as cited in U.S. EPA (2016b). No respiratory tract lesions were observed in rats or rabbits exposed to high concentrations of isopropylbenzene (up to 1,488 mg/m^3 in rats and 2,823 mg/m^3 in rabbits) during gestation Darmer et al. (1997).

Summary of Potentially Relevant Evidence

Irritative effects and asthma have been reported in humans exposed to VOC mixtures containing TMBs; however, data are insufficient to attribute these effects to TMB isomers (owing to coexposures to other compounds). No animal oral data are available for any members of the aromatic medium carbon range fraction. In inhalation studies, changes in respiratory tract pathology (such as hypertrophy of the nose, metaplasia, bronchitis, bronchopneumonia in the lung) were reported following exposures to four members of the fraction (isopropylbenzene, 1-methyl-4-ethylbenzene, and 1,2,4- and 1,2,3-TMB); LOAELs for these effects ranged between 88 and 417 mg/m^3 in rats and mice. There are insufficient data to determine if respiratory effects are consistently associated with oral or inhalation exposure to members of the aromatic medium carbon range fraction.

DEVELOPMENTAL EFFECTS

Developmental toxicity is the critical effect for the *n*-propylbenzene subchronic and chronic p-RfCs U.S. EPA (2009d). The EPA's p-RfCs for *n*-propylbenzene were developed using a read-across approach where ethylbenzene was the selected analogue.

For members of this fraction, there are no suitable human data on developmental toxicity. Studies (oral or inhalation) examining developmental endpoints in animals are available for five compounds and one mixture relevant to the aromatic medium carbon range fraction.

Human Studies

Occupational exposure to or inhalant abuse of solvents that may contain TMBs by pregnant women results in a range of cognitive, behavioral, and visual dysfunctions among their children; however, the effects cannot be solely attributed to TMBs U.S. EPA (2016b).

Animal Studies

There are available data regarding developmental effects following oral exposure for three members of the aromatic medium carbon range fraction. No significant effects on development (based on evaluations of fetal body weights and external examinations of fetuses) were reported in rats treated with 1,4-DEB at up to 750 mg/kg-day for 6 weeks MHW, 1993b as cited in Oecd (1994). Increased thymus weight was noted in female F_2 offspring of rats treated with *n*-butylbenzene at 300 mg/kg-day; other reproductive/developmental parameters evaluated in the same two-generation study (including fertility and litter endpoints) were unaffected by treatment Izumi, 2005 as cited in U.S. EPA (2010). Decreased fetal body weights and delayed skeletal ossification (incompletely or unossified thoracic vertebral centrum and sacral vertebral transverse processes) were observed following treatment of dams with HFAN at 1,250 mg/kg-day on GDs 6−15 Biodynamics, 1990c as cited in U.S. EPA (2009c).

Developmental toxicity studies via inhalation are available for four members of the aromatic medium carbon range fraction (isopropylbenzene, 1,3,5- and 1,2,4-TMB, and HFAN); significant effects were reported for each compound. Figure B-12 is an exposure-response array containing studies for which effects levels could be reliably determined. In rabbits exposed to isopropylbenzene at 2,823 mg/m^3 on GDs 6−18, increased numbers of nonviable implants and resorptions, and a decreased percentage of live fetuses were reported (not statistically significant but considered biologically significant based on the U.S. EPA assessment under the IRIS program) Darmer et al. (1997). Developmental parameters were unaffected in rats similarly exposed to isopropylbenzene at up to 1,488 mg/m^3 on GDs 6−15 Darmer et al. (1997). Reductions in fetal body weights were observed in rats following exposure to TMB isomers at ≥738 mg/m^3 on GDs 6−20 Saillenfait et al., as cited in U.S. EPA (2016b). Fetal mortality, decreased fetal body weights, or delayed skeletal development were reported following exposures to HFAN. Rabbit dams treated at 1,000 mg/m^3 on GDs 7−20 showed complete abortion Ungvary and Tatrai, 1985 as cited in U.S. EPA (2009c), mice exposed at ≥613 mg/m^3 on GDs 6−15 showed decreased fetal body weights McKee et al., 1990 as cited in U.S. EPA (2009c), rats exposed at ≥434 mg/m^3 for three generations showed decreased pup weights during lactation McKee et al., 1990 as cited in U.S. EPA (2009c), and rats treated at 600 mg/m^3 on GDs 7−15 showed delays in skeletal ossification Ungvary and Tatrai, 1985 as cited in U.S. EPA (2009c). In another study, neurobehavior was not significantly impacted in the offspring of rats treated with HFAN at up to 500 mg/m^3 on GDs 7−15 Lehotsky et al., 1985 as cited in U.S. EPA (2009c).

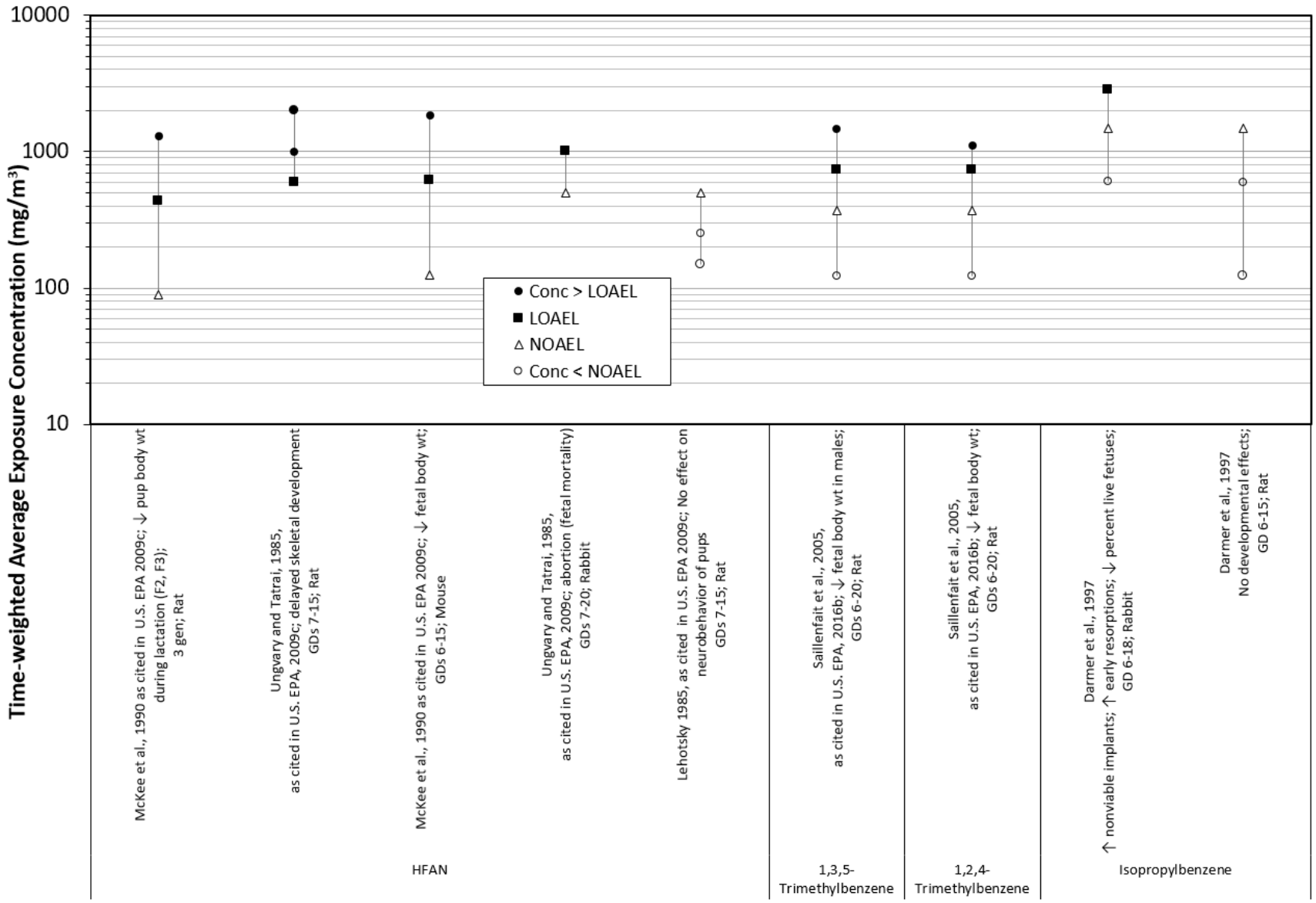


Figure B-12. Developmental Effects in Animals after Inhalation Exposure to Aromatic Medium Carbon Range

Summary of Potentially Relevant Evidence

Data from human studies indicate that exposure to solvents (presumably including members of the aromatic medium carbon range fraction) causes behavioral changes and visual and cognitive impairments. These data are insufficient to determine the specific role of aromatic medium carbon range compounds in the induction of these effects in humans. There are animal data (oral or inhalation) for six members of the fraction. Animal studies have identified effects (increased thymus weights) in rats orally exposed to *n*-butylbenzene (LOAEL of 300 mg/kg-day) and HFAN (decreased fetal body weights, delayed skeletal development) (LOAEL of 1,250 mg/kg-day). No significant effects on development were reported in rats treated with 1,4-DEB at up to 750 mg/kg-day. Inhalation toxicity studies of HFAN have likewise identified developmental effects (including decreased fetal/pup body weights and delayed skeletal ossification in rats, decreased fetal body weights in mice, and fetal death in rabbits). Similar effects (decreased numbers of live fetuses following isopropylbenzene exposure, and decreased body weights of offspring following 1,3,5- and 1,2,4-TMB exposure) were also identified after inhalation exposures to other members of the aromatic medium carbon range fraction. Lowest LOAELs (by compound or mixture) for these effects ranged from 434 to 738 mg/m^3 in rats and from 1,000 to 2,823 mg/m^3 in rabbits; a LOAEL of 613 mg/m^3 was identified for HFAN in mice (see Figure B-12). Taken together, data from oral and inhalation developmental toxicity studies consistently identify decreased fetal body weights for several members of the aromatic medium carbon range fraction.

OTHER EFFECTS

New studies identified in the PubMed searches identified reproductive effects in rats and mice exposed to isopropylbenzene by inhalation. The National Toxicology Program Ntp (2009) reported significant changes in the relative length of time spent in estrous and proestrus in treated female rats treated with isopropylbenzene for 14 weeks. In the same report, male mice treated at 907 mg/m^3 (also for 14 weeks) showed significant reductions in cauda epididymis weight and spermatid count. No other studies of sperm or estrous cycle parameters in humans or animals exposed to aromatic medium carbon range compounds or mixtures were located in the sources reviewed.

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