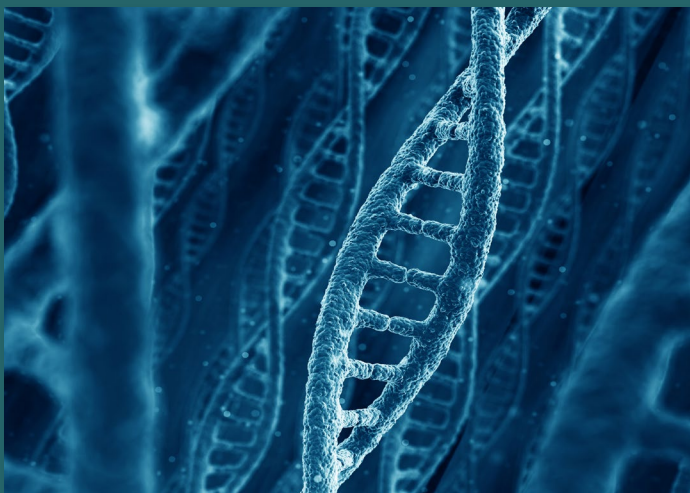


Comparison of Transcriptomic Reference Doses (TRVs) with Reference Doses/Concentrations (RfD/Cs) and Summary

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Comparison of TRVs with Traditional RfDs

- Formal statistical evaluation of concordance between TRV and RfD has primarily focused on BMD results (Reviewed in ETAP Sci Support Document, EPA 2023)
- Since the reference value is ultimately used to evaluate chemical risks, comparing traditional RfD and TRV values provides some understanding of the relative level of protection afforded by the ETAP
- 7 of the 14 chemicals used in concordance evaluation in the EPA report (EPA 2023, Gwinn et al. 2020) had EPA IRIS, EPA chronic PPRTV, or EPA Office of Water (OW) reference values

Comparison of TRVs with Traditional RfDs

- The median absolute ratio was 2.9 ± 1.4 (median absolute deviation)

Table 4-1. Comparison of Transcriptomic Reference Values (TRV) and Traditional RfD/provisional-RfD (p-RfD) Values for 7 of the 14 Chemicals Used on the Concordance Evaluation

Chemical	TRV (mg/kg-day)	RfD/ p-RfD (mg/kg-day)	TRV-to RfD Ratio	Source, Sex, Species, Study Type
Acrylamide	1.6E-04	2.0E-03	0.08	IRIS 2010; Male Rats; Chronic
Di(2-ethylhexyl) phthalate	1.1E-02	2.0E-02	0.55	IRIS 1987; Female Guinea Pigs; Subchronic-Chronic
Hexachlorobenzene	2.4E-05	8.0E-04	0.03	IRIS 1988; Male and Female Rats; Chronic
Furan	3.5E-04	1.0E-03	0.35	IRIS 1987; Male Mice; Subchronic
Perfluorooctanoic acid	3.1E-05	2.0E-05	1.55	OW 2016; Male Mice; Developmental
Tris(2-chloroisopropyl) phosphate	6.7E-03	1.0E-02	0.67	PPRTV Chronic 2012; Male Mice; Subchronic
Pentabromodiphenyl ether mixture (DE71)	4.1E-04	2.0E-03	0.21	IRIS 1987; Male Rats; Subchronic

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- Notably, the critical effect in four of the seven chemicals occurred in species other than the rat, which is the species utilized in ETAP
- For six of the seven chemicals, the TRV was lower than the RfD or provisional RfD (p-RfD), with PFOA as the only chemical with slightly higher TRV

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Additional Chemical Set

- In addition to the seven chemicals used to refine the dose response analysis parameters, 20 additional chemicals were identified from the literature review (EPA 2023) (EPA IRIS or EPA chronic PPRTV)
- A subset had multiple time points, tested species, or tissues with reported transcriptomic POD values
- Transcriptomic POD values were adjusted to HED using default body weights for species, strain, and sex used in the study (EPA 1988)
- While study designs and BMD analysis were not standardized across literature surveyed, a composite UF of 300 was used to calculate a TRV to evaluate general robustness of the approach and provide additional insight into level of protection afforded by ETAP

20 Chemicals – TRV to RfD Comparison

- A total of 22 of the 47 combinations used different species for the transcriptomic studies than the study used to derive the RfD or RfC
- A total of 28 of the 47 (~60%) combinations had TRVs that were more sensitive than the RfD/RfC; however, the relative sensitivity of the TRVs based on the open literature may be different compared with more standardized methods
- The Median Absolute Ratio = **2.3 ± 1.1** (MAD)
- Median Absolute Ratio (Non-Matched Species) = 3.2 ± 1.3 (MAD)
- Median Absolute Ratio (Matched Species) = 1.5 ± 1.1 (MAD)

Some Differences to Discuss: BDEs

- Max absolute ratio was 59-fold for 2,2',4,4'-tetrabromodiphenyl ether (BDE-47): transcriptomic changes were measured in rat liver after 5 days and the critical effect in the IRIS assessment was neurobehavioral changes in mice following single dose administration
- However, the RfD for 2,2',4,4'-tetrabromodiphenyl ether (BDE-47) used a composite UF of 3,000 to account for database uncertainties

Chemical	TRV (mg/kg-day or mg/m ³)	Exposure Duration (d)	Sex, Species, Tissue	Reference	RfD or RfC (mg/kg-day or mg/m ³)	Source, Sex, Species, Study Type	TRV-to-RfD Ratio
2,2',4,4'-Tetrabromodiphenyl ether (BDE-47)	5.9E-03	5	Male Rats, Liver	(Shockley et al. 2020)	1.0E-04	IRIS 2008, Male Mice, Single dose	58.89

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- However, the RfD for 2,2',4,4'-tetrabromodiphenyl ether (BDE-47) used a composite UF of 3,000 to account for database uncertainties
- By comparison, the absolute ratio between the TRV and RfD for 2,2',3,3',4,4',5,5',6,6'-decabromodiphenyl ether (BDE-209) was only 1.64-fold, even though the transcriptomic changes were also measured in the rat liver after 5 days and the critical effect in the IRIS assessment was also neurobehavioral changes in mice following a single dose.
- BDE-209 used a UF of 300

Chemical	TRV (mg/kg-day or mg/m ³)	Exposure Duration (d)	Sex, Species, Tissue	Reference	RfD or RfC (mg/kg-day or mg/m ³)	Source, Sex, Species, Study Type	TRV-to-RfD Ratio
2,2',3,3',4,4',5,5',6,6'-Decabromodiphenyl ether (BDE-209)	1.2E-02	5	Male Rats, Liver	(Shockley et al. 2020)	7.0E-03	IRIS 2008, Male Mice, Single dose	1.64
2,2',4,4'-Tetrabromodiphenyl ether (BDE-47)	5.9E-03	5	Male Rats, Liver	(Shockley et al. 2020)	1.0E-04	IRIS 2008, Male Mice, Single dose	58.89

Some Differences to Discuss: Naphthalene

- In addition to the bromodiphenyl ethers, the TRV value for naphthalene was approximately 19-fold higher based on the mouse lung compared with the RfC
- However, the RfC was based on adverse effects in the nasal epithelium in mice. When the TRV value for naphthalene was based on the nasal epithelium in rats, it was only 1.75-fold higher than the RfC

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Naphthalene	5.8E-02	91	Female Mice, Lung	(Thomas et al. 2011)	3.0E-03	IRIS 1998, Male and Female Mice, Chronic	19.22
Naphthalene ^a	5.2E-03	91	Male Rats, Nasal epithelium	(Clewell et al. 2014)	3.0E-03	IRIS 1998, Male and Female Mice, Chronic	1.75

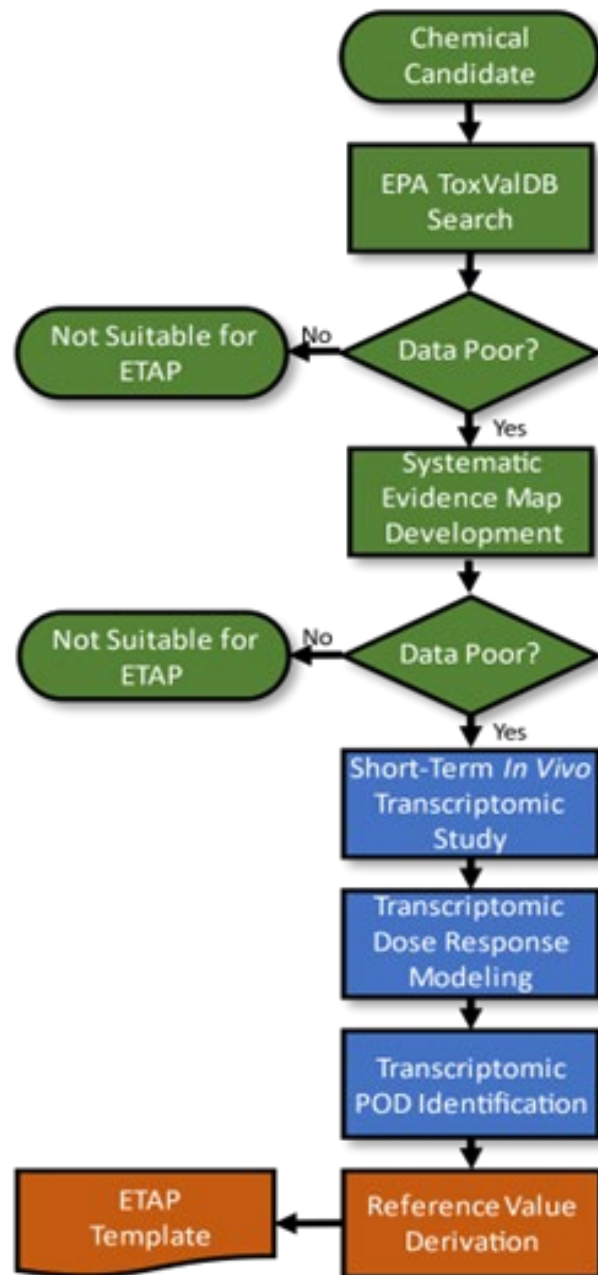
Concordance between the TRV and RfD/C

- For those combinations that used different species for the transcriptomic studies, the median absolute ratio was **3.2 + 1.3 (MAD)**,
- While those that used the same species had a median absolute ratio of **1.5 + 1.1 (MAD)**
- Overall, the results of the analysis suggest that the TRV provides a similar level of protection relative to the traditional RfD, p-RfD, and RfC values

Table 4-2: First row of the first 6 listed chemicals

Chemical	TRV (mg/kg-day or mg/m ³)	Exposure Duration (d)	Sex, Species, Tissue	Reference	RfD or RfC (mg/kg-day or mg/m ³)	Source, Sex, Species, Study Type	TRV-to-RfD Ratio
Acrylamide	1.1E-03	15	Male Mice, Lung	(Chepelev et al. 2018)	2.0E-03	IRIS 2010, Male Rats, Chronic	0.55
Allyl alcohol	6.3E-04	1	Male Rats, Liver	(Johnson et al. 2020)	5.0E-03	IRIS 1987, Male Rats, Subchronic	0.13
Benzo[a]pyrene	9.4E-05	3	Male Mice, Liver	(Moffat et al. 2015)	3.0E-04	IRIS 2017, Rats, Developmental	0.31
Bromobenzene	7.9E-03	1	Male Rats, Liver	(Johnson et al. 2020)	8.0E-03	IRIS 2009, Male Mice, Subchronic	0.99
Chloroprene ^a	1.4E-02	5	Female Mice, Lung	(Thomas et al. 2013a)	2.0E-02	IRIS 2010, Male and Female Rats, Female Mice, Chronic	0.68
Dichloroacetic acid	3.5E-02	6	Male Mice, Liver	(Cannizzo et al. 2022)	4.0E-03	IRIS 2003, Male and Female Dogs, Subchronic	8.67
...

Summary



- The ETAP provides a standardized process for systematic evidence mapping, *in vivo* exposure, sample collection, and RNA-seq analysis, transcriptomic dose response modeling, POD identification, and application of standard uncertainty factors to derive a TRV
- Comparison of transcriptomic toxicity values with traditional reference doses demonstrated similar levels of protection across a broad range of chemicals and effects
- The standardized reporting format and streamlined reporting process, founded on robust QA/QC review, enables straightforward release of ETAP results, ensuring timely delivery on ORD's human health mission objectives

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Reeder Sams

Avanti Shirke

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Jay Zhao

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