
Summary and Scientific Support Documents for Cycle 11 Recommended Groundwater Standards



WISCONSIN DEPARTMENT
of HEALTH SERVICES

P-02807 (11/2020)

Tony Evers
Governor

Andrea Palm
Secretary



State of Wisconsin
Department of Health Services

DIVISION OF PUBLIC HEALTH

1 WEST WILSON STREET
PO BOX 2659
MADISON WI 53701-2659

Telephone: 608-266-1251
Fax: 608-267-2832
TTY: 711 or 800-947-3529

November 6, 2020

Darsi Foss, Administrator
Division of Environmental Management
Department of Natural Resources
101 S Webster St, Box 7921
Madison, WI 53703

Dear Ms. Foss,

Enclosed is the 11th Cycle of Groundwater Standards Proposals. This packet includes proposals for new standards for 22 substances. Our recommendations include individual standards for 6 pesticides, individual standards for 12 per- and polyfluoroalkyl substances (PFAS), and combined standards for 4 PFAS. We did not recommend standards for 18 PFAS because of limited health information. Also enclosed with this letter is a summary table of our recommendations.

Please contact me if you have any questions regarding these recommendations. We look forward to working with you over the coming months as you use these proposals in rule-making.

Sincerely,

A handwritten signature in cursive script that reads "Stephanie Smiley".

Stephanie Smiley
Interim State Health Office and Division Administrator
Division of Public Health

Enclosures:

Summary of Cycle 11 Recommendations
Scientific Support Documents for the Cycle 11 Recommended Groundwater Standards

Summary of Cycle 11 Recommendations

Substance	Recommended Enforcement Standard ⁽ⁱ⁾	Recommended Preventive Action Limit ⁽ⁱ⁾
Metalaxyl	800 µg/L	160 µg/L
Chlorantraniliprole	16 mg/L	3.2 mg/L
Flumetsulam	10 mg/L	2 mg/L
Fomesafen	25 µg/L	5 µg/L
Hexazinone	400 µg/L	40 µg/L
Saflufenacil	460 µg/L	46 µg/L
Perfluorotridecanoic acid (PFTriA) ⁽ⁱⁱ⁾	No recommendations	
Perfluorotetradecanoic acid (PFTeA)	10 µg/L	2 µg/L
Perfluorobutanoic acid (PFBA)	10 µg/L	2 µg/L
Perfluoropentanoic acid (PFPeA) ⁽ⁱⁱ⁾	No recommendations	
Perfluorohexanoic acid (PFHxA)	150 µg/L	30 µg/L
Perfluoroheptanoic acid (PFHpA) ⁽ⁱⁱⁱ⁾	No recommendations	
Perfluorononanoic acid (PFNA)	30 ng/L	3 ng/L
Perfluorodecanoic acid (PFDA)	300 ng/L	60 ng/L
Perfluoroundecanoic acid (PFUnA)	3 µg/L	0.6 µg/L
Perfluorobutanesulfonic acid (PFBS)	450 µg/L	90 µg/L
Perfluorohexanesulfonic acid (PFHxS)	40 ng/L	4 ng/L
Perfluoroheptanesulfonic acid (PFHpS) ⁽ⁱⁱⁱ⁾	No recommendations	
Perfluorooctane sulfonamide (FOSA)	20 ng/L*	2 ng/L*
N-Ethyl Perfluorooctane sulfonamide (NEtFOSA)		
N-Ethyl perfluorooctane sulfonamidoacetic acid (NEtFOSAA)		
N-Ethyl perfluorooctane sulfonamidoethanol (NEtFOSE)		
Perfluorododecanoic acid (PFDoA)	500 ng/L	100 ng/L
6:2 Fluorotelomer sulfonic acid (6:2 FTSA) ⁽ⁱⁱ⁾	No recommendation	
8:2 Fluorotelomer sulfonic acid (8:2 FTSA) ⁽ⁱⁱ⁾	No recommendation	
Perfluorodecanesulfonic acid (PFDS) ⁽ⁱⁱⁱ⁾	No recommendations	
Perfluoropentanesulfonic acid (PFPeS) ⁽ⁱⁱⁱ⁾	No recommendations	
Hexafluoropropylene oxide dimer acid (HFPO-DA; GenX) ^(iv)	300 ng/L	30 ng/L
4:2 Fluorotelomer sulfonic acid (4:2 FTSA) ⁽ⁱⁱⁱ⁾	No recommendations	
10:2 Fluorotelomer sulfonic acid (10:2 FTSA) ⁽ⁱⁱⁱ⁾	No recommendations	
Perfluorohexadecanoic acid (PFHxDA) ⁽ⁱⁱⁱ⁾	No recommendations	
Perfluorooctadecanoic acid (PFODA)	400 µg/L	80 µg/L
4,8-Dioxa-3H-perfluorononanoic acid (DONA)	3 µg/L	0.6 µg/L

9-chlorohexanodecafluoro-3-oxanonane-1-sulfonic acid (9Cl-PF3ONS; F-53B Major) ^(iii; iv)	No recommendations
11-chloroeicosafluoro-3-oxaundecane-1-sulfonic acid (11Cl-PF3OUdS; F-53B Minor) ^(iii; iv)	No recommendations
Perfluorododecanesulfonic acid (PFDoS) ⁽ⁱⁱⁱ⁾	No recommendations
Perfluorononanesulfonic acid (PFNS) ⁽ⁱⁱⁱ⁾	No recommendations
N-Methyl Perfluorooctane sulfonamide (NMeFOSA) ⁽ⁱⁱⁱ⁾	No recommendations
N-Methyl perfluorooctane sulfonamidoacetic acid (NMetOSAA) ⁽ⁱⁱⁱ⁾	No recommendations
N-Methyl perfluorooctane sulfonamidoethanol (NMetOSE) ⁽ⁱⁱⁱ⁾	No recommendations

Notes:

- * DHS recommends a combined standard for NtFOSE, NtFOSA, NtFOSAA, FOSA, PFOS and PFOA.
- i. ng/L= nanograms per liter – equivalent to parts per trillion
µg/L = micrograms per liter – equivalent to parts per billion
mg/L = milligrams per liter – equivalent to parts per million
- ii. DHS reviewed this substance but did not recommend standards due to limited health information.
- iii. DHS did not review this substance because our screening indicated that there was not enough technical information to set a standard.
Ayers, J. F. Oct 28 2019. Revised Timeline for recommendations for state groundwater quality standards.
- iv. GenX and F-53B are a trade names.

Cycle 11 Recommendations – Table of Contents

Metalaxyl	1
Chlorantraniliprole	11
Flumetsulam	18
Fomesafen	25
Hexazinone	31
Saflufenacil	40
Perfluorotridecanoic acid (PFTriA)	47
Perfluorotetradecanoic acid (PFTeA)	61
Perfluorobutanoic acid (PFBA)	75
Perfluoropentanoic acid (PFPeA)	97
Perfluorohexanoic acid (PFHxA)	109
Perfluoroheptanoic acid (PFHpA)	131
Perfluorononanoic acid (PFNA)	141
Perfluorodecanoic acid (PFDA)	166
Perfluoroundecanoic acid (PFUnA)	200
Perfluorobutanesulfonic acid (PFBS)	216
Perfluorohexanesulfonic acid (PFHxS)	233
Perfluorooctane sulfonamide (FOSA)	263
N-Ethyl Perfluorooctane sulfonamide (NEtFOSA)	
N-Ethyl perfluorooctane sulfonamidoacetic acid (NEtFOSAA)	
N-Ethyl perfluorooctane sulfonamidoethanol (NEtFOSE)	
Perfluorododecanoic acid (PFDoA)	288
6:2 Fluorotelomer sulfonic acid (6:2 FTSA)	316
8:2 Fluorotelomer sulfonic acid (8:2 FTSA)	324
Perfluorodecanesulfonic acid (PFDS)	331
Hexafluoropropylene oxide dimer acid (HFPO-DA; GenX)	339
Perfluorooctadecanoic acid (PFODA)	364
4,8-Dioxa-3H-perfluorononanoic acid (DONA)	378

Metalaxyl | 2020

Substance Overview

Metalaxyl is a systemic pesticide that is commonly used to control plant diseases that are caused by water-mold fungi (Oomycetes).¹ Metalaxyl works by inhibiting the production of proteins in the ribosome which ultimately leads to the death of the fungi.²⁻⁵ Metalaxyl usually consists of a mixture between two molecules that are mirror images of each other, s-metalaxyl and r-metalaxyl. Both s-metalaxyl and r-metalaxyl can possess similar properties in the environment and body (Figure 1). Limited studies have suggested s-metalaxyl is more toxic than r-metalaxyl in cell culture studies.^{4,6} However, both enantiomers can display similar properties in the environment and in the body.^{4,6}

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard for metalaxyl. DHS recommends an enforcement standard of 800 micrograms per liter (µg/L) for metalaxyl. The recommended standard is based on EPA's chronic oral reference dose.⁷

DHS recommends that the preventive action limit for metalaxyl be set at 20% of the enforcement standard because metalaxyl has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture studies.⁷

Current Standards

Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

Recommended Standards

Enforcement Standard:	800 µg/L
Preventive Action Limit:	160 µg/L

Health Effects

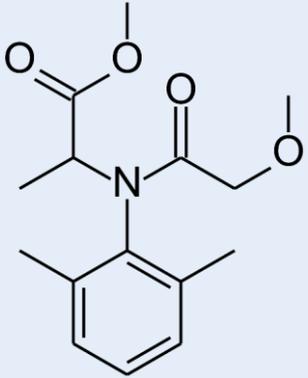
There are limited data on the health effects of metalaxyl on people. A single epidemiology study suggested metalaxyl exposure was significantly associated with non-fatal myocardial infarctions (heart attacks), but supporting evidence for this association is not available.⁸

Studies in research animals have shown limited effects of metalaxyl.^{7,9} In these research animal studies, the authors consistently found that exposure to metalaxyl can decrease body weight, decrease body weight gains during lactation, increase motor activity, decrease motor reflex, and at high concentrations metalaxyl can increase mortality of adults and their offspring.^{7,9} One subchronic study in dogs found that metalaxyl increased alkaline phosphatase levels which indicated liver damage and increased liver

weights (MRID 00071598).^{7,9} Similar results were observed in rats (MRID 00098481, 00132009, 00150185).^{7,9}

Metalaxyl has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell cultures.¹ The EPA states that metalaxyl did not show evidence of carcinogenicity for humans.^{7,9}

Chemical Profile

Metalaxyl	
Structure:	
CAS Number:	57837-19-1
Formula:	C ₁₅ H ₂₁ NO ₄
Molar Mass:	279.33 g/mol
Synonyms:	(+/-)-Metalaxyl Methyl N-(2-methoxyacetyl)-N-(2,6-xylyl)-DL-alaninate Metalaxil Metaxanin

Exposure Routes

The Wisconsin Department of Agriculture, Trade, and Consumer Protection (DATCP) has approved the use of a number of commercial and agricultural products containing metalaxyl for use in Wisconsin.¹⁰

People can be exposed to metalaxyl from contaminated food, soil, and water. Certain foods may have metalaxyl in or on them from its use as a fungicide.^{7,9} The EPA regulates how much pesticide residues can be in foods. People can get exposed to metalaxyl by touching recently treated plants or accidentally ingesting soil (dirt) near recently treated plants.^{7,9}

¹ Carcinogenic effects means the substance can cause cancer; mutagenic effects means the substance can cause DNA damage; teratogenic means the substance can cause birth defects; and interactive effects mean the substance can affect the toxicity of another substance or its toxicity can be affected by another substance.

Metalaxyl is moderately stable under normal environmental conditions and has a half-life of 400 days in water. ^{7,9} Metalaxyl is persistent and water soluble and has been detected in groundwater. ^{7,9}

Current Standard

Wisconsin does not currently have groundwater standards for Metalaxyl.¹¹

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

EPA Oral Reference Dose:	0.08 mg/(kg-d)	1994
--------------------------	----------------	------

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

None available

Literature Search

Literature Search Dates:	1994 – 2020
Total studies evaluated:	Approximately 50
Key studies found?	No
Critical studies identified?	No

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for metalaxyl.¹²

Health Advisory

The EPA has not established health advisories for metalaxyl.¹³

Drinking Water Concentration (Cancer Risk)

The EPA has not established a drinking water concentration based on a cancer risk level determination for metalaxyl.¹⁴

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for metalaxyl.¹⁵

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Information System (IRIS) program.

EPA Oral Reference Dose

In 1994, the EPA conducted a Human Health Risk Assessment as part of the registration of metalaxyl. In their assessment, EPA reviewed a number of toxicity studies.^{7,9} They selected a chronic study in dogs as the principal study (MRID 00071598). In this study, male and female dogs were exposed to different doses of metalaxyl (0, 1.6, 7.8, and 30.6 milligrams per kilogram per day or mg/kg-d) through diet for six months. From this study, the EPA selected a no-observable-effect-level (NOEL) of 7.8 mg/kg-d because negative effects on the liver were observed at higher doses in male dogs. The EPA used an uncertainty factor of 100 to account for differences between people and research animals (10) and differences among people (10). The EPA's chronic oral reference dose for metalaxyl is 0.08 mg/kg-d.⁷

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If we determine that something is carcinogenic and there is no federal number or ADI from the EPA, DHS must set the standard at a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of a metalaxyl, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of metalaxyl. If so, we look to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The International Agency for Research on Cancer (IARC) and the Joint FAO/WHO Meeting on Pesticide Residues (JMPR) have not evaluated the carcinogenicity of metalaxyl.^{16,17} The EPA has evaluated the carcinogenicity of metalaxyl in 1985 and concluded that metalaxyl did not have carcinogenic potential in research animals. The EPA classified metalaxyl as a chemical that does not show evidence of carcinogenicity for humans.^{16,17}

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for metalaxyl.^{7,9,14}

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information that was not considered when the value was established and indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For metalaxyl, we searched for values that have been published since 1994 when EPA published their reregistration document. We did not find any relevant guidance values.

Literature Search

Our literature review focused on relevant scientific literature on the health effects of metalaxyl published on or before 1994. We looked for studies related to metalaxyl toxicity or effects on a disease state in which information on exposure or dose was included as part of the study.² Ideally, relevant

² We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

Title/abstract: metalaxyl

Subject area: n/a

Language: English

We also searched online for toxicity studies published by national research programs.

studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

In our literature review, approximately 50 toxicity studies were returned by the search engines. We excluded studies on non-mammalian or cell systems, non-oral exposure routes, and those that did not evaluate health risks from further review. After applying these exclusion criteria, we did not identify any key studies.

In our literature review, we found one relevant epidemiology study that examined the effects of metalaxyl on human health (Table A1).⁸ While multiple potential exposure sources and the ability for other compounds to cause similar health effects preclude using these data to establish a health-based value, such studies are helpful in identifying the crucial effects and ensuring that the animal data used to establish the standard is relevant to people.

Critical Toxicity Studies

We did not identify any critical toxicity studies for metalaxyl.

Key health effects

We did not find any studies that suggest metalaxyl has caused carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture studies.

Discussion

Standard Selection

DHS recommends an enforcement standard of 800 µg/L for metalaxyl.

There are no federal numbers and no state drinking water standard for metalaxyl. Additionally, the EPA has not established a cancer slope factor for metalaxyl because they determined metalaxyl is non-carcinogenic to humans.

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information

However, the EPA does have an acceptable daily intake (oral reference dose) for metalaxyl.⁷ In our review, we did not find any significant technical information that was published since the EPA established their oral reference dose.⁷ Therefore, DHS calculated the recommended enforcement standard using the EPA's chronic oral reference dose, an average body weight of 10 kg, a water consumption rate of 1 L/d, and a relative source contribution factor of 100% as specified Chapter 160, Wis. Stats.

DHS recommends a preventive action limit of 160 µg/L for metalaxyl.

DHS recommends that the preventive action limit for metalaxyl be set at 20% of the enforcement standard because metalaxyl has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell cultures.⁷

Prepared by Gavin Dehnert, Ph.D. and Sarah Yang, Ph.D.

Wisconsin Department of Health Services

References

1. Liu T, Zhu L, Han Y, Wang J, Wang J, Zhao Y. The cytotoxic and genotoxic effects of metalaxyl-M on earthworms (*Eisenia fetida*). *Environmental toxicology and chemistry*. 2014;33(10):2344-2350.
2. Zhang P, Wang S, He Y, et al. Identifying Metabolic Perturbations and Toxic Effects of Rac-Metalaxyl and Metalaxyl-M in Mice Using Integrative NMR and UPLC-MS/MS Based Metabolomics. *Int J Mol Sci*. 2019;20(21).
3. Zhang P, Zhu W, Qiu J, et al. Evaluating the enantioselective degradation and novel metabolites following a single oral dose of metalaxyl in mice. *Pesticide biochemistry and physiology*. 2014;116:32-39.
4. Zhang P, Zhu W, Wang D, Yan J, Wang Y, He L. Enantioselective Effects of Metalaxyl Enantiomers on Breast Cancer Cells Metabolic Profiling Using HPLC-QTOF-Based Metabolomics. *Int J Mol Sci*. 2017;18(1).
5. Zhang R, Zhou Z. Effects of the Chiral Fungicides Metalaxyl and Metalaxyl-M on the Earthworm *Eisenia fetida* as Determined by ¹H-NMR-Based Untargeted Metabolomics. *Molecules (Basel, Switzerland)*. 2019;24(7).
6. Wang X, Zhu W, Qiu J, Zhang P, Wang Y, Zhou Z. Enantioselective metabolism and toxic effects of metalaxyl on primary hepatocytes from rat. *Environmental science and pollution research international*. 2016;23(18):18649-18656.
7. USEPA. Reregistration Eligibility Decision (RED) Metalaxyl. 1994.

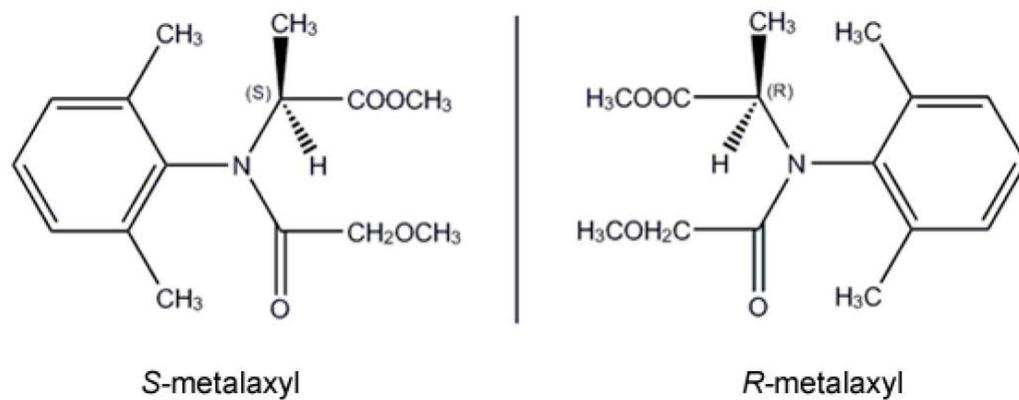
8. Dayton SB, Sandler DP, Blair A, Alavanja M, Beane Freeman LE, Hoppin JA. Pesticide use and myocardial infarction incidence among farm women in the agricultural health study. *Journal of occupational and environmental medicine*. 2010;52(7):693-697.
9. USEPA. R.E.D. Facts Metalaxyl. 1994.
10. DATCP. Pesticide Database Searches. 2016; <https://www.kellysolutions.com/wi/pesticideindex.asp>.
11. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 140*2017.
12. USEPA. National Primary Drinking Water Regulations. 2018; <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>.
13. USEPA. Drinking Water Contaminant Human Health Effects Information. 2019; <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>.
14. USEPA. IRIS Assessments. 2019; https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm.
15. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 809*2018.
16. IARC. List of Classification, Volumes 1-123. 2018; <https://monographs.iarc.fr/list-of-classifications-volumes/>. Accessed May 17, 2019.
17. JMPR. Inventory of evaluations performed by the Joint Meeting on Pesticide Residues (JMPR). 2012; <http://apps.who.int/pesticide-residues-jmpr-database>. Accessed May 24, 2019.

Appendix A. Toxicity Data

Table A-I. Metalaxyl Epidemiological Studies from Literature Review

Study Type	Population	Time period	Exposure	Outcomes	Results	Reference
Prospective Study	Females in the Agricultural Health Study Population from Iowa and North Carolina	1993-1997	Participants were exposed to the pesticides either as pesticide applicators or pesticide applicators spouses.	Myocardial infarction (heart attack)	Metalaxyl use was associated with an increase in myocardial infarctions (OR = 2.4; 95% CI 1.1, 5.3).	Dayton, 2010
OR: Odds Ratio 95% CI: 95% confidence interval						

Appendix B. Chemical structure of metalaxyl enantiomers.



Chemical structure of metalaxyl enantiomers based on figure 1 of Zhang et al. 2017.⁴

Chlorantraniliprole | 2020

Substance Overview

Chlorantraniliprole is an insecticide used to control a variety of pests on a number of plants.^{1,2} Chlorantraniliprole works by interrupting normal muscle contraction causing paralysis and death.^{1,2}

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard for chlorantraniliprole. DHS recommends an enforcement standard of 16 milligrams per liter (mg/L) for chlorantraniliprole. The recommended standard is based on based on the United States Environmental Protection Agency's (EPA's) chronic oral reference dose for chlorantraniliprole.¹

DHS recommends that the preventive action limit for chlorantraniliprole be set at 20% of the enforcement standard because chlorantraniliprole has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture studies.

Current Standards	
Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

Recommended Standards	
Enforcement Standard:	16 mg/L
Preventive Action Limit:	3.2 mg/L

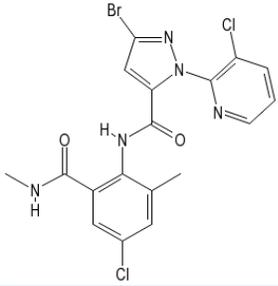
Health Effects

Studies in research animals have shown limited effects of chlorantraniliprole.^{1,2} The most consistent effect of chlorantraniliprole in these studies is induction of liver enzymes and increases in liver weight.² One long-term study in rodents found that chlorantraniliprole affected the formation of microvesicles (parts of a cell's membrane) in the adrenal gland, but did not affect adrenal gland function.²

Chlorantraniliprole has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture studies.^{1,2a} The EPA has classified chlorantraniliprole as not likely to be carcinogenic to humans.^{1,2}

a Carcinogenic effects means the substance can cause cancer; mutagenic effects means the substance can cause DNA damage; teratogenic means the substance can cause birth defects; and interactive effects mean the substance can affect the toxicity of another substance or its toxicity can be affected by another substance.

Chemical Profile

Chlorantraniliprole	
Structure:	 The chemical structure of Chlorantraniliprole is shown. It consists of a central benzene ring substituted with a methyl group, a chlorine atom, and a methylcarbamoyl group (-NH-C(=O)-CH3). This benzene ring is connected via an amide bond (-NH-C(=O)-) to a pyrazole ring. The pyrazole ring has a bromine atom at the 3-position and is further substituted at the 1-position with a 3-chloro-2-pyridinyl group.
CAS Number:	500008-45-7
Formula:	C ₁₈ H ₁₄ N ₅ O ₂ BrCl ₂
Molar Mass:	483.15 g/mol
Synonyms:	3-Bromo- <i>N</i> -[4-chloro-2-methyl-6-(methylcarbamoyl)phenyl]-1-(3-chloro-2-pyridine-2-yl)-1H-pyrazole-5-carboxamide

Exposure Routes

The Wisconsin Department of Agriculture, Trade, and Consumer Protection (DATCP) has approved the use of a number of commercial products containing chlorantraniliprole for use in Wisconsin.³

People can be exposed to chlorantraniliprole from food, soil, and water.^{1,2} Certain foods may have some chlorantraniliprole in or on them from its use as a pesticide. The EPA regulates how much pesticide residues can be in foods. People can get exposed to chlorantraniliprole by touching recently treated plants or by touching or accidentally ingesting soil (dirt) in or near recently treated plants.

Chlorantraniliprole can leach through the soil and enter the groundwater.^{1,2} Chlorantraniliprole is highly soluble in water and has the potential to travel in groundwater.^{1,2}

Current Standard

Wisconsin does not currently have groundwater standards for chlorantraniliprole.⁴

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

EPA Oral Reference Dose:	1.58 mg/kg-d	(2010)
--------------------------	--------------	--------

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

None available

Literature Search

Literature Search Dates:	2010 – 2019
Key studies found?	No
Critical studies identified?	No

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for chlorantraniliprole.⁵

Health Advisory

The EPA has not established health advisories for chlorantraniliprole.⁶

Drinking Water Concentration (Cancer Risk)

The EPA has not established drinking water concentration based cancer risk for chlorantraniliprole.⁷

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for chlorantraniliprole.⁸

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Assessment System (IRIS) program.

EPA Oral Reference Dose

In 2010, the EPA conducted a Human Health Risk Assessment as part of the registration of chlorantraniliprole. In their assessment, EPA reviewed a number of toxicity studies.^{1,2} They selected a chronic/carcinogenicity study in mice as the principal study. In this study, male and female rats were exposed to different doses of chlorantraniliprole (males: 0, 2.6, 9.2, 26.1, 158, 935 milligrams per kilogram body weight per day or mg/kg-d; females: 0, 3.34, 11.6, 32.9, 196, 1155 mg/kg-d) through diet for 18 months. From this study, the EPA selected a No Observable Adverse Effect Level (NOAEL) of 158 mg/kg-d because effects on the liver were observed at the highest dose. The EPA used a total uncertainty factor of 100 to account for differences between people and research animals (10) and differences among people (10). The EPA's chronic oral reference for chlorantraniliprole is 1.58 mg/kg-d.

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If we determine that something is carcinogenic and there is no federal number or ADI from the EPA, DHS must set the standard at a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of chlorantraniliprole, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of chlorantraniliprole. If so, we look to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA has classified chlorantraniliprole as not likely to be carcinogenic to humans.^{1,2}

The International Agency for Research on Cancer (IARC) and the Joint FAO/WHO Meeting on Pesticide Residues (JMPR) have not evaluated the carcinogenicity of chlorantraniliprole.^{9,10}

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for chlorantraniliprole.^{1,2}

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information that was not considered when the value was established and indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For chlorantraniliprole, we searched for values that have been published since 2010 when EPA published their human health risk assessment. We did not find any relevant guidance values.

Literature Search

Our literature review focused on the scientific literature published after EPA's human health risk assessment in 2010. We looked for studies related to chlorantraniliprole toxicity or effects on a disease state in which information on exposure or dose was included as part of the study.^b Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

Approximately 20 studies were returned by the search engines. We excluded monitoring studies, studies evaluating risk from non-mammalian species, and studies on the effects on plants from further review. After applying these exclusion criteria, we did not identify any key studies.

Critical Toxicity studies

We did not identify any critical toxicity studies.

Key Health effects

We did not find studies that show chlorantraniliprole has caused carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture studies.

^b We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

Title/abstract: chlorantraniliprole

Subject area: toxicology

Language: English

We also searched online for toxicity studies published by national research programs.

Standard Selection

DHS recommends an enforcement standard of 16 mg/L for chlorantraniliprole.

There are no federal numbers and no state drinking water standard for chlorantraniliprole. Additionally, the EPA has not established a cancer slope factor for chlorantraniliprole because they determined that it is not likely to be carcinogenic to humans.

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information

However, the EPA does have an acceptable daily intake (oral reference dose) for chlorantraniliprole.¹ In our review, we did not find any significant technical information that was published since the EPA established their oral reference dose. Therefore, DHS calculated the recommended enforcement standard using the EPA's oral reference dose, an average body weight of 10 kg, and a water consumption rate of 1 L/d as specified Chapter 160, Wis. Stats.

DHS recommends a preventive action limit of 3.2 mg/L for chlorantraniliprole.

DHS recommends that the preventive action limit for chlorantraniliprole be set at 20% of the enforcement standard because chlorantraniliprole has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture.^{1,2}

Prepared by Sarah Yang, Ph.D.

Wisconsin Department of Health Services

References

1. USEPA. Memorandum: Chlorantraniliprole; Human Health Risk Assessment for Proposed Use on Tobacco. In: Office of Prevention P, and Toxic Substances, ed. Vol DP#3692242010.
2. USEPA. Pesticide Factsheet: Chlorantraniliprole. In: Office of Prevention P, and Toxic Substances, ed2008.
3. DATCP. Pesticide Database Searches. 2016; <https://www.kellysolutions.com/wi/pesticideindex.asp>.
4. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 1402017*.
5. USEPA. National Primary Drinking Water Regulations. 2018; <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>.
6. USEPA. Drinking Water Contaminant Human Health Effects Information. 2019; <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>.
7. USEPA. IRIS Assessments. 2019; https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm.

8. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 809*2018.
9. Wolterink GD, V. Chlorantraniliprile. In: JMPR, ed2008.
10. IARC. List of Classification, Volumes 1-123. 2018; <https://monographs.iarc.fr/list-of-classifications-volumes/>. Accessed May 17, 2019.

Flumetsulam | 2020

Substance Overview

Flumetsulam is an herbicide in the triazolopyrimidine chemical class that is used to control broadleaf weeds and grasses.¹⁻⁴ Flumetsulam works on both pre- and post-emergent plants by inhibiting the enzyme acetolactate synthase, which regulates plant growth and eventually leads to plant death.¹⁻⁴

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard for flumetsulam. DHS recommends an enforcement standard of 10 milligrams per liter (mg/L) for flumetsulam. The recommended standard is based on based on the United States Environmental Protection Agency's (EPA's) chronic oral reference dose for flumetsulam.^{3,4}

DHS recommends that the preventive action limit for flumetsulam be set at 20% of the enforcement standard because flumetsulam has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture studies.^{3,4}

Current Standards

Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

Recommended Standards

Enforcement Standard:	10 mg/L
Preventive Action Limit:	2 mg/L

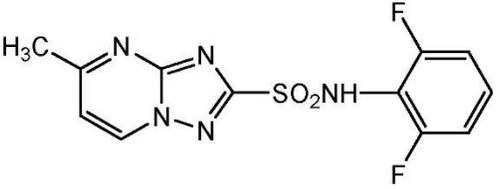
Health Effects

Studies in research animals have shown limited effects of flumetsulam.^{3,4} In these studies, flumetsulam exposure consistently shows negative impacts on kidneys. One chronic study in dogs found that flumetsulam increased the presence of kidney stones, increased inflammation in the kidney, and decreased the size of the kidney. Similar results were observed in a chronic rat study.^{3,4} In our literature review, we did not find any relevant epidemiology studies that examined the effects of flumetsulam on human health.

Flumetsulam has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture studies.^{3,4,1} The EPA states there is evidence to support that flumetsulam is non-carcinogenic to humans.^{3,4}

1 Carcinogenic effects means the substance can cause cancer; mutagenic effects means the substance can cause DNA damage; teratogenic means the substance can cause birth defects; and interactive effects mean the substance can affect the toxicity of another substance or its toxicity can be affected by another substance.

Chemical Profile

Flumetsulam	
Structure:	
CAS Number:	98967-40-9
Formula:	C ₁₂ H ₉ F ₂ N ₅ O ₂ S
Molar Mass:	325.3 g/mol
Synonyms:	N-(2,6-difluorophenyl)-5-methyl-[1,2,4]triazolo[1,5-a]pyrimidine-2-sulfonamide Broadstrike XRD 498 DE 498

Exposure Routes

The Wisconsin Department of Agriculture, Trade, and Consumer Protection (DATCP) has approved the use of a number of agricultural products containing flumetsulam for use in Wisconsin.⁵

People can be exposed to flumetsulam from contaminated food, soil, and water. Certain foods may have flumetsulam in or on them from its use as a pesticide.^{3,4} The EPA regulates how much pesticide residues can be in foods. People can get exposed to flumetsulam by touching recently treated plants or accidentally ingesting soil (dirt) near recently treated plants.

Flumetsulam is stable under photolysis and is water soluble.^{3,4} Flumetsulam can leach through soil and contaminate surface water and groundwater.^{3,4,6}

Current Standard

Wisconsin does not currently have groundwater standards for flumetsulam.⁷

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

EPA Oral Reference Dose:	1 mg/kg-d	2013
--------------------------	-----------	------

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

None available

Literature Search

Literature Search Dates:	2013 - 2020
Total studies evaluated:	Approximately 30
Key studies found?	No
Critical studies identified?	No

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for flumetsulam.⁸

Health Advisory

The EPA has not established health advisories for flumetsulam.⁹

Drinking Water Concentration (Cancer Risk)

The EPA has not established a drinking water concentration based on a cancer risk level determination for flumetsulam.¹⁰

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for flumetsulam.¹¹

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Information System (IRIS) program.

EPA Oral Reference Dose

In 2013, the EPA conducted a Preliminary Human Health Risk Assessment for flumetsulam as part of the pesticide's registration review. In their assessment, EPA reviewed a number of toxicity studies.^{3,4} They selected a chronic study in dogs as the principal study (MRID 41952103). In this study, male and female dogs were exposed to different doses of flumetsulam (0, 20, 100, and 500 milligrams per kilogram per day or mg/kg-d) through diet for one year. From this study, the EPA selected a no-observable-adverse-effect-level (NOAEL) of 100 mg/kg-d because effects on the kidney were observed at higher doses in male dogs. The EPA used an uncertainty factor of 100 to account for differences between people and research animals (10) and differences among people (10). The EPA's chronic oral reference dose for flumetsulam is 1 mg/kg-d.^{3,4}

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If we determine that something is carcinogenic and there is no federal number or ADI from the EPA, DHS must set the standard at a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of flumetsulam, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of flumetsulam. If so, we look to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA has evidence to support flumetsulam is non-carcinogenic to humans.^{3,4}

The International Agency for Research on Cancer (IARC) and the Joint FAO/WHO Meeting on Pesticide Residues (JMPR) has not evaluated the cancer potential of flumetsulam.^{12,13}

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for flumetsulam.^{3,4}

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information that was not considered when the value was established and indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For flumetsulam, we searched for values that have been published since 2013 when EPA published their Preliminary Human Health Risk Assessment for Registration Review. We did not find any relevant guidance values.

Literature Search

Our literature review focused on the scientific literature published after the Preliminary Human Health Risk Assessment for Registration Review of flumetsulam in 2013. We looked for studies related to flumetsulam toxicity or effects on a disease state in which information on exposure or dose was included as part of the study.² Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

In our literature review, we did not find any relevant epidemiology studies that examined the effects of flumetsulam on human health.

² We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

Title/abstract: Flumetsulam

Subject area: N/A

Language: English

We also searched online for toxicity studies published by national research programs.

Approximately 30 toxicity studies were returned by the search engines. We excluded studies on non-mammalian or cell systems, non-oral exposure routes, and those that did not evaluate health risks from further review. After applying these exclusion criteria, we did not identify any key studies.

Critical toxicity studies

We did not identify any critical toxicity studies.

Key Health effects

In our literature search, we did not find studies that show flumetsulam has caused carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture studies.

Standard Selection

DHS recommends an enforcement standard of 10 mg/L for flumetsulam.

There are no federal numbers and no state drinking water standard for flumetsulam. Additionally, the EPA has not established a cancer slope factor for flumetsulam because they determined there is evidence to support flumetsulam is non-carcinogenic to humans.

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information

However, the EPA does have an acceptable daily intake (oral reference dose) for flumetsulam.^{3,4} In our review, we did not find any significant technical information that was published since the EPA established their oral reference dose. Therefore, DHS calculated the recommended enforcement standard using the EPA's chronic oral reference dose, an average body weight of 10 kg, a water consumption rate of 1 L/d, and a relative source contribution factor of 100% as specified Chapter 160, Wis. Stats.

DHS recommends a preventive action limit of 2 mg/L for flumetsulam.

DHS recommends that the preventive action limit for flumetsulam be set at 20% of the enforcement standard because flumetsulam has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell cultures.^{3,4}

Prepared by Gavin Dehnert, Ph.D. and Sarah Yang, Ph.D.

Wisconsin Department of Health Services

References

1. Chen CN, Lv LL, Ji FQ, et al. Design and synthesis of N-2,6-difluorophenyl-5-methoxyl-1,2,4-triazolo 1,5-a -pyrimidine-2-sulfo namide as acetohydroxyacid synthase inhibitor. *Bioorganic & Medicinal Chemistry*. 2009;17(8):3011-3017.
2. Fontaine DD, Lehmann RG, Miller JR. SOIL ADSORPTION OF NEUTRAL AND ANIONIC FORMS OF A SULFONAMIDE HERBICIDE, FLUMETSULAM. *Journal of Environmental Quality*. 1991;20(4):759-762.
3. USEPA. Preliminary human health risk assessment for registration review. In:2013.
4. USEPA. Flumetsulam: HED risk assessment for the tolerance reassessment eligibility document (TRED). In:2004.
5. DATCP. Pesticide Database Searches. <https://www.kellysolutions.com/wi/pesticideindex.asp>. Published 2016. Accessed.
6. Tingle CH, Shaw DR, Gerard PD. Flumetsulam mobility in two Mississippi soils as influenced by irrigation timing. *Weed Science*. 1999;47(3):349-352.
7. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 140*2017.
8. USEPA. National Primary Drinking Water Regulations. <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>. Published 2018. Accessed.
9. USEPA. Drinking Water Contaminant Human Health Effects Information. <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>. Published 2019. Accessed.
10. USEPA. IRIS Assessments. https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm. Published 2019. Accessed.
11. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 809*2018.
12. JMPR. Inventory of evaluations performed by the Joint Meeting on Pesticide Residues (JMPR). <http://apps.who.int/pesticide-residues-jmpr-database>. Published 2012. Accessed May 24, 2019.
13. IARC. List of Classification, Volumes 1-123. <https://monographs.iarc.fr/list-of-classifications-volumes/>. Published 2018. Accessed May 17, 2019.

Fomesafen | 2020

Substance Overview

Fomesafen^a is an herbicide used to control a variety of weeds and grasses.¹ Fomesafen works by inhibiting the enzyme protoporphyrinogen oxidase (PPO) leading to irreversible plant cell membrane damage. Fomesafen products are used on a variety of fruit and vegetable plants.¹

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard for fomesafen. DHS recommends an enforcement standard of 25 milligrams per liter (mg/L) for fomesafen. The recommended standard is based on the United States Environmental Protection Agency's (EPA's) chronic oral reference dose for fomesafen.¹

DHS recommends that the preventive action limit for fomesafen be set at 20% of the enforcement standard because fomesafen has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture studies.

Current Standards	
Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

Recommended Standards	
Enforcement Standard:	25 µg/L
Preventive Action Limit:	5 µg/L

Health Effects

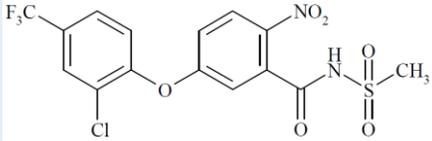
Studies in research animals have shown that high levels of fomesafen can affect food consumption, lower body weight, damage the liver, cause miscarriages, and reduce immune response.¹

Fomesafen has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture studies.^b The EPA has classified fomesafen as not likely to be carcinogenic to humans.¹

a Fomesafen products are formulated as a sodium salt and the concentration of the active ingredient in the formulation is expressed in terms of the acid equivalent. This scientific support document and the included groundwater standard recommendations apply to both the acid and salt of fomesafen.

b Carcinogenic effects means the substance can cause cancer; mutagenic effects means the substance can cause DNA damage; teratogenic means the substance can cause birth defects; and interactive effects mean the substance can affect the toxicity of another substance or its toxicity can be affected by another substance.

Chemical Profile

Fomesafen	
Structure:	
CAS Number:	72178-02-0
Formula:	C ₁₅ H ₁₀ ClF ₃ N ₂ O ₆ S
Molar Mass:	438.8 g/mol
Synonyms:	5-[2-Chloro-4-(trifluoromethyl)phenoxy]-N-(methanesulfonyl)-2-nitrobenzamide 5-[2-Chloro-α, α, α-trifluoro-p-tolyloxy]-N-(methanesulfonyl)-2-nitrobenzamide

Exposure Routes

The Wisconsin Department of Agriculture, Trade, and Consumer Protection (DATCP) has approved a number of commercial products containing fomesafen for use in Wisconsin.²

People can be exposed to fomesafen from food, air, soil, and water.¹ Certain foods may have some fomesafen in or on them from its use as a pesticide. The EPA regulates how much pesticide residue can be in foods. People can get exposed to fomesafen by walking through recently sprayed areas by breathing in air or touching sprayed soil. Fomesafen can leach through the soil and enter the groundwater.^{3,4} Fomesafen is highly soluble in water and has the potential to travel in groundwater.^{1,5}

Current Standard

Wisconsin does not currently have groundwater standards for fomesafen.⁶

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

EPA Oral Reference Dose:	0.0025 mg/kg-d	(2013)
--------------------------	----------------	--------

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

None available

Literature Search

Literature Search Dates:	2013 – 2019
Total studies evaluated:	Approximately 20

Key studies found?	No
Critical studies identified?	No

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for fomesafen.⁷

Health Advisory

The EPA has not established health advisories for fomesafen.⁸

Drinking Water Concentration (Cancer Risk)

The EPA has not established drinking water concentration based cancer risk for fomesafen.⁹

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for fomesafen.¹⁰

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Assessment System (IRIS) program.

EPA Oral Reference Dose

In 2013, the EPA conducted a Human Health Risk Assessment as part of the registration of fomesafen sodium. In their assessment, EPA reviewed a number of toxicity studies.¹ They selected a chronic/carcinogenicity study in rats as the principal study (MRID: 00142125). In this study, male and female rats were exposed to different doses of fomesafen (0, 0.25, 5, and 50 milligrams per kilogram body weight per day or mg/kg-d) through diet for 2 years. From this study, the EPA selected a No

Observable Adverse Effect Level (NOAEL) of 0.25 mg/kg-d because higher doses caused hyalinization of the liver in male animals.^c The EPA used a total uncertainty factor of 100 to account for differences between people and research animals (10) and differences among people (10). The EPA's chronic oral reference dose for fomesafen is 0.0025 mg/kg-d.

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If we determine that something is carcinogenic and there is no federal number or ADI from the EPA, DHS must set the standard at a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of fomesafen, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of fomesafen. If so, we look to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA has classified fomesafen as not likely to be carcinogenic to humans.¹

The International Agency for Research on Cancer (IARC) and the Joint FAO/WHO Meeting on Pesticide Residues (JMPR) have not evaluated the carcinogenicity of fomesafen.^{11,12}

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for fomesafen.¹

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information that was not considered when the value was established and indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For fomesafen, we searched for values that had been published since 2013 when EPA published their human health risk assessment. We did not find any relevant guidance values.

Literature Search

^c Hyalinization is a condition in which normal tissue deteriorates into a homogeneous, translucent material.

Our literature review focused on the scientific literature published after EPA’s human health risk assessment in 2013. We looked for studies related to fomesafen toxicity or effects on a disease state in which information on exposure or dose was included as part of the study.^d Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

Approximately 20 studies were returned by the search engines. We excluded monitoring studies, studies evaluating risk from non-mammalian species, and studies on the effects on plants from further review. After applying these exclusion criteria, we did not identify any key studies.

Critical toxicity studies

we did not identify any critical toxicity studies.

Key health effects

We did not find any key studies that suggest fomesafen can have carcinogenic, mutagenic, teratogenic, or interactive effects in human, animals, or cell culture. The EPA has classified fomesafen as not likely to be carcinogenic to humans.

Standard Selection

DHS recommends an enforcement standard of 25 µg/L for fomesafen.

There are no federal numbers and no state drinking water standard for fomesafen. Additionally, the EPA has not established a cancer slope factor for fomesafen because they determined that it is not likely to be carcinogenic to humans.

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information

However, the EPA does have an acceptable daily intake (oral reference dose) for fomesafen.¹ In our review, we did not find any significant technical information that was published since the EPA established their oral reference dose. Therefore, DHS calculated the recommended enforcement standard using the EPA’s oral reference dose, an average body weight of 10 kg, and a water consumption rate of 1 L/d and a relative source contribution factor of 100% as specified Chapter 160, Wis. Stats.

^d We used the National Institutes of Health’s PubMed resource and Clarivate Analytics’ Web of Science resource for this search. We used the following search terms in the literature review:

Title/abstract: Fomesafen
Subject area: toxicology
Language: English

We also searched online for toxicity studies published by national research programs.

DHS recommends a preventive action limit of 5 µg/L for fomesafen.

DHS recommends that the preventive action limit for fomesafen be set at 20% of the enforcement standard because fomesafen has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture.¹

Prepared by Sarah Yang, Ph.D.

Wisconsin Department of Health Services

References

1. USEPA. Fomesafen Sodium: Human Health Risk Assessment for the Section 3 Registration Action on Cantaloupe, Cucumber, Pea (Succulent), Pumpkin, Summer Squash, Winter Squash, Watermelon, Soybean (Succulent) and Lima Bean (Succulent). In: Prevention OoCSaP, ed. Washington, D.C.2013.
2. DATCP. Pesticide Database Searches. 2016; <https://www.kellysolutions.com/wi/pesticideindex.asp>.
3. Li X, Grey T, Price K, Vencill W, Webster T. Adsorption, desorption and persistence of fomesafen in soil. *Pest management science*. 2019;75(1):270-278.
4. Potter TL, Truman CC, Webster TM, Bosch DD, Strickland TC. Tillage, Cover-Crop Residue Management, and Irrigation Incorporation Impact on Fomesafen Runoff. *Journal of agricultural and food chemistry*. 2011;59(14):7910-7915.
5. Mills MS, Simmons ND. Assessing the ground-water contamination potential of agricultural chemicals: a flexible approach to mobility and degradation studies. *Pestic Sci*. 1998;54(4):418-434.
6. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 1402017*.
7. USEPA. National Primary Drinking Water Regulations. 2018; <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>.
8. USEPA. Drinking Water Contaminant Human Health Effects Information. 2019; <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>.
9. USEPA. IRIS Assessments. 2019; https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm.
10. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 8092018*.
11. IARC. List of Classification, Volumes 1-123. 2018; <https://monographs.iarc.fr/list-of-classifications-volumes/>. Accessed May 17, 2019.
12. JMPR. Inventory of evaluations performed by the Joint Meeting on Pesticide Residues (JMPR). 2012; <http://apps.who.int/pesticide-residues-jmpr-database>. Accessed May 24, 2019.

Hexazinone | 2020

Substance Overview

Hexazinone is a broad spectrum herbicide in the triazine family that is used to control a variety of plants.¹⁻³ Hexazinone works on post-emergent plants by inhibiting photosynthesis which eventually leads to plant death.¹⁻³

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard for hexazinone. DHS recommends an enforcement standard of 400 micrograms per liter (µg/L) for hexazinone. The recommended standard is based on the United States Environmental Protection Agency's (EPA's) lifetime health advisory that was established in 1996.⁴

DHS recommends that the preventive action limit for hexazinone be set at 10% of the enforcement standard because hexazinone has been shown to cause mutagenic effects in cell culture studies.^{1,2,4}

Current Standards

Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

Recommended Standards

Enforcement Standard:	400 µg/L
Preventive Action Limit:	40 µg/L

Health Effects

Studies in research animals have shown limited effects of hexazinone.^{1,2,4} The most consistent effects of hexazinone in these studies were a decrease in body weight gains, decrease in albumin levels, increase in alkaline phosphatase activity, and increase in liver weights. One chronic study in dogs found that hexazinone increased the presence of liver disease in both males and females.^{1,2,4} We did not find any epidemiology studies that examined the health effects of hexazinone.

Hexazinone has not been shown to be to cause carcinogenic, teratogenic, or interactive effects in people, research animals, or cell culture studies.^{1,2,4} However at this time, the Carcinogenicity Peer Review Committee determined there is not enough evidence to classify or not classify hexazinone as carcinogenic to humans. Hexazinone was found to have mutagenic effects in a cell culture study and was found to cause damage to DNA in hamster ovary cells.^{1,2,4a}

a Carcinogenic effects means the substance can cause cancer; mutagenic effects means the substance can cause DNA damage; teratogenic means the substance can cause birth defects; and interactive effects mean the substance can affect the toxicity of another substance or its toxicity can be affected by another substance.

Chemical Profile

Hexazinone	
Structure:	
CAS Number:	51235-04-2
Formula:	C ₁₂ H ₂₀ N ₄ O ₂
Molar Mass:	252.31 g/mol
Synonyms:	3-Cyclohexyl-6-(dimethylamino)-1-methyl-1,3,5-triazine-2,4(1H,3H)-dione 1,3,5-Triazine-2,4(1H,3H)-dione, 3-cyclohexyl-6-(dimethylamino)-1-methyl-Hexazinone

Exposure Routes

The Wisconsin Department of Agriculture, Trade, and Consumer Protection (DATCP) has approved the use of a number of commercial products containing hexazinone for use in Wisconsin.⁵

People can be exposed to hexazinone from food, soil, and water.^{1,2} Certain foods may have hexazinone in or on them from its use as a pesticide. The EPA regulates how much pesticide residues can be in foods. People can get exposed to hexazinone by touching recently treated plants or by touching or accidentally ingesting soil (dirt) near recently treated plants.

Hexazinone can leach through the soil and enter groundwater.^{1,2} Hexazinone is highly water soluble and has been shown to travel in surface water and groundwater.^{1,2}

Current Standard

Wisconsin does not currently have groundwater standards for hexazinone.⁶

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A	
Health Advisory:		
1 and 10-Day Child	2000 µg/L	
Longer-term Child	1000 µg/L	(1996)
Longer-term Adult	5000 µg/L	
Lifetime	400 µg/L	
Drinking Water Concentration (Cancer Risk):	N/A	

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A	
-----------------------------------	-----	--

Acceptable Daily Intake

EPA Oral Reference Dose:	0.05 mg/kg-d	(1994)
--------------------------	--------------	--------

Oncogenic Potential

EPA Cancer Slope Factor:	N/A	
--------------------------	-----	--

Guidance Values

ATSDR Oral Minimum Risk Level:	N/A	
--------------------------------	-----	--

Literature Search

Literature Search Dates:	1996 - 2019	
Total studies evaluated:	Approximately 50	
Key studies found?	No	
Critical studies identified?	No	

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for hexazinone.⁷

Health Advisory

In 1996, the EPA established several health advisories for hexazinone.⁴

1 and 10-Day Child

The EPA based the 1 and 10-Day Child Health Advisory on a 1980 study that evaluated the effects of hexazinone in a developmental study using rabbits. In this study, researchers exposed rabbits throughout days 6 to 19 of gestation to multiple doses of hexazinone (0, 20, 50, and 125 milligrams hexazinone per kilogram body weight per day or mg/kg-d) by gavage. The authors found that the highest dose decreased offspring body weight gain and delayed bone development of limbs.

The EPA selected a No Observable Adverse Effect Level (NOAEL) of 50 mg/kg-d hexazinone from this study. The EPA applied a total uncertainty factor of 300 to account for interspecies extrapolation (10), variability in human sensitivity (10), and to account for lack of short-term toxicity data in the most sensitive test species (3). They used a body weight of 10 kg, water consumption rate of 1 liter per day (L/d), and a relative source contribution of 100% to obtain a health advisory level of 2,000 micrograms per liter ($\mu\text{g/L}$).

Longer-term Child

The EPA based the Longer-term Health Advisory for Children on a 1991 study that evaluated the effects of hexazinone on rats. In this study, researchers exposed male and female rats in a two-generation reproduction study to multiple doses of hexazinone (0, 200, 2000, and 5000 parts per million (ppm) or 0, 14.3, 143, and 358 mg/kg-d) by diet. The authors found that the mid and highest doses decreased body weight and body weight gain.

The EPA selected a NOAEL of 14.3 mg/kg-d hexazinone from this study. They applied a total uncertainty factor of 100 to account for interspecies extrapolation (10) and variability in human sensitivity (10). For the Longer-term Child Health Advisory, they used a body weight of 10 kg, water consumption rate of 1 L/d, and a relative source contribution of 100% to obtain a health advisory level of 1,000 $\mu\text{g/L}$.

Longer-term Adult

The EPA based the Longer-term Health Advisory for Adults on a 1991 study that evaluated the effects of hexazinone on rats. In this study, researchers exposed male and female rats in a two-generation reproduction study to multiple doses of hexazinone (0, 200, 2000, and 5000 ppm or 0, 14.3, 143, 358 mg/kg-d) by diet. The authors found that the mid and highest doses decreased body weight and body weight gain.

The EPA selected a NOAEL of 14.3 mg/kg-d hexazinone from this study. They applied a total uncertainty factor of 100 to account for interspecies extrapolation (10) and variability in human sensitivity (10). For the Longer-term Adult Health Advisory, they used a body weight of 70 kg, water consumption rate of 2 L/d, and a relative source contribution of 100% to obtain a health advisory level of 5,000 $\mu\text{g/L}$.

Lifetime

The EPA based the Lifetime Health Advisory on their 1994 oral reference dose of 0.05 mg/kg-d for hexazinone. The EPA conducted a Human Health Risk Assessment as part of the registration of hexazinone. In their assessment, EPA reviewed a number of toxicity studies.^{1,2} The EPA selected a chronic exposure study in dogs to use as their principal study (MRID: 421623-01). In this study, male and female dogs were exposed to different doses of hexazinone (0, 200, 1500, 6000 ppm or 0, 5, 37.5, or 150 mg/kg-d) through diet for twelve months.

From the study, the EPA selected a NOAEL of 5 mg/kg-d because decreased body weight, decreased albumin levels, increased alkaline phosphatase activities, increased liver weights, and increased appearance of liver disease in histopathology were observed at higher doses. The EPA used a total uncertainty factor of 100 to account for differences between people and research animals (10) and differences among people (10). The EPA's chronic oral reference for hexazinone is 0.05 mg/kg-d.

For the Lifetime Health Advisory, the EPA used the chronic oral reference dose of 0.05 mg/kg-d, a body weight of 70 kg, water consumption rate of 2 L/d, and relative source contribution of 20% to obtain a health advisory level of 400 µg/L.

Drinking Water Concentration (Cancer Risk)

The EPA has not established a drinking water concentration based on a cancer risk level determination for hexazinone.⁸

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for hexazinone.⁹

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Information System (IRIS) program.

EPA Oral Reference Dose

In 1994, the EPA conducted a Human Health Risk Assessment as part of the registration of hexazinone. In their assessment, EPA reviewed a number of toxicity studies.^{1,2} The EPA selected a chronic exposure study in dogs to use as their principal study (see above section on the lifetime health advisory for more details on this study). From the study, the EPA selected a No Observable Adverse Effect Level (NOAEL) of 5 mg/kg-d. The EPA used a total uncertainty factor of 100 and determined the chronic oral reference for hexazinone is 0.05 mg/kg-d (for more details, see lifetime health advisory section above).

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If we determine that something is carcinogenic and there is no federal number or ADI from the EPA, DHS must set the standard at a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of hexazinone, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of hexazinone. If so, we look to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA has inadequate information to support or refute the carcinogenic potential of hexazinone to humans.^{1,2,4}

The International Agency for Research on Cancer (IARC) and the Joint FAO/WHO Meeting on Pesticide Residues (JMPR) has not evaluated the cancer potential of hexazinone.^{10,11}

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for hexazinone.^{1,2,12}

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information that was not considered when the value was established and indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For hexazinone, we searched for values that have been published since 1996 when EPA published their Drinking Water Health Advisory for hexazinone. We did not find any relevant guidance values.

Literature Search

Our literature review focused on the scientific literature published after EPA's Drinking Water Health Advisory for hexazinone in 1996. We looked for studies related to hexazinone toxicity or effects on a disease state in which information on exposure or dose was included as part of the study.^b Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

In our literature review, we did not find any relevant epidemiology studies that examined the effects of hexazinone on human health. Approximately 50 toxicity studies were returned by the search engines. We excluded studies on non-mammalian or cell systems, non-oral exposure routes, and those that did not evaluate health risks from further review. After applying these exclusion criteria, we did not identify any key studies.

Critical toxicity studies

We did not find any critical toxicity studies.

Key health effects

In our literature review, we did not find any studies that suggest hexazinone can have carcinogenic, teratogenic, or interactive effects in people, research animals, and/or cell culture studies.^{1,2,4} The EPA evaluated two animal studies for carcinogenicity. In one study, mice were fed hexazinone (200, 2500, 10000 mg/L) for two years. The authors found hexazinone caused a number of changes to the mouse liver including liver cell growth, growth of liver nodules, and liver cell death. The EPA states this hexazinone appears to be carcinogenic in this study; however, the authors concluded that hexazinone was not carcinogenic. In the other study, rats were fed hexazinone (200, 1000, and 2500 mg/L) for two years. The authors found hexazinone was not carcinogenic. The Carcinogenicity Peer Review Committee reviewed hexazinone and categorized hexazinone as a group D chemical, not classifiable as to human carcinogenicity. The CPRC concluded the animal evidence that hexazinone can be carcinogenic was equivocal, not enough evidence to support or refute hexazinone as being carcinogenic to humans.^{1,2,4}

^b We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

Title/abstract: Hexazinone

Subject area: N/A

Language: English

We also searched online for toxicity studies published by national research programs.

The EPA evaluated several cell culture studies for mutagenic effects of hexazinone and found one study that stated hexazinone was positive for mutagenicity.^{1,2,4} In that cell culture study, hamster ovary cells were exposed to hexazinone (1.58 to 19.82 millimoles (mM)). The authors found that hexazinone can cause damage to DNA.^{1,2,4} In four other cell culture studies, hexazinone was not found to be mutagenic.^{1,2,4}

Standard Selection

DHS recommends an enforcement standard of 400 µg/L for hexazinone.

DHS recommends using the EPA's Lifetime Health Advisory as the groundwater enforcement standard for hexazinone. This is the most recent federal number available. In our review, we did not find any significant technical information that was published since the EPA established their health advisories.

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information

DHS recommends a preventive action limit of 40 µg/L for hexazinone.

DHS recommends that the preventive action limit for hexazinone be set at 10% of the enforcement standard because studies have shown that hexazinone can cause mutagenic effects.^{1,2,4} Hexazinone was found to be positive for mutagenicity in one cell culture study.^{1,2,4} Hexazinone has not been shown to cause carcinogenic, teratogenic, or interactive effects.^{1,2,4}

Prepared by Gavin Dehnert, Ph.D. and Sarah Yang, Ph.D.

Wisconsin Department of Health Services

References

1. USEPA. R.E.D. Facts Pesticide Reregistration Hexazinone. 1994.
2. USEPA. Reregistration Eligibility Decision (RED) Hexazinone. 1994.
3. LeBaron HM, McFarland JE, Burnside OC. *The triazine herbicides*. Elsevier; 2008.
4. USEPA. Drinking Water Health Advisory Hexazinone. 1996.
5. DATCP. Pesticide Database Searches. 2016; <https://www.kellysolutions.com/wi/pesticideindex.asp>.
6. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 1402017*.

7. USEPA. National Primary Drinking Water Regulations. 2018; <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>.
8. USEPA. IRIS Assessments. 2019; https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm.
9. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 8092018*.
10. IARC. List of Classification, Volumes 1-123. 2018; <https://monographs.iarc.fr/list-of-classifications-volumes/>. Accessed May 17, 2019.
11. JMPR. Inventory of evaluations performed by the Joint Meeting on Pesticide Residues (JMPR). 2012; <http://apps.who.int/pesticide-residues-jmpr-database>. Accessed May 24, 2019.
12. USEPA. Hexazinone; CASRN 51235-04-2. In: System IRI, ed1987.

Saflufenacil | 2020

Substance Overview

Saflufenacil is an herbicide used to control broadleaf weeds.¹ It is applied to small grains, corn, chickpeas, cotton, edible beans, edible peas, lentils, lupine, sorghum, soybeans, and sunflowers before plants emerge from the soil, and to fruit and nut tree orchards and vineyards after plants emerge.¹ Certain foods may have saflufenacil residue in or on them from its use as a pesticide.^{1,2} Saflufenacil works by damaging plant cells and causing plant death.²

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard for saflufenacil. DHS recommends an enforcement standard of 460 micrograms per liter (µg/L) for saflufenacil. The recommended standard is based on the United States Environmental Protection Agency's (EPA's) chronic oral reference dose for saflufenacil.²

DHS recommends that the preventive action limit for saflufenacil be set at 10% of the enforcement standard because saflufenacil has been shown to cause teratogenic effects in research animals.³

Current Standards	
Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

Recommended Standards	
Enforcement Standard:	460 µg/L
Preventive Action Limit:	46 µg/L

Health Effects

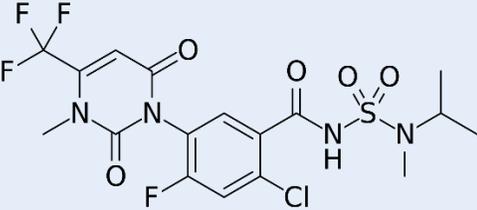
Studies in research animals have shown limited effects of saflufenacil.^{1,2} No studies of saflufenacil exposure among people were identified. Chronic and subchronic exposure to saflufenacil in research animals primarily affects the blood and development of blood cells but can also affect the liver and spleen.^{1,2} In reproductive studies, saflufenacil has shown to affect development, including decreased fetal and pre-weaning body weights, decreased viability, and skeletal changes in rodents.

Saflufenacil has not been shown to cause carcinogenic, mutagenic, or interactive effects in people, research animals, or cell culture studies.^{1,2,a} The EPA has classified saflufenacil as not likely to be

a Carcinogenic effects means the substance can cause cancer; mutagenic effects means the substance can cause DNA damage; teratogenic means the substance can cause birth defects; and interactive effects mean the substance can affect the toxicity of another substance or its toxicity can be affected by another substance.

carcinogenic to humans.^{1,2} In one reproductive study in rats, saflufenacil showed teratogenic effects (skeletal changes).³

Chemical Profile

Saflufenacil	
Structure:	
CAS Number:	372137-35-4
Formula:	C ₁₇ H ₁₇ ClF ₄ N ₄ O ₅ S
Molar Mass:	500.85 g/mol
Synonyms:	<i>N'</i> -[2-chloro-4-fluoro-5-(3-methyl-2,6-dioxo-4-(trifluoromethyl)3,6-dihydro-1(2H)-pyrimidinyl)benzyl]- <i>N</i> -isopropyl- <i>N</i> -methylsulfamide

Exposure Routes

The Wisconsin Department of Agriculture, Trade, and Consumer Protection (DATCP) has approved a number of commercial products containing saflufenacil for use in Wisconsin.⁴

People can be exposed to saflufenacil from food and drinking water.^{1,2} Certain foods may have saflufenacil residue in or on them from its use as a pesticide.^{1,2} The EPA regulates how much pesticide residues can be in foods.

People can become exposed to saflufenacil by touching recently treated plants or by touching or accidentally ingesting soil (dirt) near recently treated plants. Saflufenacil can reach surface and ground water sources of drinking water.^{1,2} Saflufenacil is soluble in water and has the potential to travel in groundwater.^{1,2}

Current Standard

Wisconsin does not currently have groundwater standards for saflufenacil.⁵

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

EPA Oral Reference Dose:	0.046 mg/kg-d	(2014)
--------------------------	---------------	--------

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

None available

Literature Search

Literature Search Dates:	2010 – 2019
Key studies found?	No
Critical studies identified?	No

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for saflufenacil.⁶

Health Advisory

The EPA has not established health advisories for saflufenacil.⁷

Drinking Water Concentration (Cancer Risk)

The EPA has not established drinking water concentration based cancer risk for saflufenacil.⁸

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for saflufenacil.⁹

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Assessment System (IRIS) program.

EPA Oral Reference Dose

In 2014, the EPA conducted a Human Health Risk Assessment as part of the registration of saflufenacil. In their assessment, EPA reviewed a number of toxicity studies.^{1,2} They selected a chronic carcinogenicity study in mice for setting their chronic reference dose for saflufenacil. In this study, male and female mice were exposed to different doses of saflufenacil (males: 0, 0.2, 0.9, 4.6, and 13.8 milligrams per kilogram body weight per day or mg/kg-d; females: 0, 1.2, 6.4, 18.9, and 38.1 mg/kg-d) through diet for 18 months.¹⁰ The authors noted decreased blood parameters such as red blood cells, hemoglobin, and hematocrit. These effects were seen at lower doses in males compared to females.

From this study, the EPA selected a No Observable Adverse Effect Level (NOAEL) of 4.6 mg/kg-d due to effects seen on several blood parameters at the highest dose in males. The EPA used a total uncertainty factor of 100 to account for differences between people and research animals (10) and differences among people (10). The EPA's chronic oral reference dose for saflufenacil is 0.046 mg/kg-d.

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If we determine that something is carcinogenic and there is no federal number or ADI from the EPA, DHS must set the standard at a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of saflufenacil, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of saflufenacil. If so, we look to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA has classified saflufenacil as not likely to be carcinogenic to humans.^{1,2}

The International Agency for Research on Cancer (IARC) has not evaluated the carcinogenicity of saflufenacil.¹¹

The Joint FAO/WHO Meeting on Pesticide Residues (JMPR) evaluated the carcinogenicity of saflufenacil in 2011 and found that it demonstrated no carcinogenic potential up to the highest dose levels tested.¹²

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for saflufenacil.^{1,2}

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information that was not considered when the value was established and indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For saflufenacil, we searched for values that have been published since 2010 when EPA published their human health risk assessment. We did not find any relevant guidance values.

Literature Search

Our literature review focused on the scientific literature published after EPA's human health risk assessment in 2010. We looked for studies related to saflufenacil toxicity or effects on a disease state in which information on exposure or dose was included as part of the study.^b Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

Approximately 20 studies were returned by the search engines. We excluded monitoring studies, studies evaluating risk from non-mammalian species, and studies on the effects on plants from further review. After applying these exclusion criteria, we did not identify any key studies.

Critical toxicity studies

^b We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

Title/abstract: saflufenacil

Language: English

We also searched online for toxicity studies published by national research programs.

We did not identify any critical toxicity studies.

Key health effects

We did not find any studies indicating that saflufenacil can cause carcinogenic, mutagenic or interactive effects in people, research animals, or cell culture. However, we found one study indicating that saflufenacil can cause teratogenic effects in research animals.³ The reproductive study in rats showed that Salfufenacil increased skeletal variations (NOAEL = 5 mg/kg/day LOAEL = 20 mg/kg/day).

Standard Selection

DHS recommends an enforcement standard of 460 µg/L for saflufenacil.

There are no federal numbers and no state drinking water standard for saflufenacil. Additionally, the EPA has not established a cancer slope factor for saflufenacil because they determined that it is not likely to be carcinogenic to humans.

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information

However, the EPA does have an acceptable daily intake (oral reference dose) for saflufenacil.² In our review, we did not find any significant technical information that was published since the EPA established their oral reference dose. Therefore, DHS calculated the recommended enforcement standard using the EPA's oral reference dose, an average body weight of 10 kg, and a water consumption rate of 1 L/d as specified Chapter 160, Wis. Stats.

DHS recommends a preventive action limit of 46 µg/L for saflufenacil.

DHS recommends that the preventive action limit for saflufenacil be set at 10% of the enforcement standard because saflufenacil has been shown to cause teratogenic effects in research animals. This is based on a reproductive study in rats which showed that saflufenacil caused abnormal skeletal development.³

Prepared by Amanda Koch, MPH, Gavin Dehnert, Ph.D., and Sarah Yang, Ph.D.

Wisconsin Department of Health Services

References

1. USEPA. *Pesticide Factsheet: Saflufenacil*. 2009.

2. USEPA. *Memorandum: Saflufenacil; Human-Health Risk Assessment in Support of Tolerances for Residues in/on Barley, Wheat, Grass, and Olives*. 2014. DP#D418587.
3. Schneider S. *BAS 800 H: Two-generation reproduction toxicity study in Wistar rats—Administration via the diet*. Ludwigshafen/Rhein, Germany, BASF AG2007. Unpublished report No. 2007/1043413.
4. DATCP. Pesticide Database Searches. <https://www.kellysolutions.com/wi/pesticideindex.asp>. Published 2016. Accessed.
5. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 140*2017.
6. USEPA. National Primary Drinking Water Regulations. <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>. Published 2018. Accessed.
7. USEPA. Drinking Water Contaminant Human Health Effects Information. <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>. Published 2019. Accessed.
8. USEPA. IRIS Assessments. https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm. Published 2019. Accessed.
9. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 809*2018.
10. Kamp H. *BAS 800 H—Carcinogenicity study in C57BL/6NCrl mice—Administration via the diet over 18 months*. Ludwigshafen/Rhein, Germany, BASF AG2007.
11. IARC. List of Classification, Volumes 1-123. <https://monographs.iarc.fr/list-of-classifications-volumes/>. Published 2018. Accessed May 17, 2019.
12. PV Shah MT. *SAFLUFENACIL*. 2011.

PFTriA | 2020

Substance Overview

Perfluorotridecanoic acid (PFTriA) is a chemical in a group of contaminants called per- and polyfluoroalkyl substances (PFAS). Because of its chemical properties, PFTriA can be found as an impurity in stain repellants in commercial products like carpet and fabric and as a coating for packaging, or as an ingredient in fire-fighting foam.^{1,2} PFAS can persist in the environment and in the human body for long periods of time.¹

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard for PFTriA. DHS cannot recommend an enforcement standard for PFTriA due to insufficient technical information currently available.

Health Effects

Studies investigating the health effects of PFTriA are very limited. A few epidemiologic studies among people found that higher levels of PFTriA were associated with changes in thyroid hormones and relevant antibodies, including a decrease in T₄ levels.³⁻⁵ An *in vitro* study found that PFTriA binds with two important human thyroid hormone proteins, suggesting the thyroid may be a target of PFTriA toxicity.⁶ Two epidemiologic studies among toddlers and infants found that prenatal exposure to PFTriA was associated with a decrease in the risk of allergic diseases, though one study found this negative association in boys only and the other in girls only.^{7,8} One toxicity study in research animals found that a mixture of PFAS substances, including PFTriA, resulted in liver effects, including changes in liver biochemistry parameters and higher absolute and relative liver weights.⁹ However, it is unclear how much PFTriA exposure contributed to those effects in this study. Only one epidemiologic study among people looked at the relationship between PFTriA and liver effects; this study found that PFTriA was associated with abnormal prealbumin and total bilirubin levels.¹⁰ Supporting evidence for the association between PFTriA and additional liver effects has not been observed in published literature. PFTriA has not been shown to cause carcinogenic (cancer), mutagenic (DNA damage), teratogenic (birth defects), or interactive effects

Current Standards

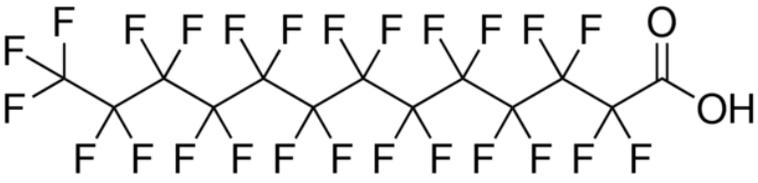
Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

Recommended Standards

Enforcement Standard:	N/A
Preventive Action Limit:	N/A

in people, research animals, or cell culture.^{1,11,a} The EPA has not evaluated the carcinogenicity of PFTriA.¹²

Chemical Profile

PFTriA	
Structure:	
CAS Number:	72629-94-8
Formula:	C ₁₃ HF ₂₅ O ₂
Molar Mass:	664.1 g/mol
Synonyms:	Perfluorotridecanoic acid Pentacosafuorotridecanoic acid 2,2,3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,11,11,12,12,13,13,13-pentacosafuorotridecanoic acid PFTriDA

Exposure Routes

PFAS, including PFTriA, can be released directly into the environment during the manufacture and use of PFAS and can be found in water or soil. PFAS can move between groundwater and surface water. Once in water, PFAS can travel long distances.^{1,2}

People can be exposed to PFAS by drinking water, eating food, and breathing in or accidentally swallowing soil or dust containing PFAS.¹

Current Standard

Wisconsin does not currently have a groundwater standard for PFTriA.¹³

a Carcinogenic effects means the substance can cause cancer; mutagenic effects means the substance can cause DNA damage; teratogenic means the substance can cause birth defects; and interactive effects mean the substance can affect the toxicity of another substance or its toxicity can be affected by another substance.

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

EPA Oral Reference Dose:	N/A
--------------------------	-----

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

None available

Literature Search

Literature Search Dates:	1900 - 2019
Key studies found?	Yes
Critical studies identified?	No

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for PFTriA.¹⁴

Health Advisory

The EPA has not established health advisories for PFTriA.¹⁵

Drinking Water Concentration (Cancer Risk)

The EPA has not established a drinking water concentration based on a cancer risk level determination for PFTriA.¹²

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for PFTriA.¹⁶

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Information System (IRIS) program.

EPA Oral Reference Dose

The EPA does not have an oral reference dose for PFTriA.¹²

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If DHS determines that something is carcinogenic and there is no federal number or ADI from the EPA, the standard must be set at a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of PFTriA, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of PFTriA. If so, we looked to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA and International Agency for Research on Cancer (IARC) have not evaluated the carcinogenicity of PFTriA.^{12,17}

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for PFTriA.¹²

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information that was not considered when the value was established and indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For PFTriA, we searched for values that have been published during or before December 2019. While the Agency for Toxic Substances and Disease Registry (ATSDR) reviewed the toxicity of 14 PFAS compounds in 2018, they did not review nor establish any guidance values for PFTriA.¹

Literature Search

Our literature review focused on relevant scientific literature on the health effects of PFTriA published during or before December 2019. We looked for studies related to PFTriA toxicity or effects on a disease state in which information on exposure or dose was included as part of the study.^b Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

Seven toxicity studies were returned by the search engines. We excluded studies on non-mammalian or cell systems, non-oral exposure routes, and those that did not evaluate health risks from further review. After applying these exclusion criteria, we located one key toxicity study on PFTriA (see Table A-1).⁹ To be considered a critical toxicity study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.^{c-d} The identified key study did not meet the criteria to be considered a critical toxicity study, as this study involved oral dosing of a mixture of PFAS substances instead of PFTriA alone (Table A-2).

In our search, we also located ten epidemiology studies (Table A-3). While multiple potential exposure sources and the ability for other PFAS compounds to cause similar health effects preclude using these data to establish a health-based value, such studies are helpful in identifying the crucial effects and ensuring that the animal data used to establish the standard is relevant to people. One *in vitro* study

b We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

Title/abstract: "PFTrDA" or "perfluorotridecanoic acid"

Subject area: toxicology

Language: English

We also searched online for toxicity studies published by national research programs.

c Appropriate toxicity values include the no observable adverse effect level (NOAEL), lowest observable adverse effect level (LOAEL), and benchmark dose (BMD).

d Due to the limited availability of data for this substance, we considered a study to be of an appropriate duration if animals were exposed for at least 28 days or if the exposure occurred during pregnancy. We accounted for differences in exposure duration through the use of an uncertainty factor.

examining the potential toxicity of PFTriA to the human thyroid was also identified and included as supportive information along with several epidemiologic studies evaluating the association between PFTriA exposure and thyroid hormone levels (Table A-4).

Critical toxicity studies

We did not identify any critical toxicity studies.

Key health effects

We did not find literature that suggest PFTriA has carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture.

Standard Selection

DHS does not recommend an enforcement standard for PFTriA.

The available health information on PFTriA is very limited. There are no federal numbers, nor a state drinking water standard, for PFTriA. Additionally, the EPA has not evaluated the carcinogenicity or established an ADI (oral reference dose) for PFTriA.

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information

Only a handful of epidemiologic studies have evaluated the risk of PFTriA. No critical studies evaluating the toxicity of PFTriA alone in research animals were located.

DHS does not recommend a preventive action limit for PFTriA.

Due to the lack of a critical toxicity study and supportive epidemiological studies, we have concluded that there is insufficient evidence to establish a preventive action limit for PFTriA.

Prepared by Amanda Koch, MPH and Sarah Yang, Ph.D.

Wisconsin Department of Health Services

References

1. ATSDR. Toxicological Profile for Perfluoroalkyls - Draft for Public Comment. In: Registry AftSaD, ed. Atlanta, GA2017.
2. ITRC. Environmental Fate and Transport for Per- and Polyfluoroalkyl Substances. In: Council ITR, ed2018.

3. Ji K, Kim S, Kho Y, et al. Serum concentrations of major perfluorinated compounds among the general population in Korea: dietary sources and potential impact on thyroid hormones. *Environ Int*. 2012;45:78-85.
4. Kim DH, Kim UJ, Kim HY, Choi SD, Oh JE. Perfluoroalkyl substances in serum from South Korean infants with congenital hypothyroidism and healthy infants--Its relationship with thyroid hormones. *Environ Res*. 2016;147:399-404.
5. Kim S, Choi K, Ji K, et al. Trans-placental transfer of thirteen perfluorinated compounds and relations with fetal thyroid hormones. *Environ Sci Technol*. 2011;45(17):7465-7472.
6. Ren XM, Qin WP, Cao LY, et al. Binding interactions of perfluoroalkyl substances with thyroid hormone transport proteins and potential toxicological implications. *Toxicology*. 2016;366-367:32-42.
7. Goudarzi H, Miyashita C, Okada E, et al. Effects of prenatal exposure to perfluoroalkyl acids on prevalence of allergic diseases among 4-year-old children. *Environ Int*. 2016;94:124-132.
8. Okada E, Sasaki S, Kashino I, et al. Prenatal exposure to perfluoroalkyl acids and allergic diseases in early childhood. *Environ Int*. 2014;65:127-134.
9. Mertens JJ, Sved DW, Marit GB, et al. Subchronic toxicity of S-111-S-WB in Sprague Dawley rats. *Int J Toxicol*. 2010;29(4):358-371.
10. Nian M, Li QQ, Bloom M, et al. Liver function biomarkers disorder is associated with exposure to perfluoroalkyl acids in adults: Isomers of C8 Health Project in China. *Environ Res*. 2019;172:81-88.
11. Buhrke T, Kibellus A, Lampen A. In vitro toxicological characterization of perfluorinated carboxylic acids with different carbon chain lengths. *Toxicol Lett*. 2013;218(2):97-104.
12. USEPA. IRIS Assessments. 2019; https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm.
13. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 140*2017.
14. USEPA. National Primary Drinking Water Regulations. 2018; <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>.
15. USEPA. Drinking Water Contaminant Human Health Effects Information. 2019; <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>.
16. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 809*2018.
17. IARC. List of Classification, Volumes 1-123. 2018; <https://monographs.iarc.fr/list-of-classifications-volumes/>. Accessed May 17, 2019.
18. Cao W, Liu X, Liu X, et al. Perfluoroalkyl substances in umbilical cord serum and gestational and postnatal growth in a Chinese birth cohort. *Environ Int*. 2018;116:197-205.

19. Kim JH, Park HY, Jeon JD, et al. The modifying effect of vitamin C on the association between perfluorinated compounds and insulin resistance in the Korean elderly: a double-blind, randomized, placebo-controlled crossover trial. *Eur J Nutr.* 2016;55(3):1011-1020.
20. Niu J, Liang H, Tian Y, et al. Prenatal plasma concentrations of Perfluoroalkyl and polyfluoroalkyl substances and neuropsychological development in children at four years of age. *Environ Health.* 2019;18(1):53.
21. Tian Y, Liang H, Miao M, et al. Maternal plasma concentrations of perfluoroalkyl and polyfluoroalkyl substances during pregnancy and anogenital distance in male infants. *Hum Reprod.* 2019;34(7):1356-1368.

Appendix A. Toxicity Data

Table A-I. PFTriA Toxicity Studies from Literature Review

Study Type	Species	Duration	Doses (mg/kg-d)	Route	Key Findings	Toxicity Value (mg/kg-d)	Reference
Sub-chronic	Rat	90 days	0, 0.025, 0.125, 0.6* <i>*Doses of S-111-S-WB, a mixture of perfluoro fatty acid ammonium salts of different carbon chain lengths (C₆-C₁₃), including PFTriA (C₁₃).</i>	Gavage	<p>Among males at 0.6 mg/kg-d: Lower body weight, higher mean blood clotting time, higher mean bilirubin and urea nitrogen concentrations, lower mean reticulocyte, total protein, and globulin concentrations</p> <p>Among males at 0.125 and 0.6 mg/kg-d: Higher albumin to globulin ratios and alkaline phosphatase concentrations, higher absolute and relative liver weights</p> <p>Among females at 0.6 mg/kg-d: Higher albumin to globulin ratios and alkaline phosphatase concentrations, relative liver weights</p> <p>In addition to these findings, liver hypertrophy was observed in 10 of 10 examined rats.</p>	<p>PFTriA NOAEL: N/A</p> <p>S-111-S-WB NOAEL: 0.025 mg/kg-d (males), 0.125 mg/kg-d (females)</p>	Mertens et al., 2010 ⁹

Table A-2. Critical Study Selection for PFTriA

Reference	Appropriate duration?	Effects consistent with other studies?	Effects relevant to humans?	Number of Doses	Toxicity value identifiable?	Critical study?
Mertens et al., 2010 ⁹	✓	✓	✓	3	⊘	No

To be considered a critical study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.

Table A-3. PFTriA Epidemiological Studies from Literature Review*

Study Type	Population	Time period	Exposure	Outcomes	Results	Other PFASs evaluated	Reference
Prospective cohort	Chinese infants	2013–2015	PFAS concentrations in umbilical cord blood	Gestational and postnatal growth	No associations between PFTriA and birth weight or length, or postnatal weight or length were identified.	PFOA, PFNA, PFDA, PFUnA, PFDoA, PFTeDA, PFHxDA, PFHxS, PFOS, PFDS	Cao et al., 2018 ¹⁸
Prospective cohort	4-year-old Japanese children	2003–2013	PFAS concentrations in maternal plasma	Prevalence of total allergic diseases (TAD)* <i>*TAD includes wheezing, eczema, and rhinoconjunctivitis symptoms</i>	A marginal negative association between PFTriA and TAD (highest vs. lowest quartile) among boys was identified: AOR=0.647, 95% CI: 0.416, 0.1.00; $p=0.017$ <i>PFTriA was not associated with TAD in girls.</i>	PFHxS, PFOS, PFHxA, PFHpA, PFOA, PFNA, PFDA, PFUnA, PFDoA, PFTeDA	Gourdazi et al., 2016 ⁷
Cross-sectional	Korean general population	2008	PFAS concentrations in serum	Thyroid hormone levels	Among females, a 1 ng/mL increase in PFTriA was associated with a 0.037 $\mu\text{g}/\text{dL}$ decrease in T_4 levels (95% CI: -0.071, -0.003) and a 0.016 $\mu\text{IU}/\text{mL}$ increase in TSH levels (95% CI: 0.001, 0.035). <i>No associations between PFTriA and thyroid hormone levels among males were identified.</i>	PFHxS, PFHpS, PFOS, PFDS, PFOA, PFNA, PFDA, PFUnA, PFTriA, PFTeDA, NMeFOSAA, EtPFOSAA	Ji et al., 2012 ³
Cross-sectional	Korean pregnant women	2008–2009	PFAS concentrations in maternal and fetal cord serum	Birth weight, fetal thyroid hormone levels	A 1 ng/mL increase in maternal PFTriA was associated with a 0.380 ng/dL decrease in fetal T_3 ($p<0.05$ in adjusted model) and a 0.441 $\mu\text{IU}/\text{mL}$ decrease in fetal T_4 ($p<0.05$ in adjusted model). A 1 ng/mL increase in fetal PFTriA was associated with a 0.391 $\mu\text{IU}/\text{mL}$ decrease in fetal T_4 ($p<0.05$ in unadjusted model only). <i>No associations between maternal or fetal PFTriA and birth weight were identified.</i>	PFHxS, PFHpS, PFOS, PFDS, PFOA, PFNA, PFDA, PFUnA, PFDoA, PFTeDA, MePFOSAA, EtPFOSAA	Kim et al., 2011 ⁵

Case-control	Korean infants with congenital hypothyroidism (CH) and those without	2009–2010	PFAS concentrations in infant serum	Thyroid hormone levels and levels of relevant antibodies	PFTriA was negatively correlated with antibodies related to metabolic disease among infants with CH compared to infants without CH ($r = -0.478$, $p < 0.05$).	PFBA, PFPeA, PFHxA, PFHpA, PFOA, PFNA, PFDA, PFUnA, PFDoA, PFTeDA, PFBS, PFHxS, PFHpS, PFOS, PFDS	Kim et al., 2016 ⁴
Prospective cohort	Korean elderly	2011–2012	PFAS concentrations in serum	Insulin resistance	<i>No association between PFTriA and insulin resistance identified.</i>	PFBS, PFHxS, PFOS, PFDS, PFBA, PFPeA, PFHxA, PFHpA, PFOA, PFNA, PFDA, PFUnA, PFDoA, PFTeDA	Kim et al., 2016 ¹⁹
Cross-sectional	Chinese adults	2015–2016	PFAS concentrations in serum	Liver function biomarker levels	PFTriA was associated with an abnormal level of prealbumin (AOR: 1.30, 95% CI: 1.12, 1.50) and total bilirubin (AOR: 1.19, 95% CI: 1.03, 1.36).	PFHxS, PFOS, PFDS, PFBA, PFPeA, PFHxA, PFOA, PFNA, PFDA, PFUnA, PFDoA, PFTeDA	Nian et al., 2019 ¹⁰
Prospective cohort	4-year-old Chinese children	2012	PFAS concentrations in maternal plasma	Neuro-psychological development	A natural log unit increase in prenatal PFTriA was associated with a marginally higher risk of developmental problems in communication among boys (highest vs. lowest tertile RR = 1.83, 95% CI: 1.08, 3.12; $p < 0.05$). <i>No association between prenatal PFTriA and risk of developmental problems in communication among girls identified.</i>	PFOS, PFOA, PFHxS, PFNA, PFDA, PFUnA, PFDoA	Niu et al., 2019 ²⁰
Prospective cohort	Japanese mothers and their infants	2003–2009	PFAS concentrations in maternal plasma	Infant allergic diseases (eczema, wheezing, and allergic rhinoconjunctivitis symptoms)	Prenatal exposure to PFTrDA was associated with a decrease in the risk of developing infant allergic diseases in early childhood in female infants (highest vs. lowest quartile AOR=0.51, 95% CI: 0.35, 0.75). Prenatal exposure to PFTrDA was associated with a decrease in the risk of developing eczema in early childhood in female infants (highest vs. lowest quartile AOR=0.39, 95% CI: 0.23, 0.64).	PFHxA, PFHpA, PFOA, PFNA, PFDA, PFUnA, PFTriA, PFTeDA, PFHxS, PFOS	Okada et al., 2014 ⁸

No associations between prenatal PFDoA exposure and the risk of TAD or eczema in male infants were identified.

Prospective cohort	Chinese pregnant women and their male infants	2012	PFAS concentrations in maternal plasma	Anogenital distance (AGD)	Per unit increase in maternal PFTriA, AGD decreased on average by 1.11 cm (95% CI: -2.17, -0.06) in male infants at 6 months of age (but not at birth nor at 12 months).	PFHxS, PFOS, PFOA, PFNA, PFDA, PFDS, PFUnA, PFDoA, PFTeDA, PFHxDA	Tian et al., 2019 ²¹
--------------------	---	------	--	---------------------------	--	---	---------------------------------

**This literature review is not exhaustive as the primary purpose of the search was to identify epidemiological studies that support toxicological findings.*

Epidemiologic terms: OR=odds ratio; AOR=adjusted odds ratio; RR=relative risk; 95% CI=95% confidence interval; r=Spearman correlation coefficient; β =regression coefficient
PFAS acronyms: PFOA=perfluorooctanoic acid, PFNA=perfluorononanoic acid, PFDA=perfluorodecanoic acid, PFUnA= perfluoroundecanoic acid, PFDoA=perfluorododecanoic acid, PFTeDA=perfluorotetradecanoic acid, PFHxDA=perfluorohexadecanoic acid, PFHxS=perfluorohexane sulfonate, PFOS=perfluorooctane sulfonic acid, PFDS=perfluorodecanesulfonate, PFBS=perfluorobutane sulfonate, PFBA=perfluorobutanoic acid, PFPeA=perfluoropentanoic acid, PFHxA=perfluorohexanoic acid, PFHpA=perfluoroheptanoic acid, 6:2 FTSA=6:2 fluorotelomer sulfonates, NMeFOSAA=N-methyl perfluorooctanesulfonamidoacetate, EPAH=2-(N-ethyl-perfluorooctane sulfonamido)acetate, MPAH=2-(N-methyl-perfluorooctane sulfonamido) acetate, PFSA=perfluorooctane sulfonamide; PFHpS=perfluoroheptane sulfonic acid; PFDS=perfluorodecane sulfonic acid; EtPFOSAA= 2-(Nethyl-perfluorooctane sulfonamido) acetic acid

Table A-4. PFTriA *In vitro* Toxicity Studies from Literature Review

Species	Cell Line	Duration (hr)	Doses (mM)	Other PFASs evaluated	Results	Reference
Human	Thyroid hormone binding protein (TTR and TBG) assay	N/A	20, 50	PFBA, PFHxA, PFHpA, PFOA, PFNA, PFDA, PFUdA, PFDoA, PFTeDA, PFBS, PFHxS, PFOS, FTOH-6:2, FTOH-8:2, FTOH-10:2	PFASs with a medium chain length and a sulfonate acid group are optimal for TTR binding, and PFASs with lengths longer than 12 carbons, like PFTriA, are optimal for TBG binding. While most PFASs, including PFTriA, bound TTR, only PFTriA and PFTdA acid bound TBG.	Ren et al., 2016 ⁶
<p><i>In vitro</i> terms: TTR=transthyretin, TBG=thyroxine-binding globulin</p> <p><i>PFAS acronyms:</i> PFOA=perfluorooctanoic acid, PFNA=perfluorononanoic acid, PFDA=perfluorodecanoic acid, PFA=perfluoroundecanoic acid, PFDoA=perfluorododecanoic acid, PFTeDA=perfluorotetradecanoic acid, PFHxDA=perfluorohexadecanoic acid, PFHxS=perfluorohexane sulfonate, PFOS=perfluorooctane sulfonic acid, PFDS=perfluorodecanesulfonate, PFBS=perfluorobutane sulfonate, PFBA=perfluorobutanoic acid, PFPeA=perfluoropentanoic acid, PFHxA=perfluorohexanoic acid, PFHpA=perfluoroheptanoic acid, 6:2 FTSA=6:2 fluorotelomer sulfonates, NMeFOSAA=N-methyl perfluorooctanesulfonamidoacetate, EPAH=2-(N-ethyl-perfluorooctane sulfonamido)acetate, MPAH=2-(N-methyl-perfluorooctane sulfonamido) acetate, PFSA=perfluorooctane sulfonamide; PFHpS=perfluoroheptane sulfonic acid; PFDS=perfluorodecane sulfonic acid; EtPFOSAA= 2-(Nethyl-perfluorooctane sulfonamido) acetic acid</p>						

PFTeA | 2020

Substance Overview

Perfluorotetradecanoic acid (PFTeA) is a chemical in a group of contaminants called per- and polyfluoroalkyl substances (PFAS). Because of its chemical properties, PFTeA can be found as an impurity in stain repellants in commercial products like carpet and fabric, as a coating for packaging, or as an ingredient in fire-fighting foam.^{1,2} PFAS, like PFTeA, can persist in the environment and in the human body for long periods of time.¹

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard for PFTeA. DHS recommends an enforcement standard of 10 micrograms per liter (µg/L) for PFTeA. The recommended standard is based on a study that found that PFTeA can significantly decrease body weight gain in pregnant rats and their offspring.³

DHS recommends that the preventive action limit for PFTeA be set at 20% of the enforcement standard because PFTeA has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture.

Health Effects

At this time, very few studies have evaluated the effects of PFTeA in people. The available studies have found that PFTeA may be associated with abnormal levels of pre-albumin (a protein made in the liver to help carry thyroid hormone and Vitamin A through your bloodstream), increased total cholesterol, and increased low-density lipoprotein (LDL) levels. Studies in research animals have shown that high levels of PFTeA can affect the blood, cause damage to the spleen, thymus, and liver, reduce body weight gain in offspring, and alter the weights of the pituitary gland, seminal vesicle, and liver.

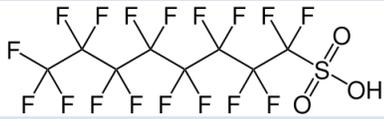
PFTeA has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture.^{1,4,a} The EPA has not evaluated the carcinogenicity of PFTeA.⁵

a Carcinogenic effects means the substance can cause cancer; mutagenic effects means the substance can cause DNA damage; teratogenic means the substance can cause birth defects; and interactive effects mean the substance can affect the toxicity of another substance or its toxicity can be affected by another substance.

Current Standards	
Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

Recommended Standards	
Enforcement Standard:	10 µg/L
Preventive Action Limit:	2 µg/L

Chemical Profile

PFTeA	
Structure:	
CAS Number:	376-06-7
Formula:	C ₁₄ HF ₂₇ O ₂
Molar Mass:	714.1 g/mol
Synonyms:	Perfluorotetradecanoic acid Heptacosafuorotetradecanoic acid Tetradecanoic acid, 2,2,3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,11,11,12, 12,13,13,14,14,14-heptacosafuoro- PFTeDA

Exposure Routes

PFAS, including PFTeA, can be released directly into the environment during the manufacture and use of PFAS and can be found in water or soil. PFAS can move between groundwater and surface water. Once in water, PFAS can travel long distances.^{1,2}

People can be exposed to PFAS by drinking water, eating food, and breathing in or accidentally swallowing soil or dust containing PFAS.¹

Current Standard

Wisconsin does not currently have a groundwater standard for PFTeA.⁶

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

EPA Oral Reference Dose:	N/A
--------------------------	-----

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

None available

Literature Search

Literature Search Dates:	1900 - 2020
Key studies found?	Yes
Critical studies identified?	Yes

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for PFTeA.⁷

Health Advisory

The EPA has not established health advisories for PFTeA.⁸

Drinking Water Concentration (Cancer Risk)

The EPA has not established a drinking water concentration based on cancer risk for PFTeA.⁵

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for PFTeA.⁹

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Information System (IRIS) program.

EPA Oral Reference Dose

The EPA does not have an oral reference dose for PFTeA.⁵

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If DHS determines that something is carcinogenic and there is no federal number or ADI from the EPA, the standard must be set at a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of PFTeA, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of PFTeA. If so, we looked to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA and International Agency for Research on Cancer (IARC) have not evaluated the carcinogenicity of PFTeA.^{5,10}

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for PFTeA.⁵

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information that was not considered when the value was established and indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For PFTeA, we searched for values that been published during or before April 2020. While the Agency for Toxic Substances and Disease Registry (ATSDR) reviewed the toxicity of 14 PFAS compounds in 2018, they did not review nor establish any guidance values for PFTeA.¹

Literature Search

Our literature review focused on relevant scientific literature on the health effects of PFTeA published during or before May 2020. We looked for studies related to PFTeA toxicity or effects on a disease state

in which information on exposure or dose was included as part of the study.^b Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

Approximately fifteen toxicity studies were returned by the search engines. We excluded studies on non-mammalian or cell systems, non-oral exposure routes, and those that did not evaluate health risks from further review. After applying these exclusion criteria, we located one key study on PFTeA (summarized in Table A-1).¹¹ To be considered a critical toxicity study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.^{c-d} The identified key study met the criteria to be considered a critical toxicity study (see Table A-2).

In our search, we located three epidemiology studies (See Table A-3 for a summary). While multiple potential exposure sources and the ability for other PFAS compounds to cause similar health effects preclude using these data to establish a health-based value, such studies are helpful in identifying the crucial effects and ensuring that the animal data used to establish the standard is relevant to people.

Critical Toxicity Studies

To compare between results in the critical studies, we calculated an acceptable daily intake (ADI) for each study/effect. The ADI is the estimated amount of PFTeA that a person can be exposed to every day and not experience health effects. As such, we calculated ADI by dividing a toxicity value from either a no-observed adverse effect level (NOAEL), lowest observed adverse effect level (LOAEL), or benchmark dose (BMD) identified in a study by a factor accounting for various sources of scientific uncertainty.^{e,12} Uncertainty factors were included, as appropriate, to account for differences between humans and animals, differences between healthy and sensitive human populations, using data from short-term experiments to protect against effects from long-term exposure, and using data where a health effect was observed to estimate the level that does not cause an effect. To ensure appropriate protection, we

^b We used the National Institutes of Health's PubMed resource, Clarivate Analytics' Web of Science resource, and Google Scholar for this search. We used the following search terms in the literature review:

Title/abstract: "PFTeDA" or "perfluorotetradecanoic acid" or "PFTeA"

Subject area: toxicology

Language: English

We also searched online for toxicity studies published by national research programs.

^c Appropriate toxicity values include the no observable adverse effect level (NOAEL), lowest observable adverse effect level (LOAEL), and benchmark dose (BMD).

^d Due to the limited availability of data for this substance, we considered a study to be of an appropriate duration if animals were exposed for at least 28 days or if the exposure occurred during pregnancy. We accounted for differences in exposure duration through the use of an uncertainty factor.

^e The NOAEL is the highest dose tested that did not cause an adverse effect, the LOAEL is the lowest dose tested that caused an adverse effect, and the BMD is an estimation of the dose that would cause a specific level of response (typically 5 or 10%).

have chosen to not use studies that have significant uncertainty as the basis for the recommended enforcement standards.^f This approach is consistent with that taken by EPA when establishing oral reference doses.¹³

Hirata-Koizumi et al., 2015

Hirata-Koizumi et al. exposed male and female rats to different concentrations of PFTeA (0, 1, 3, and 10 milligrams of PFTeA per kilogram body weight per day or mg/kg-d) by gavage for 42 days in males and from 14 days pre-mating through 5 days after lactation in females.¹¹ A subset of males and females in the 0 and 10 mg/kg-d dose groups were withheld treatment for an additional 14 day recovery period (see Hirata-Koizumi et al. for information on recovery group effects).

In males, PFTeA affected blood and biochemistry parameters and liver weights at the highest dose and the pituitary gland and seminal vesicle organ weight at the low, mid-, and highest dose (Table 1). PFTeA increases liver and thyroid cell damage in male rats treated at 3 and 10 mg/kg-d.

Table 1. Statistically Significant Effects Observed in Males in Hirata-Koizumi et al., 2015

Effects observed in males		Dose (mg/kg-d)		
		1	3	10
Strength	Decreased hindlimb grip strength		✓	✓
Blood	Decreased Activated Partial Thromboplastin Time			✓
	Decreased Total Protein			✓
	Decreased β-Globulin fraction of protein			✓
	Increased alkaline phosphatase			✓
	Increased blood urea nitrogen			✓
Liver	Increased absolute weight			✓
	Increased relative weight			✓
	Increased histopathology cell damage		✓	✓
Pituitary Gland	Decreased absolute weight		✓	✓
	Decreased relative weight		✓	✓
Seminal Vesicle	Decreased absolute weight	*	✓	✓
Thyroid	Increased histopathology cell damage		✓	✓

*The researchers indicated that there was decreased absolute weight of the seminal vesicle across all dose groups but indicated the NOAEL was 3 mg/kg-d. There was no additional information providing justification for why seminal vesicle weight was not considered toxicologically meaningful in determining the NOAEL. (see Hirata-Koizumi et al., 2015 for more details).¹¹

From this study, we identified a lowest observable adverse effect level (LOAEL) of 1 mg/kg-d based on the dose-dependent decrease in absolute weight of the seminal vesicle observed in male rats. We selected this value instead of the of No Observed Adverse Effect Level (NOAEL) identified by the authors because thyroid effects and decreased seminal vesicle weights are consistent with findings, such as

^f DHS considers a study to have significant uncertainty if the total uncertainty factors is equal to or greater than 3,000.

male reproductive effects and endocrine effects, in other PFAS toxicity studies in research animals.^{8,14} We determined a candidate ADI of 30 nanograms per kilogram per day (ng/kg-d) using this LOAEL and a total uncertainty factor of 30,000 to account for differences between humans and research animals (10), differences among people (10), using a short-term study to extrapolate to long-term exposures (3), using a LOAEL instead of a NOAEL (10), and the limited availability of information (10). While we obtained a candidate ADI for PFTeA from this study, this study was not used to establish the recommended enforcement standard due to significant uncertainty.

In females, PFTeA affected blood chloride levels, relative liver weights, and the β -globulin fraction of protein – an early indicator of potential immune activity-- at the highest dose (Table 2). In addition to these statistically significant findings, PFTeA also caused liver, spleen, and thymus cell damage in female rats treated at the highest dose.

Table 2. Statistically Significant Effects Observed in Females in Hirata-Koizumi et al., 2015

Effects observed in females		Dose (mg/kg-d)		
		1	3	10
Maternal	Lower body weights		✓	✓
Offspring	Lower body weights			✓
Blood	Increased chloride levels			✓
Liver	Increased relative weight			✓
	Decreased β -Globulin fraction of protein			✓

From this study, the authors identified a NOAEL of 1 mg/kg-d based on the dose-dependent decrease in maternal body weights during pregnancy. Decreased body weight gain in pregnant females and in offspring is consistent with findings in other PFAS toxicity studies in research animals.¹⁵ No significant changes in reproductive parameters and no abnormalities were found in neonates. We determined a candidate ADI of 1000 ng/kg-d based on the NOAEL (10) and a total uncertainty factor of 1,000 to account for differences between humans and research animals (10), differences among people (10), short term exposure in mating and pregnant rats (1), and the limited availability of information (10).

Key health effects

We did not find any studies that suggest PFTeA has caused carcinogenic, mutagenic, teratogenic, or interactive effects in people or research animals.

Discussion

g Hirata-Koizumi et al. identified a NOAEL of 1 mg/kg-d based on hepatocellular hypertrophy, alterations in liver biomarkers and weights, increased hypertrophy of thyroid cells, and significantly decreased hind limb grip strength at higher doses.

In research animals, PFTeA has been shown to impact the liver and spleen, decrease the weight of offspring at birth and during early development, and alter a variety of blood and biochemistry parameters, consistent with similar effects seen in other long-chain PFAS.^{1,16,17}

While studies on the effects of PFTeA among people are limited and results are mixed, data from available studies suggest that PFTeA may impact total cholesterol levels, LDL levels, and abnormal pre-albumin levels.¹⁸⁻²⁰

Other studies have demonstrated associations between other long-chain PFAS and birth weight, development, and male infertility in both humans and research animals.¹ The evidence in animals and limited evidence in humans suggests that PFTeA may have consistent effects to these other PFAS. Related developmental effects have been observed in research animals, including delays in hormone and motor development, though these effects have either not been seen or not been studied for PFTeA.¹

A number of studies have demonstrated that liver effects caused by PFAS occur primarily through activation of the nuclear hormone receptor, PPAR α (peroxisome proliferator-activated receptor alpha).²¹⁻²⁵ Nuclear receptors regulate gene expression and PPAR α regulates the expression of genes involved in lipid and cholesterol metabolism.²⁶ While PPAR α receptors are found in rodents and humans, levels of these receptors are much higher in rats and mice than in monkeys and humans.²⁶ This means that effects caused by activation of PPAR α are more likely to occur in rodents than they are to occur in people. As such, the effects on the liver observed in the critical study reviewed here are likely not relevant to humans.

Standard Selection

DHS recommends an enforcement standard of 10 μ g/L PFTeA.

There are no federal numbers and no state drinking water standard for PFTeA. Additionally, the EPA has not established a cancer slope factor or ADI (oral reference dose) for PFTeA. However, we found several critical studies evaluating the toxicity of PFTeA.

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information

However, we found a study that evaluated the toxicity of PFTeA in male and female rats. To calculate the ADI as specified by Ch. 160.13, Wisc, Statute, DHS used a NOAEL of 1 mg/kg-d based on adverse effects on body weight gain in pups.³ We selected this endpoint because low birth weight has been demonstrated to increase risk of newborn mortality and increase the risk for other diseases later in life, such as diabetes, cardiovascular disease, and asthma.²⁷ The findings of endocrine effects, developmental effects (body weight gain in offspring), and decreased body weight gain in pregnancy are consistent with

animal findings with possible human links in other PFAS compounds including PFOA and PFOS.^{11,15} To determine the enforcement standard, DHS used an ADI of 1000 ng/kg-day and a body weight of 10 kg, a water consumption rate of 1 L/d, and a relative source contribution of 100% as required by Ch. 160, Wis. Stats.

DHS recommends a preventive action limit of 2 µg/L PFTeA.

DHS recommends that the preventive action limit for PFTeA be set at 20% of the enforcement standard because PFTeA has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture.

Prepared by Brita Kilburg-Basnyat, Ph.D. and Sarah Yang, Ph.D.

Wisconsin Department of Health Services

References

1. ATSDR. Toxicological Profile for Perfluoroalkyls - Draft for Public Comment. In: Registry AfTSaD, ed. Atlanta, GA2017.
2. ITRC. Environmental Fate and Transport for Per- and Polyfluoroalkyl Substances. In: Council ITR, ed2018.
3. Hirata-Koizumi M, Fujii S, Furukawa M, Ono A, Hirose A. Repeated dose and reproductive/developmental toxicity of perfluorooctadecanoic acid in rats. *The Journal of toxicological sciences*. 2012;37(1):63-79.
4. Buhrke T, Kibellus A, Lampen A. In vitro toxicological characterization of perfluorinated carboxylic acids with different carbon chain lengths. *Toxicol Lett*. 2013;218(2):97-104.
5. USEPA. IRIS Assessments. https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm. Published 2019. Accessed.
6. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 140*2017.
7. USEPA. National Primary Drinking Water Regulations. <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>. Published 2018. Accessed.
8. USEPA. Drinking Water Contaminant Human Health Effects Information. <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>. Published 2019. Accessed.
9. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 809*2018.

10. IARC. List of Classification, Volumes 1-123. <https://monographs.iarc.fr/list-of-classifications-volumes/>. Published 2018. Accessed May 17, 2019.
11. Hirata-Koizumi M, Fujii S, Furukawa M, Ono A, Hirose A. Repeated dose and reproductive/developmental toxicity of perfluorooctadecanoic acid in rats. *J Toxicol Sci*. 2012;37(1):63-79.
12. Zhang Y, Beesoon S, Zhu L, Martin JW. Biomonitoring of perfluoroalkyl acids in human urine and estimates of biological half-life. *Environ Sci Technol*. 2013;47(18):10619-10627.
13. USEPA. A Review of the Reference Dose and Reference Concentration Processes. 2002(EPA/630/P-02/002F).
14. ITRC. Human and Ecological Health Effects of select PFAS. In: Council ITR, ed2020.
15. ITRC. PFAS Fact Sheets: PFAS Water and Soil Values Table Excel File. In: Council ITR, ed2020.
16. Takahashi M, Ishida S, Hirata-Koizumi M, Ono A, Hirose A. Repeated dose and reproductive/developmental toxicity of perfluoroundecanoic acid in rats. *J Toxicol Sci*. 2014;39(1):97-108.
17. (NTP) NTP. NTP technical report on the toxicity studies of perfluoroalkyl carboxylates (perfluorohexanoic acid, perfluorooctanoic acid, perfluorononanoic acid, and perfluorodecanoic acid) administered by gavage to Sprague Dawley (Hsd:Sprague Dawley SD) rats. In. Vol Toxicity Report 97. Research Triangle Park, NC: National Toxicology Program2019.
18. Nian M, Li Q-Q, Bloom M, et al. Liver function biomarkers disorder is associated with exposure to perfluoroalkyl acids in adults: Isomers of C8 Health Project in China. *Environmental research*. 2019;172:81-88.
19. Zeng X-W, Qian Z, Emo B, et al. Association of polyfluoroalkyl chemical exposure with serum lipids in children. *Science of the Total Environment*. 2015;512:364-370.
20. Cao W, Liu X, Liu X, et al. Perfluoroalkyl substances in umbilical cord serum and gestational and postnatal growth in a Chinese birth cohort. *Environment international*. 2018;116:197-205.
21. Das KP, Wood CR, Lin MT, et al. Perfluoroalkyl acids-induced liver steatosis: Effects on genes controlling lipid homeostasis. *Toxicology*. 2017;378:37-52.
22. Rosen MB, Das KP, Rooney J, Abbott B, Lau C, Corton JC. PPAR alpha-independent transcriptional targets of perfluoroalkyl acids revealed by transcript profiling. *Toxicology*. 2017;387:95-107.
23. Albrecht PP, Torsell NE, Krishnan P, et al. A species difference in the peroxisome proliferator-activated receptor alpha-dependent response to the developmental effects of perfluorooctanoic acid. *Toxicol Sci*. 2013;131(2):568-582.

24. Palkar PS, Anderson CR, Ferry CH, Gonzalez FJ, Peters JM. Effect of prenatal peroxisome proliferator-activated receptor alpha (PPARalpha) agonism on postnatal development. *Toxicology*. 2010;276(1):79-84.
25. Wolf DC, Moore T, Abbott BD, et al. Comparative hepatic effects of perfluorooctanoic acid and WY 14,643 in PPAR-alpha knockout and wild-type mice. *Toxicol Pathol*. 2008;36(4):632-639.
26. Hall AP, Elcombe CR, Foster JR, et al. Liver hypertrophy: a review of adaptive (adverse and non-adverse) changes--conclusions from the 3rd International ESTP Expert Workshop. *Toxicol Pathol*. 2012;40(7):971-994.
27. (WHO) WHO. *WHA Global Nutrition Targets 2025: Low Birth Weight Policy Brief*. 2014.

Appendix A. Toxicity Data

Table A-I. PFTeA Toxicity Studies from Literature Review

Study Type	Species	Duration	Doses (mg/kg-d)	Route	Endpoints	Toxicity Value (mg/kg-d)	Reference
Long-term	Sprague-Dawley Rats	42 days with 14 days of recovery	0, 1, 3, 10	Gavage	Hepatocyte hypertrophy and/or fatty changes in the liver and follicular cell hypertrophy in the thyroid at mid- and high doses.	NOAEL: N/A LOAEL: 1	Hirata-Koizumi et al., 2015 ¹¹
Developmental	Sprague-Dawley Rats	14 days pre-mating to day 5 of lactation (41-46 days)	0, 1, 3, 10	Gavage	Inhibited postnatal body weight gain pups	NOAEL: 1 LOAEL: 3	Hirata-Koizumi et al., 2015 ¹¹

Table A-2. Critical Study Selection for PFTeA

Reference	Appropriate duration?	Effects consistent with other studies?	Effects relevant to humans?	Number of Doses	Toxicity value identifiable?	Critical study?
Hirata-Koizumi et al., 2015 (males)	✓	✓	✓	3	✓	Yes
Hirata-Koizumi et al., 2015 (females)	✓	✓	✓	3	✓	Yes

To be considered a critical study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.

Table A-3. PFTeA Epidemiological Studies from Literature Review*

Study Type	Population	Time period	Exposure	Outcomes	Results	Other PFASs evaluated	Reference
Prospective cohort	Chinese infants	2013–2015	PFAS concentrations in umbilical cord blood	Gestational and postnatal growth	No associations between PFTeA and birth weight or length, or postnatal weight or length were identified.	PFOA, PFNA, PFDA, PFUnA, PFDoA, PFTriA, PFHxDA, PFHxS, PFOS, PFDS	Cao et al., 2018 ²⁰
Cross-sectional	Taiwanese children	2009–2010	PFAS concentrations in serum	TC, HDL, LDL, TG	In the quartile analysis, PFTeA was associated with elevations in TC (p=0.002) and LDL (p=0.004) with arithmetical means of 30.7 ng/ml in boys and 27.4 ng/ml in girls.	PFOS, PFOA, PFBS, PFNA	Zeng et al., 2015 ¹⁹
Cross-sectional	Chinese adults	2015–2016	PFAS concentrations in serum	Liver function biomarker levels	PFTeA was associated with an abnormal level of prealbumin (AOR: 1.28, 95% CI: 1.05, 1.56).	PFHxS, PFOS, PFDS, PFBA, PFPeA, PFHxA, PFOA, PFNA, PFDA, PFUnA, PFDoA, PFTrDA	Nian et al., 2019 ¹⁸

**This literature review is not exhaustive as the primary purpose of the search was to identify epidemiological studies that support toxicological findings.*

Epidemiologic terms: OR=odds ratio; AOR=adjusted odds ratio; RR=relative risk; 95% CI=95% confidence interval; r=Spearman correlation coefficient; β=regression coefficient
PFAS acronyms: PFOA=perfluorooctanoic acid, PFNA=perfluorononanoic acid, PFDA=perfluorodecanoic acid, PFUnA= perfluoroundecanoic acid, PFDoA=perfluorododecanoic acid, PFTriA=perfluorotetradecanoic acid, PFHxDA=perfluorohexadecanoic acid, PFHxS=perfluorohexane sulfonate, PFOS=perfluorooctane sulfonic acid, PFDS=perfluorodecanesulfonate, PFBS=perfluorobutane sulfonate, PFBA=perfluorobutanoic acid, PFPeA=perfluoropentanoic acid, PFHxA=perfluorohexanoic acid, PFHpA=perfluoroheptanoic acid, 6:2 FTSA=6:2 fluorotelomer sulfonates, NMeFOSAA=N-methyl perfluorooctanesulfonamidoacetate, EPAH=2-(N-ethyl-perfluorooctane sulfonamido)acetate, MPAH=2-(N-methyl-perfluorooctane sulfonamido) acetate, PFSA=perfluorooctane sulfonamide; PFHpS=perfluoroheptane sulfonic acid; PFDS=perfluorodecane sulfonic acid; EtPFOSAA= 2-(Nethyl-perfluorooctane sulfonamido) acetic acid

PFBA | 2020

Substance Overview

Perfluorobutanoic acid^a (PFBA) is a chemical in a group of contaminants called per- and polyfluoroalkyl substances (PFAS). Because of its unique chemical properties, PFBA is used in textile manufacturing and commercial and industrial products such as paper wrapping, food packaging, and fire-fighting foams.¹ PFBA can also be formed when other PFAS are broken down in the environment or the body.²⁻⁶ Because PFBA has a molecular structure and chemical properties highly similar to those of perfluorooctanoic acid (PFOA), it has been used as an alternative to PFOA after restrictions were put in place for the manufacturing and use of PFOA.

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard for PFBA. DHS recommends an enforcement standard of 20 micrograms per liter (µg/L) for PFBA. The recommended standard is based on two studies that found that PFBA affected the thyroid, blood, cholesterol levels, and liver in male rats.^{7,8}

DHS recommends that the preventive action limit for PFBA be set at 20% of the enforcement standard because PFBA has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell cultures.

Current Standards

Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

Recommended Standards

Enforcement Standard:	10 µg/L
Preventive Action Limit:	2 µg/L

Health Effects

Most PFAS can persist in the environment and in the human body for long periods of time.⁹ PFBA has been shown to leave the body much faster than other PFAS, such as PFOA, which can stay in the body for years. The half-life of PFBA is estimated to be 72–87 hours.¹⁰

While information about the health effects of PFBA exposure is limited, studies in research animals indicate that PFBA can affect health. These studies have found that high levels of PFBA can affect the liver, thyroid, and several blood parameters in rats and mice, with health effects seen more often in

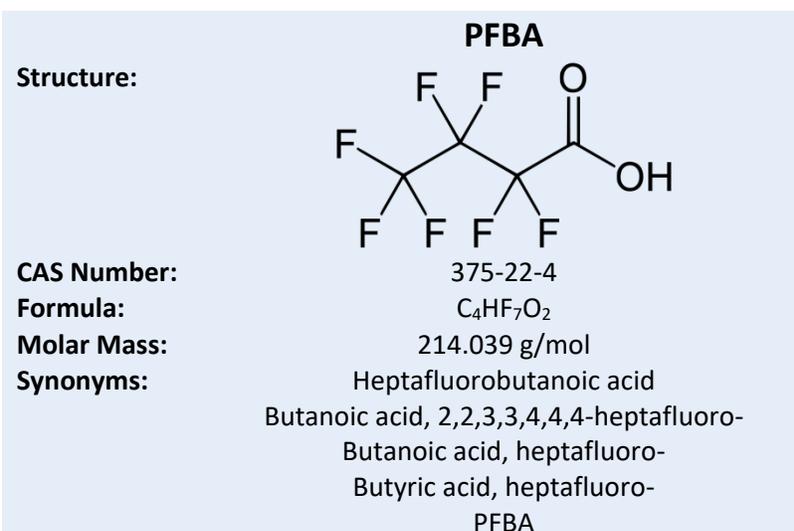
^a This scientific support document and the included groundwater standard recommendations also apply to anion salts of perfluorobutanoic acid.

males than in females.^{7,8,11-15} One study found that high levels of PFBA can cause developmental delays in mice.¹⁶

A few studies have looked at the relationship between PFBA exposure and health effects in humans (summarized in Table A-3). Of these, one study showed an association between higher levels of PFBA and an increased risk for hypertension in men and women.¹⁷ Another study found a correlation between PFBA and lower hepatitis B virus-related antibody levels, suggesting that PFBA may decrease immunity to disease.¹⁸ Additional studies did not find associations between PFBA and other health-related endpoints, including serum lipid concentrations, congenital hypothyroidism, and insulin resistance.¹⁹⁻²¹

PFBA has not been shown to cause carcinogenic (cancer), mutagenic (DNA damage), teratogenic (birth defects), or interactive effects in people, research animals, or cell cultures.⁹ The EPA has not evaluated the carcinogenicity of PFBA.

Chemical Profile



Exposure Routes

PFAS, including PFBA, can be released directly into the environment during the manufacture and use of PFAS and can be found in water or soil. PFAS can move between groundwater and surface water. Once in water, PFAS can travel long distances.^{9,22}

People can be exposed to PFAS, like PFBA, by drinking water, eating food, and breathing in or accidentally swallowing soil or dust containing PFAS.⁹

Current Standard

Wisconsin does not currently have groundwater standards for PFBA.²³

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

EPA Oral Reference Dose:	N/A
--------------------------	-----

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

ATSDR Oral Minimum Risk Level:	N/A
--------------------------------	-----

Literature Search

Literature Search Dates:	1900–2019
Key studies found?	Yes
Critical studies identified?	Yes

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for PFBA.²⁴

Health Advisory

The EPA has not established health advisories for PFBA.²⁵

Drinking Water Concentration (Cancer Risk)

The EPA has not established a drinking water concentration based on a cancer risk level determination for PFBA.²⁶

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for PFBA.²⁷

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides or for use by the Integrated Risk Information System (IRIS) program.

EPA Oral Reference Dose

The EPA does not have an oral reference dose for PFBA.²⁶

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If we determine that something is carcinogenic and there is no federal number or ADI from the EPA, DHS must set the standard at a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of PFBA, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of PFBA. If so, we look to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA and International Agency for Research on Cancer (IARC) have not evaluated the carcinogenicity of PFBA.

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for PFBA.²⁶

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information that was not considered when the value was established and that indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For PFBA, we searched for values that been published on or before January 2020. While the Agency for Toxic Substances and Disease Registry (ATSDR) reviewed the toxicity of PFBA in 2018, they did not establish any guidance values for PFBA.^{b,9}

Literature Search

Our literature review focused on relevant scientific literature on the health effects of PFBA published on or before January 2020. We looked for studies related to PFBA toxicity or effects on a disease state in which information on exposure or dose was included as part of the study.^c Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

Approximately 475 total studies on PFBA were returned by the search engines. We excluded studies on non-mammalian or cell systems, non-oral exposure routes, and those that did not evaluate health risks from further review. After applying these exclusion criteria, we located nine key toxicity studies on PFBA (summarized in Table A-1). To be considered a critical toxicity study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with

b ATSDR did not identify any chronic-duration oral studies for PFBA in their literature review. While they located several intermediate-duration studies, they did not establish a minimum risk level (MRL) for PFBA.²² Although the available studies had examined potentially sensitive endpoints and developmental toxicity, the database was missing a reliable estimate of elimination half-life in humans; ATSDR did not consider the available half-life information adequate for derivation of an MRL due to lack of data from females.

c We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

Title/abstract: "perfluorobutanoic acid" or "perfluorobutanoate" or "perfluorobutyric acid" or "heptafluorobutyric acid"

Subject area: N/A

Language: English

We also searched online for toxicity studies published by national research programs.

other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.^{d-e} Five of these studies met the criteria to be considered a critical toxicity study (see Table A-2).

In our search, we also located several epidemiology studies (summarized in Table A-3). While multiple potential exposure sources and the ability for other PFAS compounds to cause similar health effects preclude using these data to establish a health-based value, such studies are helpful in identifying the critical effects and ensuring that the animal data used to establish the standard are relevant to people.

Critical Toxicity Studies

To compare between results from recently found studies and the study used to set the current enforcement standard, we calculated an acceptable daily intake (ADI) for each study/effect. The ADI is the estimated amount of PFBS that a person can be exposed to every day and not experience health impacts. As such, we calculated ADI by dividing a toxicity value from either a no-observed adverse effect level (NOAEL), lowest observed adverse effect level (LOAEL), or benchmark dose lower confidence level (BMDL) identified in a study by a factor accounting for various sources of scientific uncertainty.^f We included uncertainty factors to account for differences between humans and animals, differences between healthy and sensitive human populations, using data from short-term experiments to protect against effects from long-term exposure, and using data where a health effect was observed to estimate the level that does not cause an effect, as appropriate. To ensure appropriate protection, we have chosen to not use studies that have significant scientific uncertainty as the basis for the recommended enforcement standards.^g This approach is consistent with that taken by EPA when establishing oral reference doses.²⁸

Butenhoff et al., 2012a

Butenhoff et al. exposed male and female rats to different concentrations of PFBA (0, 6, 30, and 150 milligrams of PFBA per kilogram body weight per day or mg/kg-d) through gavage for 28 days.⁷ The

d Appropriate toxicity values include the no observable adverse effect level (NOAEL), lowest observable adverse effect level (LOAEL), and benchmark dose (BMD).

e Due to the limited availability of data for this substance, we considered a study to be of an appropriate duration if animals were exposed for at least 28 days or if the exposure occurred during pregnancy. We accounted for differences in exposure duration through the use of an uncertainty factor.

f The NOAEL is the highest dose tested that did not cause an adverse effect, the LOAEL is the lowest dose tested that caused an adverse effect, and the BMDL is the lower limit of a one-sided 95% confidence interval established using benchmark dose modeling. Benchmark dose modeling is considered the state of the science for establishing health-based values like an acceptable daily intake. Benchmark dose modeling takes account of all of the data for a particular effect from a particular experiment, allows for increased consistency, and can better account for statistical uncertainties.

g DHS considers a study to have significant uncertainty if the total uncertainty factors is greater than 3,000.

authors found that PFBA did not affect females in a dose-response manner, but increased absolute liver weight and lowered serum total cholesterol and thyroxine in males (Table 1). In addition to these statistically significant findings, the study observed minimal liver cell damage at the highest dose and minimal to slight enlargement of the thyroid at all doses and with increasing incidence in a dose-dependent manner.

Table 1. Statistically Significant Effects Observed in Male and Female Rats in Butenhoff et al., 2012a

Effects observed in males		Dose (mg/kg-d)		
		6	30	150
Liver	Higher absolute liver weight		✓	✓
	Lower serum total cholesterol		✓	✓
Thyroid	Higher absolute thyroid weight	*	*	
	Lower serum total thyroxine	✓	✓	✓
	Lower serum free thyroxine	✓	✓	✓
Clinical chemistry	Higher serum potassium			*
	Higher inorganic phosphate			*
Effects observed in females				
Thymus	Higher absolute thymus weight	*		

*The authors did not consider this finding to be toxicologically meaningful (see Butenhoff et al., 2012a for more details).⁷

From this study, we identified a lowest observable adverse effect level (LOAEL) of 6 mg/kg-d based on effects on thyroid hormones (lower serum thyroxine) in male rats. We selected this value instead of the higher NOAEL identified by the authors because thyroid effects are consistent with findings in other PFAS toxicity studies in research animals.^{h,9} We estimated a candidate ADI of 0.002 mg/kg-d from this study based on the LOAEL and the total uncertainty factor of 100,000 to account for differences between people and research animals (10), differences among people (10), use of a shorter duration study to protect against effects from long-term exposure (10), the use of a LOAEL instead of a No Observed Adverse Effect Level (NOAEL) (10), and the limited availability of information (10). While we obtained a candidate ADI for PFBA from this study, this study was not used to establish the recommended enforcement standard due to significant uncertainty.

Butenhoff et al., 2012b

Butenhoff et al. exposed male and female rats to different concentrations of PFBA (0, 1.2, 6, and 30 mg/kg-d) through gavage for 90 days.⁷ The authors found that PFBA increased absolute liver weight and alkaline phosphatase levels and lowered serum total thyroxine and other blood parameters in males at 30 mg/kg-d (Table 2). In addition to these statistically significant findings, the study observed slight to

^h Butenhoff et al. identified a NOAEL of 6 mg/kg-d because thyroid effects resolved after cessation of exposure.

minimal liver cell damage at the highest dose and slight to minimal enlargement of the thyroid at all doses and with increasing incidence in a dose-dependent manner.

Table 2. Statistically Significant Effects Observed in Male and Female Rats in Butenhoff et al., 2012b

Effects observed in males		Dose (mg/kg-d)		
		1.2	6	30
Blood	Lower red blood cell concentration			✓
	Lower hemoglobin concentration			✓
	Lower hematocrit			✓
	Higher red cell distribution width			✓
Clinical chemistry	Lower serum calcium			*
Liver	Higher alkaline phosphatase			*
	Lower serum total protein			*
	Lower serum total bilirubin		*	*
	Higher absolute liver weight			✓
Thyroid	Lower serum total thyroxine			✓
Effects observed in females				
Liver	Lower serum total bilirubin			*

*The authors did not consider this finding to be toxicologically meaningful (see Butenhoff et al., 2012b for more details).

From this study, the authors identified a NOAEL of 6 mg/kg-d based on effects on the liver, thyroid, calcium levels, and blood in males at the highest dose. We estimated a candidate ADI of 0.002 mg/kg-d from this study based on the NOAEL and a total uncertainty factor of 3,000 to account for differences between people and research animals (10), differences among people (10), the use of a long-term study duration (3), and the limited availability of information (10).

In 2018, the Minnesota Department of Health established a benchmark dose lower confidence level (BMDL) of 3 mg/kg-d based on this study and van Otterdijk, 2007b described below.^{7,8,46} We estimated a candidate ADI of 0.001 mg/kg-d PFBS from this study based on the BMDL established by the Minnesota Department of Health and a total uncertainty factor of 3,000 to account for differences between humans and research animals (10), differences among people (10), use of a long-term study duration (3), and the limited availability of information (10).

Das et al., 2008

Das et al. exposed pregnant mice to different concentrations of PFBA (0, 35, 175, and 350 mg/kg-d) through gavage during gestation day (GD) 1–17 and examined effects in them and in their pups through postnatal day (PD) 21 and 291.¹⁶ The authors found that PFBA increased absolute and relative liver weights in mothers at 175 and 350 mg/kg-d and caused full-litter losses at the highest dose (Table 3). In

offspring, PFBA affected growth by delaying eye opening at all three doses and delaying the onset of puberty at 175 and 350 mg/kg-d.

Table 3. Statistically Significant Effects Observed in Pregnant Female Mice in Das et al., 2008

Effects observed in mothers		Dose (mg/kg-d)		
		35	175	350
Liver	Higher absolute liver weight at term		✓	✓
	Higher relative liver weight at term		✓	✓
	Higher absolute liver weight at postweaning age		✓	✓
	Higher relative liver weight at postweaning age		✓	✓
Reproduction	Increase in full-litter loss*			✓
Effects observed in offspring				
Growth	Delayed eye opening	✓	✓	✓
	Delayed vaginal opening		✓	✓
	Delayed preputial separation			✓

*Mice that were positive for implantations but did not have any fetuses at term were categorized as having experienced full-litter losses.

From this study, we identified a LOAEL of 35 mg/kg-d based on delayed eye opening in offspring at all doses. We estimated a candidate ADI of 0.012 mg/kg-d based on the LOAEL and a total uncertainty factor of 10,000 to account for differences between humans and research animals (10), differences among people (10), the use of a LOAEL instead of a NOAEL (10), and the limited availability of information (10). While we obtained a candidate ADI for PFBA from this study, this study was not used to establish the recommended enforcement standard due to significant uncertainty.

Van Otterdijk, 2007a

Van Otterdijk exposed male and female rats to different concentrations of PFBA (0, 6, 30, and 150 mg/kg-d) through gavage for 28 days.¹⁴ In the study, the authors found that PFBA increased absolute and relative liver weights at 150 mg/kg-d and affected several clinical biochemistry parameters in males, including increased potassium and organic phosphate levels at 150 mg/kg-d and reduced cholesterol at 30 and 150 mg/kg-d (Table 4). In addition to these statistically significant findings, the authors saw other effects in males, including minimal enlargement of the liver and delayed bilateral pupillary reflex at 150 mg/kg-d and minimal to slight enlargement of the thyroid gland at 30 and 150 mg/kg-d.

Table 4. Statistically Significant Effects Observed in Rats in van Otterdijk, 2007a

Effects observed in males		Dose (mg/kg-d)		
		6	30	150
Clinical biochemistry	Increased potassium levels			✓
	Increased organic phosphate levels			✓

Liver	Increased absolute and relative liver weights		✓	✓
	Reduced cholesterol levels		✓	✓
Thyroid	Increased absolute and relative thyroid weights	*	*	
Effects observed in females				
Thymus	Higher absolute thymus weight	*		

*The authors did not consider this finding to be toxicologically meaningful (see van Otterdijk, 2007a for more details).¹⁴

From this study, the authors identified a NOAEL of 6 mg/kg-d based on minimal to slight enlargement of the thyroid gland, increased liver weights and lower cholesterol levels in males at 30 and 150 mg/kg-d PFBA. We estimated a candidate ADI of 0.002 mg/kg-d based on the NOAEL and a total uncertainty factor of 10,000 to account for differences between humans and research animals (10), differences among people (10), use of a shorter duration study to protect against effects from long-term exposure (10), and the limited availability of information (10). While we obtained a candidate ADI for PFBA from this study, this study was not used to establish the recommended enforcement standard due to significant uncertainty.

Van Otterdijk, 2007b

Van Otterdijk exposed male and female rats to different concentrations of PFBA (0, 1.2, 6, and 30 mg/kg-d) through oral gavage for 90 days.⁸ In the study, the authors found that PFBA affected the liver and blood in males at 30 mg/kg-d (Table 5). In addition to these statistically significant findings, the authors observed delayed pupillary reflex in males and females and minimal to slight enlargement of the liver and thyroid in males at 30 mg/kg-d PFBA.

Table 5. Statistically Significant Effects Observed in Rats in van Otterdijk, 2007b

Effects observed in males		Dose (mg/kg-d)		
		1.2	6	30
Blood	Reduced red blood cell count			✓
	Increased red blood cell distribution width			✓
	Reduced hemoglobin level			✓
	Reduced hematocrit level			✓
	Increased white blood cell count	*		
Liver	Increased absolute and relative liver weights			✓
	Increased alkaline phosphatase activity levels			✓
	Reduced total protein levels			✓
	Reduced total bilirubin levels		*	*
	Reduced alanine aminotransferase activity levels	*		
	Reduced aspartame aminotransferase activity levels	*		
Clinical biochemistry	Reduced calcium levels			✓
	Increased sodium levels	*		

	Increased inorganic phosphate levels	*	
Thymus	Increased absolute and relative thymus weights		*
Effects observed in females			
Blood	Reduced mean corpuscular hemoglobin		*
	Reduced mean corpuscular hemoglobin concentration		*
Liver	Reduced total bilirubin levels		*
Clinical biochemistry	Increased sodium levels	*	

*The authors did not consider this finding to be toxicologically meaningful (see van Otterdijk, 2007b for more details).⁸

From this study, the authors identified a NOAEL of 6 mg/kg-d based on minimal to slight enlargement of the thyroid gland, increased liver weights and lower cholesterol levels in males at 30 and 150 mg/kg-d PFBA. We estimated a candidate ADI of 0.002 mg/kg-d based on the NOAEL and a total uncertainty factor of 3,000 to account for differences between humans and research animals (10), differences among people (10), use of a long-term study duration (3), and the limited availability of information (10).

In 2018, the Minnesota Department of Health established a benchmark dose lower confidence level (BMDL) of 3 mg/kg-d based on this study and Butenhoff et al., 2012b described above.^{7,8,46} We estimated a candidate ADI of 0.001 mg/kg-d PFBS from this study based on the BMDL established by the Minnesota Department of Health and a total uncertainty factor of 3,000 to account for differences between humans and research animals (10), differences among people (10), use of a long-term study duration (3), and the limited availability of information (10).

Key health effects

We did not find literature that suggest PFBA has carcinogenic, mutagenic, teratogenic or interactive effects in people, research animals, or cell culture.

Discussion

Studies in research animals show that PFBA can affect the thyroid, blood, cholesterol levels, and liver in male rats, a finding consistent with other PFAS studies in people and research animals that have also shown associations with these effects.^{7-9,14,16} In addition, several new studies among people exposed to high levels of PFAS suggest that PFBA exposure is associated with risk for hypertension and lower immunity to disease.^{17,18}

Studies in research animals and people have shown that PFBA, as well as other PFAS, can affect the levels of thyroid hormones.⁹ Thyroid hormones are crucial for development, energy balance, and metabolism in all species.²⁹ In people, thyroid hormones play an important role in the development of the brain, lungs, and heart.²⁹ Scientists have learned that certain PFAS, including PFBA, can bind to

transport proteins involved in moving thyroid hormones throughout the body.³⁰ Scientists are still learning how this effect occurs and its impact on health.

Studies in research animals have shown that PFBA, as well as other PFAS, can affect the blood (e.g., red blood cell concentrations, hemoglobin level, hematocrit level).⁹ Testing whether these components fall within a normal range can help determine the health of a human or an animal and whether disease is present or there are issues with normal organ function.³¹ While PFAS, including PFBA, can be found in the blood of exposed individuals, epidemiologic studies among people have not found associated changes in blood components.^{9,32}

Studies in rodents have shown that PFBA, as well as other PFAS, can reduce cholesterol levels.⁹ However, studies in people have shown that PFAS exposure is associated with increased total cholesterol and LDL levels.^{9,33} Scientists are learning more about the ways in which PFAS reduce cholesterol levels in rodents and believe that they are associated with activation of nuclear hormone receptors.³⁴ Because nuclear receptors, like PPAR α , are important regulators of lipid metabolism, changes in their expression patterns can affect lipid transport and alter cholesterol levels. However, at this time, it is unclear whether PFAS might act the same way in people as they do in animals.

A number of studies have demonstrated that liver effects caused by PFAS, like PFBA, occur primarily through activation of the nuclear hormone receptor, PPAR α (peroxisome proliferator-activated receptor alpha).³⁵⁻⁴⁴ Nuclear receptors regulate gene expression and PPAR α regulates the expression of genes involved in lipid and cholesterol metabolism.⁴⁵ While PPAR α receptors are found in rodents and humans, levels of these receptors are much higher in rats and mice than in monkeys and humans.⁴⁵ This means that effects caused by activation of PPAR α are more likely to occur in rodents than they are to occur in people. As such, the effects on the liver observed in the critical studies reviewed here are likely not relevant to humans.

Standard Selection

DHS recommends an enforcement standard of 10 $\mu\text{g/L}$ for PFBA.

There are no federal numbers and no state drinking water standard for PFBA. Additionally, the EPA has not evaluated the carcinogenicity or established an ADI (oral reference dose) for PFBA.

However, we found several studies evaluating the toxicity of PFBA in research animals. To calculate the ADI as specified in s. 160.13, Wisc. Statute, DHS selected critical studies by Butenhoff et al., 2012b and van Otterdijk, 2007b.^{7,8} We selected these studies because they are long-term studies with agreeing NOAELs which found that PFBA affected the thyroid,

Basis for Enforcement Standard

- Federal Number
 - Cancer Potential
 - EPA Acceptable Daily Intake
 - Technical information
-

blood, cholesterol, and liver in male rats. In 2018, the Minnesota Department of Health established a benchmark dose lower confidence level (BMDL) of 3.0 mg/kg-d for PFBA based on these studies.^{46,i} We used this BMDL and a total uncertainty factor of 3000 to obtain an ADI of 0.001 mg/kg-d. This uncertainty factor accounts for differences between humans and research animals (10), differences among people (10), use of a long-term study duration (3), and the limited availability of information (10). To determine the recommended enforcement standard, DHS used the ADI, and, as required by Ch. 160, Wis. Stats., a body weight of 10 kg, a water consumption rate of 1 L/d, and a relative source contribution of 100%.

DHS recommends a preventive action limit of 2 µg/L for PFBA.

DHS recommends that the preventive action limit for PFBA be set at 20% of the enforcement standard because PFBA has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell cultures.⁹

Prepared by Amanda Koch, MPH and Sarah Yang, Ph.D.

Wisconsin Department of Health Services

References

1. Kissa E. *Fluorinated Surfactants and Repellants*. New York, NY: Marcel Dekker; 2001.
2. Abada B, Alivio TEG, Shao Y, et al. Photodegradation of fluorotelomer carboxylic 5:3 acid and perfluorooctanoic acid using zinc oxide. *Environ Pollut*. 2018;243(Pt A):637-644.
3. Arakaki A, Ishii Y, Tokuhisa T, et al. Microbial biodegradation of a novel fluorotelomer alcohol, 1H,1H,2H,2H,8H,8H-perfluorododecanol, yields short fluorinated acids. *Appl Microbiol Biotechnol*. 2010;88(5):1193-1203.

i The BMDL is the lower limit of a one-sided 95% confidence interval established using benchmark dose modeling. Benchmark dose modeling is considered the state of the science for establishing health-based values like an acceptable daily intake. Benchmark dose modeling takes account of all of the data for a particular effect from a particular experiment, allows for increased consistency, and can better account for statistical uncertainties.^{29,41-42}

4. Taniyasu S, Yamashita N, Yamazaki E, Petrick G, Kannan K. The environmental photolysis of perfluorooctanesulfonate, perfluorooctanoate, and related fluorochemicals. *Chemosphere*. 2013;90(5):1686-1692.
5. Arakaki A, Nakata S, Tokuhisa T, et al. Quantitative and time-course analysis of microbial degradation of 1H,1H,2H,2H,8H,8H-perfluorododecanol in activated sludge. *Appl Microbiol Biotechnol*. 2017;101(22):8259-8266.
6. Zhao L, Folsom PW, Wolstenholme BW, Sun H, Wang N, Buck RC. 6:2 fluorotelomer alcohol biotransformation in an aerobic river sediment system. *Chemosphere*. 2013;90(2):203-209.
7. Butenhoff JL, Bjork JA, Chang SC, et al. Toxicological evaluation of ammonium perfluorobutyrate in rats: Twenty-eight-day and ninety-day oral gavage studies. *Reproductive Toxicology*. 2012;33(4):513-530.
8. Otterdijk FMv. *Repeated Dose 90-Day Oral Toxicity Study With MTDID 8391 By Daily Gavage In The Rat Followed By A 3-Week Recovery Period*. 3M;2007. 3M Study No. 06-398.
9. ATSDR. Toxicological Profile for Perfluoroalkyls - Draft for Public Comment. In: Registry AFTSaD, ed. Atlanta, GA2017.
10. Chang SC, Das K, Ehresman DJ, et al. Comparative pharmacokinetics of perfluorobutyrate in rats, mice, monkeys, and humans and relevance to human exposure via drinking water. *Toxicological Sciences*. 2008;104(1):40-53.
11. Crebelli R, Caiola S, Conti L, et al. Can sustained exposure to PFAS trigger a genotoxic response? A comprehensive genotoxicity assessment in mice after subacute oral administration of PFOA and PFBA. *Regul Toxicol Pharmacol*. 2019;106:169-177.
12. Ikeda T, Aiba K, Fukuda K, Tanaka M. The induction of peroxisome proliferation in rat liver by perfluorinated fatty acids, metabolically inert derivatives of fatty acids. *J Biochem*. 1985;98(2):475-482.
13. Just WW, Gorgas K, Hartl FU, Heinemann P, Salzer M, Schimassek H. Biochemical effects and zonal heterogeneity of peroxisome proliferation induced by perfluorocarboxylic acids in rat liver. *Hepatology*. 1989;9(4):570-581.
14. Otterdijk FMv. *Repeated Dose 28-Day Oral Toxicity Study With MTDID-8391 By Daily Gavage In The Rat, Followed By A 21-Day Recovery Period*. 3M; 21 June 2007 2007. 3M Study No. 06-226.
15. Permadi H, Lundgren B, Andersson K, DePierre JW. Effects of perfluoro fatty acids on xenobiotic-metabolizing enzymes, enzymes which detoxify reactive forms of oxygen and lipid peroxidation in mouse liver. *Biochem Pharmacol*. 1992;44(6):1183-1191.
16. Das KP, Grey BE, Zehr RD, et al. Effects of perfluorobutyrate exposure during pregnancy in the mouse. *Toxicol Sci*. 2008;105(1):173-181.

17. Bao WW, Qian ZM, Geiger SD, et al. Gender-specific associations between serum isomers of perfluoroalkyl substances and blood pressure among Chinese: Isomers of C8 Health Project in China. *Sci Total Environ*. 2017;607-608:1304-1312.
18. Zeng XW, Li QQ, Chu C, et al. Alternatives of perfluoroalkyl acids and hepatitis B virus surface antibody in adults: Isomers of C8 Health Project in China. *Environ Pollut*. 2019;259:113857.
19. Fu Y, Wang T, Fu Q, Wang P, Lu Y. Associations between serum concentrations of perfluoroalkyl acids and serum lipid levels in a Chinese population. *Ecotoxicol Environ Saf*. 2014;106:246-252.
20. Kim DH, Kim UJ, Kim HY, Choi SD, Oh JE. Perfluoroalkyl substances in serum from South Korean infants with congenital hypothyroidism and healthy infants--Its relationship with thyroid hormones. *Environ Res*. 2016;147:399-404.
21. Kim JH, Park HY, Jeon JD, et al. The modifying effect of vitamin C on the association between perfluorinated compounds and insulin resistance in the Korean elderly: a double-blind, randomized, placebo-controlled crossover trial. *Eur J Nutr*. 2016;55(3):1011-1020.
22. ITRC. Environmental Fate and Transport for Per- and Polyfluoroalkyl Substances. In: Council ITR, ed2018.
23. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 140*2017.
24. USEPA. National Primary Drinking Water Regulations. 2018; <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>.
25. USEPA. Drinking Water Contaminant Human Health Effects Information. 2019; <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>.
26. USEPA. IRIS Assessments. 2019; https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm.
27. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 809*2018.
28. USEPA. A Review of the Reference Dose and Reference Concentration Processes. 2002(EPA/630/P-02/002F).
29. Calsolaro V, Pasqualetti G, Niccolai F, Caraccio N, Monzani F. Thyroid Disrupting Chemicals. *Int J Mol Sci*. 2017;18(12):17.
30. Ren XM, Qin WP, Cao LY, et al. Binding interactions of perfluoroalkyl substances with thyroid hormone transport proteins and potential toxicological implications. *Toxicology*. 2016;366-367:32-42.
31. National Heart L, and Blood Institute. Blood Tests. <https://www.nhlbi.nih.gov/health-topics/blood-tests>. Accessed 4/9/2020.
32. Calafat AM, Kato K, Hubbard K, Jia T, Botelho JC, Wong LY. Legacy and alternative per- and polyfluoroalkyl substances in the U.S. general population: Paired serum-urine data from the 2013-2014 National Health and Nutrition Examination Survey. *Environ Int*. 2019;131:105048.

33. Dong Z, Wang H, Yu YY, Li YB, Naidu R, Liu Y. Using 2003-2014 U.S. NHANES data to determine the associations between per- and polyfluoroalkyl substances and cholesterol: Trend and implications. *Ecotoxicol Environ Saf.* 2019;173:461-468.
34. Bijland S, Rensen PCN, Pieterman EJ, et al. Perfluoroalkyl Sulfonates Cause Alkyl Chain Length-Dependent Hepatic Steatosis and Hypolipidemia Mainly by Impairing Lipoprotein Production in APOE*3-Leiden CETP Mice. *Toxicological Sciences.* 2011;123(1):290-303.
35. Das KP, Wood CR, Lin MT, et al. Perfluoroalkyl acids-induced liver steatosis: Effects on genes controlling lipid homeostasis. *Toxicology.* 2017;378:37-52.
36. Rosen MB, Das KP, Rooney J, Abbott B, Lau C, Corton JC. PPAR alpha-independent transcriptional targets of perfluoroalkyl acids revealed by transcript profiling. *Toxicology.* 2017;387:95-107.
37. Albrecht PP, Torsell NE, Krishnan P, et al. A species difference in the peroxisome proliferator-activated receptor alpha-dependent response to the developmental effects of perfluorooctanoic acid. *Toxicological sciences : an official journal of the Society of Toxicology.* 2013;131(2):568-582.
38. Palkar PS, Anderson CR, Ferry CH, Gonzalez FJ, Peters JM. Effect of prenatal peroxisome proliferator-activated receptor alpha (PPARalpha) agonism on postnatal development. *Toxicology.* 2010;276(1):79-84.
39. Wolf DC, Moore T, Abbott BD, et al. Comparative hepatic effects of perfluorooctanoic acid and WY 14,643 in PPAR-alpha knockout and wild-type mice. *Toxicologic pathology.* 2008;36(4):632-639.
40. Rosenmai AK, Ahrens L, le Godec T, Lundqvist J, Oskarsson A. Relationship between peroxisome proliferator-activated receptor alpha activity and cellular concentration of 14 perfluoroalkyl substances in HepG2 cells. *J Appl Toxicol.* 2018;38(2):219-226.
41. Wolf CJ, Takacs ML, Schmid JE, Lau C, Abbott BD. Activation of mouse and human peroxisome proliferator-activated receptor alpha by perfluoroalkyl acids of different functional groups and chain lengths. *Toxicol Sci.* 2008;106(1):162-171.
42. Foreman JE, Chang SC, Ehresman DJ, et al. Differential hepatic effects of perfluorobutyrate mediated by mouse and human PPAR-alpha. *Toxicol Sci.* 2009;110(1):204-211.
43. Mahapatra CT, Damayanti NP, Guffey SC, Serafin JS, Irudayaraj J, Sepulveda MS. Comparative in vitro toxicity assessment of perfluorinated carboxylic acids. *J Appl Toxicol.* 2017;37(6):699-708.
44. Bjork JA, Wallace KB. Structure-activity relationships and human relevance for perfluoroalkyl acid-induced transcriptional activation of peroxisome proliferation in liver cell cultures. *Toxicol Sci.* 2009;111(1):89-99.
45. Hall AP, Elcombe CR, Foster JR, et al. Liver hypertrophy: a review of adaptive (adverse and non-adverse) changes--conclusions from the 3rd International ESTP Expert Workshop. *Toxicol Pathol.* 2012;40(7):971-994.

46. Health MDo. Toxicological Summary for: Perfluorobutanoate. In:2018.
47. Committee ES, Hardy A, Benford D, et al. Update: use of the benchmark dose approach in risk assessment. *EFSA Journal*. 2017;15(1):e04658.
48. Haber LT, Dourson ML, Allen BC, et al. Benchmark dose (BMD) modeling: current practice, issues, and challenges. *Critical Reviews in Toxicology*. 2018;48(5):387-415.
49. USEPA. Benchmark Dose Technical Guidance. In. Washington, DC 204602012.

Appendix A. Toxicity Data

Table A-I. PFBA Toxicity Studies from Literature Review

Study Type	Species	Duration	Doses	Route	Key Findings	Toxicity Value (mg/kg-d)	Reference
Short-Term	Rat	28 days	6, 30, 150 mg/kg-d	Gavage	Males: Higher serum potassium and inorganic phosphate at 150 mg/kg-d, higher absolute liver weight at 30 and 150 mg/kg-d, and lower serum total cholesterol and thyroxine levels at all doses Females: Higher absolute thymus weight at 6 mg/kg-d only	LOAEL: 30 (males); N/A (females) NOAEL (reported): 6 (males); 150 (females)	Butenhoff et al., 2012a ⁷
Long-Term	Rat	90 days	1.2, 6, 30 mg/kg-d	Gavage	Males: Higher absolute liver weight, lowered serum total thyroxine and other blood parameters, and altered clinical chemistry at 30 mg/kg-d; lower serum total bilirubin at 6 and 30 mg/kg-d Females: Lower serum total bilirubin at 30 mg/kg-d	LOAEL: 30 (males); N/A (females) NOAEL (reported): 6 (males); 30 (females)	Butenhoff et al., 2012b ⁷
Short-Term	Rat	5 weeks	5 mg/kg body weight (corresponding to 28 mg/L)	Drinking water	Mild liver hypertrophy with no genotoxicity	LOAEL: 5 NOAEL: N/A	Crebelli et al., 2019 ¹¹
Developmental	Mouse	GD 1–17	35, 175, 350 mg/kg-d	Gavage	Mothers: Greater incidence of full-litter loss at 350 mg/kg-d; higher maternal liver weights at 175 and 350 mg/kg-d. Offspring: Delays in eye-opening at all three doses; delays in vaginal opening at 175 and 350 mg/kg-d; delays in preputial opening at the highest dose	LOAEL: 35 NOAEL: N/A	Das et al., 2008 ¹⁶
Short-Term	Rat	2 weeks	0.02% PFBA (14 days in chow)	Diet (chow)	Significant increase in liver cell enzyme activity; no significant effect on relative liver weights	LOAEL: 0.02% NOAEL: N/A	Ikeda et al., 1985 ¹²
Short-Term	Rat	14 days	0.25% PFBA (14 days in chow)	Diet (chow)	Significantly increased liver size, significant changes in liver cell enzyme activity	LOAEL: 0.25% NOAEL: N/A	Just et al., 1989 ¹³

Short-Term	Mouse	10 days	78 mg/kg/day	Diet (chow)	Significant increase in absolute and relative liver weight accompanied by changes in enzymes involved in drug metabolism and/or in deactivation of reactive oxygen species; no significant effect on parameters of peroxisomal fatty acid β -oxidation	LOAEL: 78 NOAEL: N/A	Permadi et al., 1992 ¹⁵
Short-Term	Rat	28 days	6, 30, 150 mg/kg-d	Gavage	Increased absolute and relative liver weights at 150 mg/kg-d; increased potassium and organic phosphate levels at 150 mg/kg-d and reduced cholesterol at 30 and 150 mg/kg-d	LOAEL: 30 (males); N/A (females) NOAEL (reported): 6 (males); 150 (females)	van Otterdijk, 2007a ¹⁴
Long-Term	Rat	90 days	1.2, 6, 30 mg/kg-d	Gavage	Males at 30 mg/kg-d: Reduced red blood cell count, blood hemoglobin and hematocrit levels; increased red blood cell distribution width; increased absolute and relative liver weights; increased alkaline phosphatase activity levels; reduced total protein levels; reduced alanine aminotransferase and aspartame aminotransferase activity levels; reduced calcium levels	LOAEL: 30 (males); N/A (females) NOAEL (reported): 6 (males); 30 (females)	van Otterdijk, 2007b ⁸

GD=gestation day, PND=postnatal day, IP=intraperitoneal

Table A-2. Critical Study Selection for PFBA

Reference	Appropriate duration?	Effects consistent with other studies?	Effects relevant to humans?	Number of Doses	Toxicity value identifiable?	Critical study?
Butenhoff et al., 2012a ⁷	✓	✓	✓	3	✓	Yes
Butenhoff et al., 2012b ⁷	✓	✓	✓	3	✓	Yes
Crebelli et al., 2019 ¹¹	⊘	✓	✓	1	⊘	No
Das et al., 2008 ¹⁶	✓	✓	✓	3	✓	Yes
Ikeda et al., 1985 ¹²	⊘	✓	✓	1	✓	No
Just et al., 1989 ¹³	⊘	✓	✓	1	⊘	No
Permadi et al., 1992 ¹⁵	⊘	✓	✓	1	✓	No
Van Otterdijk, 2007a ¹⁴	✓	✓	✓	3	✓	Yes
Van Otterdijk, 2007b ⁸	✓	✓	✓	3	✓	Yes

To be considered a critical study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.

Table A-3. PFBA Epidemiological Studies from Literature Review*

Study Type	Population	Time period	Exposure	Outcomes	Results	Other PFAS evaluated	Reference
Cross-sectional	Chinese adults	2015–2016	PFBA concentrations in blood serum	Hypertension, blood pressure	<p>A 1-ln-unit increase of PFBA concentration (ng/ml) was significantly associated with an increased risk of hypertension (aOR=1.10, 95% CI: 1.04, 1.17) among males and females. When stratified by gender, the association remained significant for both genders.</p> <p>A 1-ln-unit increase of PFBA concentration (ng/ml) was not significantly associated with a change in diastolic blood pressure.</p>	PFNA, PFHxS, PFDA, PFHxA, PFDoA, PFTeA, PFTrA, PFUnA, PFBS, PFDS, PFHpA, PFPA	Bao et al., 2017 ¹⁷
Cross-sectional	Chinese general population (age range: 0.3–88 years)	2011	PFBA concentrations in blood serum	Serum lipid concentrations	PFBA was not associated with total cholesterol, triglycerides, high-density lipoprotein cholesterol or low-density lipoprotein cholesterol.	PFBS, PFHxS, PFOS, PFPeA, PFHxA, PFHpA, PFOA, PFNA, PFDA, PFUnA, PFDoA	Fu et al., 2014 ¹⁹
Case-control	South Korean infants	2009–2010	PFBA concentrations in blood serum	Congenital hypothyroidism	No statistically significant differences in PFBA exposure levels between infants with and without congenital hypothyroidism were identified.	PFPeA, PFHxA, PFHpA, PFOA, PFNA, PFDA, PFUnA, PFOS, PFBS, PFDoA, PFTrDA, PFTeDA, PFHxS, PFDS, PFHpS,	Kim et al., 2016 ²⁰
Prospective cohort	Korean elderly	2011–2012	PFAS concentrations in blood serum	Insulin resistance	No relationship between insulin resistance and those with varying PFBA levels was identified.	PFBS, PFHxS, PFOS, PFDS, PFBA, PFPeA, PFHxA, PFHpA, PFOA, PFNA, PFDA, PFUnA, PFTrDA, PFTeDA	Kim et al., 2016 ²¹

Cross-sectional	Chinese adults?	PFAS concentrations in blood serum	Hepatitis B virus surface antibody levels in blood serum	Lower serum Hepatitis B virus surface antibody levels (log mIU/mL) were observed for each log-unit increase in linear PFBA ($\beta = -0.18$, 95% CI: 0.28, -0.08).	Zeng et al., 2019 ¹⁸
-----------------	-----------------	------------------------------------	--	--	---------------------------------

***This literature review is not exhaustive as the primary purpose of the search was to identify epidemiological studies that support toxicological findings.**

Epidemiologic terms: OR=odds ratio; AOR=adjusted odds ratio; RR=relative risk; 95% CI=95% confidence interval; r=Spearman correlation coefficient; β =regression coefficient
PFAS acronyms: PFOA=perfluorooctanoic acid, PFNA=perfluorononanoic acid, PFDA=perfluorodecanoic acid, PFUnA= perfluoroundecanoic acid, PFTrDA=perfluorotridecanoic acid, PFTeDA=perfluorotetradecanoic acid, PFHxDA=perfluorohexadecanoic acid, PFHxS=perfluorohexane sulfonate, PFOS=perfluorooctane sulfonic acid, PFDS=perfluorodecanesulfonate, PFBS=perfluorobutane sulfonate, PFBA=perfluorobutanoic acid, PFPeA=perfluoropentanoic acid, PFHxA=perfluorohexanoic acid, PFHpA=perfluoroheptanoic acid, 6:2 FTSA=6:2 fluorotelomer sulfonates, NMeFOSAA=N-methyl perfluorooctanesulfonamidoacetate, EPAH=2-(N-ethyl-perfluorooctane sulfonamido)acetate, MPAH=2-(N-methyl-perfluorooctane sulfonamido) acetate, PFSA=perfluorooctane sulfonamide; PFHpS=perfluoroheptane sulfonic acid; PFDS=perfluorodecane sulfonic acid; EtPFOSAA= 2-(Nethyl-perfluorooctane sulfonamido) acetic acid

PFPeA | 2020

Substance Overview

Perfluoropentanoic acid^a (PFPeA) is a chemical in a group of contaminants called per- and polyfluoroalkyl substances (PFAS). As PFAS with longer carbon chains are phased out of production, PFPeA, with only five carbons, is increasingly found in stain repellants in commercial products like carpet and fabric, and in some fire-fighting foams.¹ PFPeA persists in the environment for decades.¹

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard or Preventive Action Limit for PFPeA.

DHS found insufficient information to recommend an enforcement standard or preventive action limit for PFPeA.

Health Effects

Studies investigating the health effects of PFPeA are limited.

Data from *in vitro* studies suggest that the liver may be a target of PFPeA toxicity; this effect is consistent with those seen with other PFAS.²⁻⁴ Limited studies in people suggest that PFPeA may also impact thyroid hormone levels and sperm motility.⁵⁻⁷

There is insufficient evidence to determine if PFPeA causes carcinogenic (cancer), mutagenic (DNA damage), teratogenic (birth defects), or interactive effects in people or research animals. The EPA has not evaluated the carcinogenicity of PFPeA.⁸

Current Standards

Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

Recommended Standards

Enforcement Standard:	N/A
Preventive Action Limit:	N/A

a This scientific support document also applies to anion salts of perfluoropentanoic acid.

Chemical Profile

Acronym	
Structure:	
CAS Number:	2706-90-3
Formula:	C ₅ HF ₉ O ₂
Molar Mass:	264.05 g/mol
Synonyms:	Pentanoic acid, 2,2,3,3,4,4,5,5,5-nonafluoro-; Undecafluorohexanoic acid; Perfluorovaleric acid

Exposure Routes

PFAS, including PFPeA, can be released directly into the environment during the manufacture and use of PFAS and can be found in water or soil. PFAS can move between groundwater and surface water. Once in water, PFAS can travel long distances.^{9,10} Short-chain PFAS, such as PFPeA, are more mobile and can travel longer distances than longer-chain PFAS.¹

People can be exposed to PFAS by drinking water, eating food, and breathing in or accidentally swallowing soil or dust containing PFAS.¹⁴

Current Standard

Wisconsin does not currently have groundwater standards for PFPeA.¹¹

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

EPA Oral Reference Dose:	N/A
--------------------------	-----

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

None Available

Literature Search

Literature Search Dates:	1900-2019
Key studies found?	No
Critical studies identified?	No

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for PFPeA.¹²

Health Advisory

The EPA has not established health advisories for PFPeA.¹³

Drinking Water Concentration (Cancer Risk)

The EPA has not established drinking water concentrations based on a cancer risk level determination for PFPeA.⁸

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for PFPeA.¹⁴

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Information System (IRIS) program.

EPA Oral Reference Dose

The EPA does not have an oral reference dose for PFPeA.⁸

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If we determine that something is carcinogenic and there is no federal number or ADI from the EPA, DHS must set the standard at a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of PFPeA, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of PFPeA. If so, we look to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA and International Agency for Research on Cancer (IARC) have not evaluated the carcinogenicity of PFPeA.^{8,15}

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for PFPeA.⁸

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information that was not considered when the value was established and that indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For PFPeA, we searched for values that have been published on or before Dec 2019. As of Dec 2019, no agencies or organizations have reviewed the toxicity of PFPeA or developed guidance values for PFPeA.

Literature Search

Our literature review focused on relevant scientific literature on the health effects of PFPeA published on or before Dec 2019. We looked for studies related to PFPeA toxicity or effects on a disease state in which information on exposure or dose was included as part of the study.^b Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

Seven toxicity studies were returned by the search engines. We excluded studies on non-mammalian or cell systems, non-oral exposure routes, and those that did not evaluate health risks from further review. After applying these exclusion criteria, we located one key toxicity study on PFPeA (summarized in Table A-1). To be considered a critical toxicity study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.^{c,d} After

^b We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

Title/abstract: PFPeA OR 2706-90-3 OR "Perfluoropentanoic acid" OR "perfluoropentanoate"

Subject area: toxicology

Language: English

We also searched online for toxicity studies published by national research programs.

^c Appropriate toxicity values include the no observable adverse effect level (NOAEL), lowest observable adverse effect level (LOAEL), and benchmark dose (BMD). The NOAEL is the highest dose tested that did not cause an adverse effect, the LOAEL is the lowest dose tested that caused an adverse effect, and the BMD is an estimation of the dose that would cause a specific level of response (typically 5 or 10%).

^d Due to the limited availability of data for this substance, we considered a study to be of an appropriate duration if animals were exposed for at least 28 days or if the exposure occurred during pregnancy. We accounted for differences in exposure duration through the use of an uncertainty factor.

reviewing the key study, we determined that it did not meet the criteria to be considered a critical toxicity study (see Table A-2). We did locate one *in vitro* study evaluating the toxicity of PFPeA in mammalian cell lines (see Table A-3 for details on this study).

In our search, we also located four epidemiology studies (see Table A-4 for a summary). While multiple potential exposure sources and the ability for other PFAS compounds to cause similar health effects preclude using these data to establish a health-based value, such studies are helpful in identifying the crucial effects and ensuring that the animal data used to establish the standard are relevant to people.

Critical toxicity studies

We did not identify any critical toxicity studies.

Key health effects

We did not find any studies indicating that PFPeA can cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture.

Standard Selection

DHS found insufficient technical information to recommend an enforcement standard for PFPeA.

We did not identify sufficient technical information to recommend an enforcement standard for PFPeA at this time. There are no federal numbers and no state drinking water standard for PFPeA. Additionally, the EPA has not evaluated the carcinogenicity or established an ADI (oral reference dose) for PFPeA.

We did not find any critical studies evaluating the toxicity of PFPeA in research animals. While a handful of epidemiological studies have evaluated the risk of PFPeA, the results are inconsistent. There is limited

evidence from human studies that PFPeA may impact thyroid hormones, but *in vitro* studies have not supported that evidence.^{5,6,17} Available information from one cell culture study suggests that the toxic potency of PFPeA is between that of perfluorobutanoic acid (PFBA, four carbons) and perfluorohexanoic acid (PFHxA, six carbons), but limited scientific information precludes us from combining PFPeA with either chemical.⁴

Basis for Enforcement Standard

- Federal Number
 - Cancer Potential
 - EPA Acceptable Daily Intake
 - Technical information
-

DHS does not recommend a preventive action limit for PFPeA.

We did not identify sufficient technical information to recommend a preventive action limit for PFPeA at this time.

Prepared by Nathan Kloczko, MPH and Sarah Yang, Ph.D.

Wisconsin Department of Health Services

References

1. Brendel S, Fetter É, Staude C, Vierke L, Biegel-Engler A. Short-chain perfluoroalkyl acids: environmental concerns and a regulatory strategy under REACH. *Environ Sci Eur.* 2018;30(1):9-9.
2. Wolf CJ, Schmid JE, Lau C, Abbott BD. Activation of mouse and human peroxisome proliferator-activated receptor- α (PPAR α) by perfluoroalkyl acids (PFAAs): further investigation of C4-C12 compounds. *Reprod Toxicol.* 2012;33(4):546-551.
3. Rosenmai AK, Ahrens L, le Godec T, Lundqvist J, Oskarsson A. Relationship between peroxisome proliferator-activated receptor α activity and cellular concentration of 14 perfluoroalkyl substances in HepG2 cells. *J Appl Toxicol.* 2018;38(2):219-226.
4. Rand AA, Rooney JP, Butt CM, Meyer JN, Mabury SA. Cellular toxicity associated with exposure to perfluorinated carboxylates (PFCAs) and their metabolic precursors. *Chem Res Toxicol.* 2014;27(1):42-50.
5. Li Y, Cheng Y, Xie Z, Zeng F. Perfluorinated alkyl substances in serum of the southern Chinese general population and potential impact on thyroid hormones. *Sci Rep.* 2017;7:43380.
6. Shah-Kulkarni S, Kim BM, Hong YC, et al. Prenatal exposure to perfluorinated compounds affects thyroid hormone levels in newborn girls. *Environ Int.* 2016;94(Elsevier):607-613.
7. Song X, Tang S, Zhu H, et al. Biomonitoring PFAAs in blood and semen samples: Investigation of a potential link between PFAAs exposure and semen mobility in China. *Environ Int.* 2018;113:50-54.
8. USEPA. IRIS Assessments. https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm. Published 2019. Accessed.
9. ATSDR. Toxicological Profile for Perfluoroalkyls - Draft for Public Comment. In: Registry AftSaD, ed. Atlanta, GA2017.
10. ITRC. Environmental Fate and Transport for Per- and Polyfluoroalkyl Substances. In: Council ITR, ed2018.
11. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 140*2017.
12. USEPA. National Primary Drinking Water Regulations. <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>. Published 2018. Accessed.
13. USEPA. Drinking Water Contaminant Human Health Effects Information. <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>. Published 2019. Accessed.

14. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 809*2018.
15. IARC. List of Classification, Volumes 1-123. <https://monographs.iarc.fr/list-of-classifications-volumes/>. Published 2018. Accessed May 17, 2019.
16. Ritter L, Totman C, Krishnan K, Carrier R, Vezina A, Morisset V. Deriving uncertainty factors for threshold chemical contaminants in drinking water. *Journal of toxicology and environmental health Part B, Critical reviews*. 2007;10(7):527-557.
17. Croce L, Coperchini F, Tonacchera M, Imbriani M, Rotondi M, Chiovato L. Effect of long- and short-chain perfluorinated compounds on cultured thyroid cells viability and response to TSH. *J Endocrinol Invest*. 2019;42(11):1329-1335.
18. Upham BL, Park JS, Babica P, et al. Structure-activity-dependent regulation of cell communication by perfluorinated fatty acids using in vivo and in vitro model systems. *Environ Health Perspect*. 2009;117(4):545-551.
19. Nian M, Li QQ, Bloom M, et al. Liver function biomarkers disorder is associated with exposure to perfluoroalkyl acids in adults: Isomers of C8 Health Project in China. *Environ Res*. 2019;172:81-88.

Appendix A. Toxicity Data

Table A-I. PFPeA Toxicity Studies from Literature Review

Study Type	Species	Duration	Doses (mg/kg-d)	Route	Endpoints	Toxicity Value (mg/kg-d)	Reference
Short-term	Rats	1 week	0, 32.3	Dietary, 0.02%	No inhibition of gap-junctional intercellular communications No increase in liver weight	NOAEL: 32.3	Upham et al., 2009 ⁽¹⁸⁾
NOAEL: No observed adverse effect level							

Table A-3. Critical Study Selection for PFPeA

Reference	Appropriate duration?	Effects consistent with other studies?	Effects relevant to humans?	Number of Doses	Toxicity value identifiable?	Critical study?
Upham et al., 2009	⊘	⊘	⊘	1	✓	No

To be considered a critical study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.

Table A-3. PFPeA In Vitro Toxicity Studies from Literature Review

Species	Cell Line	Duration (hr)	Doses (mM)	Other PFAS evaluated	Key Findings	Reference
Human	THLE-2 (liver epithelial)	24	0.050 – 4	PFBA, PFHpA PFHxA, PFOA, PFNA, PFDA	Perfluorocarboxylic acids increase in toxicity with increasing chain length; PFPeA between PFBS and PFHxA	Rand, 2013 (4)
<p>PFAS acronyms: PFOA=perfluorooctanoic acid, PFNA=perfluorononanoic acid, PFDA=perfluorodecanoic acid, PFUnA= perfluoroundecanoic acid, PFDoA=perfluorododecanoic acid, PFTriA=perfluorotetradecanoic acid, PFHxDA=perfluorohexadecanoic acid, PFHxS=perfluorohexane sulfonate, PFOS=perfluorooctane sulfonic acid, PFDS=perfluorodecanesulfonate, PFBS=perfluorobutane sulfonate, PFBA=perfluorobutanoic acid, PFPeA=perfluoropentanoic acid, PFHxA=perfluorohexanoic acid, PFHpA=perfluoroheptanoic acid, 6:2 FTSA=6:2 fluorotelomer sulfonates, NMeFOSAA=N-methyl perfluorooctanesulfonamidoacetate, EPAH=2-(N-ethyl-perfluorooctane sulfonamido)acetate, MPAH=2-(N-methyl-perfluorooctane sulfonamido) acetate, PFSA=perfluorooctane sulfonamide; PFHpS=perfluoroheptane sulfonic acid; PFDS= perfluorodecane sulfonic acid; EtPFOSAA= 2-(Nethyl-perfluorooctane sulfonamido) acetic acid</p>						

Table A-4. PFPeA Epidemiological Studies from Literature Review

Study Type	Population	Time period	Exposure	Outcomes	Results	Other PFAS Studied	Reference
Cross-sectional	202 Chinese adults	2013-2014	Serum PFAS concentrations	Serum thyroid hormone levels	PFPeA was positively correlated with TGAb and TMAb in all samples. In a subgroup with hyperthyroidism, PFPeA was associated with a 1.2-8% decrease in TSH.	PFOS, PFOA, PFHxS, PFPrA, PFBA, PFHxA	Li et al., 2017 ⁽⁵⁾
Cross-sectional	1605 highly exposed Chinese adults	2015-2016	Serum PFAS concentrations	Liver function biomarkers	PFPeA was associated with elevated odds of abnormal prealbumin levels (OR = 1.25, 95% CI: 1.07, 1.46)	PFBA, PFHxA, PFHxS, PFNA, PFDA, PFDS, PFUnA, PFDoA, PFTrA, PFTeA	Nian et al., 2019 ⁽¹⁹⁾
Cohort	279 Korean mother-daughter pairs	2006-2010	Cord blood PFAS concentrations	Thyroid hormone levels	Serum PFPeA concentrations were positively associated with T4 concentrations in adjusted analyses (β = 0.27, 95% CI: 0.04, 0.49).	PFOS, PFOA, PFNA, PFDA, PFDoA, PFTrA, PFTeA, PFUnA, PFHxS	Shah-Kulkarni et al., 2016 ⁽⁶⁾
Cross-sectional	103 male Chinese adults	2012-2013	Blood and semen PFAS concentrations	Semen quality	PFPeA concentrations in semen were negatively associated with progressive motility (r = -0.344), but PFPeA concentrations in blood were positively associated with progressive motility (r = 0.235).	PFBA, PFHxA, PFBS, PFHpA, PFOA, PFHS, PFOS	Song et al., 2018 ⁽⁷⁾
<p>Epidemiologic terms: OR=odds ratio; AOR=adjusted odds ratio; RR=relative risk; 95% CI=95% confidence interval; r=Spearman correlation coefficient; β=regression coefficient</p> <p>PFAS acronyms: PFOA=perfluorooctanoic acid, PFNA=perfluorononanoic acid, PFDA=perfluorodecanoic acid, PFUnA= perfluoroundecanoic acid, PFDoA=perfluorododecanoic acid, PFTriA=perfluorotetradecanoic acid, PFHxDA=perfluorohexadecanoic acid, PFHxS=perfluorohexane sulfonate, PFOS=perfluorooctane sulfonic acid, PFDS=perfluorodecanesulfonate, PFBS=perfluorobutane sulfonate, PFBA=perfluorobutanoic acid, PFPeA=perfluoropentanoic acid, PFHxA=perfluorohexanoic acid, PFHpA=perfluoroheptanoic acid, 6:2 FTSA=6:2 fluorotelomer sulfonates, NMeFOSAA=N-methyl perfluorooctanesulfonamidoacetate, EPAH=2-(N-ethyl-perfluorooctane sulfonamido)acetate, MPAH=2-(N-methyl-perfluorooctane sulfonamido) acetate, PFSA=perfluorooctane sulfonamide; PFHpS=perfluoroheptane sulfonic acid; PFDS= perfluorodecane sulfonic acid; EtPFOSAA= 2-(Nethyl-perfluorooctane sulfonamido) acetic acid</p>							

PFHxA | 2020

Substance Overview

Perfluorohexanoic acid^a (PFHxA) is a chemical in a group of contaminants called per- and polyfluoroalkyl substances (PFAS). Because of its chemical properties, PFHxA can be found as an impurity in stain repellants in commercial products like carpet and fabric, as a coating for packaging, and in some fire-fighting foams.^{1,2} PFHxA can persist in the environment for decades.¹

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard for PFHxA. DHS recommends an enforcement standard of 150 micrograms per liter (µg/L) for PFHxA. The recommended standard is based on a study that found that high levels of PFHxA caused a number of health effects in rats after long-term exposure.³

DHS recommends that the preventive action limit for PFHxA be set at 20% of the enforcement standard because PFHxA has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture.

Current Standards

Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

Recommended Standards

Enforcement Standard:	150 µg/L
Preventive Action Limit:	30 µg/L

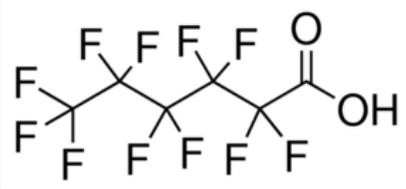
Health Effects

At this time, very few studies have evaluated the effects of PFHxA in people.^{1,4-8} The available studies have found that PFHxA may be associated with decreased testosterone levels and downregulated protective immune proteins in males.^{4,7,8} Studies in research animals have shown that high levels of PFHxA can impact development, alter thyroid hormone levels, affect the blood, cause kidney and liver damage, and reduce survival.^{1,9-12}

PFHxA has not been shown to cause carcinogenic (cancer), mutagenic (DNA damage), teratogenic (birth defects), or interactive effects in people, research animals, or cell culture.¹ The EPA has not evaluated the carcinogenicity of PFHxA.¹³

^a This scientific support document and the included groundwater standard recommendations also apply to anion salts of perfluorohexanoic acid.

Chemical Profile

PFHxA	
Structure:	
CAS Number:	307-24-4
Formula:	C ₆ HF ₁₁ O ₂
Molar Mass:	314.1 g/mol
Synonyms:	Undecafluorohexanoic acid 2,2,3,3,4,4,5,5,6,6,6-undecafluoro- hexanoic acid Perfluoro-1-pentanecarboxylic acid Undecafluorocaproic acid Perfluoro-n-hexanoic acid

Exposure Routes

People can be exposed to PFHxA by drinking contaminated water, swallowing contaminated soil, eating food that was packaged in material that contains PFHxA, and breathing in or swallowing dust that contains PFHxA.¹

In the environment, PFHxA can be found in water or soil as an impurity from the use of other PFAS in manufacturing and consumer products.¹ It can also get into the water or soil from the use of fire-fighting foam.¹⁴ PFHxA can move between groundwater and surface water. Once in groundwater, PFHxA can travel long distances.¹

Current Standard

Wisconsin does not currently have groundwater standards for PFHxA.¹⁵

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

EPA Oral Reference Dose:	N/A
--------------------------	-----

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

None available

Literature Search

Literature Search Dates:	2000 – 2019
Number of studies found ^b :	Approximately 270
Key studies found?	Yes
Critical studies identified?	Yes

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for PFHxA.¹⁶

Health Advisory

The EPA has not established health advisories for PFHxA.¹⁷

^b We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

Title/abstract: PFHxA or "perfluorohexanoic acid"

Subject area: toxicology

Language: English

We also searched online for toxicity studies published by national research programs.

Drinking Water Concentration (Cancer Risk)

The EPA has not established a drinking water concentration based on a cancer risk level determination for PFHxA.¹³

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for PFHxA.¹⁸

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, Ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Information System (IRIS) program.

EPA Oral Reference Dose

The EPA does not have an oral reference dose for PFHxA.¹³

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If we determine that something is carcinogenic and there is no federal number or ADI from the EPA, DHS must set the standard at a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of PFHxA, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of PFHxA. If so, we look to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA and International Agency for Research on Cancer (IARC) have not evaluated the carcinogenicity of PFHxA.^{13,19}

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for PFHxA.¹³

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information that was not considered when the value was established and indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For PFHxA, we searched for values that had been published on or before September 2019. While the Agency for Toxic Substances and Disease Registry (ATSDR) reviewed the toxicity of PFHxA in 2018, they did not establish any guidance values for PFHxA.^{1, c}

Literature Search

Our literature review focused on relevant scientific literature on the health effects of PFHxA published on or before September 2019. We looked for studies related to PFHxA toxicity or effects on a disease state in which information on exposure or dose was included as part of the study. Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

Approximately 270 toxicity studies were returned by the search engines. We excluded studies on non-mammalian species or cell systems, non-oral exposure routes, and those that did not evaluate health risks from further review. After applying these exclusion criteria, we located five key toxicity studies on PFHxA (summarized in Table A-1). To be considered a critical toxicity study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an

c ATSDR stated that they did not identify any intermediate-duration oral studies for PFHxA in their literature review. While they located one chronic duration study, they did not establish a minimum risk level for PFHxA because this study did not measure serum levels and a half-life for PFHxA in humans was not available.

identifiable toxicity value.^{d-e} All of these studies met the criteria to be considered a critical toxicity study (see Table A-2).

In our search, we also located a handful of epidemiology studies in our search (See Table A-3 for a summary). While multiple potential exposure sources and the ability for other PFAS compounds to cause similar health effects preclude using these data to establish a health-based value, such studies are helpful in identifying the crucial effects and ensuring that the animal data used to establish the standard is relevant to people.

Critical Toxicity Studies

To compare between results from recently found studies, we calculated an acceptable daily intake (ADI) for each study/effect. The ADI is the estimated amount of PFHxA that a person can be exposed to every day and not experience health impacts. Limited data suggest that the half-life of PFHxA in people is much shorter (several months) than that of other PFAS (years for PFOS, PFOA, and PFHxS).^{21,22} As such, we calculated ADI by dividing a toxicity value from either a no-observed adverse effect level (NOAEL), lowest observed adverse effect level (LOAEL), or benchmark dose (BMD) identified in a study by a factor accounting for various sources of scientific uncertainty.^f Uncertainty factors were included, as appropriate, to account for differences between humans and animals, differences between healthy and sensitive human populations, using data from short-term experiments to protect against effects from long-term exposure, and using data where a health effect was observed to estimate the level that does not cause an effect. To ensure appropriate protection, we have chosen to not use studies that have significant uncertainty as the basis for the recommended enforcement standards.^g This approach is consistent with that taken by EPA when establishing oral reference doses.²⁰

Chengelis et al., 2009

Chengelis et al. exposed male and female rats to different concentrations of PFHxA (0, 10, 50, and 200 milligrams of PFHxA per kilogram body weight per day or mg/kg-d) for 90 days through gavage.⁹ PFHxA affected growth and caused liver damage in males and altered some biochemistry parameters in males

d Appropriate toxicity values include the no observable adverse effect level (NOAEL), lowest observable adverse effect level (LOAEL), and benchmark dose (BMD).²⁰

e Due to the limited availability of data for this substance, we considered a study to be of an appropriate duration if animals were exposed for at least 28 days or if the exposure occurred during pregnancy. We accounted for differences in exposure duration through the use of an uncertainty factor.

f The NOAEL is the highest dose tested that did not cause an adverse effect, the LOAEL is the lowest dose tested that caused an adverse effect, and the BMD is an estimation of the dose that would cause a specific level of response (typically 5 or 10%).

g DHS considers a study to have significant uncertainty if the total uncertainty factors is greater than 3,000.

and females (Table 1). In addition to these statistically significant effects, PFHxA also caused pathological effects in the livers in males at 200 mg/kg-d.

Table 1. Statistically Significant Effects Observed in Chengelis et al., 2009

Effects observed in males before recovery period		Dose (mg/kg-d)		
		10	50	200
Kidney	Elevated relative kidney weight	✓	✓	✓
Growth	Reduced body weight		*	*
Blood	Elevated platelet counts	*	*	
	Reduced red blood cell count			*
	Reduced hemoglobin level			*
	Reduced hematocrit level			*
	Elevated reticulocyte counts			*
Biochemistry	Reduced cholesterol levels		✓	✓
	Reduced calcium levels		✓	✓
	Reduced total protein			✓
	Reduced globulin			✓
	Elevated alanine aminotransferase (ALT)			✓
	Elevated alkaline phosphatase (ALP)			✓
	Elevated peroxisomal β -oxidation activity			✓
Liver	Elevated relative liver weight			*
	Elevated liver to brain weight			*
Effects observed in females before recovery period		Dose (mg/kg-d)		
		10	50	200
Kidney	Elevated relative kidney weight		✓	
Blood	Reduced red blood cell count			*
Biochemistry	Reduced globulin			✓

*The authors did not consider this effect to be adverse (see Chengelis et al., 2009 for more details).

From this study, we identified a LOAEL of 10 mg/kg-d based on elevated relative kidney weight at all doses. We selected this value instead of the NOAEL identified by the authors because this effect was observed in males at all doses. While data on this effect are limited, and some studies among people have shown that other PFAS may impact kidney function.^{1,23} We applied a total uncertainty factor of 3000 to account for differences between people and research animals (10), differences among people (10), using data from a short-term study to protect from long-term effects (3), and the limited availability of information (10). We obtained a candidate ADI of 0.0033 mg/kg-d.

Iwai et al., 2019

In this study, Iwai et al. re-evaluated the findings from a study that they published in 2014.^{10,24} In the 2014 study, they exposed pregnant mice to different concentrations of PFHxA during pregnancy (gestational days 6 to 18) through gavage in two separate experiments.²⁴ In the first experiment (Phase I), animals were exposed to 0, 100, 350, and 500 mg/kg-d PFHxA. In the second experiment (Phase II), animals were exposed to 0, 7, 35, and 175 mg/kg-d PFHxA. In these experiments, PFHxA affected offspring viability, development, and liver weight (Table 2). In the 2019 study, the authors evaluated the statistically significant effects observed in 2014 to determine if they are related to PFHxA exposure and should be considered adverse. In this evaluation, the authors considered these effects relative to pooled controls from phases I and II, historical controls from other studies at the same laboratory, and data from the literature.

Table 2. Statistically Significant Effects Observed in Iwai et al., 2014⁽²⁴⁾

Effects in Phase I		Dose (mg/kg-d)		
		100	350	500
Survival	Increased percent of dams with no stillbirths			✓
	Increased percent of dams with all pups dying on PND 0-3			✓
	Decreased percent of liveborn			✓
	Increased number of pups found dead on PND 0			✓
	Increased number of pups found dead on PNDs 1-4		✓	✓
	Decreased viability on Day 4			✓
	Decreased viability on Day 7			✓
Development	Decreased percent of pups meeting eye opening criterion on PND 13		✓	✓
	Delayed eye opening (later criterion day)		✓	
Growth	Decreased pup weight per litter on PND 0	*	✓	✓
	Reduced body weight gain in parental females on PNDs 0-4		✓	✓
	Reduced offspring male body weight at PND 21	✓	✓	✓
	Reduced offspring female body weight at PNDs 21, 28, 35	✓	✓	
	Reduced offspring female body weight at PND 41 (termination)		✓	
Liver	Decreased relative liver weight in offspring males			✓
Effects in Phase II		Dose (mg/kg-d)		
		7	35	175
Survival	Decreased lactation index (number)	*		
	Increased stillborn			*
	Increased number of pups found dead on PND 0			*

Growth	Decreased pup weight per litter on PND 0		*
Development	Decreased preputial separation	*	

*The authors did not consider this effect to be treatment-related or did not consider it adverse.

In their re-evaluation, the authors determined that the stillbirths reported in 2014 were unlikely to be related to PFHxA exposure. They also determined that the effects on mortality at PND 0 are not adverse because they did not occur in a dose-dependent manner and were not sustained throughout the duration of the experiment. As such, the authors identified an unbounded NOAEL of 175 mg/kg-d because they did not observe any statistically significant adverse effects related to PFHxA exposure at all levels evaluated in Phase II.

While effects on body weight and body weight gain were observed in offspring at several time points, these effects were not observed at the end of the exposure period and did not occur in a dose-dependent manner. As such, we used the unbounded NOAEL and applied a total uncertainty factor of 1000 to account for differences between people and research animals (10), differences among people (10), and the limited availability of information (10). We obtained a candidate ADI of 0.175 mg/kg-d.

Klaunig et al., 2015

Klaunig et al. exposed male and female rats to different concentrations of PFHxA (0, 10, 50, and 200 mg/kg-d) for two years through gavage.²⁵ PFHxA caused clinical toxicity and altered some biochemistry parameters in males and females and caused kidney damage in females (Table 3). In addition to these statistically significant effects, PFHxA also caused pathological effects in the kidneys in females at 200 mg/kg-d.

Table 3. Statistically Significant Effects Observed in Klaunig et al., 2015⁽³⁾

Effects in males		Dose (mg/kg-d)		
		2.5	15	100
Biochemistry	Increased phosphorus levels		*	*
	Decreased triglycerides*	*		*
	Decreased free fatty acid	*		*
	Increased sodium level			*
Urine	Increased urine specific gravity		✓	
	Increased urobilinogen level		✓	
	Decreased urine pH			*
Clinical	Abnormal lung sounds (rales)			✓
	Yellow material on body (ventral trunk, anogenital, urogenital areas)			✓
Effects in females		Dose (mg/kg-d)		
		5	30	200
Biochemistry	Decreased red blood cell levels			*

	Decreased hemoglobin levels	*
	Increased reticulocyte level	*
	Increased triglycerides	*
	Decreased low density lipoprotein (LDL) and very low density lipoprotein (VLDL)	*
Urine	Increased urine specific gravity	✓
	Increased urine total volume	✓
Clinical	Decreased survival rate	*
	Abnormal lung sounds (rales)	✓
	Yellow material on body (ventral trunk, anogenital, urogenital areas)	✓

*The authors did not consider this effect to be treatment-related or did not consider it adverse.

From this study, we identified a NOAEL of 15 mg/kg-d based on clinical toxicity effects in male rats at the highest dose. We applied a total uncertainty factor of 1000 to account for differences between people and research animals (10), differences among people (10), and the limited availability of information (10). We obtained a candidate ADI of 0.015 mg/kg-d.

Loveless et al., 2009

Loveless et al. evaluated the subchronic, reproductive, and developmental toxicity of PFHxA in rats.¹¹ In these experiments, they exposed male and female rats to different concentrations of PFHxA (0, 20, 100, and 500 mg/kg-d) through gavage. Animals were exposed for 90 days in the subchronic study; for 126 days in the reproductive study which includes cohabitation, gestation, and lactation; and from gestation days 6 to 22 during the developmental study.

Subchronic Study

In the subchronic study, PFHxA affected body weight, blood parameters, and the liver, kidneys, and thyroid (Table 4A). In addition to these statistically significant effects, PFHxA also caused pathological changes in the nose, liver, and thyroid. More specifically, PFHxA caused degeneration of the olfactory epithelium and other changes to the nasal passage in males and females at doses of 100 and 500 mg/kg-d; hepatocellular hypertrophy in males at 100 and 500 mg/kg-d and females at 500 mg/kg-d; and hypertrophy of the thyroid follicular epithelium in males and females and 500 mg/kg-d.

Table 4A. Significant Subchronic Effects Observed in Loveless et al., 2009 ⁽¹¹⁾

Effects in males		Dose (mg/kg-d)		
		20	100	500
Growth	Decreased body weight on days 42 to 105			✓
Biochemistry	Decreased calcium			*
	Decreased sodium			*
	Increased potassium			*
	Increased chloride		*	*
Urine	Increased volume			✓
	Decreased osmolality			✓
	Decreased protein			✓
	Increased fluoride			*
Blood	Increased red blood cells			*
	Decreased hemoglobin			✓
	Decreased hematocrit			✓
	Decreased mean corpuscular hemoglobin concentration			✓
	Increased reticulocyte			*
	Increased platelet			*
	Increased neutrophils	*	*	*
	Decreased eosinophils			*
	Decreased basophils			*
	Decreased activated partial prothrombin time	*	*	*
Kidney	Increased relative kidney weight		*	*
	Decreased creatinine			*
Brain	Increased relative brain weight			✓
Spleen	Decreased absolute spleen weight			✓
Thymus	Decreased absolute thymus weight			✓
Reproduction	Increased relative testes weight			✓
Liver	Increased absolute and relative liver weight			*
	Increased peroxisomal β -oxidation activity			*
	Increased aspartate aminotransferase (AST)		*	*
	Increased alanine aminotransferase (ALT)	*	*	*
	Decreased sorbitol dehydrogenase (SDH)			*
	Increased alkaline phosphatase (ALP)			*
	Decreased bilirubin		*	*
	Increased blood urea nitrogen (BUN)			*

	Decreased cholesterol		*	
	Decreased total protein		*	*
	Decreased globulin		*	*
		Dose (mg/kg-d)		
Effects in females		20	100	500
Urine	Increased volume			✓
	Decreased protein			✓
	Increased fluoride			*
Blood	Increased red blood cells			*
	Decreased hemoglobin			✓
	Decreased hematocrit			✓
	Increased mean corpuscular volume			*
	Increased mean corpuscular hemoglobin			*
	Decreased mean corpuscular hemoglobin concentration			✓
	Increased reticulocyte			*
	Increased platelets			*
	Decreased prothrombin time			*
	Kidney	Increased relative kidney weight		
Decreased creatinine				*
Thyroid	Increased absolute thyroid gland weight	✓		
Liver	Increased absolute and relative liver weight			*
	Increased peroxisomal β -oxidation activity			*
	Decreased bilirubin		*	*

* The authors did not consider this finding to be adverse, biologically meaningful, or related to PFHxA-treatment (see Loveless et al., 2009 for more details).¹¹

From the subchronic study, the researchers identified a NOAEL of 20 mg/kg-d based on nasal lesions observed at higher doses. We applied a total uncertainty factor of 3000 to account for differences between people and research animals (10), differences among people (10), using data from a short-term study to protect from long-term effects (3), and the limited availability of information (10). We obtained a candidate ADI of 0.0067 mg/kg-d.

Reproductive Study

In the reproductive study, PFHxA affected body weight in parents and offspring and caused clinical toxicity in parents (Table 4B). In addition to these statistically significant effects, PFHxA also caused stained skin/fur in males and females at 500 mg/kg-d.

Table 4B. Significant Reproductive Effects Observed in Loveless et al., 2009 ⁽¹¹⁾

Dose (mg/kg-d)

Effects in parental males		20	100	500
Weight	Reduced body weight gain		✓	✓
	Reduced body weight gain in parental males			✓
Effects in parental females				
Weight	Reduced body weight gain during the first week of pregnancy			✓
	Increased body weight gain during lactation			✓
Effects in offspring				
Weight	Reduced mean weight during lactation			✓

From the reproductive study, the researchers identified a parental NOAEL of 20 mg/kg-d based on reduced body weight in males at higher doses. We applied a total uncertainty factor of 1000 to account for differences between people and research animals (10), differences among people (10), and the limited availability of information (10). We obtained a candidate ADI of 0.02 mg/kg-d.

Developmental Study

In the developmental study, PFHxA affected body weight in parental mothers (Table 4C).

Table 4C. Significant Developmental Effects Observed in Loveless et al., 2009⁽¹¹⁾

Effects in mothers		Dose (mg/kg-d)		
		20	100	500
Weight	Reduced total weight gain from GD 6 to 21			✓
	Reduced overall net weight gain			✓

From the developmental study, the researchers identified a parental NOAEL of 100 mg/kg-d based on reduced maternal body weight at higher doses. We applied a total uncertainty factor of 1000 to account for differences between people and research animals (10), differences among people (10), and the limited availability of information (10). We obtained a candidate ADI of 0.1 mg/kg-d.

NTP, 2019

The National Toxicology Program (NTP) exposed male and female rats to different concentrations of PFHxA (0, 62.6, 125, 250, 500, or 1000 mg/kg-d) for 28 days through gavage.²⁵ PFHxA affected thyroid hormone levels in males, altered blood and biochemistry parameters in males and females, and caused liver damage in males and females (Table 5).

Table 5. Statistically Significant Effects Observed in NTP, 2019⁽¹²⁾

Effects in males		Dose (mg/kg-d)				
		62.6	125	250	500	1000
Thyroid	Decreased triiodothyronine (T3)	✓	✓	✓	✓	✓
	Decreased free and total thyroxine (T4)	✓	✓	✓	✓	✓

Blood	Decreased hematocrit and hemoglobin	✓	✓	✓	✓	✓
	Decreased erythrocytes	✓	✓	✓	✓	✓
	Increased mean cell volume			✓	✓	✓
	Increased platelets				✓	✓
	Increased reticulocytes				✓	✓
	Decreased mean cell hemoglobin volume					✓
Biochemistry	Decreased cholesterol	✓	✓	✓	✓	✓
	Increased total protein		✓	✓	✓	✓
	Decreased globulin		✓	✓	✓	✓
	Decreased albumin		✓			✓
	Increased albumin/globulin ratio			✓	✓	✓
	Increased ALT, ALP, and AST				✓	✓
	Increased sorbitol dehydrogenase					✓
	Increased bile salts/acids					✓
Liver	Increased relative liver weight			✓	✓	✓
	Liver damage				✓	✓
Other	Damage to olfactory epithelium			✓	✓	✓
	Spleen damage				✓	✓
	Increased relative kidney weight				✓	✓
	Decreased body weight at necropsy					✓
	Decreased sperm per cauda epididymis					✓
		Doses (mg/kg-d)				
Effects in females		62.6	125	250	500	1000
Blood	Decreased hematocrit and hemoglobin			✓	✓	✓
	Decreased erythrocytes			✓	✓	✓
	Increased reticulocytes				✓	✓
	Increased mean cell volume				✓	✓
Biochemistry	Increased AST and ALT				✓	✓
	Decreased globulin					✓
	Increased albumin/globulin ratio					✓
	Decreased total protein					✓
	Increased ALP					✓
	Increased bile salts/acids					✓
Liver	Increased relative liver weight				✓	✓
	Liver damage					✓
Other	Olfactory epithelium damage			✓	✓	✓
	Increased relative kidney weight					✓

ALT = alanine aminotransferase; AST = aspartate aminotransferase; ALP = alkaline phosphatase

From this study, we identified a LOAEL of 62.6 mg/kg-d based on effects on thyroid hormone levels and blood parameters in males. We applied the a total uncertainty factor of 100,000 to account for differences between people and research animals (10), differences among people (10), using a LOAEL instead of a NOAEL (10), using data from a short-term study to protect from long-term effects (10), and the limited availability of information (10). While we obtained a candidate ADI of 0.0006 mg/kg-d for PFHxA from this study. However, this study was not used to establish the recommended enforcement standard due to significant uncertainty.

Key Health Effects

In our literature review, we did not find any studies indicating that PFHxA can cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture studies.

Discussion

PFHxA has been shown to impact development, alter thyroid hormone levels, affect the blood, and cause liver damage in research animals. While studies on the effects of PFHxA in people are limited, PFHxA has been shown to be associated with decreased testosterone levels in adolescent males, decreased levels of CC16 (a protein involved in protecting the respiratory tract against inflammation), and decreased bilirubin production.⁴⁻⁸ The effects of PFHxA are consistent with those observed with other PFAS compounds, but generally occur at higher doses. These differences may be due to differences in pharmacokinetics. The half-life of PFHxA in rodents is very short.^{22,26,27} Unlike other PFAS, this trend appears to be true for people as well. One study estimated the mean half-life in people to be 32 days and many exposure studies have detected little to no PFHxA in human blood/serum.^{21,28} While scientists are still learning how PFHxA and other PFAS causes toxicity, studies have shown that both chain length and functional group can affect toxic potency.²⁹⁻³⁵

Standard Selection

DHS recommends an enforcement standard of 150 µg/L for PFHxA.

There are no federal numbers and no state drinking water standard for PFHxA. Additionally, the EPA has not evaluated the carcinogenicity or established an ADI (oral reference dose) for PFHxA.

However, we found several studies evaluating the toxicity of PFHxA. To calculate the ADI as specified in Ch. 160.13, Wisc. Statute, DHS selected the 2015 study by Klaunig et al. as the critical study.³ We selected this study because it evaluated long-term effects on PFHxA on male and female animals and used a broad range of doses (2 to 200 mg/kg-d). From

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information

this study, we selected a NOAEL of 15 mg/kg-d because of clinical effects (rales and yellow material on fur) observed in male rats at the highest dose. We applied a total uncertainty factor of 1000 to account for differences between people and research animals (10), differences among people (10), and the limited availability of information (10). To determine the recommended enforcement standard, DHS used the ADI, and, as required by Ch. 160, Wis. Stats., a body weight of 10 kg, a water consumption rate of 1 L/d, and a relative source contribution of 100%.

DHS recommends a preventive action limit of 30 µg/L for PFHxA.

DHS recommends that the preventive action limit for PFHxA be set at 20% of the enforcement standard because PFHxA has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture.

Prepared by Gavin Dehnert, Ph.D. and Sarah Yang, Ph.D.

Wisconsin Department of Health Services

References

1. ATSDR. Toxicological Profile for Perfluoroalkyls - Draft for Public Comment. In: Registry AftSaD, ed. Atlanta, GA2017.
2. Rice PA. C6-Perfluorinated Compounds: The New Greaseproofing Agents in Food Packaging. *Current environmental health reports*. 2015;2(1):33-40.
3. Klaunig JE, Shinohara M, Iwai H, et al. Evaluation of the Chronic Toxicity and Carcinogenicity of Perfluorohexanoic Acid (PFHxA) in Sprague-Dawley Rats. *Toxicologic Pathology*. 2015;43(2):209-220.
4. Fan HM, Ducatman A, Zhang JJ. Perfluorocarbons and Gilbert syndrome (phenotype) in the C8 Health Study Population. *Environmental research*. 2014;135:70-75.
5. Nian M, Li QQ, Bloom M, et al. Liver function biomarkers disorder is associated with exposure to perfluoroalkyl acids in adults: Isomers of C8 Health Project in China. *Environmental research*. 2019;172:81-88.
6. Siebenaler R, Cameron R, Butt CM, Hoffman K, Higgins CP, Stapleton HM. Serum perfluoroalkyl acids (PFAAs) and associations with behavioral attributes. *Chemosphere*. 2017;184:687-693.
7. Zhou Y, Bao WW, Qian ZM, et al. Perfluoroalkyl substance exposure and urine CC16 levels among asthmatics: A case-control study of children. *Environmental research*. 2017;159:158-163.

8. Zhou Y, Hu LW, Qian ZM, et al. Association of perfluoroalkyl substances exposure with reproductive hormone levels in adolescents: By sex status. *Environment international*. 2016;94:189-195.
9. Chengelis CP, Kirkpatrick JB, Radovsky A, Shinohara M. A 90-day repeated dose oral (gavage) toxicity study of perfluorohexanoic acid (PFHxA) in rats (with functional observational battery and motor activity determinations). *Reproductive Toxicology*. 2009;27(3-4):342-351.
10. Iwai H, Hoberman AM, Goodrum PE, Mendelsohn E, Anderson JK. Addendum to Iwai and Hoberman (2014)-Reassessment of Developmental Toxicity of PFHxA in Mice. *International Journal of Toxicology*. 2019;38(3):183-191.
11. Loveless SE, Slezak B, Serex T, et al. Toxicological evaluation of sodium perfluorohexanoate. *Toxicology*. 2009;264(1-2):32-44.
12. NTP. NTP Technical Report on the Toxicity Studies of Perfluoroalkyl Carboxylates (PFHxA, PFOA, PFNA, and PFDA) administered by gavage to sprague-dawley rats. In: Program NT, ed. Vol NTP Tox 972019.
13. USEPA. IRIS Assessments. 2019; https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm.
14. ITRC. Environmental Fate and Transport for Per- and Polyfluoroalkyl Substances. In: Council ITR, ed2018.
15. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 140*2017.
16. USEPA. National Primary Drinking Water Regulations. 2018; <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>.
17. USEPA. Drinking Water Contaminant Human Health Effects Information. 2019; <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>.
18. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 809*2018.
19. IARC. List of Classification, Volumes 1-123. 2018; <https://monographs.iarc.fr/list-of-classifications-volumes/>. Accessed May 17, 2019.
20. USEPA. A Review of the Reference Dose and Reference Concentration Processes. 2002(EPA/630/P-02/002F).
21. Russell MH, Nilsson H, Buck RC. Elimination kinetics of perfluorohexanoic acid in humans and comparison with mouse, rat and monkey. *Chemosphere*. 2013;93(10):2419-2425.
22. Russell MH, Himmelstein MW, Buck RC. Inhalation and oral toxicokinetics of 6:2 FTOH and its metabolites in mammals. *Chemosphere*. 2015;120:328-335.
23. Jain RB, Ducatman A. Perfluoroalkyl acids serum concentrations and their relationship to biomarkers of renal failure: Serum and urine albumin, creatinine, and albumin creatinine ratios across the spectrum of glomerular function among US adults. *Environ Res*. 2019;174:143-151.

24. Iwai H, Hoberman AM. Oral (Gavage) Combined Developmental and Perinatal/Postnatal Reproduction Toxicity Study of Ammonium Salt of Perfluorinated Hexanoic Acid in Mice. *International Journal of Toxicology*. 2014;33(3):219-237.
25. Butenhoff JL, Chang SC, Ehresman DJ, York RG. Evaluation of potential reproductive and developmental toxicity of potassium perfluorohexanesulfonate in Sprague Dawley rats. *Reprod Toxicol*. 2009;27(3-4):331-341.
26. Chengelis CP, Kirkpatrick JB, Myers NR, Shinohara M, Stetson PL, Sved DW. Comparison of the toxicokinetic behavior of perfluorohexanoic acid (PFHxA) and nonafluorobutane-1-sulfonic acid (PFBS) in cynomolgus monkeys and rats. *Reproductive Toxicology*. 2009;27(3-4):400-406.
27. Lwabuchi K, Senzaki N, Mazawa D, et al. Tissue toxicokinetics of perfluoro compounds with single and chronic low doses in male rats. *Journal of Toxicological Sciences*. 2017;42(3):301-317.
28. Nilsson H, Karrman A, Westberg H, Rotander A, van Bavel B, Lindstrom G. A time trend study of significantly elevated perfluorocarboxylate levels in humans after using fluorinated ski wax. *Environmental science & technology*. 2010;44(6):2150-2155.
29. Gomis MI, Vestergren R, Borg D, Cousins IT. Comparing the toxic potency in vivo of long-chain perfluoroalkyl acids and fluorinated alternatives. *Environment international*. 2018;113:1-9.
30. Emerce E, Cetin O. Genotoxicity assessment of perfluoroalkyl substances on human sperm. *Toxicology and Industrial Health*. 2018;34(12):884-890.
31. Eriksen KT, Raaschou-Nielsen O, Sorensen M, Roursgaard M, Loft S, Moller P. Genotoxic potential of the perfluorinated chemicals PFOA, PFOS, PFBS, PFNA and PFHxA in human HepG2 cells. *Mutation Research-Genetic Toxicology and Environmental Mutagenesis*. 2010;700(1-2):39-43.
32. Gorrochategui E, Perez-Albaladejo E, Casas J, Lacorte S, Porte C. Perfluorinated chemicals: Differential toxicity, inhibition of aromatase activity and alteration of cellular lipids in human placental cells. *Toxicology and Applied Pharmacology*. 2014;277(2):124-130.
33. Mulkiewicz E, Jastorff B, Skladanowski AC, Kleszczynski K, Stepnowski P. Evaluation of the acute toxicity of perfluorinated carboxylic acids using eukaryotic cell lines, bacteria and enzymatic assays. *Environmental Toxicology and Pharmacology*. 2007;23(3):279-285.
34. Nobels I, Dardenne F, De Coen W, Blust R. Application of a multiple endpoint bacterial reporter assay to evaluate toxicological relevant endpoints of perfluorinated compounds with different functional groups and varying chain length. *Toxicology in Vitro*. 2010;24(6):1768-1774.
35. Wolf CJ, Takacs ML, Schmid JE, Lau C, Abbott BD. Activation of mouse and human peroxisome proliferator-activated receptor alpha by perfluoroalkyl acids of different functional groups and chain lengths. *Toxicological Sciences*. 2008;106(1):162-171.

Appendix A. Toxicity Data

Table A-I. PFHxA Toxicity Studies from Literature Review

Study Type	Species	Duration	Doses (mg/kg-d)	Form ⁱ	Route	Key Findings	Toxicity Value (mg/kg-d)	Reference
Sub-chronic	Rat	90 d	0, 10, 50, 200	PFHxA	Gavage	Lower body weights (not dose-responsive) in males at 50 and 200 mg/kg-d. Lower red blood cell parameters, higher reticulocyte counts, and lower globulin at 200 mg/kg-d in males and females. Higher liver enzymes in males at 50 and 200 mg/kg-d. Lower total protein and higher albumin to protein ratio, lower cholesterol and calcium levels in males at 200 mg/kg-d. Liver effects (centrilobular hepatocellular hypertrophy, higher liver weights) at 200 mg/kg-d in males.	NOAEL: 50 (males) LOAEL: 200 (males)	Chengelis et al., 2009 ⁽⁹⁾
Re-evaluation	Mouse	GD 6 to 18	Phase 1: 0, 100, 350, 500 Phase 2: 0, 7, 35, 175	NH ₄ PFHx	Gavage	Compared results from 2014 study to pooled and historical controls. Determined that effects on viability and growth were not adverse/significant. In the 2014 study, PFHxA caused orality, excess salivation, and changes in body weight occurred in mothers at 350 and 500 mg/kg-d. It also increased stillbirths, reduction in viability indices, and delays in physical development in offspring at 350 and 500. At 175 mg/kg-d, increased incidence of stillbirths, pup death (PPD1) and reduced pup weights (PPD1) were observed.	NOAEL: 175 LOAEL: not reported	Iwai et al., 2019 ⁽¹⁰⁾
Subchronic	Rat	90 d	0, 20, 100, 500	NaPFHx	Gavage	PFHxA decreased body weight in males and reduced red blood cell counts, hemoglobin, and hematocrit values and increased platelets in males and females at 500 mg/kg-d. PFHxA increased liver weights in males at 100 and 500	NOAEL: 20 LOAEL: 100	Loveless et al., 2009 ⁽¹¹⁾

						mg/kg-d and females at 500 mg/kg-d and increased kidney weights in males and females at 100 and 500 mg/kg-d. PFHxA caused histological changes in the nose, liver, and thyroid gland.		
Reproduction	Rat	126 d ⁱⁱ	0, 20, 100, 500	NaPFHx	Gavage	PFHxA caused stained fur in males and females at 500 mg/kg-d. PFHxA reduced body weight in males at 100 and 500 mg/kg-d. PFHxA also reduced mean maternal body weight gain during the first week of pregnancy and reduced mean pup weight during lactation at 500 mg/kg-d.	Parental NOAEL: 20 LOAEL: 100 Reproduction NOAEL: 100 LOAEL: 500	Loveless et al., 2009 ⁽¹¹⁾
Development	Rat	126 d	0, 20, 100, 500	NaPFHx	Gavage	PFHxA reduced maternal total weight gain from GDs 6 to 21 and overall net weight gain and reduced food consumption at 500 mg/kg-d.	Maternal NOAEL: 100 LOAEL: 500	Loveless et al., 2009 ⁽¹¹⁾
Chronic	Rat	2 years	Males: 0, 2.5, 15, 100 Females: 0, 5, 30, 200	PFHxA	Gavage	Dose-dependent decrease in survival in females. Histological changes in kidneys of females at 200 mg/kg-d.	NOAEL: 100 (males)	Klaunig et al., 2015* (³)
Sub-chronic	Rat	28 d	0, 62.6, 125, 250, 500, 1000	PFHxA	Gavage	In males, PFHxA decreased thyroid hormone, hematocrit, hemoglobin, erythrocytes, and cholesterol levels at all doses and caused liver damage in males at 500 and 1000 mg/kg-d. In females, PFHxA decreased hematocrit, hemoglobin, erythrocytes at levels of 250 mg/kg-d and higher. Other effects occurred at the highest dose. See Table 4 for more details.	LOAEL: 62.6	NTP, 2019 ⁽¹²⁾

i. PFHxA = perfluorohexanoic acid; NaPFHx = Sodium perfluorohexanoate; NH₄PFHx = ammonium salt of perfluorohexanoic acid

ii. Animals were exposed for 70 days prior to cohabitation through gestation and lactation.

* Study was reviewed by ATSDR ⁽¹⁾

Table A-2. Critical Study Selection for PFHxA

Reference	Appropriate duration?	Effects consistent with other studies?	Effects relevant to humans?	Number of Doses	Toxicity value identifiable?	Critical study?
Chengelis et al., 2009	✓	✓	✓	3	✓	Yes
Iwai et al., 2019	✓	✓	✓	6 (over 2 phases)	✓	Yes
Klaunig et al., 2015	✓	✓	✓	3	✓	Yes
Loveless et al., 2009 (subchronic)	✓	✓	✓	3	✓	Yes
Loveless et al., 2009 (reproduction)	✓	✓	✓	3	✓	Yes
Loveless et al., 2009 (development)	✓	✓	✓	3	✓	Yes
NTP, 2019	✓	✓	✓	5	✓	Yes

To be considered a critical study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.

Table A-3. PFHxA Epidemiological Studies from Literature Review

Study Type	Population	Time period	Exposure	Outcomes	Results	Other PFAS Studied	Reference
Cross-sectional	225 Taiwanese 11-15 year olds	2009-2010	Serum PFAS levels	Reproductive hormone concentrations	Among males, PFHxA was negatively associated with ln(testosterone): ($\beta = -0.3095$, 95%CI: $-0.5942, -0.0248$)	PFOS, PFOA, PFBS, PFDA, PFDoA, PFHxA, PFHxS, PFNA, PFTA	Zhou, 2016 ⁽⁸⁾
Cross-sectional	43,996 adults > 18 years from C8 Health Project, mid-Ohio Valley	2005-2006	Serum PFAS levels	Gilbert Syndrome (low bilirubin production)	After full adjustment, serum PFHxA concentrations remained higher in the Gilbert Syndrome phenotype population.	PFHxA, PFHxS, PFOS, PFNA, PFDA, PFPeA, PFHpA, PFOA, PFOA, PFOA, PFDoA	Fan, 2014 ⁽⁴⁾
Case-control	231 Taiwanese 10- to 15-year-olds	2009-2010	Serum PFAS levels	Clara cell secretory protein (CC16, respiratory tract immune protein)	Urinary CC16 was significantly, negatively associated with PFHxA ($\beta = -0.310$, 95% CI: $-0.455, -0.165$) among asthmatic boys. Significant interaction effects between asthma and PFHxA were found	PFOS, PFOA, PFBS, PFDA, PFDoA, PFHxA, PFHxS, PFNA, PFTA	Zhou, 2017 ⁽⁷⁾
<p>Epidemiologic terms: OR=odds ratio; AOR=adjusted odds ratio; RR=relative risk; 95% CI=95% confidence interval; r=Spearman correlation coefficient; β=regression coefficient PFAS acronyms: PFOA=perfluorooctanoic acid, PFNA=perfluorononanoic acid, PFDA=perfluorodecanoic acid, PFUnA= perfluoroundecanoic acid, PFDoA=perfluorododecanoic acid, PFTriA=perfluorotetradecanoic acid, PFHxDA=perfluorohexadecanoic acid, PFHxS=perfluorohexane sulfonate, PFOS=perfluorooctane sulfonic acid, PFDS=perfluorodecanesulfonate, PFBS=perfluorobutane sulfonate, PFBA=perfluorobutanoic acid, PFPeA=perfluoropentanoic acid, PFHxA=perfluorohexanoic acid, PFHpA=perfluoroheptanoic acid, 6:2 FTSA=6:2 fluorotelomer sulfonates, NMeFOSAA=N-methyl perfluorooctanesulfonamidoacetate, EPAH=2-(N-ethyl-perfluorooctane sulfonamido)acetate, MPAH=2-(N-methyl-perfluorooctane sulfonamido) acetate, PFSA=perfluorooctane sulfonamide; PFHpS=perfluoroheptane sulfonic acid; PFDS= perfluorodecane sulfonic acid; EtPFOSAA= 2-(Nethyl-perfluorooctane sulfonamido) acetic acid</p>							

PFHpA | 2020

Substance Overview

Perfluoroheptanoic acid^a (PFHpA) is a chemical in a group of contaminants called per- and polyfluoroalkyl substances (PFAS). Because of its chemical properties, PFHpA can be found as an impurity in stain repellants in commercial products like carpet and fabric and as a coating for packaging, or as an ingredient in fire-fighting foam.^{1,2} PFHpA can persist in the environment for decades.¹

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard or Preventive Action Limit for PFHpA.

DHS did not identify sufficient technical information to recommend an enforcement standard or preventive action limit for PFHpA.

Health Effects

Studies investigating the health effects of PFHpA are limited. Data from *in vitro* studies suggest that the liver or thyroid may be targets of PFHpA toxicity.^{3,4} One epidemiological study has demonstrated a limited association between PFHpA concentrations and the risk of gestational diabetes, but supporting evidence for that association has not yet been observed.⁵

PFHpA has not been shown to cause carcinogenic (cancer), mutagenic (DNA damage), teratogenic (birth defects), or interactive effects in people, research animals, or cell culture.^{1,6} The EPA has not evaluated the carcinogenicity of PFHpA.⁷

Current Standards

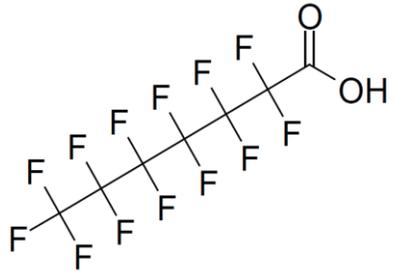
Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

Recommended Standards

Enforcement Standard:	N/A
Preventive Action Limit:	N/A

a This scientific support document also applies to anion salts of perfluoroheptanoic acid.

Chemical Profile

PFHpA	
Structure:	
CAS Number:	375-85-9
Formula:	C ₇ HF ₁₃ O ₂
Molar Mass:	364.06 g/mol
Synonyms:	perfluoro-n-heptanoic acid; tridecafluoro-1-heptanoic acid; heptanoic acid, 2,2,3,3,4,4,5,5,6,6,7,7, 7-tridecafluoro-

Exposure Routes

People can be exposed to PFHpA by drinking contaminated water, swallowing contaminated soil, eating food that was packaged in material that contains PFHpA, and breathing in or swallowing dust that contains PFHpA.^{1,2}

In the environment, PFHpA can be found in water or soil as an impurity from the use of other PFAS in manufacturing and consumer products.^{1,2} PFHpA can move between groundwater and surface water. PFHpA has shown more affinity for surface water than groundwater.² Once in groundwater, PFHpA can travel long distances.²

Current Standard

Wisconsin does not currently have a groundwater standard for PFHpA.⁸

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

EPA Oral Reference Dose:	N/A
--------------------------	-----

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

None available

Literature Search

Literature Search Dates:	1900 - 2019
Total studies evaluated:	Approximately 200
Key studies found?	No
Critical studies identified?	No

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for PFHpA.⁹

Health Advisory

The EPA has not established health advisories for PFHpA.¹⁰

Drinking Water Concentration (Cancer Risk)

The EPA has not established a drinking water concentration based on a cancer risk level determination for PFHpA.⁷

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for PFHpA.¹¹

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Information System (IRIS) program.

EPA Oral Reference Dose

The EPA does not have an oral reference dose for PFHpA.⁷

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If DHS determines that something is carcinogenic and there is no federal number or ADI from the EPA, the standard must be set at a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of PFHpA, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of PFHpA. If so, we looked to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA and International Agency for Research on Cancer (IARC) have not evaluated the carcinogenicity of PFHpA.^{7,12}

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for PFHpA.⁷

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information that was not considered when the value was established and indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For PFHpA, we searched for values that had been published on or before September 2019. While the Agency for Toxic Substances and Disease Registry (ATSDR) reviewed the toxicity of PFHpA in 2018, they did not establish any guidance values for PFHpA.^{1,b}

Literature Search

Our literature review focused on relevant scientific literature on the health effects of PFHpA published on or before September 2019. We looked for studies related to PFHpA toxicity or effects on a disease state in which information on exposure or dose was included as part of the study.^c Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

Twelve toxicity studies were returned by the search engines. We excluded studies on non-mammalian or cell systems, non-oral exposure routes, and those that did not evaluate health risks from further review. After applying these exclusion criteria, we did not locate any key toxicity studies on PFHpA. We did locate four *in vitro* studies evaluating the toxicity of PFHpA in mammalian cell lines (see Table A-1 for details on these studies).

In our search, we also located four epidemiology studies (See Table A-2 for a summary). While multiple potential exposure sources and the ability for other PFAS compounds to cause similar health effects preclude using these data to establish a health-based value, such studies are helpful in identifying the critical effects and ensuring that the animal data used to establish the standard is relevant to people.

b ATSDR stated that they did not identify any acute, intermediate, or chronic oral studies for PFHpA in their literature review.

c We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

Title/abstract: PFHpA or "perfluoroheptanoic acid" or "perfluoroheptanoate"

Subject area: toxicology

Language: English

We also searched online for toxicity studies published by national research programs.

Critical toxicity studies

We did not identify any critical toxicity studies.

Key health effects

We did not find studies that suggest PFHpA has caused carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture.

Standard Selection

DHS did not identify sufficient technical information to recommend an enforcement standard for PFHpA.

The available health information on PFHpA is very limited. There are no federal numbers and no state drinking water standard for PFHpA. Additionally, the EPA has not evaluated the carcinogenicity or established an ADI (oral reference dose) for PFHpA.

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information

While a handful of epidemiological studies have evaluated the risk of PFHpA, the results are inconsistent. For instance, one study found an association between elevated risk of gestational diabetes among women with a family history of type 2 diabetes and plasma PFHpA levels,⁵ but another study investigating the association between diabetes and PFHpA was inconclusive.¹³ Additionally, we did not find any critical studies evaluating the toxicity of PFHpA in research animals. Available information from cell culture studies suggest that the toxic potency of PFHpA is between that of perfluorooctanoic acid (PFOA, 8 carbons) and perfluorohexanoic acid (PFHxA, 6 carbons) (see Table A-2 for more details on these studies). Likewise, limited data suggest that the half-life of PFHpA in people (1.2-1.5 years) is between that of other perfluorocarboxylic acids (2.3 years for PFOA; 1-3 months for PFHxA).¹⁴ However, the major differences of half-life and toxicity between PFHxA and PFOA and the relative difference of PFHpA to both of those chemicals precludes us from combining PFHpA with either of those standards.

Due to limited scientific information available, we have concluded that there is insufficient evidence to establish an enforcement standard for PFHpA at this time.

DHS does not recommend a preventive action limit for PFHpA.

Due to limited scientific information available, we have concluded that there is insufficient evidence to establish a preventive action limit for PFHpA at this time.

Prepared by Nathan Kloczko, MPH and Sarah Yang, Ph.D.

Wisconsin Department of Health Services

References

1. ATSDR. Toxicological Profile for Perfluoroalkyls - Draft for Public Comment. In: Registry AftSaD, ed. Atlanta, GA2017.
2. ITRC. Environmental Fate and Transport for Per- and Polyfluoroalkyl Substances. In: Council ITR, ed2018.
3. Wolf CJ, Schmid JE, Lau C, Abbott BD. Activation of mouse and human peroxisome proliferator-activated receptor-alpha (PPAR alpha) by perfluoroalkyl acids (PFAAs): Further investigation of C4-C12 compounds. *Reproductive Toxicology*. 2012;33(4):546-551.
4. Weiss JM, Andersson PL, Lamoree MH, Leonards PEG, van Leeuwen SPJ, Hamers T. Competitive Binding of Poly- and Perfluorinated Compounds to the Thyroid Hormone Transport Protein Transthyretin. *Toxicological Sciences*. 2009;109(2):206-216.
5. Rahman ML, Zhang C, Smarr MM, et al. Persistent organic pollutants and gestational diabetes: A multi-center prospective cohort study of healthy US women. *Environ Int*. 2019;124:249-258.
6. Buhrke T, Kibellus A, Lampen A. In vitro toxicological characterization of perfluorinated carboxylic acids with different carbon chain lengths. *Toxicol Lett*. 2013;218(2):97-104.
7. USEPA. IRIS Assessments. 2019; https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm.
8. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 140*2017.
9. USEPA. National Primary Drinking Water Regulations. 2018; <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>.
10. USEPA. Drinking Water Contaminant Human Health Effects Information. 2019; <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>.
11. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 809*2018.
12. IARC. List of Classification, Volumes 1-123. 2018; <https://monographs.iarc.fr/list-of-classifications-volumes/>. Accessed May 17, 2019.
13. Lind L, Zethelius B, Salihovic S, van Bavel B, Lind PM. Circulating levels of perfluoroalkyl substances and prevalent diabetes in the elderly. *Diabetologia*. 2014;57(3):473-479.
14. Zhang Y, Beesoon S, Zhu L, Martin JW. Biomonitoring of perfluoroalkyl acids in human urine and estimates of biological half-life. *Environ Sci Technol*. 2013;47(18):10619-10627.

15. Hoyer BB, Bonde JP, Tottenborg SS, et al. Exposure to perfluoroalkyl substances during pregnancy and child behaviour at 5 to 9years of age. *Horm Behav.* 2018;101:105-112.
16. Mattsson K, Rignell-Hydbom A, Holmberg S, et al. Levels of perfluoroalkyl substances and risk of coronary heart disease: Findings from a population-based longitudinal study. *Environ Res.* 2015;142:148-154.
17. Wang B, Zhang R, Jin F, et al. Perfluoroalkyl substances and endometriosis-related infertility in Chinese women. *Environ Int.* 2017;102:207-212.
18. Kleszczynski K, Gardzielewski P, Mulkiewicz E, Stepnowski P, Skladanowski AC. Analysis of structure-cytotoxicity in vitro relationship (SAR) for perfluorinated carboxylic acids. *Toxicology in Vitro.* 2007;21(6):1206-1211.
19. Mulkiewicz E, Jastorff B, Skladanowski AC, Kleszczynski K, Stepnowski P. Evaluation of the acute toxicity of perfluorinated carboxylic acids using eukaryotic cell lines, bacteria and enzymatic assays. *Environmental Toxicology and Pharmacology.* 2007;23(3):279-285.
20. Rand AA, Rooney JP, Butt CM, Meyer JN, Mabury SA. Cellular toxicity associated with exposure to perfluorinated carboxylates (PFCAs) and their metabolic precursors. *Chem Res Toxicol.* 2014;27(1):42-50.

Appendix A. Toxicity Data

Table A-I. PFHpA *In vitro* Toxicity Studies from Literature Review

Species	Cell Line	Duration (hr)	Doses (mM)	Other PFAS evaluated	Key Findings	Reference
Human	HepG2 (liver carcinoma)	72	0.001 – 2	PFBA, PFHxA, PFOA, PFNA, PFDA, PFDoDA	PFCAs increase in toxicity with increasing chain length; PFHpA between PFHxA and PFOA. PFHpA not mutagenic in bacterial tests	Buhrke, 2013 ⁽⁶⁾
Human	HCT116 (colon cancer)	4, 24, 72	0.001 – 5	PFHxA, PFOA, PFNA, PFDA, PFDoDA, PFTeDA, PFHxDA, PFOcDA	PFCAs increase in toxicity with increasing chain length; PFHpA between PFHxA and PFOA	Kleszczynski, 2007 ⁽¹⁸⁾
Rat	IPC-81 (leukemia)	44	0.002 – 20	PFHxA, PFOA, PFNA, PFDA	PFHpA more toxic than PFHxA, less toxic than PFOA, PFNA, PFDA	Mulkiewicz, 2007 ⁽¹⁹⁾
Rat	C6 (brain glial)	44	0.002 – 10	PFHxA, PFOA, PFNA, PFDA	PFHpA more toxic than PFHxA, less toxic than PFOA, PFNA, PFDA	Mulkiewicz, 2007 ⁽¹⁹⁾
Human	THLE-2 (liver epithelial)	24	0.050 – 4	PFBA, PFPeA, PFHxA, PFOA, PFNA, PFDA	PFCAs increase in toxicity with increasing chain length; PFHpA between PFHxA and PFOA	Rand, 2013 ⁽²⁰⁾
N/A	TTR-binding assay	8	0.01 – 10	PFBA, PFHxA, PFOA, PFNA, PFDCa, PFUnA, PFDoA, PFTdA, PFBS, PFHxS, PFOS	PFHpA second highest TTR binding potential of PFCAs after PFOA	Weiss, 2009 ⁽⁴⁾

***In vitro* terms:** TTR = Human thyroid hormone transport protein transthyretin

PFAS acronyms: PFOA=perfluorooctanoic acid, PFNA=perfluorononanoic acid, PFDA=perfluorodecanoic acid, PFUnA= perfluoroundecanoic acid, PFDoA=perfluorododecanoic acid, PFTriA=perfluorotetradecanoic acid, PFHxDA=perfluorohexadecanoic acid, PFHxS=perfluorohexane sulfonate, PFOS=perfluorooctane sulfonic acid, PFDS=perfluorodecanesulfonate, PFBS=perfluorobutane sulfonate, PFBA=perfluorobutanoic acid, PFPeA=perfluoropentanoic acid, PFHxA=perfluorohexanoic acid, PFHpA=perfluoroheptanoic acid, 6:2 FTSA=6:2 fluorotelomer sulfonates, NMeFOSAA=N-methyl perfluorooctanesulfonamidoacetate, EPAH=2-(N-ethyl-perfluorooctane sulfonamido)acetate, MPAH=2-(N-methyl-perfluorooctane sulfonamido) acetate, PFSA=perfluorooctane sulfonamide; PFHpS=perfluoroheptane sulfonic acid; PFDS= perfluorodecane sulfonic acid; EtPFOSAA= 2-(Nethyl-perfluorooctane sulfonamido) acetic acid

Table A-2. PFHpA Epidemiological Studies from Literature Review

Study Type	Population	Time period	Exposure	Outcomes	Results	Other PFAS Evaluated	Reference
Cohort	1,023 Mother/Child Pairs from Greenland and Ukraine	2002-2004	Serum PFAS levels at median 25 weeks pregnant	Child behavior (SDQ-total) and hyperactivity at 5-9 years	No association between perfluoroheptanoic acid (PFHpA) and behavior	PFHxS, PFNA, PFDA	Høyer, 2018 ⁽¹⁵⁾
Case-control	231 male farmers with CHD	1990-2009	Serum PFAS levels	Coronary heart disease (CHD)	There was a significant association between PFHpA and CHD (OR=2.58; 95% CI: 1.39, 4.78) for the 3rd quartile compared to the lowest quartile. This association was attenuated in the 4 th quartile (OR = 1.73; 95% CI: 0.94 – 3.16).	PFOA, PFOS, PFNA, PFDA, PFHxS, PFUnDA, PFDoDA	Mattsson, 2015 ⁽¹⁶⁾
Cohort	2,292 18-40 year-old US women	2009-2013	Plasma PFAS levels	Gestational diabetes	PFHpA was associated with a higher risk of gestational diabetes among women with a family history of type 2 diabetes (RR = 1.22, 95% CI: 1.12 - 1.34) and women with a normal pre-pregnancy BMI (RR = 1.22, 95% CI: 1.11 - 1.33).	N-MeFOSAA, PFDA, PFDoDA, PFDS, PFHxS, PFNA, PFOA, PFOS, PFOSA, PFUnDA	Rahman, 2019 ⁽⁵⁾
Case-control	157 20-45 year-old Chinese Women	2014-2015	Plasma PFAS levels	Endometriosis-related infertility	Plasma concentrations of PFHpA were associated with lower odds of endometriosis-related infertility (OR = 0.48, 95% CI: 0.26 - 0.86). This finding was attenuated in sensitivity analyses.	PFDoA, PFUnA, PFDA, PFNA, PFOSA, PFOS, PFOA, PFHxS, PFBS	Wang, 2017 ⁽¹⁷⁾
<p>Epidemiologic terms: OR=odds ratio; AOR=adjusted odds ratio; RR=relative risk; 95% CI=95% confidence interval; r=Spearman correlation coefficient; β=regression coefficient</p> <p>PFAS acronyms: PFOA=perfluorooctanoic acid, PFNA=perfluorononanoic acid, PFDA=perfluorodecanoic acid, PFUnA= perfluoroundecanoic acid, PFDoA=perfluorododecanoic acid, PFTriA=perfluorotetradecanoic acid, PFHxDA=perfluorohexadecanoic acid, PFHxS=perfluorohexane sulfonate, PFOS=perfluorooctane sulfonic acid, PFDS=perfluorodecanesulfonate, PFBS=perfluorobutane sulfonate, PFBA=perfluorobutanoic acid, PFPeA=perfluoropentanoic acid, PFHxA=perfluorohexanoic acid, PFHpA=perfluoroheptanoic acid, 6:2 FTSA=6:2 fluorotelomer sulfonates, NMeFOSAA=N-methyl perfluorooctanesulfonamidoacetate, EPAH=2-(N-ethyl-perfluorooctane sulfonamido)acetate, MPAH=2-(N-methyl-perfluorooctane sulfonamido) acetate, PFSA=perfluorooctane sulfonamide; PFHpS=perfluoroheptane sulfonic acid; PFDS= perfluorodecane sulfonic acid; EtPFOSAA= 2-(Nethyl-perfluorooctane sulfonamido) acetic acid</p>							

PFNA | 2020

Substance Overview

Perfluorononanoic acid (PFNA) is a chemical in a group of contaminants called per- and polyfluoroalkyl substances (PFAS).¹ PFAS are manmade chemicals that have been used in industry and consumer products since the 1940s. Because of their unique physical and chemical properties, PFAS can be found in a variety of commercial products such as paper and textile coatings, food packaging, surfactants, repellants, and fire-fighting foams.^{1,2} PFAS with long carbon chains, like PFNA, cannot be easily broken down and excreted from the body and they can persist in the environment and in the human body for long periods of time.^{1,2}

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard for PFNA. DHS recommends an enforcement standard of 30 nanograms per liter (ng/L) for PFNA. The recommended standard is based on a study that found that PFNA can cause reproductive effects in research animals.³

DHS recommends that the preventive action limit for PFNA be set at 10% of the enforcement standard because PFNA has been shown to cause interactive effects in research animals and humans.^{4,5}

Current Standards	
Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

Recommended Standards	
Enforcement Standard:	30 ng/L
Preventive Action Limit:	3 ng/L

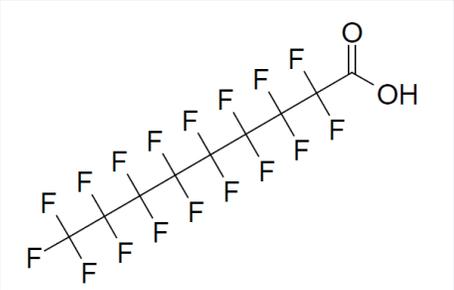
Health Effects

Studies among people exposed to PFAS have shown that high levels of PFNA are associated with increased serum lipids, particularly total cholesterol and low-density lipoprotein (LDL) cholesterol, decreased antibody response to vaccines, and increased incidence of other immunological deficiencies, such as wheezing and asthma.⁵⁻¹⁰ Additional epidemiology studies among people have demonstrated associations between PFNA and delayed physical and hormonal development.¹¹⁻¹⁴ Studies in research animals have found that high levels of PFNA can alter thyroid hormones and impact fertility, and may cause organ damage and impact body weight.^{3,15,16}

We found limited information that PFNA may cause interactive effects in research animals. One study in rats found that co-exposure to PFNA and a mixture of endocrine-disrupting chemicals increased levels of several reproductive hormones over the levels seen in animals exposed to PFNA alone.⁴ PFNA has not

been shown to cause carcinogenic, mutagenic, or teratogenic effects.^a The Environmental Protection Agency (EPA) has not evaluated the carcinogenicity of PFNA.¹⁷

Chemical Profile

PFNA	
Structure:	
CAS Number:	375-95-1
Formula:	C ₉ HF ₁₇ O ₂
Molar Mass:	464.08 g/mol
Synonyms:	PFNA; perfluoro-n-nonanoic acid; perfluorononan-1-oic acid; hepta-decafluoro-nonanoic acid; nonanoic acid, 2,2,3,3,4,4,5,5,6,6,7,7,8,8,9,9,9-hepta-decafluoro-

Exposure Routes

People can be exposed to PFAS, including PFNA, by drinking contaminated water, swallowing contaminated soil, eating fish from contaminated lakes, eating food that was packaged in material that contains PFNA, and breathing in or swallowing dust that contains PFNA.^{1,18,19} Babies born to mothers exposed to PFAS, like PFNA, may be exposed to PFNA during pregnancy and breastfeeding.^{1,20,21}

In the environment, PFNA can be found in water or soil as an impurity from the use of other PFAS in manufacturing and consumer products and fire-fighting foam. PFNA can move between groundwater and surface water. Once in groundwater, PFNA can travel long distances.^{1,22}

Current Standard

Wisconsin does not currently have groundwater standards for PFNA.²³

^a Carcinogenic effects means the substance can cause cancer; mutagenic effects means the substance can cause DNA damage; teratogenic means the substance can cause birth defects; and interactive effects mean the substance can affect the toxicity of another substance or its toxicity can be affected by another substance.

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

EPA Oral Reference Dose:	N/A
--------------------------	-----

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

ATSDR Draft Oral Minimum Risk Level:	3 ng/kg-d	(2018)
--------------------------------------	-----------	--------

Literature Search

Literature Search Dates:	1900 - 2019
Total studies evaluated	Approximately 55
Key studies found?	Yes
Critical studies identified?	Yes

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for PFNA.²⁴

Health Advisory

The EPA has not established health advisories for PFNA.²⁵

Drinking Water Concentration (Cancer Risk)

The EPA has not established a drinking water concentration based on a cancer risk level determination for PFNA.¹⁷

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for PFNA.²⁶

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Information System (IRIS) program.

EPA Oral Reference Dose

The EPA does not have an oral reference dose for PFNA.¹⁷

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If we determine that something is carcinogenic and there is no federal number or ADI from the EPA, DHS must set the standard at a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of PFNA, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of PFNA. If so, we look to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA and International Agency for Research on Cancer (IARC) have not evaluated the carcinogenicity of PFNA.^{17,27}

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for PFNA.¹⁷

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information that was not considered when the value was established and indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For PFNA, we searched for values that had been published on or before October 2019. We found a relevant guidance value from the Agency for Toxic Substances and Disease Registry (ATSDR), which reviewed the toxicity of PFNA in 2018.¹

ATSDR Intermediate Oral Minimum Reference Level

In 2018, the ATSDR established an intermediate oral minimum risk level (MRL) of 3 ng/kg-d for PFNA.¹ The ATSDR evaluated three studies on PFNA toxicity in research animals: Wolf et al. (2010), Rogers et al., (2014), and Das et al., (2015) (see table A-1 for more detail on these studies).^{3,16,28}

ATSDR selected the 2015 study by Das et al. as the critical study. In this study mice were exposed to several concentrations of PFNA (0, 1, 3, 5, and 10 milligrams of PFNA per kilogram body weight per day or mg/kg-d) during pregnancy (gestational days 1 to 17) via gavage. ATSDR identified a no observable adverse effect level (NOAEL) of 1 mg/kg-d based on effects on development at higher doses. Due to differences between rodents and humans, the ATSDR did not consider the observed liver effects to be relevant to humans.^b To account for differences in the half-life of PFNA in people versus research animals, the ATSDR estimated a human equivalent dose from measured serum concentrations in animals using the trapezoid rule.³⁰ To obtain the MRL, they used a human equivalent dose of 0.001 mg/kg-d and applied a total uncertainty factor of 300 to account for the differences between people and research animals (3), differences among people (10), and the limited availability of data (10).

Literature Search

Our literature review focused on relevant scientific literature on the health effects of PFNA published on or before August 2020. We looked for studies related to PFNA toxicity or effects on a disease state in which information on exposure or dose was included as part of the study or studies related to modeling PFNA exposure or dose using pharmacokinetics in animals or humans.^c Ideally, relevant studies used *in*

^b For more details on this determination, see Hall et al., 2012 and section 2.9 of ATSDR's Toxicological Profile.^{1,24}

^c We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

vivo (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

Approximately 55 toxicity studies were returned by the search engines. We excluded studies on non-mammalian or cell systems, non-oral exposure routes, and those that did not evaluate health risks from further review. After applying these exclusion criteria, we located nine key toxicity studies on PFNA (summarized in Table A-1). To be considered a critical toxicity study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.^{31d-e} Three of these studies met the criteria to be considered a critical toxicity study (see Table A-2).

To be considered a critical pharmacokinetics study, the study must model oral exposure in humans or rodents. One of the key studies met the criteria to be considered a critical pharmacokinetic study (the section below has more details on these studies).

In our search, we also located over 90 epidemiology studies in our search (See Table A-3 for the most relevant epidemiology studies not reviewed by the ATSDR). While multiple potential exposure sources and the ability for other PFAS compounds to cause similar health effects preclude using these data to establish a health-based value, such studies are helpful in identifying the crucial effects and ensuring that the animal data used to establish the standard is relevant to people.

Critical Toxicity Studies

To compare between results from recently found studies and the study used to set the current enforcement standard, we calculated an acceptable daily intake (ADI) for each study/effect. The ADI is the estimated amount of PFNA that a person can be exposed to every day and not experience health impacts. As such, we calculated ADI by dividing a toxicity value from either a no-observed adverse effect level (NOAEL), lowest observed adverse effect level (LOAEL), or benchmark dose (BMD) identified in a study by a factor accounting for various sources of scientific uncertainty.^f Uncertainty factors were

Title/abstract: PFNA or “perfluorononanoic acid” or “perfluorononanoate” or 375-95-1
Subject area: toxicology
Language: English

We also searched online for toxicity studies published by national research programs.

d Appropriate toxicity values include the no observable adverse effect level (NOAEL), lowest observable adverse effect level (LOAEL), and benchmark dose (BMD).

e Due to the limited availability of data for this substance, we considered a study to be of an appropriate duration if animals were exposed for at least 28 days or if the exposure occurred during pregnancy. We accounted for differences in exposure duration through the use of an uncertainty factor.

f The NOAEL is the highest dose tested that did not cause an adverse effect, the LOAEL is the lowest dose tested that caused an adverse effect, and the BMD is an estimation of the dose that would cause a specific level of response (typically 5 or 10%).

included, as appropriate, to account for differences between humans and animals, differences between healthy and sensitive human populations, using data from short-term experiments to protect against effects from long-term exposure, and using data where a health effect was observed to estimate the level that does not cause an effect. To ensure appropriate protection, we have chosen to not use studies that have significant uncertainty as the basis for the recommended enforcement standards.^g This approach is consistent with that taken by EPA when establishing oral reference doses.³¹

Das et al., 2015

Das et al. exposed female mice to different concentrations of PFNA (1, 3, 5, or 10 milligrams of PFNA per kilogram body weight per day or mg/kg-d) during pregnancy (gestational days 1-17) through gavage.³ Additional details of this study are described in the *ATSDR Intermediate Oral Minimum Reference Level (Draft)* section above.

We identified a NOAEL of 1 mg/kg-d, as also identified by ATSDR, based on decreased weight gain and developmental delays in the offspring. We obtained a human equivalent dose of 0.0011 mg/kg-d by estimating the time-weight average serum concentration at the NOAEL, as also identified by ATSDR, and applied an uncertainty factor of 300 to account for differences between people and research animals (3), lack of information on sensitive subgroups (10), and the limited availability of information (10) as also completed by ATSDR (see Table A-4). We obtained an ADI of 3 ng/kg-d for PFNA from this study.

Wolf et al., 2010

Wolf et al. exposed female mice to different concentrations of PFNA (0, 0.83, 1.1, 1.5, and 2.0 mg/kg-d) during pregnancy (gestational days 1-18) through gavage.¹⁶ PFNA decreased the number of live pups per litter at 1.1 and 2.0 mg/kg-d and caused decreased weight gain at 2.0 mg/kg-d. PFNA also caused decreased absolute and relative liver weight in offspring at all doses and in mothers at 1.1, 1.5, and 2.0 mg/kg-d. (Table 2).

Table 2. Statistically Significant Effects Observed in Wolf et al., 2010

		Dose (mg/kg-d)			
		0.83	1.1	1.5	2.0
Effects observed in mothers					
Reproduction	Decreased number of live pups per litter		✓		✓
Liver	Decreased absolute and relative liver weight		✓	✓	✓
Effects observed in offspring		0.83	1.1	1.5	2.0
Development	Decreased pups with eyes open				✓
Growth	Decreased weight gain				✓
Liver	Decreased absolute and relative liver weight	✓	✓	✓	✓

^g DHS considers a study to have significant uncertainty if the total uncertainty factors is greater than 3,000.

We identified a NOAEL of 0.83 mg/kg-d based on fewer live pups per litter at 1.1 and 2.0 mg/kg-d. The researchers observed that the dose at 1.5 mg/kg-d did not reach statistical significance, despite survival and the number of live pups at birth were still compromised. The authors confirmed that the dosing had been done correctly, and proposed that this finding was due to natural biological variability. We obtained a human equivalent dose by estimating the time-weight average serum concentration at the NOAEL, and applied an uncertainty factor of 300 to account for differences between people and research animals (3), differences among people (10), and the limited availability of information (10) (see Table A-4). We obtained a candidate ADI of 3 ng/kg-d for PFHxS from this study.

NTP, 2019

The National Toxicology Program (NTP) exposed male and female rats to different concentrations of PFNA (males: 0, 0.625, 1.25, 2.5, 5, 10 mg/kg-d; females: 0, 1.56, 3.12, 6.25, 12.5, 25 mg/kg-d) through gavage for 28 days.¹⁵ PFNA affected thyroid hormones, affected blood and biochemical parameters, and caused liver damage in both males and females (Table 3 and Table 4).

Table 3. Statistically Significant Effects Observed in male rats.¹⁵

Effects observed in males		Dose (mg/kg-d)		
		0.625	1.25	2.5
Blood	Decreased reticulocytes	✓	✓	✓
	Decreased mean cell volume			✓
	Increased mean cell hemoglobin concentration			✓
	Decreased leukocytes			✓
	Decreased lymphocytes			✓
	Bone marrow cell decrease			✓
Biochemistry	Decreased total protein	✓	✓	✓
	Increased sorbitol dehydrogenase		✓	✓
Thyroid	Decreased TSH		✓	✓
	Decreased free & total thyroxine		✓	✓
	Decreased testosterone			✓
	Increased relative thyroid gland weight			✓
Liver	Increased relative liver weight	✓	✓	✓
	Liver damage	✓	✓	✓
	Increased ALT, ALP		✓	
	Increased AST		✓	✓
	Decreased albumin		✓	✓
	Decreased globulin	✓	✓	✓
	Increased albumin/globulin ratio	✓	✓	✓

	Increased total bilirubin	✓	✓
	Increased direct bilirubin	✓	✓
	Increased indirect bilirubin		✓
	Decreased cholesterol	✓	✓
	Decreased triglycerides	✓	✓
	Increased bile salts/acids	✓	✓
	Increased urea nitrogen	✓	✓
	Decreased creatinine		✓
	Decreased glucose		✓
Reproduction	Decreased relative testis weight		✓
	Decreased sperm per cauda epididymis	✓	✓
	Testes damage		✓
Thymus	Decreased relative thymus weight	✓	✓
	Thymus atrophy		✓
Kidney	Increased relative adrenal gland weight		✓
	Increased relative kidney weight	✓	✓
Spleen	Decreased relative spleen weight	✓	✓
Gastrointestinal tract	Stomach damage		✓

ALT = alanine aminotransferase; AST = aspartate aminotransferase; ALP = alkaline phosphatase; TSH = thyroid stimulating hormone

Table 4. Statistically Significant Effects Observed in female rats.¹⁵

Effects observed in females		Dose (mg/kg-d)		
		1.56	3.12	6.25
Biochemistry	Increased urea nitrogen		✓	✓
	Increased total protein	✓	✓	✓
	Increased sorbitol dehydrogenase	✓	✓	✓
Thyroid	Decreased free & total thyroxine		✓	✓
Liver	Increased relative liver weight	✓	✓	✓
	Liver damage	✓	✓	✓
	Increased albumin	✓	✓	✓
	Decreased globulin	✓	✓	✓
	Increased albumin/globulin ratio	✓	✓	✓
	Increased direct bilirubin			✓
	Increased ALT & ALP		✓	✓
	Increased bile salts/acids		✓	✓
Spleen	Decreased relative spleen weight			✓

Kidney	Increased relative kidney weight	✓	✓	✓
Hormone	Increased testosterone	✓	✓	✓

ALT = alanine aminotransferase; AST = aspartate aminotransferase; ALP = alkaline phosphatase; TSH = thyroid stimulating hormone

From this study, we identified a LOAEL of 0.625 mg/kg-d based on changes to several biochemistry parameters including increased urea nitrogen, decreased total protein and increased relative kidney weight in males. We applied an uncertainty factor of 100,000 to account for differences between people and research animals (10), differences among people (10), using a LOAEL instead of a NOAEL (10), using data from a short-term study to protect from long-term effects (10), and the limited availability of information (10). While we obtained a candidate ADI of 0.00625 ng/kg-d for PFHxS from this study, this study was not used to establish to recommended enforcement standard due to significant uncertainty.

Critical Pharmacokinetic Studies

A physiologically-based pharmacokinetic (PBPK) model can be used to relate the amount of a chemical exposure to the amount of a chemical found in different organs at different time points.³² PBPK models are developed using mathematical values and equations that describe characteristics and processes of the body, such as body weight, blood flow rate, and metabolism rate. The PBPK model uses a NOAEL and known animal serum levels to estimate a human point of departure (POD_{Human}). POD_{Human} is defined as the point on a toxicological dose-response curve established from experimental data that corresponds to an estimated no effect level. The POD_{Human} and PBPK models can be used to better understand what animal toxicity data means for human health and PBPK models provide a critical link between chemical toxicity and exposure information, as well as an important tool for using animal experiments to inform evaluations of the health effects of chemicals on humans.^{32,33}

Kim et al., 2019

In 2019, Kim et al., published a study in which they developed a physiology-based pharmacokinetic (PBPK) model to estimate PFNA serum levels in male and female rats from exposure to the NOAEL used in Fang et al.^{32,34}

The researchers obtained a point of departure for humans (POD_{Human}) of 8.81 µg/kg-d for males and 8.89 µg/kg-d for females (Table 5).³² We used a uncertainty factor of 1000 to account for differences among people (10), using data from a short-term study to protect from long-term effects (10), and the limited availability of data (10).³² We established candidate ADIs of 8 ng/kg-d for males and 9 ng/kg-d for females.

Table 5. Toxicity parameters and ADIs from Kim et al. 2019.

Sex	NOAEL (mg/kg-d)	Endpoint	POD _{Human} (µg/kg-d)	Total Uncertainty Factor	ADI (ng/kg-d)
Female	1	Liver toxicity*	8.89	1000**	9
Male	1	Liver toxicity*	8.81	1000**	8

* Toxicity endpoint from Fang et al.
** DHS selected a total uncertainty factor of 1000 to account for differences among people (10), use of a shorter duration study to protect against effects from long-term exposure (10), and the limited availability of data (10).

Key health effects

At this time, we did not find studies to show that PFNA has caused carcinogenic, mutagenic, or teratogenic effects in humans, research animals, or culture cells. We found limited information that PFNA may cause interactive effects in research animals.⁴

One study exposed male rats to different concentrations of PFNA (0, 0.0125, 0.25, 5 mg/kg-d), to a mixture of endocrine-disrupting chemicals^h (2.5 mg/kg-d), and to the combination of PFNA and the mixture of chemicals (0.0125, 0.25, 5 mg/kg-d PFNA; 2.5 mg/kg-d mixture at all PFNA levels) via gavage for 14 days.⁴ PFNA and the mixture of chemicals increased a variety of hormone levels in a dose-independent fashion as compared to PFNA alone or the mixture of chemicals alone. Specifically, PFNA and the mixture of chemicals increased levels of androstenedione, testosterone, and dihydrotestosterone (reproductive hormones) over the levels seen in PFNA alone. This was supported at the molecular level as PFNA and the mixture of chemicals increased the levels of proteins and enzymes related to these reproductive hormones as compared to PFNA alone. Additionally, the mixture caused a 2.8-fold increase in serum PFNA concentrations at the lowest dose, suggesting that interaction with different chemicals may increase PFNA absorption into the body.⁴

Discussion

In research animals, PFNA has been shown to impact litter size and pup survival, weight gain, and suppress immunologic impacts in people and research animals.¹ In our additional review, we also found that PFNA can cause liver damage, modify a variety of circulating biochemical factors, decrease thyroid hormones, and cause damage to other organs in people and research animals.^{5,35-37}

Studies in research animals and people have shown that PFNA, as well as other PFAS, can affect the levels of thyroid hormones.¹ Thyroid hormones are crucial for development, energy balance, and metabolism in all species.³⁸ In people, thyroid hormones play an important role in the development of the brain, lungs, and heart.³⁸ Scientists have learned that certain PFAS, including PFNA, can bind to

^h This mixtures including pesticides, plasticizers, preservatives, sun filters, and licorice and grapefruit constituents.

transport proteins involved in moving thyroid hormones throughout the body.³⁹ Scientists are still learning how this effect occurs and its impact on health.

Other studies in humans have demonstrated associations between elevated PFNA blood levels and a variety of immunologic responses, such as increased risk of upper and lower respiratory tract infections and atopic dermatitis.^{1,9,20,40} An inverse association between PFNA concentrations and anti-rubella antibodies has also been demonstrated.⁹ Epidemiological studies have also demonstrated associations between high maternal PFNA concentrations and modified physical and mental developmental outcomes, though these associations have been mixed.^{12-14,41-45} Many of these outcomes are consistent with other effects seen in humans for other long-chain PFAS, such as PFOA.¹

A number of studies have demonstrated that liver effects caused by PFNA, as well as other PFAS, occur primarily through activation of the nuclear hormone receptor, PPAR α (peroxisome proliferator-activated receptor alpha).^{46,47} Nuclear receptors regulate gene expression and PPAR α regulates the expression of genes involved in lipid and cholesterol metabolism.^{46,47} While PPAR α receptors are found in rodents and humans, levels of these receptors are much higher in rats and mice than in monkeys and humans.²⁹ Additionally, PFNA specifically was not shown to activate human PPAR α .⁴⁶ This suggests that effects caused by activation of PPAR α are more likely to occur in rodents than they are to occur in people. As such, the effects on the liver observed in the critical studies reviewed here are likely not relevant to humans. This conclusion is supported by the lack of associations between PFAS exposure and liver disease in epidemiological studies (Table A-3).

Standard Selection

DHS recommends an enforcement standard of 30 ng/L for PFNA.

There are no federal numbers and no state drinking water standard for PFNA. Additionally, the EPA has not evaluated the carcinogenicity or established an ADI (oral reference dose) for PFNA. However, we found several critical toxicity and pharmacokinetic modeling studies evaluating the toxicity of PFNA.^{3,15,16,32}

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information

As such, DHS recommends using available scientific information to establish an ADI through the procedures specified in Ch. 160, Wis. Stats. To establish the ADI, we used the 2015 study by Das et al. as the critical study and effects on delayed reproductive development and offspring birth weight as the critical effects. Toxicity studies in animals continue to show that developmental endpoints are critical effects for PFAS, including PFNA, with effects occurring in offspring after exposure during pregnancy and lactation. Additionally, epidemiology studies continue to associate maternal PFNA exposure to

developmental problems in their babies, as PFNA has been shown to cross the placenta during pregnancy.^{1,20,21}

We obtained an ADI of 3 ng/kg-d from a human equivalent dose of 1000 ng/kg-d and a total uncertainty factor of 300. To determine the recommended enforcement standard, DHS used the ADI, and, as required by Ch. 160, Wis. Stats., a body weight of 10 kg, a water consumption rate of 1 L/d, and a relative source contribution of 100%.

DHS recommends a preventive action limit of 3 ng/L for PFNA.

DHS recommends that the preventive action limit for PFNA be set at 10% of the enforcement standard because PFNA has been shown to cause interactive effects in research animals and people.^{4,5} At this time, PFHxS has not been shown to cause carcinogenic, mutagenic, or teratogenic effects in people, research animals, or cell culture studies.

Prepared by Gavin Dehnert, Ph.D., and Sarah Yang, Ph.D.

Wisconsin Department of Health Services

References

1. ATSDR. Toxicological Profile for Perfluoroalkyls - Draft for Public Comment. In: Registry AFTSaD, ed. Atlanta, GA2017.
2. (ITRC) ITRC. History and Use of Per- and Polyfluoroalkyl Substances (PFAS). https://pfas-1.itrcweb.org/wp-content/uploads/2017/11/pfas_fact_sheet_history_and_use_11_13_17.pdf. Published 2017. Accessed Oct 2019.
3. Das KP, Grey BE, Rosen MB, et al. Developmental toxicity of perfluorononanoic acid in mice. *Reprod Toxicol*. 2015;51:133-144.
4. Hadrup N, Pedersen M, Skov K, et al. Perfluorononanoic acid in combination with 14 chemicals exerts low-dose mixture effects in rats. *Arch Toxicol*. 2016;90(3):661-675.
5. Zhou Y, Hu LW, Qian ZM, et al. Interaction effects of polyfluoroalkyl substances and sex steroid hormones on asthma among children. *Sci Rep*. 2017;7(1):899.
6. Khalil N, Ebert JR, Honda M, et al. Perfluoroalkyl substances, bone density, and cardio-metabolic risk factors in obese 8-12 year old children: A pilot study. *Environ Res*. 2018;160:314-321.

7. Lin PD, Cardenas A, Hauser R, et al. Per- and polyfluoroalkyl substances and blood lipid levels in pre-diabetic adults-longitudinal analysis of the diabetes prevention program outcomes study. *Environ Int.* 2019;129:343-353.
8. Salihovic S, Stubleski J, Karrman A, et al. Changes in markers of liver function in relation to changes in perfluoroalkyl substances - A longitudinal study. *Environ Int.* 2018;117:196-203.
9. Granum B, Haug LS, Namork E, et al. Pre-natal exposure to perfluoroalkyl substances may be associated with altered vaccine antibody levels and immune-related health outcomes in early childhood. *J Immunotoxicol.* 2013;10(4):373-379.
10. Buser MC, Scinicariello F. Perfluoroalkyl substances and food allergies in adolescents. *Environ Int.* 2016;88:74-79.
11. Ernst A, Brix N, Lauridsen LLB, et al. Exposure to Perfluoroalkyl Substances during Fetal Life and Pubertal Development in Boys and Girls from the Danish National Birth Cohort. *Environ Health Perspect.* 2019;127(1):17004.
12. Lind DV, Priskorn L, Lassen TH, et al. Prenatal exposure to perfluoroalkyl substances and anogenital distance at 3 months of age in a Danish mother-child cohort. *Reprod Toxicol.* 2017;68:200-206.
13. Meng Q, Inoue K, Ritz B, Olsen J, Liew Z. Prenatal Exposure to Perfluoroalkyl Substances and Birth Outcomes; An Updated Analysis from the Danish National Birth Cohort. *Int J Environ Res Public Health.* 2018;15(9).
14. Starling AP, Adgate JL, Hamman RF, et al. Perfluoroalkyl Substances during Pregnancy and Offspring Weight and Adiposity at Birth: Examining Mediation by Maternal Fasting Glucose in the Healthy Start Study. *Environ Health Perspect.* 2017;125(6):067016.
15. (NTP) NTP. NTP technical report on the toxicity studies of perfluoroalkyl carboxylates (perfluorohexanoic acid, perfluorooctanoic acid, perfluorononanoic acid, and perfluorodecanoic acid) administered by gavage to Sprague Dawley (Hsd:Sprague Dawley SD) rats. In. Vol Toxicity Report 97. Research Triangle Park, NC: National Toxicology Program 2019.
16. Wolf CJ, Zehr RD, Schmid JE, Lau C, Abbott BD. Developmental effects of perfluorononanoic Acid in the mouse are dependent on peroxisome proliferator-activated receptor-alpha. *PPAR Res.* 2010;2010.
17. USEPA. IRIS Assessments. https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm. Published 2019. Accessed.
18. Christensen KY, Raymond M, Blackowicz M, et al. Perfluoroalkyl substances and fish consumption. *Environ Res.* 2017;154:145-151.
19. Christensen KY, Raymond M, Thompson BA, Anderson HA. Perfluoroalkyl substances in older male anglers in Wisconsin. *Environ Int.* 2016;91:312-318.

20. Chen Q, Huang R, Hua L, et al. Prenatal exposure to perfluoroalkyl and polyfluoroalkyl substances and childhood atopic dermatitis: a prospective birth cohort study. *Environ Health*. 2018;17(1):8.
21. Preston EV, Webster TF, Oken E, et al. Maternal Plasma per- and Polyfluoroalkyl Substance Concentrations in Early Pregnancy and Maternal and Neonatal Thyroid Function in a Prospective Birth Cohort: Project Viva (USA). *Environ Health Perspect*. 2018;126(2):027013.
22. ITRC. Environmental Fate and Transport for Per- and Polyfluoroalkyl Substances. In: Council ITR, ed2018.
23. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 140*2017.
24. USEPA. National Primary Drinking Water Regulations. <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>. Published 2018. Accessed.
25. USEPA. Drinking Water Contaminant Human Health Effects Information. <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>. Published 2019. Accessed.
26. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 809*2018.
27. IARC. List of Classification, Volumes 1-123. <https://monographs.iarc.fr/list-of-classifications-volumes/>. Published 2018. Accessed May 17, 2019.
28. Rogers JM, Ellis-Hutchings RG, Grey BE, et al. Elevated Blood Pressure in Offspring of Rats Exposed to Diverse Chemicals During Pregnancy. *Toxicological Sciences*. 2013;137(2):436-446.
29. Hall AP, Elcombe CR, Foster JR, et al. Liver hypertrophy: a review of adaptive (adverse and non-adverse) changes--conclusions from the 3rd International ESTP Expert Workshop. *Toxicologic pathology*. 2012;40(7):971-994.
30. ATSDR. Calculation of AUC and TWA serum concentrations of perfluoroalkyls from serum concentration data using the trapezoid rule. In: DHS, ed2019.
31. USEPA. A Review of the Reference Dose and Reference Concentration Processes. 2002(EPA/630/P-02/002F).
32. Kim SJ, Choi EJ, Choi GW, Lee YB, Cho HY. Exploring sex differences in human health risk assessment for PFNA and PFDA using a PBPK model. *Arch Toxicol*. 2019;93(2):311-330.
33. USEPA. Physiologically-based pharmacokinetic (PBPK) models: Scientific models to help evaluate health effects of chemicals. 2018.
34. Fang X, Gao G, Xue H, Zhang X, Wang H. Exposure of perfluorononanoic acid suppresses the hepatic insulin signal pathway and increases serum glucose in rats. *Toxicology*. 2012;294(2-3):109-115.
35. Zhang Y, Zhang Y, Klaassen CD, Cheng X. Alteration of Bile Acid and Cholesterol Biosynthesis and Transport by Perfluorononanoic Acid (PFNA) in Mice. *Toxicol Sci*. 2018;162(1):225-233.

36. Zhou W, Zhang L, Tong C, et al. Plasma Perfluoroalkyl and Polyfluoroalkyl Substances Concentration and Menstrual Cycle Characteristics in Preconception Women. *Environ Health Perspect.* 2017;125(6):067012.
37. Singh S, Singh SK. Prepubertal exposure to perfluorononanoic acid interferes with spermatogenesis and steroidogenesis in male mice. *Ecotoxicol Environ Saf.* 2019;170:590-599.
38. Andersen LG, Holst C, Michaelsen KF, Baker JL, Sorensen TI. Weight and weight gain during early infancy predict childhood obesity: a case-cohort study. *International journal of obesity (2005).* 2012;36(10):1306-1311.
39. Sharma D, Shastri S, Sharma P. Intrauterine Growth Restriction: Antenatal and Postnatal Aspects. *Clinical medicine insights Pediatrics.* 2016;10:67-83.
40. Impinen A, Nygaard UC, Lodrup Carlsen KC, et al. Prenatal exposure to perfluoroalkyl substances (PFASs) associated with respiratory tract infections but not allergy- and asthma-related health outcomes in childhood. *Environ Res.* 2018;160:518-523.
41. Starling AP, Adgate JL, Hamman RF, Kechris K, Calafat AM, Dabelea D. Prenatal exposure to per- and polyfluoroalkyl substances and infant growth and adiposity: the Healthy Start Study. *Environ Int.* 2019;131:104983.
42. Wang Y, Adgent M, Su PH, et al. Prenatal Exposure to Perfluorocarboxylic Acids (PFCAs) and Fetal and Postnatal Growth in the Taiwan Maternal and Infant Cohort Study. *Environ Health Perspect.* 2016;124(11):1794-1800.
43. Vuong AM, Braun JM, Yolton K, et al. Prenatal and childhood exposure to perfluoroalkyl substances (PFAS) and measures of attention, impulse control, and visual spatial abilities. *Environ Int.* 2018;119:413-420.
44. Vuong AM, Yolton K, Xie C, et al. Prenatal and childhood exposure to poly- and perfluoroalkyl substances (PFAS) and cognitive development in children at age 8 years. *Environ Res.* 2019;172:242-248.
45. Zhang H, Yolton K, Webster GM, et al. Prenatal and childhood perfluoroalkyl substances exposures and children's reading skills at ages 5 and 8 years. *Environ Int.* 2018;111:224-231.
46. Wolf CJ, Schmid JE, Lau C, Abbott BD. Activation of mouse and human peroxisome proliferator-activated receptor-alpha (PPARalpha) by perfluoroalkyl acids (PFAAs): further investigation of C4-C12 compounds. *Reprod Toxicol.* 2012;33(4):546-551.
47. Cheng X, Klaassen CD. Critical role of PPAR-alpha in perfluorooctanoic acid- and perfluorodecanoic acid-induced downregulation of Oatp uptake transporters in mouse livers. *Toxicol Sci.* 2008;106(1):37-45.
48. Fang X, Zhang L, Feng Y, Zhao Y, Dai J. Immunotoxic effects of perfluorononanoic acid on BALB/c mice. *Toxicol Sci.* 2008;105(2):312-321.

49. Fang X, Gao G, Zhang X, Wang H. Perfluorononanoic acid disturbed the metabolism of lipid in the liver of streptozotocin-induced diabetic rats. *Toxicol Mech Methods*. 2015;25(8):622-627.
50. Feng Y, Shi Z, Fang X, Xu M, Dai J. Perfluorononanoic acid induces apoptosis involving the Fas death receptor signaling pathway in rat testis. *Toxicol Lett*. 2009;190(2):224-230.
51. Olsen GW, Burris JM, Ehresman DJ, et al. Half-life of serum elimination of perfluorooctanesulfonate, perfluorohexanesulfonate, and perfluorooctanoate in retired fluorochemical production workers. *Environmental health perspectives*. 2007;115(9):1298-1305.
52. Sundstrom M, Chang SC, Noker PE, et al. Comparative pharmacokinetics of perfluorohexanesulfonate (PFHxS) in rats, mice, and monkeys. *Reprod Toxicol*. 2012;33(4):441-451.
53. Kim SJ, Heo SH, Lee DS, Hwang IG, Lee YB, Cho HY. Gender differences in pharmacokinetics and tissue distribution of 3 perfluoroalkyl and polyfluoroalkyl substances in rats. *Food and chemical toxicology : an international journal published for the British Industrial Biological Research Association*. 2016;97:243-255.

Appendix A. Toxicity Data

Table A-I. PFNA Toxicity Studies from Literature Review

Study Type	Species	Duration	Doses (mg/kg-d)	Route	Significant Endpoints	Toxicity Value (mg/kg-d)	Reference
Reproduction	Mouse	GD 1-17	0, 1, 3, 5, 10	Gavage	PFNA decreased body weight gain, delayed eye opening, preputial separation, and vaginal opening at 3 mg/kg-d. PFNA decreased postnatal survival starting at 5 mg/kg-d.	NOAEL: 1 LOAEL: 3	Das, 2015 (3)*
Acute	Mouse	14 d	0, 1, 3, 5	Gavage	PFNA decreased body weight and decreased absolute and relative thymus weight lower starting at 3 mg/kg-d. PFNA increased cell death in the thymus at 5 mg/kg-d.	NOAEL: 1 LOAEL: 3	Fang, 2008 (48)
Acute	Rat	14 d	0, 0.2, 1, 5	Gavage	PFNA decreased HDL starting at 0.2mg/kg-d, increased glucose concentrations at 1 mg/kg-d, and altered protein expression of genes related to glucose metabolism at 5 mg/kg-d.	NOAEL: N/A LOAEL: 0.2	Fang, 2012 (34)
Acute	Rat	7 d	0, 0.2, 1, 5	Gavage	PFNA increased ALT levels in the liver and increased concentrations of total cholesterol starting at 1 mg/kg-d and increased liver lipid concentrations at 5 mg/kg-d.	NOAEL: 0.2 LOAEL: 1	Fang, 2015 (49)
Acute	Rat	14 d	0, 1, 3, 5	Gavage	PFNA increased cell death in male testis starting at 3 mg/kg-d.	NOAEL: 1 LOAEL: 3	Feng, 2009 (50)
Interactive	Rat	14 d	0, 0.0125, 0.25, 5	Gavage	PFNA mixed with multiple hormones increased kidney transport protein expression levels starting at 0.0125 mg/kg-d.	NOAEL: n/a LOAEL: 0.0125	Hadrup, 2016 ⁽⁴⁾
Sub-chronic	Rat	28 d	Males: 0, 0.625, 1.25, 2.5, 5, 10; Females: 0, 1.56, 3.12, 6.25, 12.5, 25	Gavage	PFNA decreased thyroid levels starting at 1.25mg/kg-d in males and 3.12 mg/kg-d for females. PFNA impacted reproductive endpoints starting at 1.25 mg/kg-d in males.	NOAEL: n/a LOAEL: 0.625	NTP, 2019 (15)
Reproduction	Rat	GD 1-20	0, 5	Gavage	PFNA decreased birth weight, increased blood pressure at 10 weeks, and increased kidney damage at 5 mg/kg-d.	NOAEL: n/a LOAEL: 5	Rogers, 2014 ^{(28)*}
Acute	Mouse	14 d	0, 2, 5	Gavage	PFNA decreased testosterone and modified testicular protein expression starting at 2 mg/kg-d and decreased weight gain at 5 mg/kg-d.	NOAEL: n/a LOAEL: 2	Singh, 2019 (37)
Reproduction	Mouse	GD 1-18	0, 0.83, 1.1, 1.5, 2	Gavage	PFNA decreased litter size and pup survival, and pup weight gain starting at 1.1 mg/kg-d.	NOAEL: 0.83	Wolf, 2010 (16)*

* = Reviewed by ATSDR

Table A-2. Critical Study Selection for PFNA

Reference	Appropriate duration?	Effects consistent with other studies?	Effects relevant to humans?	Number of Doses	Toxicity value identifiable?	Critical study?
Das, 2015*	✓	✓	✓	4	✓	Yes
Fang, 2008	⊘	✓	✓	3	✓	No
Fang, 2012	⊘	✓	✓	3	✓	No
Fang, 2015	⊘	✓	✓	3	✓	No
Feng, 2009	⊘	✓	✓	3	✓	No
Hadrup, 2016	⊘	✓	✓	3	⊘	No
NTP, 2019	✓	✓	✓	5	✓	Yes
Rogers, 2014*	✓	✓	✓	1	✓	No
Singh, 2019	⊘	✓	✓	2	✓	No
Wolf, 2010*	✓	✓	✓	4	✓	Yes

To be considered a critical study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.

* = Reviewed by ATSDR

Table A-3. Recent PFNA Epidemiological Studies from Literature Review

Study Type	Population	Time period	Exposure	Outcomes	Results	Other PFAS Studied	Reference
Nested Cohort	445 adolescents from the Danish Puberty Cohort	2012-2017	Maternal plasma PFAS concentrations	Multiple puberty indicators	PFNA was associated with earlier age of puberty onset in girls (8.78 months earlier, 95% CI: 16.55, 1.02), and higher age of puberty onset in boys (4.45 months, 95% CI: -1.3 to 10.21).	PFOS, PFOA, PFHxS, PFHpS, PFDA	Ernst et al., 2019 ⁽¹¹⁾
Nested Cohort	99 Norwegian pregnant women & children	2007-2008	Maternal plasma PFAS concentrations	Anti-vaccine antibody levels, common infectious disease, allergy-related outcomes	Maternal PFNA concentrations were associated with a higher number of episodes of common cold in the child's first three years of birth, and were associated with lower anti-vaccine antibodies against Rubella	PFOS, PFOA, PFHxS	Granum et al., 2013 ⁽⁹⁾
Cross-sectional	48 8-12y obese children from Ohio	2016	Serum PFAS concentrations	Bone health, blood lipids	PFNA levels were associated with a decreased bone health, raised blood pressure, raised LDL, and raised total cholesterol	PFOA, PFOS, PFHxS	Khalil et al., 2018 ⁽⁶⁾
Cohort	888 prediabetic adults	1996-2014	Plasma PFAS concentrations	Blood lipids	PFNA levels were significantly associated with higher total cholesterol and LDL. Likewise, the prospective risk of hypercholesterolemia and hypertriglyceridemia was significantly elevated for higher PFNA levels	PFOS, PFOA, PFHxS, EtFOSAA, MeFOSAA	Lin et al., 2019 ⁽⁷⁾
Cohort	638 Danish children from Odense Child Cohort	2010-2014	Maternal serum PFAS concentrations	Anogenital distance in children	PFNA was associated with a decreased anogenital distance in girls.	PFOS, PFOA, PFHxS, PFDA	Lind et al., 2017 ⁽¹²⁾
Cohort	2137 mother-infant pairs from Danish National Birth Cohort	1996-2002	Maternal plasma PFAS concentrations	Low birth weight, preterm birth	Maternal PFNA exposure was associated with lower birth weight (-36.3 g per doubling of PFNA, 95% CI: -70.6, -2) and elevated risk of preterm birth (-1.0 days per doubling, 95% CI: -1.7, -0.3).	PFOA, PFOS, PFHpS, PFDA	Meng et al., 2018 ⁽¹³⁾

Cohort	1002 Swedish adults > 70y	2001-2014	Plasma PFAS concentrations	Liver function	Higher PFNA levels were associated with lower circulating bilirubin and higher levels of ALT, ALP, and GGT, indicating impact on liver function.	PFHpA, PFOA, PFDA, PFUnA, PFHxS, PFOS, PFOSA	Salihovic et al., 2018 ⁽⁸⁾
Cohort	628 mother-infant pairs from Healthy Start Cohort	2009-2014	Maternal serum PFAS concentrations	Child weight and adiposity at birth	The highest percentile concentrations of PFNA were associated with reduced birth weight (-92.1g, 95% CI: -150.6, -33.6) and reduced offspring adiposity (-0.85%, 95% CI: -1.46, -0.24), as compared to the lowest exposures.	PFOA, PFOS, PFDA, PFHxS	Starling et al., 2017 ⁽¹⁴⁾
Nested Case-Control	231 asthmatic Taiwanese adolescents (225 control)	2009-2010	Serum PFAS concentrations	Reproductive hormone levels and associated asthma	Among those with and without asthma, PFNA levels were associated with higher estradiol levels. PFNA was also shown to interact with estradiol levels and asthma differentially by sex (e.g. OR for asthma among girls with low estradiol = 0.82; OR for asthma among girls with high estradiol = 4.16.	PFHxS, PFOS, PFOA, PFDA	Zhou et al., 2017a ⁽⁵⁾
Cross-sectional	950 Chinese women of childbearing age	2013-2015	Plasma PFAS concentrations	Menstrual cycle characteristics	A log-unit increase in PFNA increased odds of irregular menstrual cycle by 50% (95% CI: 1.03,2.07), and a higher PFNA concentration increased odds of long menstrual cycle (OR = 1.49, 95% CI = 1.05, 2.11)	PFOA, PFOS, PFHxS, PFDA, PFUnA, PFBS, PFDoA, PFHpA, PFOSA	Zhou et al., 2017b ⁽³⁶⁾

Epidemiologic terms: OR=odds ratio; AOR=adjusted odds ratio; RR=relative risk; 95% CI=95% confidence interval; r=Spearman correlation coefficient; β=regression coefficient

PFAS acronyms: PFOA=perfluorooctanoic acid, PFNA=perfluorononanoic acid, PFDA=perfluorodecanoic acid, PFUnA= perfluoroundecanoic acid, PFDoA=perfluorododecanoic acid, PFTriA=perfluorotetradecanoic acid, PFHxDA=perfluorohexadecanoic acid, PFHxS=perfluorohexane sulfonate, PFOS=perfluorooctane sulfonic acid, PFDS=perfluorodecanesulfonate, PFBS=perfluorobutane sulfonate, PFBA=perfluorobutanoic acid, PFPeA=perfluoropentanoic acid, PFHxA=perfluorohexanoic acid, PFHpA=perfluoroheptanoic acid, 6:2 FTSA=6:2 fluorotelomer sulfonates, NMeFOSAA=N-methyl perfluorooctanesulfonamidoacetate, EPAH=2-(N-ethyl-perfluorooctane sulfonamido)acetate, MPAH=2-(N-methyl-perfluorooctane sulfonamido) acetate, PFSA=perfluorooctane sulfonamide; PFHpS=perfluoroheptane sulfonic acid; PFDS= perfluorodecane sulfonic acid; EtPFOSAA= 2-(Nethyl-perfluorooctane sulfonamido) acetic acid

Table A-4. PFNA Estimated Acceptable Daily Intakes (ADIs) for the Identified Critical Toxicity Studies

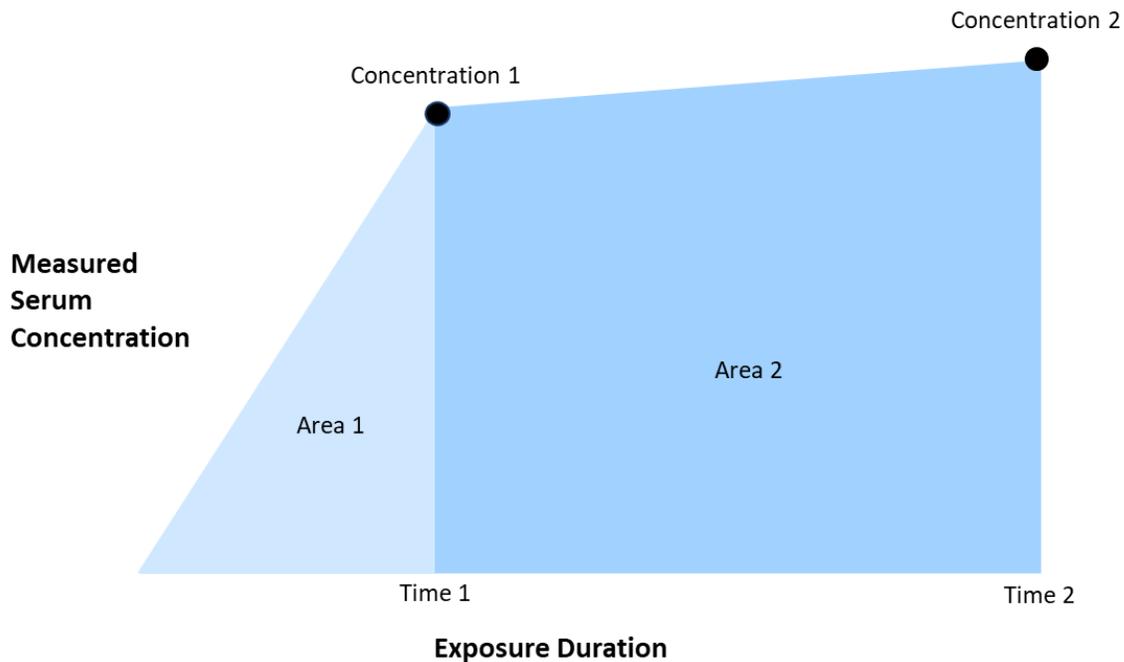
Reference	Toxicity Value (mg/kg-d)	Measured serum concentration at toxicity value (µg/L)	Area under the curve	Time-weight average serum concentration (µg/L)	Human equivalent dose (mg/kg-d)	Total Uncertainty Factor	ADI (ng/kg-d)
Das, 2015	1	13.67	61.58	6.84	0.0011	300	3
Wolf, 2010	0.83	8.91	98.25	4.47	0.0007	300	3
NTP, 2019	0.625	56.73	794.98	28.39	0.0044	3000	2

GD = gestation day; PND = postnatal day; ADI = acceptable daily intake

Appendix B. Calculation of Human Equivalent Dose

To calculate the human equivalent dose for PFNA, we followed a three-step process.

1. We first calculated the area under the curve at the selected toxicity value using the trapezoid rule.



In this mathematical approach, the area under the curve is divided into one or more trapezoids and area of each trapezoid is calculated (Equation B-1).

$$\text{Equation B-1} \quad \text{Area} = \frac{h}{2}(p+q)$$

Where:

- h = The difference in time between the data points.
- q = Measured serum concentration at first time point
- p = Measured serum concentration at second time point

The areas of all of the trapezoids are summed to give the area under the curve (Equation B-2).

$$\text{Equation B-2} \quad \text{AUC} = \text{Area}_1 + \text{Area}_2 + \dots + \text{Area}_n$$

2. We then calculated the time-weight average serum concentration as a surrogate for the steady-state serum concentration (Equation B-3).

$$\text{Equation B-3} \quad \text{TWA} = \frac{\text{AUC}}{\text{ED}}$$

Where:

- AUC = The difference in time between the data points.
- ED = Exposure duration (days)

3. Finally, we calculated the human equivalent dose (HED) by accounting for the long half-life in people.

$$\text{HED} = \frac{\text{TWA} \times \frac{\ln 2}{t_{1/2}} \times V_d}{\text{AF}}$$

Where:

- $t_{1/2}$ = half-life in days⁹
We selected a half-life of 3 years to consistent with other studies⁵¹
- V_d = Volume of distribution¹⁰
While data on the V_d of PFHxS in people are not available, one study has estimated V_d for nonhuman primates.⁵² Because the critical effect that we identified occurred in male animals, we used the V_d for male nonhuman primates (0.287 L/kg).
- AF = Gastrointestinal absorption fraction.¹¹
Data on the AF of PFHxS in people are not available. A few studies have estimated AF in research animals.^{52,53} Because PFHxS has been shown to be completely bioavailable in some of these studies, we used an AF of 1.

These values were also used by ATSDR in establishing their draft intermediate oral minimum risk level.¹

9 The half-life is a measure of elimination rate. The longer the half-life, the longer it takes a chemical to be removed from the body.

10 The volume of distribution is the theoretical volume needed to contain the amount of the chemical administered at the measured serum concentration.

11 The gastrointestinal absorption factor accounts for the bioavailability of the chemical after oral exposure. In other words, it is a measure how much of the chemical is available to cause harm within the body.

PFDA | 2020

Substance Overview

Perfluorodecanoic acid (PFDA)³ is a chemical in a group of contaminants called per- and polyfluoroalkyl substances (PFAS). PFAS are manmade chemicals that have been used in industry and consumer products since the 1940s. Because of their unique physical and chemical properties, PFAS can be found in a variety of commercial products such as paper and textile coatings, food packaging, surfactants, repellants, and fire-fighting foams.^{1,2} PFDA is highly stable and therefore can persist in the environment for decades.¹

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard for PFDA. DHS recommends an enforcement standard of 300 nanograms per liter (ng/L) for PFDA. The recommended standard is based on a study that found PFDA exposure during pregnancy decreased fetal weight at birth in mice.³

DHS recommends that the preventive action limit for PFDA be set at 10% of the enforcement standard because it has been shown to have carcinogenic and mutagenic effects in people, research animals, and cell culture assays.⁴⁻⁹

Current Standards

Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

Recommended Standards

Enforcement Standard:	300 ng/L
Preventive Action Limit:	30 ng/L

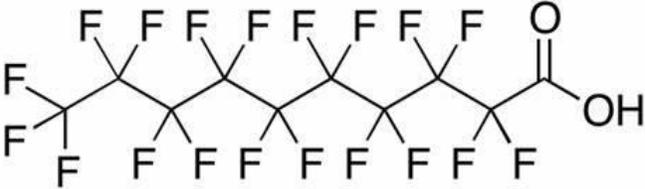
Health Effects

Studies among people exposed to high levels of PFAS have shown that PFDA is associated with a decrease in fetal body weight, a decrease in child growth parameters, an increase in thyroid hormone levels, an increase in cholesterol levels, a decreased antibody response to vaccines, and may affect sperm.^{1,5,6,10-21} Studies in research animals have found that high levels of PFDA can contribute to an increase in fetal mortality, a decrease in fetal weight gain, a decrease in body weight gain, altered immune cell populations, increased thyroid hormone levels, altered cholesterol levels, and can cause liver damage.^{1-3,7,22-28}

a This scientific support document and the included groundwater standard recommendations also apply to the anion salt of perfluorodecanoic acid (PFDA).

Limited information is available about the carcinogenic, mutagenic, teratogenic, and interactive effects of PFDA.^{b 1} The United States Environmental Protection Agency (EPA) and the International Agency for Research on Cancer (IRAC) have not assessed the carcinogenicity of PFDA. However, some studies have shown carcinogenic and mutagenic effects of PFDA in people, research animals, and cell culture assays.^{8,9,29,30} Additionally, limited research suggests PFDA may have interactive effects with endocrine disrupting chemicals in research animals and humans.⁴⁻⁷ PFDA has not been shown to have teratogenic effects in humans, research animals, and cell culture assays.

Chemical Profile

Perfluorodecanoic acid	
Structure:	
CAS Number:	335-76-2
Formula:	$C_{10}HF_{19}O_2$
Molar Mass:	514.09 g/mol
Synonyms:	Nonadecafluorodecanoic acid Decanoic acid, 2,2,3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,10- nonadecafluoro- Perfluoro-1-nonanecarboxylic acid Perfluorocapric acid

Exposure Routes

People can be exposed to PFDA by drinking contaminated water, swallowing contaminated soil, eating fish from contaminated lakes, eating food that was packaged in material that contains PFDA, and breathing in or swallowing dust that contains PFDA.^{1,31-33} Babies born to mothers exposed to PFDA can themselves be exposed to PFDA during pregnancy and breastfeeding.^{1,34,35}

In the environment, PFDA can be found in water or soil as an impurity from the use of other PFAS in manufacturing and consumer products and fire-fighting foam. PFDA can move between groundwater and surface water. Once in groundwater, PFDA can travel long distances.¹

^b Carcinogenic effects means the substance can cause cancer; mutagenic effects means the substance can cause DNA damage; teratogenic means the substance can cause birth defects; and interactive effects mean the substance can affect the toxicity of another substance or its toxicity can be affected by another substance.

Current Standard

Wisconsin does not currently have groundwater standards for PFDA.³⁶

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

EPA Oral Reference Dose:	N/A
--------------------------	-----

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

ATSDR Oral Minimum Risk Level:	N/A
--------------------------------	-----

Literature Search

Literature Search Dates:	1900 - 2020
Total studies evaluated:	Approximately 175
Key studies found?	Yes
Critical studies identified?	Yes

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for PFDA.³⁷

Health Advisory

The EPA has not established health advisories for PFDA.³⁸

Drinking Water Concentration (Cancer Risk)

The EPA has not established drinking water concentration based on a cancer risk level determination for PFDA.³⁹

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for PFDA.⁴⁰

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Information System (IRIS) program.

EPA Oral Reference Dose

The EPA does not have an oral reference dose for PFDA.³⁹

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If we determine that something is carcinogenic and there is no federal number or ADI from the EPA, DHS must set the standard at a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of PFDA, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of PFDA. If so, we look to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA and International Agency for Research on Cancer (IARC) have not evaluated the carcinogenicity of PFDA.^{39,41}

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for PFDA.³⁹

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information that was not considered when the value was established and indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For PFDA, we searched for values that had been published on or before February 2020.^a While the Agency for Toxic Substances and Disease Registry (ATSDR) reviewed the toxicity of PFDA in 2018, they did not establish any guidance values for PFDA.¹

Literature Search

Our literature review focused on relevant scientific literature on the health effects of PFDA published on or before February 2020. We looked for studies related to PFDA toxicity or effects on a disease state in which information on exposure or dose was included as part of the study or studies related to modeling PFOA exposure or dose using pharmacokinetics in animals or humans.^c Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

Approximately 175 toxicity studies were returned by the search engines. We excluded studies on non-mammalian or cell systems, non-oral exposure routes, and those that did not evaluate health risks from further review. After applying these exclusion criteria, we located 12 key toxicity studies on PFDA (summarized in Table A-1). To be considered a critical toxicity study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable

^c We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

Title/abstract: "PFDA" or "PFDeA" or "Perfluorodecanoic acid"

Subject area: N/A

Language: English

We also searched online for toxicity studies published by national research programs.

toxicity value.^{d-e} Two of these studies met the criteria to be considered a critical toxicity study (see Table A-2).

To be considered a critical pharmacokinetics study, the study must model oral exposure in humans or rodents. One of the key studies met the criteria to be considered a critical pharmacokinetic study (the section below has more details on these studies).

In our search, we also located epidemiology studies in our search (See Table A-3 for a summary). While multiple potential exposure sources and the ability for other PFAS compounds to cause similar health effects preclude using these data to establish a health-based value, such studies are helpful in identifying the crucial effects and ensuring that the animal data used to establish the standard is relevant to people.

Critical Toxicity Studies

To compare between results from recently found studies and the study used to set the current enforcement standard, we calculated an acceptable daily intake (ADI) for each study/effect. The ADI is the estimated amount of PFDA that a person can be exposed to every day and not experience health impacts. As such, we calculated the ADI by dividing a toxicity value from either a no-observed adverse effect level (NOAEL), lowest observed adverse effect level (LOAEL), or benchmark dose (BMD) identified in a study by a factor accounting for various sources of scientific uncertainty.^f Uncertainty factors were included, as appropriate, to account for differences between humans and animals, differences between healthy and sensitive human populations, using data from short-term experiments to protect against effects from long-term exposure, and using data where a health effect was observed to estimate the level that does not cause an effect. To ensure appropriate protection, we have chosen to not use studies that have significant uncertainty as the basis for the recommended enforcement standards.^g This approach is consistent with that taken by EPA when establishing oral reference doses.⁴²

Harris and Birnbaum, 1989

d Appropriate toxicity values include the no observable adverse effect level (NOAEL), lowest observable adverse effect level (LOAEL), and benchmark dose (BMD).

e Due to the limited availability of data for this substance, we considered a study to be of an appropriate duration if animals were exposed for at least 28 days or if the exposure occurred during pregnancy. We accounted for differences in exposure duration through the use of an uncertainty factor.

f The NOAEL is the highest dose tested that did not cause an adverse effect, the LOAEL is the lowest dose tested that caused an adverse effect, and the BMD is an estimation of the dose that would cause a specific level of response (typically 5 or 10%).

g DHS considers a study to have significant uncertainty if the total uncertainty factors is greater than 3,000.

Harris and Birnbaum exposed female mice over different gestational periods to different concentrations of PFDA in two separate experiments. Harris and Birnbaum exposed female mice to different concentrations of PFDA in two separate experiments.³ In the first experiment, they exposed mice to 0, 0.25, 0.5, 1, 2, 4, 8, 16, and 32 milligrams of PFDA per kilogram body weight per day or mg/kg-d through gavage for gestational days 10 to 13. They found that PFDA decreased maternal body weight, decreased fetal body weight, and increased absolute and relative maternal liver weights (Table 1A).

Table 1A. Statistically Significant Effects Observed After Exposure from Gestation Days 10-13 in Harris and Birnbaum, 1989

		Dose (mg/kg-d)							
		0.25	0.5	1	2	4	8	16	32
Growth	Lower maternal body weight net gain							✓	✓
	Lower fetal body weight		✓	✓	✓	✓	✓	✓	✓
Liver	Higher maternal absolute liver weight			✓	✓	✓	✓	✓	✓
	Higher maternal relative liver weight		✓	✓	✓	✓	✓	✓	✓

From this experiment, we identified a no-observed-adverse-effect-concentration (NOAEL) of 0.25 mg/kg-d based on effects on fetal body weight at higher doses. We estimate a candidate ADI of 250 nanograms of PFDA per kilogram body weight per day or ng/kg-d based on this NOAEL and a total uncertainty factor of 1000 to account for differences between people and research animals (10), difference among people (10), and the limited availability of information (10).

In the second experiment, the researchers exposed female mice to 0, 0.03, 0.3, 1, 3, 6.4, and 12.8 mg/kg-d PFDA through gavage for gestational days 6 to 15. They found that PFDA increased fetal mortality, decreased fetal body weight, decreased maternal body weight, and increased absolute and relative maternal liver weights (Table 1B).

Table 1B. Statistically Significant Effects Observed After Exposure from Gestation Days 6-15 in Harris and Birnbaum, 1989

		Dose (mg/kg-d)						
		0.03	0.1	0.3	1	3	6.4	12.8
Survival	Decreased fetal survival							✓
	Percentage resorptions per liter							✓
Growth	Lower maternal body weight net gain						✓	✓
	Lower fetal body weight		✓	✓	✓	✓	✓	✓
Liver	Higher maternal absolute liver weight				✓	✓	✓	✓
	Higher maternal relative liver weight				✓	✓	✓	✓

From this experiment, we identified a NOAEL of 0.03 mg/kg-d based on effects on fetal body weight at higher doses. We estimate a candidate ADI of 30 ng/kg-d PFDA from this study based on the NOAEL and

a total uncertainty factor of 1000 to account for differences between people and research animals (10), difference among people (10), and the limited availability of information (10).

Frawley et al., 2018

Frawley et al. exposed female rats and female mice to different concentrations of PFDA in two separate experiments.⁴³ In the first experiment, the researchers exposed female rats to different concentrations of PFDA (0, 0.125, 0.25, 0.5, 1, and 2 mg/kg-d) through gavage for 28 days. They found that PFDA decreased body weight, increased thymus weight, increased immune system activity in the thymus, increased kidney weight, and increased liver weight (Table 2A). They also found PFDA induced histological changes in liver cells.

Table 2A. Statistically Significant Effects Observed Female Rats in Frawley et al., 2018

Effects in rats		Dose (mg/kg-d)				
		0.125	0.25	0.5	1	2
Growth	Decreased body weight				✓	✓
Liver	Increased liver weight	✓	✓	✓	N/A	N/A
Thymus	Increased thymus weight	✓	✓		N/A	N/A
	Increase immune activity in the thymus			*	N/A	N/A
Kidney	Increased kidney weight		✓	✓	N/A	N/A
Blood	Decreased mean corpuscular hemoglobin concentrations		*	*	N/A	N/A

*The authors did not consider this finding to be adverse, biologically meaningful, or related to PFDA treatment (see Frawley et al., 2018 for more details). N/A – Authors indicated that rats which received 1 or 2 mg PFDA/kg-d were excluded from further evaluation due to adverse clinical observations in accordance with NTP laboratory Animal Management guidelines (see Frawley et al., 2018 for more details).

From this experiment, we identified a lowest-observable-adverse-effect-level (LOAEL) of 0.125 mg/kg-d based on effects on thymus and liver weights at higher doses. We estimate a candidate ADI of 1.25 ng/kg-d PFDA from this study based on the LOAEL and a total uncertainty factor of 100,000 to account for differences between people and research animals (10), difference among people (10), study duration (10), LOAEL (10), and the limited availability of information (10). While we obtained a candidate ADI for PFDA from this study, this study was not used to establish a recommended enforcement standard due to significant uncertainty.

In the second experiment, Frawley et al. exposed female mice to different concentrations of PFDA (0, 0.05, 0.09, 0.18, 0.36, and 0.71 mg/kg-d) through gavage for 28 days. They found that PFDA decreased body weight, increased liver weight, and decreased spleen weight. Additionally, the researchers found PFDA decreased the numbers of certain spleen cells indicating immunotoxicity potential (decreased

total spleen numbers, b-cells, t-cells, helper-t-cells, cytotoxic t-cells, immature t-cells, natural killer cells, and macrophage cells) (Table 2B).

Table 2B. Statistically Significant Effects Observed in Frawley et al., 2018

Effects observed in female mice		Dose (mg/kg-d)				
		0.05	0.09	0.18	0.36	0.71
Growth	Decreased body weight gain					✓
Liver	Increased liver weight		✓	✓	✓	✓
Spleen	Decreased spleen weight			✓	✓	✓
	Decreased total number of Spleen cells					✓
	Decrease number of Ig ⁺ B-cells					✓
	Decrease number of CD3 ⁺ T-cells			✓	✓	✓
	Decrease number of CD4 ⁺ CD8 ⁻ Helper T-cells			✓	✓	✓
	Decrease number of CD4 ⁻ CD8 ⁺ Cytotoxic T-cells		✓	✓	✓	✓
	Decrease number of CD4 ⁺ CD8 ⁺ Immature T-cells					✓
	Decrease number of Nk1.1 ⁺ CD3 ⁻ Natural Killer cell			✓		✓
	Decrease number of Mac3 ⁺ macrophage		✓	✓	✓	✓

From this experiment, we identified a NOAEL of 0.09 mg/kg-d based on decreased number of macrophage and cytotoxic t-cells in the spleen and increased liver weight at higher doses. We estimate a candidate ADI of 9 ng/kg-d PFDA from this study based on the NOAEL and a total uncertainty factor of 10,000 to account for differences between people and research animals (10), difference among people (10), study duration (10), and the limited availability of information (10). While we obtained a candidate ADI for PFDA from this study, this study was not used to establish a recommended enforcement standard due to significant uncertainty.

Critical Pharmacokinetic Studies

A physiologically-based pharmacokinetic (PBPK) model can be used to relate the amount of a chemical exposure to the amount of a chemical found in different organs at different time points.^{44,45}

PBPK models are developed using mathematical values and equations that describe characteristics and processes of the body, such as body weight, blood flow rate, and metabolism rate. The PBPK model uses a NOAEL and known rat serum levels to estimate a human point of departure (POD_{Human}). POD_{Human} is defined as the point on a toxicological dose-response curve established from experimental data that corresponds to an estimated no effect level. The POD_{Human} and PBPK models can be used to better understand what animal toxicity data mean for human health and PBPK models can provide a critical link between chemical toxicity and exposure information, as well as an important tool for using animal experiments to inform evaluations of the health effects of chemicals on humans.⁴⁴

Kim et al., 2019

,

In 2019, Kim et al., published a study in which they developed a physiology-based pharmacokinetic (PBPK) model to estimate PFDA serum levels in male and female rats from exposure to the NOAEL used in Kawashima et al.²⁴

The researchers obtained a point of departure for humans (POD_{Human}) of 28.4 µg/kg-d for males and 67.6 µg/kg-d for females (Table 3).⁴⁴ To obtain the candidate ADIs, we used an uncertainty factor of 1000 to account for differences among people (10), using data from a short-term study to protect from long-term effects (10), and the limited availability of data (10). We established a candidate ADI of 30 ng/kg-d for males and 70 ng/kg-d for females.

Table 3. Toxicity parameters and ADIs from Kim et al. 2019.⁴⁴

Sex	NOAEL (mg/kg-d)	Endpoint	POD _{Human} (µg/kg-d)	Total Uncertainty Factor	ADI (ng/kg-d)
Female	1	Liver toxicity*	67.6	1000**	70
Male	1	Liver toxicity*	28.4	1000**	30

* Toxicity endpoint from Kawashima et al.²⁴
 ** DHS selected a total uncertainty factor of 1000 to account for differences among people (10), use of a shorter duration study to protect against effects from long-term exposure (10), and the limited availability of data (10).

Key Health Effects

In our literature review, we found studies that indicate PFDA may cause carcinogenic and mutagenic effects in people, research animals, and cell culture assays.⁴⁻⁹ We did not find any studies indicating PFDA can cause teratogenic or interactive effects in people, research animals, or cell culture assays. We found limited information that PFDA may cause carcinogenic effects in humans.⁹ One case-control study among 161 Inuit women of Greenland showed that an increase in PFDA serum levels was associated with an increased risk of breast cancer.⁹

We found limited information that PFDA may cause mutagenic effects.⁸ One study that used the comet assay showed that PFDA exposure can lead to DNA damage and bind to DNA molecules by groove binding in mouse liver cell cultures. The authors suggest that PFDA can cause toxicity to genes by inducing oxidative stress.⁸

Discussion

While studies on the effects of PFDA among people are limited, PFDA has been associated with decreased baby weight, decreased childhood growth parameters, altered thyroid hormone levels, and altered cholesterol levels.^{1,5,6,11,12,14-16,20,21,46} In research animals, PFDA has been shown to decrease offspring survival, fetal weight, body and liver weights and increase liver lipid levels.¹ In our review, we

found that PFDA can also decrease spleen weight and specific spleen cells, increase thymus and kidney weight, and cause histological changes in the liver in research animals.^{3,7,22-24,26-28,43}

A number of studies have demonstrated associations between long-chain PFAS and birth weight and development in people and research animals. Multiple studies using research animals have noted a decrease in fetal body weight and a decrease in body weight gain as a common endpoint when exposed to PFDA.^{2,7,23-28} Similarly, some recent epidemiology studies show associations between increased PFDA concentrations and decreases in baby birth weights in humans.^{14,15,18,19,21} Scientists have associated multiple health issues in adulthood with lower weight at birth such as adult obesity, unfavorable metabolic outcomes, and increased risk of cardiovascular disease.^{21,47-49} Scientists are still continuing to learn how lower birth weight can affect adult human health.

Studies in research animals and people have shown that PFDA, as well as other PFAS, can affect the levels of thyroid hormones.¹ Thyroid hormones are crucial for development, energy balance, and metabolism in all species.⁴⁸ In people, thyroid hormones play an important role in the development of the brain, lungs, and heart.⁴⁸ Scientists have learned that certain PFAS, including PFDA, can bind to transport proteins involved in moving thyroid hormones throughout the body.⁵⁰ Scientists are still learning how this effect occurs and its impact on health.

A number of studies have demonstrated that liver effects caused by PFDA, as well as other PFAS, occur primarily through activation of the nuclear hormone receptor, PPAR α (peroxisome proliferator-activated receptor alpha).^{51,52} Nuclear receptors regulate gene expression and PPAR α regulates the expression of genes involved in lipid and cholesterol metabolism.⁵⁰ While PPAR α receptors are found in rodents and humans, levels of these receptors are much higher in rats and mice than in monkeys and humans.⁵⁰ Additionally, PFDA specifically was not shown to activate human PPAR α .⁵¹ This suggests that effects caused by activation of PPAR α are more likely to occur in rodents than they are to occur in people. As such, the effects on the liver observed in the critical studies reviewed here are likely not relevant to humans. This conclusion is supported by the lack of associations between PFAS exposure and liver disease in epidemiological studies (Table A-3).

Standard Selection

DHS recommends an enforcement standard of 300 ng/L for PFDA.

There are no federal numbers and no state drinking water standard for PFDA. Additionally, the EPA has not evaluated the carcinogenicity of PFDA or established an ADI (oral reference dose) for PFDA.

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information

However, we found critical toxicity and pharmacokinetic modeling studies for PFDA. To calculate the ADI as specified in s. 160.13, Wisc. Statute, DHS selected the 1989 study by Harris and Birnbaum as the critical study.³ We selected this study because it evaluated the effects of PFDA exposure during reproductive development and evaluated both maternal and offspring endpoints. Toxicity studies in animals continue to show that developmental endpoints are critical effects for PFAS, including PFDA, with effects occurring in offspring after exposure during pregnancy and lactation.^{3,7} Additionally, epidemiology studies continue to associate maternal PFDA exposure to developmental problems in offspring, as PFDA has been shown to cross the placenta during pregnancy.^{10,11,14,15,19,46 1,34,35} We choose fetal body weight as the critical effect as it can be associated with health issues such as adult obesity, unfavorable metabolic outcomes, and increased risk of cardiovascular disease.^{21,47-49}

As described above, we obtained an ADI of 30 ng/kg-d from a NOAEL of 0.03 mg/kg-d and a total uncertainty factor of 1000. To determine the recommended enforcement standard, DHS used the ADI, and, as required by Ch. 160, Wis. Stats., a body weight of 10 kg, a water consumption rate of 1 L/d, and a relative source contribution of 100%.

DHS recommends a preventive action limit of 30 ng/L for PFDA.

DHS recommends that the preventive action limit for PFDA be set at 10% of the enforcement standard. PFDA has been shown to cause carcinogenic and mutagenic effects in people, research animals, and cell culture assays.⁴⁻⁹ At this time, PFDA has not been shown to have teratogenic or interactive effects in people, research animals, or cell culture studies.

Prepared by Gavin Dehnert, Ph.D. and Sarah Yang, Ph.D.

Wisconsin Department of Health Services

References

1. ATSDR. Toxicological Profile for Perfluoroalkyls - Draft for Public Comment. In: Registry AftSaD, ed. Atlanta, GA2017.
2. Kawabata K, Matsuzaki H, Nukui S, et al. Perfluorododecanoic Acid Induces Cognitive Deficit in Adult Rats. *Toxicological sciences : an official journal of the Society of Toxicology*. 2017;157(2):421-428.
3. Harris MW, Birnbaum LS. Developmental toxicity of perfluorodecanoic acid in C57BL/6N mice. *Fundamental and applied toxicology : official journal of the Society of Toxicology*. 1989;12(3):442-448.

4. Kim DH, Kim UJ, Kim HY, Choi SD, Oh JE. Perfluoroalkyl substances in serum from South Korean infants with congenital hypothyroidism and healthy infants--Its relationship with thyroid hormones. *Environmental research*. 2016;147:399-404.
5. Aimuzi R, Luo K, Chen Q, et al. Perfluoroalkyl and polyfluoroalkyl substances and fetal thyroid hormone levels in umbilical cord blood among newborns by prelabor caesarean delivery. *Environment international*. 2019;130:104929.
6. Inoue K, Ritz B, Andersen SL, et al. Perfluoroalkyl Substances and Maternal Thyroid Hormones in Early Pregnancy; Findings in the Danish National Birth Cohort. *Environmental health perspectives*. 2019;127(11):117002.
7. Harris MW, Uraih LC, Birnbaum LS. Acute toxicity of perfluorodecanoic acid in C57BL/6 mice differs from 2,3,7,8-tetrachlorodibenzo-p-dioxin. *Fundamental and applied toxicology : official journal of the Society of Toxicology*. 1989;13(4):723-736.
8. Xu MC, Zhang T, Lv C, et al. Perfluorodecanoic acid-induced oxidative stress and DNA damage investigated at the cellular and molecular levels. *Ecotoxicology and environmental safety*. 2019;185.
9. Wielsoe M, Kern P, Bonefeld-Jorgensen EC. Serum levels of environmental pollutants is a risk factor for breast cancer in Inuit: a case control study. *Environmental health : a global access science source*. 2017;16(1):56.
10. Ernst A, Brix N, Lauridsen LLB, et al. Exposure to Perfluoroalkyl Substances during Fetal Life and Pubertal Development in Boys and Girls from the Danish National Birth Cohort. *Environmental health perspectives*. 2019;127(1):17004.
11. Gyllenhammar I, Diderholm B, Gustafsson J, et al. Perfluoroalkyl acid levels in first-time mothers in relation to offspring weight gain and growth. *Environment international*. 2018;111:191-199.
12. Jain RB, Ducatman A. Roles of gender and obesity in defining correlations between perfluoroalkyl substances and lipid/lipoproteins. *The Science of the total environment*. 2019;653:74-81.
13. Louis GM, Chen Z, Schisterman EF, et al. Perfluorochemicals and human semen quality: the LIFE study. *Environmental health perspectives*. 2015;123(1):57-63.
14. Kashino I, Sasaki S, Okada E, et al. Prenatal exposure to 11 perfluoroalkyl substances and fetal growth: A large-scale, prospective birth cohort study. *Environment international*. 2020;136:105355.
15. Kwon EJ, Shin JS, Kim BM, et al. Prenatal Exposure to Perfluorinated Compounds Affects Birth Weight Through GSTM1 Polymorphism. *Journal of occupational and environmental medicine*. 2016;58(6):e198-205.

16. Mora AM, Fleisch AF, Rifas-Shiman SL, et al. Early life exposure to per- and polyfluoroalkyl substances and mid-childhood lipid and alanine aminotransferase levels. *PLoS medicine*. 2018;111:1-13.
17. Seo SH, Son MH, Choi SD, Lee DH, Chang YS. Influence of exposure to perfluoroalkyl substances (PFASs) on the Korean general population: 10-year trend and health effects. *Environment international*. 2018;113:149-161.
18. Tian Y, Liang H, Miao M, et al. Maternal plasma concentrations of perfluoroalkyl and polyfluoroalkyl substances during pregnancy and anogenital distance in male infants. *Human reproduction (Oxford, England)*. 2019;34(7):1356-1368.
19. Wang Y, Adgent M, Su PH, et al. Prenatal Exposure to Perfluorocarboxylic Acids (PFCAs) and Fetal and Postnatal Growth in the Taiwan Maternal and Infant Cohort Study. *Environmental health perspectives*. 2016;124(11):1794-1800.
20. Wang Y, Rogan WJ, Chen PC, et al. Association between maternal serum perfluoroalkyl substances during pregnancy and maternal and cord thyroid hormones: Taiwan maternal and infant cohort study. *Environmental health perspectives*. 2014;122(5):529-534.
21. Wikstrom S, Lin PI, Lindh CH, Shu H, Bornehag CG. Maternal serum levels of perfluoroalkyl substances in early pregnancy and offspring birth weight. *Pediatric research*. 2019.
22. Yamamoto A, Kawashima Y. Perfluorodecanoic acid enhances the formation of oleic acid in rat liver. *The Biochemical journal*. 1997;325 (Pt 2):429-434.
23. Takagi A, Sai K, Umemura T, Hasegawa R, Kurokawa Y. Short-term exposure to the peroxisome proliferators, perfluorooctanoic acid and perfluorodecanoic acid, causes significant increase of 8-hydroxydeoxyguanosine in liver DNA of rats. *Cancer letters*. 1991;57(1):55-60.
24. Kawashima Y, Kobayashi H, Miura H, Kozuka H. Characterization of hepatic responses of rat to administration of perfluorooctanoic and perfluorodecanoic acids at low levels. *Toxicology*. 1995;99(3):169-178.
25. Brewster DW, Birnbaum LS. The biochemical toxicity of perfluorodecanoic acid in the mouse is different from that of 2,3,7,8-tetrachlorodibenzo-p-dioxin. *Toxicology and applied pharmacology*. 1989;99(3):544-554.
26. Permadi H, Lundgren B, Andersson K, DePierre JW. Effects of perfluoro fatty acids on xenobiotic-metabolizing enzymes, enzymes which detoxify reactive forms of oxygen and lipid peroxidation in mouse liver. *Biochemical pharmacology*. 1992;44(6):1183-1191.
27. Permadi H, Lundgren B, Andersson K, Sundberg C, DePierre JW. Effects of perfluoro fatty acids on peroxisome proliferation and mitochondrial size in mouse liver: dose and time factors and effect of chain length. *Xenobiotica; the fate of foreign compounds in biological systems*. 1993;23(7):761-770.

28. Wang D, Gao Q, Wang T, et al. Green tea polyphenols and epigallocatechin-3-gallate protect against perfluorodecanoic acid induced liver damage and inflammation in mice by inhibiting NLRP3 inflammasome activation. *Food research international (Ottawa, Ont)*. 2020;127:108628.
29. Dong T, Peng Y, Zhong N, et al. Perfluorodecanoic acid (PFDA) promotes gastric cell proliferation via sPLA2-IIA. *Oncotarget*. 2017;8(31):50911-50920.
30. Cheng X, Klaassen CD. Perfluorocarboxylic acids induce cytochrome P450 enzymes in mouse liver through activation of PPAR-alpha and CAR transcription factors. *Toxicological sciences : an official journal of the Society of Toxicology*. 2008;106(1):29-36.
31. Hu XDC, Dassuncao C, Zhang XM, et al. Can profiles of poly- and Perfluoroalkyl substances (PFASs) in human serum provide information on major exposure sources? *Environ Health*. 2018;17:15.
32. Christensen KY, Raymond M, Thompson BA, Anderson HA. Perfluoroalkyl substances in older male anglers in Wisconsin. *Environment international*. 2016;91:312-318.
33. Motas Guzman M, Clementini C, Perez-Carceles MD, et al. Perfluorinated carboxylic acids in human breast milk from Spain and estimation of infant's daily intake. *The Science of the total environment*. 2016;544:595-600.
34. Yao Q, Shi R, Wang C, et al. Cord blood Per- and polyfluoroalkyl substances, placental steroidogenic enzyme, and cord blood reproductive hormone. *Environment international*. 2019;129:573-582.
35. Shi Y, Yang L, Li J, et al. Occurrence of perfluoroalkyl substances in cord serum and association with growth indicators in newborns from Beijing. *Chemosphere*. 2017;169:396-402.
36. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 140*2017.
37. USEPA. National Primary Drinking Water Regulations. <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>. Published 2018. Accessed.
38. USEPA. Drinking Water Contaminant Human Health Effects Information. <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>. Published 2019. Accessed.
39. USEPA. IRIS Assessments. https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm. Published 2019. Accessed.
40. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 809*2018.
41. IARC. List of Classification, Volumes 1-123. <https://monographs.iarc.fr/list-of-classifications-volumes/>. Published 2018. Accessed May 17, 2019.
42. USEPA. A Review of the Reference Dose and Reference Concentration Processes. 2002(EPA/630/P-02/002F).

43. Frawley RP, Smith M, Cesta MF, et al. Immunotoxic and hepatotoxic effects of perfluoro-n-decanoic acid (PFDA) on female Harlan Sprague-Dawley rats and B6C3F1/N mice when administered by oral gavage for 28 days. *Journal of immunotoxicology*. 2018;15(1):41-52.
44. Kim SJ, Choi EJ, Choi GW, Lee YB, Cho HY. Exploring sex differences in human health risk assessment for PFNA and PFDA using a PBPK model. *Archives of toxicology*. 2019;93(2):311-330.
45. USEPA. Physiologically-based pharmacokinetic (PBPK) models: Scientific models to help evaluate health effects of chemicals. 2018.
46. Lee YA, Kim JH, Jung HW, et al. The serum concentrations of perfluoroalkyl compounds were inversely associated with growth parameters in 2-year old children. *The Science of the total environment*. 2018;628-629:226-232.
47. Barker DJ. The developmental origins of adult disease. *Journal of the American College of Nutrition*. 2004;23(6 Suppl):588s-595s.
48. Andersen LG, Holst C, Michaelsen KF, Baker JL, Sorensen TI. Weight and weight gain during early infancy predict childhood obesity: a case-cohort study. *International journal of obesity (2005)*. 2012;36(10):1306-1311.
49. Sharma D, Shastri S, Sharma P. Intrauterine Growth Restriction: Antenatal and Postnatal Aspects. *Clinical medicine insights Pediatrics*. 2016;10:67-83.
50. Hall AP, Elcombe CR, Foster JR, et al. Liver hypertrophy: a review of adaptive (adverse and non-adverse) changes--conclusions from the 3rd International ESTP Expert Workshop. *Toxicologic pathology*. 2012;40(7):971-994.
51. Wolf CJ, Takacs ML, Schmid JE, Lau C, Abbott BD. Activation of mouse and human peroxisome proliferator-activated receptor alpha by perfluoroalkyl acids of different functional groups and chain lengths. *Toxicological sciences : an official journal of the Society of Toxicology*. 2008;106(1):162-171.
52. Cheng X, Klaassen CD. Critical role of PPAR-alpha in perfluorooctanoic acid- and perfluorodecanoic acid-induced downregulation of Oatp uptake transporters in mouse livers. *Toxicological sciences : an official journal of the Society of Toxicology*. 2008;106(1):37-45.
53. Beck IH, Timmermann CAG, Nielsen F, et al. Association between prenatal exposure to perfluoroalkyl substances and asthma in 5-year-old children in the Odense Child Cohort. *Environmental health : a global access science source*. 2019;18(1):97.
54. Blake BE, Pinney SM, Hines EP, Fenton SE, Ferguson KK. Associations between longitudinal serum perfluoroalkyl substance (PFAS) levels and measures of thyroid hormone, kidney function, and body mass index in the Fernald Community Cohort. *Environmental pollution (Barking, Essex : 1987)*. 2018;242(Pt A):894-904.

55. Louis GM, Sapra KJ, Barr DB, Lu Z, Sundaram R. Preconception perfluoroalkyl and polyfluoroalkyl substances and incident pregnancy loss, LIFE Study. *Reproductive toxicology (Elmsford, NY)*. 2016;65:11-17.
56. Chen Q, Huang R, Hua L, et al. Prenatal exposure to perfluoroalkyl and polyfluoroalkyl substances and childhood atopic dermatitis: a prospective birth cohort study. *Environmental health : a global access science source*. 2018;17(1):8.
57. Christensen KY, Raymond M, Meiman J. Perfluoroalkyl substances and metabolic syndrome. *International journal of hygiene and environmental health*. 2019;222(1):147-153.
58. Dalsager L, Christensen N, Husby S, et al. Association between prenatal exposure to perfluorinated compounds and symptoms of infections at age 1-4years among 359 children in the Odense Child Cohort. *Environment international*. 2016;96:58-64.
59. Fleisch AF, Rifas-Shiman SL, Mora AM, et al. Early-Life Exposure to Perfluoroalkyl Substances and Childhood Metabolic Function. *Environmental health perspectives*. 2017;125(3):481-487.
60. Huang M, Jiao J, Zhuang P, Chen X, Wang J, Zhang Y. Serum polyfluoroalkyl chemicals are associated with risk of cardiovascular diseases in national US population. *Environment international*. 2018;119:37-46.
61. Jensen RC, Glintborg D, Gade Timmermann CA, et al. Prenatal exposure to perfluorodecanoic acid is associated with lower circulating concentration of adrenal steroid metabolites during mini puberty in human female infants. The Odense Child Cohort. *Environmental research*. 2020;182:109101.
62. Jensen RC, Glintborg D, Timmermann CAG, et al. Perfluoroalkyl substances and glycemic status in pregnant Danish women: The Odense Child Cohort. *Environment international*. 2018;116:101-107.
63. Jensen TK, Andersen LB, Kyhl HB, Nielsen F, Christesen HT, Grandjean P. Association between perfluorinated compound exposure and miscarriage in Danish pregnant women. *PloS one*. 2015;10(4):e0123496.
64. Kvaalem HE, Nygaard UC, Lodrup Carlsen KC, Carlsen KH, Haug LS, Granum B. Perfluoroalkyl substances, airways infections, allergy and asthma related health outcomes - implications of gender, exposure period and study design. *Environment international*. 2020;134:105259.
65. Lind DV, Priskorn L, Lassen TH, et al. Prenatal exposure to perfluoroalkyl substances and anogenital distance at 3 months of age in a Danish mother-child cohort. *Reproductive toxicology (Elmsford, NY)*. 2017;68:200-206.
66. Lind PM, Salihovic S, van Bavel B, Lind L. Circulating levels of perfluoroalkyl substances (PFASs) and carotid artery atherosclerosis. *Environmental research*. 2017;152:157-164.

67. Lum KJ, Sundaram R, Barr DB, Louis TA, Buck Louis GM. Perfluoroalkyl Chemicals, Menstrual Cycle Length, and Fecundity: Findings from a Prospective Pregnancy Study. *Epidemiology (Cambridge, Mass)*. 2017;28(1):90-98.
68. Meng Q, Inoue K, Ritz B, Olsen J, Liew Z. Prenatal Exposure to Perfluoroalkyl Substances and Birth Outcomes; An Updated Analysis from the Danish National Birth Cohort. 2018;15(9).
69. Niu J, Liang H, Tian Y, et al. Prenatal plasma concentrations of Perfluoroalkyl and polyfluoroalkyl substances and neuropsychological development in children at four years of age. *Environmental health : a global access science source*. 2019;18(1):53.
70. Starling AP, Adgate JL, Hamman RF, et al. Perfluoroalkyl Substances during Pregnancy and Offspring Weight and Adiposity at Birth: Examining Mediation by Maternal Fasting Glucose in the Healthy Start Study. *Environmental health perspectives*. 2017;125(6):067016.
71. Timmermann CA, Budtz-Jorgensen E, Jensen TK, et al. Association between perfluoroalkyl substance exposure and asthma and allergic disease in children as modified by MMR vaccination. *Journal of immunotoxicology*. 2017;14(1):39-49.
72. Yang L, Li J, Lai J, et al. Placental Transfer of Perfluoroalkyl Substances and Associations with Thyroid Hormones: Beijing Prenatal Exposure Study. *Scientific reports*. 2016;6:21699.
73. Zeeshan M, Yang Y, Zhou Y, et al. Incidence of ocular conditions associated with perfluoroalkyl substances exposure: Isomers of C8 Health Project in China. *Environment international*. 2020;137:105555.

Appendix A. Toxicity Data

Table A-I. PFDA Toxicity Studies from Literature Review

Study Type	Species	Duration	Doses (mg/kg-d)	Route	Endpoints	Toxicity Value (mg/kg-d)	Reference
Long Term	Mouse	30 Days ^c	0, 40, 80, 100, 120, 160	Gavage	PFDA decreased body weight at 80 mg/kg. PFDA increased liver weight at 40 mg/kg.	NOAEL: N/A LOAEL: 40	Brewster and Birnbaum, 1989 ²⁵
Short Term	Rat	28 Days	0, 0.125, 0.25, 0.5, 1.0, 2.0	Gavage	PFDA decreased body weight and weight gain at 1.0 and 2.0 mg/kg-d; increased absolute and relative liver weights at 0.25 and 0.5 mg/kg-d; increased absolute and relative thymus weights at 0.125 and 0.25 mg/kg-d; increased absolute and relative kidney weights at 0.5 mg/kg-d. PFDA induced centrilobular, single cell, and necrosis in the liver at 0.5 mg/kg/d.	NOAEL: N/A LOAEL: 0.125	Frawley et al., 2018a ⁴³
Short Term	Mouse	28 days	0, 0.045, 0.089, 0.18, 0.35, 0.71	Gavage ^A	PFDA decreased body weight and weight gain at 0.71 mg/kg-d; increased absolute and relative liver weights at 0.089, 0.18, 0.35, and 0.71 mg/kg-d; decreased relative spleen weight at 0.18, 0.35, and 0.71 mg/kg-d; decreased overall spleen weight at 0.71 mg/kg-d.	NOAEL: 0.045 LOAEL: 0.089	Frawley et al., 2018b ⁴³

Development	Mouse	Day 10-13 of gestation	0, 0.25, 0.5, 1.0, 2.0, 4.0, 8.0, 16.0, 32.0	Gavage	PFDA decreased maternal body weight at 16 and 32 mg/kg-d; increased relative and absolute liver weight at 1.0, 2.0, 4.0, 8.0, 16.0, and 32.0 mg/kg-d. PFDA decreased fetal body weight at 0.5, 1.0, 2.0, 4.0, 8.0, 16.0, and 32.0 mg/kg-d.	NOAEL: 0.025 LOAEL: 0.5	Harris and Birnbaum, 1989 ³
Development	Mouse	Day 6-15 of gestation	0, 0.03, 0.1, 0.3, 1.0, 3.0, 6.4, 12.8	Gavage	PFDA decreased maternal body weight at 6.4 and 12.8 mg/kg-d; increased relative and absolute liver weight at 1.0, 3.0, 6.4, and 12.8 mg/kg-d. PFDA decreased fetal body weight at 0.1, 0.3, 1.0, 3.0, 6.4, 12.8 mg/kg-d; increased fetus mortality and fetus resorption at 12.8 mg/kg-d.	NOAEL: 0.03 LOAEL: 0.1	Harris and Birnbaum, 1989 ³
Long Term	Mouse	30 Days ^c	0, 20, 40, 80, 160, 320 mg/kg	Gavage	PFDA increased mortality at 160 and 320 mg/kg; decreased body weight at 80 mg/kg. PFDA increased relative and absolute liver weight in a dose-dependent manner; decreased absolute and relative spleen and heart weights in a dose dependent manner. PDFA increased T3 levels at 80 mg/kg and increased T4 levels at 20, 40, and 80 mg/kg. PFDA increased bile duct hyperbilia duct hyperplasia and hepatocellular necrosisplasia in a dose dependent mann.	NOAEL: N/A LOAEL:20 mg/kg	Harris et al., 1989 ⁷

Long Term	Mouse	30 Days ^c	0, 40, 80, 100, 120, 160 mg/kg	Gavage	PFDA increased mortality at 120 and 160 mg/kg; decreased body weight at 80 mg/kg. PFDA increased relative and absolute liver weight in a dose dependent manor; decreased absolute and relative spleen and thymus in a dose dependent manor. PFDA increased hypertrophy in liver cells at 40 mg/kg; increased necrosis of individual parenchymal cells at 40 mg/kg.	NOAEL: N/A LOAEL: 40 mg/kg	Harris et al., 1989 ⁷
Short Term	Rat	9 Days ^c	0, 50mg/kg	Gavage	PFDA decreased body weight at 50 mg/kg. PFDA decreased absolute and relative liver weight at 50mg/kg.	NOAEL: N/A LOAEL: 50mg/kg	Kawabata et al., 2017 ²
Short Term	Rat	7 Days	0, 0.00125, 0.0025, 0.005 0.01% (w/w)	Diet	PFDA decreased body weight at 0.01%. PFDA increased liver weight at 0.0025 and 0.005% and relative liver weight at 0.00125, 0.0025, 0.005, and 0.01%. PFDA induced three peroxisome proliferator-responsive parameters in a dose dependent manor. PFDA increased liver concentration of triacylglycerol in a dose dependent manor and increased cholesterol at 0.01%; increased cell size and proliferations of peroxisomes, and small lipid droplets in liver cells at 0.01%.	NOAEL: N/A LOAEL: 0.00125%	Kawashima et al., 1995 ²⁴

Short term	Mouse	10 Days	0, 0.02% (w/w)	Diet	PFDA decreased body weight at 0.02% (w/w). PFDA decreased absolute and relative liver weight at 0.02% (w/w). PFDA decreased Gluthathione peroxidase at 0.02% (w/w); increased Superioxide dismutase and Epoxide hydrolase at 0.02% (w/w).	NOAEL: N/A LOAEL: 0.02%	Permadi et al., 1991 ²⁶
Short term	Mouse	10 Days	0, 0.02% (w/w)	Diet	PFDA decreased body weight at 0.02% (w/w). Decreased absolute and relative liver weight at 0.02% (w/w). PFDA increased Lauroyl-CoA oxidase and Palmitoyl-CoA oxidation at 0.02% (w/w).	NOAEL: N/A LOAEL: 0.02%	Permadi et al., 1992 ²⁷
Short term	Rat	14 Days	0, 0.01, 0.02% (w/w)	Diet	PFDA decreased body weight gain and food consumption at 0.01% and 0.02% (w/w); increased liver and kidney weight at 0.01%, 0.02% (w/w).	NOAEL: N/A LOAEL: 0.01%	Takagi et al., 1991 ²³
Long Term	Mouse	35 Days ^B	0, 0.05 mM	Drinking Water	PFDA increased mortality at 0.05mM; decreased body weights 0.05mM.	NOAEL: N/A LOAEL: 0.05mM	Wang et al., 2019a ²⁸

Short Term	Mouse	14 Days	0, 0.1, 0.05mM	Drinking Water	PFDA decreased body weights 0.1mM; increased the levels of plasma Alanine transaminase and Aspartate transaminase. PFDA caused necrosis, steatosis, edema, and degeneration in the liver at 0.1mM; decreased total-super-oxide dismutase, catalase, and glutathione peroxidase activities and increased reactive oxygen species levels, Thiobarbituric acid, and malondialdehyde contents in liver tissues at 0.1mM.	NOAEL: N/A LOAEL: 0.05mM	Wang et al., 2019b ²⁸
Short Term	Mouse	7 Days	0, 0.005% (w/w)	Diet	PFDA increased liver weight liver at 0.005% (w/w). PFDA increased activity of microsomal stearyl-CoA desaturation in the liver at 0.005% (w/w); induced activity of peroxisomal beta-oxidation in the liver at 0.005% (w/w).	NOAEL: N/A LOAEL: 0.005% (w/w)	Yamamoto and Kawashima, 1997 ²²

^A All animals were orally gavaged once per week

^B All animals died at this point in the study

^C All animals were given a single dose

Table A-2. Critical Study Selection for PFDA

Reference	Appropriate duration?	Effects consistent with other studies?	Effects relevant to humans?	Number of Doses	Toxicity value identifiable?	Critical study?
Frawley et al., 2018a	✓	✓	✓	5	✓	Yes
Frawley et al., 2018b	✓	✓	✓	5	✓	Yes
Harris and Brinbaum, 1989a	✓	✓	✓	8	✓	Yes
Harris and Brinbaum, 1989b	✓	✓	✓	7	✓	Yes
Kawashima et al., 1991	⊘	✓	✓	4	✓	No
Permadi et al., 1991	⊘	✓	✓	1	✓	No
Permadi et al., 1992	⊘	✓	✓	1	✓	No
Takagi et al., 1991	⊘	✓	✓	2	✓	No
Wang et al., 2019a	✓	✓	✓	1	✓	No
Wang et al., 2019b	⊘	✓	✓	1	✓	No
Yamamoto and Kawashima, 1997	⊘	✓	✓	2	✓	No

To be considered a critical study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.

Table A-3. PFDA Epidemiological Studies from Literature Review

Study Type	Population	Time period	Exposure	Outcomes	Results	Other PFAS Studied	Reference
Cohort	568 mother-child pairs from the Shanghai Obesity and Allergy Cohort	2012-2013	Cord plasma PFAS concentrations	Thyroid disruption	An increase in PFDA concentrations (ng/mL natural log transformed per unit) was associated with an increase in free T4 levels (pmol/L) among boys.	PFOA, PFOS, PFNA, PFUnA, PFHxS, PFDoA, PFBS, PFOSA, PFHpA	Aimuzi et al., 2019 ⁵
Cohort	981 mother-child pairs within the Odense Child Cohort	2010-2012	Maternal PFAS serum concentrations	Childhood asthma	PFDA was not associated with the wheeze test, self-reported asthma, or doctor-diagnosed asthma in children.	PFOS, PFOA, PFHxS, PFNA	Beck et al., 2019 ⁵³
Cohort	210 community members (median age 38)	1990-2008	Drinking water exposure; serum PFAS concentrations	Thyroid disruption, kidney function, and BMI	Interquartile PFDA increase was associated with a 2.20% estimated glomerular filtration rate decrease. PFDA had no associations with thyroid disruption or BMI.	PFOS, PFOA, PFNA, PFHxS, PFOSA, Me-PFOA, Et-PFOA	Blake et al., 2018 ⁵⁴
Cohort	501 mothers from Michigan and Texas	2005-2009	Maternal PFAS serum concentrations	Pregnancy loss	PFDA was not associated with an increase in pregnancy loss.	PFNA, PFOS, PFOA, PFOSA, Et-PFOSE-AcOH, Me-PFOA-AcOH	Buck Louis et al., 2016 ⁵⁵
Cohort	501 fathers from Michigan and Texas	2005-2009	Paternal PFAS serum concentrations	Semen parameters	An increase in PFDA (1-unit ln transformed ng/mL) was associated with a decrease in sperm head length (um); $\beta = -0.155$ (95% CI: -0.304, -0.006). An increase in PFDA (1-unit ln transformed ng/mL) was associated with a decrease in the percentage of sperm coiled tails; $\beta = -7.603$ (95% CI: -14.014, -1.193).	PFNA, PFOS, PFOA, PFOSA, Me-PFOA-AcOH	Buck Louis et al., 2015 ¹³

Cohort	687 mother-child pairs from Shanghai, China	2012-2015	Cord serum PFAS concentrations	Atopic dermatitis	PFDA was associated with a two-fold increase atopic dermatitis risk; AOR = 2.67 (95% CI: 1.00-4.57).	PFOA, PFOS, PFNA, PFUnA, PFHxS, PFOSA, PFDoA, PFBS, PFHpA	Chen et al., 2018 ⁵⁶
Cross sectional study	2975 participants from National Health and Nutrition Examination Survey	2007-2008, 2009-2010, 2011-2012, 2013-2014	Serum PFAS concentrations	Metabolic syndrome	PFDA was associated with a decreased risk with metabolic syndrome; OR = 0.72 (Q1 vs Q4) (95% CI: 0.53, 0.99). PFDA was associated with a decrease in triglycerides; OR = 0.45 (Q1 vs Q4) (95% CI: 0.25, 0.80). PFDA was associated with a decrease in high-density lipoprotein cholesterol; OR = 0.48 (Q1 vs Q4) (95% CI: 0.30, 0.77).	PFOA, PFOS, MPAH, PFHxS, PFNA, PFUnDA	Christensen et al., 2019 ⁵⁷
Cohort	359 mother-child pairs in the Odense Child Cohort	2010-2012	Maternal serum PFAS concentrations	Infection symptoms	PFDA was not associated with the number of days with symptoms or with an increase in risk of having symptoms.	PFOS, PFOA, PFHxS, PFNA	Dalsager et al., 2016 ⁵⁸
Nested Cohort	445 mother-child pairs within the Danish National Birth Cohort	2012-2017	PFAS in maternal plasma from early gestation	Hormonal developmental outcomes	PFDA (ng/mL) was associated with 3.60 month earlier average age at onset for the combined puberty indicator in girls; (95% CI: -9.03, 1.83). PFDA (ng/mL) was associated with a 4.59 month older age at onset for the combined puberty indicator in boys; (95% CI: -0.93, 10.11).	PFOS, PFOA, PFHxS, PFHpS, PFNA,	Ernst et al., 2019 ¹⁰
Cohort	665 mother-child pairs in Boston, MA; Project Viva	1999-2002 Maternal: 2007-2010 Child	Maternal plasma PFAS concentrations at first prenatal visit (mean: 9.6 weeks gestation); Child plasma PFAS	Metabolic assessment, and homeostatic model assessment of insulin resistance (HOMA-IR)	HOMA-IR was 14.5% lower per interquartile range increment in PFDA (ng/mL); (95% CI: -24.7, -2.9). PFDA was not associated with metabolic biomarkers (e.g. Leptin and Adiponectin).	PFOA, PFOS, PFNA, PFHxS	Fleisch et al., 2017 ⁵⁹

			concentrations at (mean:7.7 years)				
Cohort	381 mother-child pairs from Uppsala County, Sweden.	1996-2011	Maternal serum PFAS concentrations	Birth outcomes; growth and development	Maternal PFDA concentrations (ng/g) were inversely associated with birth weight scores in the children. Maternal PFDA concentrations were not associated with child growth parameters from age 3 months to 5 years.	PFOA, PFNA, PFUnDA, PFBS, PFHxS, PFOS,	Gyllenhammar et al., 2018 ¹¹
Cohort	10859 participants from NHANES (National Health and Nutritional Examination Survey)	1999-2014	Serum PFAS concentrations	Prevalence of cardiovascular diseases (CVD).	PFDA was positively associated with coronary heart disease: p trend = 0.0107; AOR Q4 vs Q1 = 1.84 (95%CI: 1.26-2.69).	PFOA, PFOS, PFHxS, PFNA, EPAH, MPAH, PFBS, PFHP, PFDO, n-PFOA, Sb-PFOA, n-PFOS, Sm-PFOS	Huang et al., 2018 ⁶⁰
Cross-sectional study	1366 pregnancies in the Danish National Birth Cohort	1996-2002	Maternal PFAS serum concentrations	Thyroid disruption	An increase in PFDA (per interquartile range) was associated with higher free T4 concentrations before gestational week 10; OR=1.46 (95% CI: 1.04, 2.05).	PFOS, PFOA, PFHxS, PFNA, PFHpS	Inoue et al., 2019 ⁶
Cohort	3629 participants from NHANES	2005-2014	Serum PFAS concentrations	Total cholesterol (TC), low density lipoprotein cholesterol (LDL), high density lipoprotein cholesterol (HDL), and triglycerides.	An increase in PFDA (ng/mL) concentrations was associated with an increase in adjusted concentrations of TC for obese females; $\beta = 0.0247$, $p = 0.048$. An increase in PFDA (ng/mL) concentrations was positively associated with an increase in HDL and LDL levels for obese females; HDL: $\beta=0.0442$, $p=0.01$; LDL: $\beta=0.0397$, $p=0.047$. An increase in PFDA (ng/mL) concentrations was	Et-FOSAA, Me-FOSSA, PFOA, PFOS, PFHxS, PFHpA, PFBS, PFNA, PFDoA, PFUnDA,	Jain and Ducatman, 2019 ¹²

					negatively associated with triglycerides levels for obese females; $\beta = -0.0791$, $p = 0.01$. An increase in PFDA (ng/mL) concentrations was negatively associated with triglyceride levels in obese males ($\beta = -0.0508$, $p = 0.03$).		
Cohort	373 mother-child pairs from the Odense Child Cohort	2010-2012	Maternal serum PFAS concentrations	Hormonal developmental outcomes	A two-fold increase in maternal PFDA concentration was associated with a decrease by 19.6% (95% CI: -32.9%, -3.8%) in DHEA concentration among 4 month old girls. PFDA had no associations with concentrations of androgens or gonadotropins during early stages of puberty.	PFHxS, PFOS, PFOA, PFNA	Jensen et al., 2020 ⁶¹
Cohort	318 pregnant women within the Odense Child Cohort	2010-2012	Serum PFAS concentrations	Glycemic status (plasma glucose, serum insulin, and serum C-peptide)	PFDA was not associated with any glucose related outcomes.	PFHxS, PFOS, PFOA, PFNA	Jensen et al., 2018 ⁶²
Case-Control Study	2874 pregnant women within the Odense Child Cohort	2010-2012	Maternal serum PFAS concentrations	Miscarriage	PFDA was associated with a 2.67 increased risk of miscarriage (tertile 3 vs tertile 1 95% CI: 1.31, 5.44).	PFOA, PFOS, PFHxS, PFNA	Jensen et al., 2015 ⁶³
Cohort	1985 mother-infant pairs from Hokkaido, Japan	2003-2009	Maternal serum PFAS concentrations	Birth outcomes	PFDA (per Log10 unit) was inversely associated with birth weight (g); $\beta = -72.2$ (95% CI: -138.1, -6.3).	PFHxS, PFHxA, PFHpA, PFOS, PFOA, PFNA, PFUnDA, PFDoDA, PFTrDA, PFTeDA	Kashino et al., 2020 ¹⁴
Cross sectional study	378 ten-year-old's from Oslo, Norway.	1992-2009	Serum PFAS concentrations	Health outcomes	PFDA was not associated with lung function or asthma. PFDA (ng/mL) was inversely associated with	PFBA, PFPeA, PFHxA, PFDoDA, PFTrDA, PFDS,	Kvalem et al., 2020 ⁶⁴

					atopic dermatitis in girls; RR = 0.64 (95% CI: 0.42,0.98) per IQR.	MeFOSA, EtFOSA, PFOSA, PFOA, PFNA, PFUnDA, PFHxS, PFHpS	
Cohort	268 mother-child pairs from Ewha Birth and Growth Retrospective Cohort from Seoul, Korea	2006-2010	Cord serum PFAS concentrations	Genomic DNA and birth outcomes	Maternal PFDA concentrations (ng/mL, log transformed) were negatively associated with neonatal birth weight (g); adjusted $\beta = -101.24$ (95% CI: -184.80, -17.67), $p = 0.02$.	PFHxS, PFOA, PFOS, PFNA, PFUnDA, PFDoDA, PFTTrDA	Kwon et al., 2016 ¹⁵
Cohort	361 Children from the Environment and Development of Children Cohort	2012-2013	Serum PFAS concentrations	Growth outcome measurements	PFDA concentrations were inversely related to height at 2 years old for boys and girls combined (adjusted β per ln unit -0.44 (95% CI: -0.77, -0.10)). PFDA was not associated to weight for 2 year old boys and girls.	PFBS, PFHxS, PFOS, PFDS, PFBA, PFPeA, PFHxA, PFHpA, PFOA, PFNA, PFUnDA, PFDoDA, PFTTrDA, PFTeDA	Lee et al., 2018 ⁴⁶
Cohort	638 mother-child pairs from Odense Child Cohort	2010-2012	Maternal serum PFAS concentrations	Birth outcomes and anogenital distance (AGD)	PFDA was associated with a decrease in AGD (mm) in girls; p trend <0.05, $\beta = -1.3$ (95% CI: -2.8, 0.2). PFDA was not associated with AGD in boys.	PFOS, PFOA, PFHxS, PFNA,	Lind et al., 2017 ⁶⁵
Case study	1016 seventy-years-old participants from Uppsala, Sweden	2001-2004	Plasma PFAS concentrations	Cardiovascular outcomes	PFDA was not associated with plaques in the carotid artery. PFDA (ng/mL natural log transformed) was associated with intima-median grey scale median an indicator of atherosclerosis; $\beta = 7.456$ (95% CI: 2.622, 12.47).	PFHpA, PFNA, PFUnDA, PFHxS, PFOA, PFOSA, PFOS	Lind et al., 2017 ⁶⁶

Cohort	501 couples from Michigan and Texas	2005-2009	Serum PFAS concentrations	Menstrual cycle and fecundity	PFDA was associated with a 3% increase in average cycle length (+1 day); Acceleration Factor = 1.03 (95% Credible Interval: 1.00, 1.05). PFDA was not associated with fecundity.	PFNA, PFOA, PFOS, PFOSA, Et-PFOSA-AcOH, Me-PFOSA-AcOH	Lum et al., 2017 ⁶⁷
Cohort	3535 mother-child pairs from Danish National Birth Cohort	1996-2002	Maternal Plasma PFAS concentrations	Birth outcomes	PFDA was not associated with birth weight or gestational age.	PFOS, PFOA, PFHxS, PFNA, PFHpS,	Meng et al., 2018 ⁶⁸
Cohort	682 mother-child pairs from Boston, MA	1999-2002 Maternal: 2007-2010 Child	Maternal plasma PFAS concentrations at first prenatal visit (mean: 9.6 weeks gestation); Child plasma PFAS concentrations at (mean:7.7 years)	Total cholesterol (TC), high density lipoprotein cholesterol (HDL-C), low-density lipoprotein cholesterol (LDL-C) triglycerides (TG), and alanine aminotransferase (ALT).	PFDA was positively associated with a higher TC among children; β per IQR increment =6.8 mg/dL (95% CI: 3.6, 10.1). PFDA was positively associated with a higher LDL-C among children; β per IQR increment = 3.2mg/dL (95% CI: 0.6, 5.8). PFDA was associated with higher HDL-C among children; β per IQR increment = 4.3mg/dL (95% CI: 2.6, 6.0). PFDA was associated with a lower TC/HDL-C ratio among children; β per IQR increment = -10.1 (95% CI: -18.2, -2.1).	PFOS, PFOA, PFNA, PFHxS, EtFOSAA, MeFOSAA,	Mora et al., 2018 ¹⁶
Cohort	533 mother-child pairs from Shanghai-Minhang Birth Cohort	April-December 2012	Maternal serum PFAS concentrations at 12-16 weeks' gestation	Infant neuropsychological development	An increase in maternal PFDA concentrations (natural log transformed) were associated with an increased risk of the children having a development problem with their personal social skills RR = 1.66 (95% CI: 0.99, 1.36).	PFHxS, PFOS, PFOA, PFNA, PFUnA, PFDoA, PFTrDA	Niu et al., 2019 ⁶⁹
Cohort	656 mother-child pairs	1997 and 2000	Maternal serum PFAS concentrations;	Children's behavioral development	PFDA concentrations were associated with a 0.78 point increase in strength and difficulties	PFOA, PFOS, PFHxS, PFNA	Oulhote et al., 2016

	from the Faroe Islands		Child (age 5 and age 7) serum PFAS concentrations		questionnaire scores at five years old (95% CI: 0.01, 1.55).		
Cross sectional study	786 participants from Seoul, South Korea	2006-2015	Serum PFAS concentrations	Body weight, height, thyroid disruption, cholesterol levels, diabetes.	PFDA (ng/mL) was positively associated with total cholesterol (mg/dL) ($p < 0.05$ spearman's rank correlation). PFDA was not associated with hypercholesterolemia in men or women. PFDA was not associated with uric acid concentrations in men or women. PFDA was positively associated with free T4 concentrations (mg/dL) in men and women ($p < 0.05$ spearman's rank correlation). PFDA was not associated with diabetes in men or women.	PFBA, PFHxA, PFOA, PFNA, PFUnDA, PFDoA, PFHxS, PFOS	Seo et al., 2018 ¹⁷
Case Study	170 mother-child pairs from Haidian Maternal and Child Health Hospital in Beijing	February-June 2012	Cord serum PFAS concentrations	Birth outcomes	PFDA was not associated with birth weight, birth length, or ponderal index (measure of leanness based on mass and height).	PFHxS, PFOS, PFOA, PFNA, PFUnA, PFDoA	Shi et al., 2017 ³⁵
Cohort	628 mother-child from the healthy start cohort University of Colorado Hospital	2009-2014	Maternal serum PFAS concentrations	Birth outcomes, glucose levels, cholesterol, triglycerides	PFDA was not associated with birth weight or child adiposity. PFDA concentrations (ng/mL log-unit) were inversely associated with maternal glucose levels β value = -0.016 (95% CI: -0.026, -0.006).	PFHxS, PFOA, PFOS, PFNA	Starling et al., 2016 ⁷⁰
Cohort	500 mother-child pairs from Shanghai, China	April-December 2012	Maternal serum PFAS concentrations	Birth outcomes and anogenital distance (AGD)	PFDA concentrations (per ln unit) were inversely associated with anoscrotal distance (mm) at birth; β value = -0.58 (95% CI: -1.11, -	PFHxS, PFOS, PFOA, PFNA, PFUnA, PFDoA, PFTrDA,	Tian et al., 2019 ¹⁸

					0.06). PFDA concentrations (per ln unit) were inversely associated with anopenile distance (mm) at birth; β value = -0.63 (95% CI: -1.24, -0.01).	PFTeDA, PFHxDA,	
Cohort	559 children from Children's Health and Environment in the Faroe Islands.	1997-2000	Maternal serum PFAS concentration, childhood serum concentrations at age 5 and 13	Childhood asthma and allergies	A doubling of PFDA concentrations was associated with increased odds of asthma at age 5. PFDA had no associations with total Immunoglobulin E, positive skin prick test, allergic rhinoconjunctivitis, or atopic eczema.	PFOA, PFOS, PFHxS, PFNA	Timmerman et al., 2017 ⁷¹
Cohort	285 mothers and their infants from Taiwan	2000-2001	Maternal serum PFAS concentrations at third trimester	Thyroid disruption	1-ng/mL increase in PFDA was associated with a 0.002 uIU/mL increase of maternal total T3 concentrations; (95% CI: 0.000, 0.003). 1-ng/mL increase in PFDA was associated with a 0.017ng/dL decrease in cord total T3 concentrations; (95% CI: -0.028, -0.005). PFDA had no associations with maternal free T4, total T4, and TSH concentrations.	PFHxS, PFOA, PFOS, PFNA, PFUnDA, PFDoDA, PFHxA, PFHpA	Wang et al., 2014 ²⁰
Cohort	223 mothers and their term infants born in Taiwan	2000-2001	Maternal serum PFAS concentration from third-trimester	Small for gestational age (SGA) Mean childhood height and weight	1–ln-unit increase in prenatal PFDA (ng/mL) was associated with a 0.14 kg lower birth weight among infant girls; (95% CI: -0.26, -0.02). 1–ln-unit increase in prenatal PFDA concentrations (ng/mL) were associated with small for gestational age in infant girls; OR: 3.14 (1.07, 9.19). PFDA was not associated to birth weight, birth length, birth head circumference, or Small for	PFNA, PFOA, PFUnDA, PFDoDA	Wang et al., 2016 ¹⁹

					gestational age among infant boys. 1-In-unit increase in prenatal PFDA concentration (ng/mL) were associated with lower weight and height scores during early childhood in girls: weight z score; β value = -0.32 (95% CI: -0.63, 0.00) and height z score; B value = -0.52 (95% CI: -0.80, -0.24). PFDA was not associated to weight or height scores during early childhood in boys.		
Case control study	161 Inuit women of Greenland	2000-2003 or 2011-2014	Serum PFAS concentrations	Breast cancer risk	An increase in PFDA was associated with an increased risk of breast cancer risk; tertile 3 vs tertile 1; OR = 2.36 (95% CI: 1.04, 5.36).	PFHpA, PFOA, PFNA, PFUnA, PFDoA, PFTrA, PFSA, PFHxS, PFPeA, PFTeA, PFBS, PFHpS, PFDS	Wielsoe et al., 2017 ⁹
Cohort	1533 mother-child pairs from Swedish Environmental, Longitudinal, Mother and child, Asthma and allergy (SELMA) Cohort	2007 to 2010	Maternal serum PFAS concentrations	Birth outcomes	PFDA concentrations (ng/mL per ln-unit) were negatively associated with body weight (g); β value = -58 (95% CI: -103, -13) for boys and girls. PFDA (ng/mL per ln-unit) was positively associated with SGA; OR = 1.46 (95% CI: 1.06, 2.01) for boys and girls.	PFOS, PFOA, PFHxS, PFNA, PFUnDA, PFHpA, PFDoDA	Wikstrom et al., 2019 ²¹
Case study	157 mother-child pairs from Beijing	January – March 2013	Maternal and cord Serum PFAS concentrations	Thyroid disruption	PFDA was inversely associated with TSH concentrations after adjusted for influential covariates.	PFHxA, PFOA, PFNA, PFUnA, PFDoA, PFHxS, PFOS	Yang et al., 2016 ⁷²
Cohort	351 mother-child pairs	2010-2013	Cord serum PFAS concentrations	Reproductive hormones,	PFDA was not associated with estradiol levels or testosterone	PFOS, PFOA, PFBS, PFDoA,	Yao et al., 2019 ³⁴

	from Laizhou, China			steroidogenic enzymes	levels. PFDA (ng/mL) was associated with higher placental P450arom levels (ng/mL) (a steroidogenic enzymes); β value = 0.18 (95% CI: 0.03, 0.33).	PFHpA, PFHxS, PFNA, PFOSA, PFUnA	
Cross sectional study	1202 participants from Shenyang, China	July-October 2016	Serum PFAS concentrations	Ophthalmic disruption (vision)	PFDA was not associated with eye disease.	PFHxA, PFDoDA, PFOS, PFBA, PFPA, PFHxS, PFHpA, PFNA, PFDS, PFUnDA, PFTeDA, PFTrDA	Zeeshan et al., 2020 ⁷³
<p>*This literature review is not exhaustive as the primary purpose of the search was to identify epidemiological studies that support toxicological findings.</p> <p>Epidemiologic terms: OR=odds ratio; AOR=adjusted odds ratio; RR=relative risk; 95% CI=95% confidence interval; r=Spearman correlation coefficient; β=regression coefficient</p> <p>PFAS acronyms: PFOA=perfluorooctanoic acid, PFNA=perfluorononanoic acid, PFDA=perfluorodecanoic acid, PFUnA= perfluoroundecanoic acid, PFTrDA=perfluorotridecanoic acid, PFTeDA=perfluorotetradecanoic acid, PFHxDA=perfluorohexadecanoic acid, PFHxS=perfluorohexane sulfonate, PFOS=perfluorooctane sulfonic acid, PFDS=perfluorodecanesulfonate, PFBS=perfluorobutane sulfonate, PFBA=perfluorobutanoic acid, PFPeA=perfluoropentanoic acid, PFHxA=perfluorohexanoic acid, PFHpA=perfluoroheptanoic acid, 6:2 FTSA=6:2 fluorotelomer sulfonates, NMeFOSAA=N-methyl perfluorooctanesulfonamidoacetate, EPAH=2-(N-ethyl-perfluorooctane sulfonamido)acetate, MPAH=2-(N-methyl-perfluorooctane sulfonamido) acetate, PFSA=perfluorooctane sulfonamide; PFHpS=perfluoroheptane sulfonic acid; PFDS=perfluorodecane sulfonic acid; EtPFOSAA= 2-(Nethyl-perfluorooctane sulfonamido) acetic acid</p>							

PFUnA | 2020

Substance Overview

Perfluoroundecanoic acid^a (PFUnA) is a chemical in a group of contaminants called per- and polyfluoroalkyl substances (PFAS). Because of its chemical properties of having both a water-loving group and water- and oil-repellant group on the same molecule, along with extreme stability under heat and pressure, PFUnA was introduced as an alternative to perfluorooctanoic acid (PFOA) once PFOA was phased out of production in 2004, to be used as a surfactant in chemical manufacturing and as an oil repellent in consumer products.^{1,2} From these uses, it can be found as an impurity in stain and grease repellants in commercial products like carpet and fabric, as a coating for packaging, and in some fire-fighting foams.^{1,3} PFAS can persist in the environment and in the human body for long periods of time.¹ PFAS with carbon chains consisting of seven or more carbon atoms, like PFUnA, cannot be easily broken down and excreted from the body; these compounds have shown to build up more and persist longer in the body than shorter chain PFAS.⁴⁻⁶ The half-life of PFUnA is estimated to be similar to other long-chain PFAS, with estimates ranging from 4-12 years.⁷

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard for PFUnA. DHS recommends an enforcement standard of 3 micrograms per liter (µg/L) for PFUnA. The recommended standard is based on a study that found that PFUnA can reduce birth weight and weight gain in newborn research animals.

DHS recommends that the preventive action limit for PFUnA be set at 20% of the enforcement standard because PFUnA has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture.

Current Standards	
Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

Recommended Standards	
Enforcement Standard:	3 µg/L
Preventive Action Limit:	0.6 µg/L

Health Effects

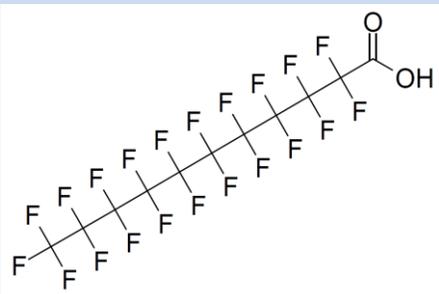
Some studies in humans exposed to multiple PFAS have shown that PFUnA may impact the immune system, heart and blood health, and physical development.^{1,8-13} Studies in research animals have

^a This scientific support document and the included groundwater standard recommendations also apply to anion salts of perfluoroundecanoic acid.

supported the findings, observing that PFUnA can have a negative impact on growth and development.^{2,14}

There is insufficient evidence to determine if PFUnA is carcinogenic (cancer), mutagenic (DNA damage), teratogenic (birth defects), or causes interactive effects in people or research animals. The EPA has not evaluated the carcinogenicity of PFUnA.¹⁵

Chemical Profile

PFUnA	
Structure:	
CAS Number:	2058-94-8
Formula:	C ₁₁ HF ₂₁ O ₂
Molar Mass:	564.09 g/mol
Synonyms:	PFUA; PFUnDA; C11-PFCA; perfluoro-n-undecanoic acid; heneicosaf fluoroundecanoic acid; 2,2,3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,11,11,11-heneicosaf fluoroundecanoic acid

Exposure Routes

PFAS, including PFUnA, can be released directly into the environment during the manufacture and use of PFAS and can be found in water or soil. PFAS can move between groundwater and surface water. Once in water, PFAS can travel long distances.^{1,16}

People can be exposed to PFAS by drinking water, eating food, and breathing in or accidentally swallowing soil or dust containing PFAS.¹⁴

Current Standard

Wisconsin does not currently have groundwater standards for PFUnA.¹⁷

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

EPA Oral Reference Dose:	N/A
--------------------------	-----

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

None Available

Literature Search

Literature Search Dates:	1900 – 2019
Key studies found?	Yes
Critical studies identified?	Yes

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for PFUnA.¹⁸

Health Advisory

The EPA has not established health advisories for PFUnA.¹⁹

Drinking Water Concentration (Cancer Risk)

The EPA has not established a drinking water concentration based on a cancer risk level determination for PFUnA.¹⁵

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for PFUnA.²⁰

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Information System (IRIS) program.

EPA Oral Reference Dose

The EPA does not have an oral reference dose for PFUnA.¹⁵

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If we determine that something is carcinogenic and there is no federal number or ADI from the EPA, DHS must set the standard at a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of PFUnA, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of PFUnA. If so, we look to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA and International Agency for Research on Cancer (IARC) have not evaluated the carcinogenicity of PFUnA.^{15,21}

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for PFUnA.¹⁵

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information that was not considered when the value was established and indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For PFUnA, we searched for values that had been published on or before November 2019. While the Agency for Toxic Substances and Disease Registry (ATSDR) reviewed the toxicity of PFUnA in 2018, they did not establish any guidance values for PFUnA, as the identified critical studies did not record animal serum levels, thus a human equivalent dose could not be calculated.^{1b}

Literature Search

Our literature review focused on relevant scientific literature on the health effects of PFUnA published on or before November 2019. We looked for studies related to PFUnA toxicity or effects on a disease state in which information on exposure or dose was included as part of the study.^c Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

Eight toxicity studies were returned by the search engines. We excluded studies on non-mammalian or cell systems, non-oral exposure routes, and those that did not evaluate health risks from further review. After applying these exclusion criteria, we located one key toxicity study on PFUnA (summarized in Table A-1). To be considered a critical toxicity study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and

b ATSDR stated that they did not identify any intermediate-duration oral studies for PFUnA in their literature review. While they located one chronic duration study, they did not establish a minimum risk level for PFUnA because this study did not measure serum levels.

c We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

Title/abstract: PFUA or PFUnA or PFUnDA or 2058-94-8 or "perfluoroundecanoic acid" or "perfluoroundecanoate"

Language: English

We also searched online for toxicity studies published by national research programs.

relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.^{d,e} We reviewed the key study and confirmed that it met the criteria to be considered a critical toxicity study (see Table A-2).

In our search, we also located 40 epidemiology studies (See Table A-3 for a summary of recent studies). While multiple potential exposure sources and the ability for other PFAS compounds to cause similar health effects preclude using these data to establish a health-based value, such studies are helpful in identifying the crucial effects and ensuring that the animal data used to establish the standard is relevant to people.

Critical Toxicity Studies

We calculated an acceptable daily intake (ADI) for each study/effect. The ADI is the estimated amount of PFUnA that a person can be exposed to every day and not experience health effects. Since there is only one study investigating the half-life in humans (4.5 – 12 years we calculated ADI by dividing a toxicity value from either a no-observed adverse effect level (NOAEL), lowest observed adverse effect level (LOAEL), or benchmark dose (BMD) identified in a study by a factor accounting for various sources of scientific uncertainty.^{f,7} Uncertainty factors were included, as appropriate, to account for differences between humans and animals, differences between healthy and sensitive human populations, using data from short-term experiments to protect against effects from long-term exposure, and using data where a health effect was observed to estimate the level that does not cause an effect. To ensure appropriate protection, we have chosen to not use studies that have significant uncertainty as the basis for the recommended enforcement standards.^g This approach is consistent with that taken by EPA when establishing oral reference doses.²³

Takahashi et al., 2014

Takahashi et al. exposed male and female rats to different concentrations of PFUnA (0, 0.1, 0.3, and 1.0 milligrams of PFUnA per kilogram body weight per day or mg/kg-d) by gavage for 42 days in males and from 14 days pre-mating through 4 days after lactation in females.² A subset of males and females in the 0 and 1.0 mg/kg-d dose groups were withheld treatment for an additional 14 day recovery period (see

d Appropriate toxicity values include the no observable adverse effect level (NOAEL), lowest observable adverse effect level (LOAEL), and benchmark dose (BMD).

e Due to the limited availability of data for this substance, we considered a study to be of an appropriate duration if animals were exposed for at least 28 days or if the exposure occurred during pregnancy. We accounted for differences in exposure duration through the use of an uncertainty factor.

f The NOAEL is the highest dose tested that did not cause an adverse effect, the LOAEL is the lowest dose tested that caused an adverse effect, and the BMD is an estimation of the dose that would cause a specific level of response (typically 5 or 10%)

g DHS considers a study to have significant uncertainty if the total uncertainty factors is greater than 3,000.

Takahashi et al. for information on recovery group effects). In males, PFUnA affected blood and biochemistry parameters and impacted the liver, spleen, and stomach at the highest dose (Table 1). In addition to these statistically significant findings, the study observed liver cell damage and stomach erosion in rats treated with 1 mg/kg-d PFUnA.

Table 1. Statistically Significant Effects Observed in Males in Takahashi et al., 2014

Effects observed in males		Dose (mg/kg-d)		
		0.1	0.3	1.0
Blood	Decreased cell volume			✓
	Decreased hemoglobin			✓
	Increased platelet count	*		✓
	Decreased coagulation			✓
	Decreased fibrinogen			✓
Metabolism	Decreased serum phospholipid		*	
	Decreased serum calcium			✓
Liver	Increased absolute liver weight			✓
	Increased relative liver weight		✓	✓
	Increased ALT			✓
	Increased ALP			✓
	Decreased serum total cholesterol		*	
	Decreased serum albumin			✓
	Increased serum albumin/globulin ratio		*	
	Decreased serum total protein			✓
Kidney	Increased blood urea nitrogen			✓
Spleen	Decreased absolute and relative spleen weight			✓
ALT = alanine aminotransferase; ALP = alkaline phosphatase				

*The authors did not consider this effect to be treatment-related, biologically relevant, or adverse.

From this repeated dose experiment in males, the authors identified a NOAEL of 0.1 mg/kg-d based on hepatocellular hypertrophy induced by PPAR α at higher doses. Other effects seen at 0.1 or 0.3 mg/kg-d, such as the increased platelet count, or increased albumin/globulin ratio were disregarded as incidental, since they were very slight or did not occur in a dose-dependent manner. We determined the ADI using this NOAEL and a total uncertainty factor of 10,000 to account for differences between humans and research animals (10), differences among people (10), using a short-term study to extrapolate to long-term exposures (10), and the limited availability of information (10). While we obtained a candidate ADI of 10 ng/kg-d for PFUnA for males, this study was not used to establish to recommended enforcement standard due to significant uncertainty.

In females, PFUnA affected blood and biochemistry parameters and impacted the liver, spleen, and stomach at the highest dose (Table 2). In addition to these statistically significant findings, the study observed liver cell damage in rats treated at the highest dose.

Table 2. Statistically Significant Effects Observed in Females in Takahashi et al., 2014

Effects observed in females		Dose (mg/kg-d)		
		0.1	0.3	1.0
Blood	Increased cell volume			✓
	Increased hemoglobin			✓
	Decreased fibrinogen			✓
Metabolism	Decreased serum phospholipid		*	
	Increased serum chlorine			✓
Liver	Increased absolute and relative liver weight			✓
	Decreased serum total cholesterol		*	
	Decreased serum total protein	*		✓
	Increased blood urea nitrogen			✓
Development	Decreased body weight of male and female pups at PNDs 0 and 4			✓
PND = postnatal day				

*The authors did not consider this effect to be treatment-related, biologically relevant, or adverse.

From this reproductive and developmental experiment in females, the authors identified a NOAEL of 0.3 mg/kg-d due to the reduced body weight in pups at the highest dose. As above, the other significant effects (for example, decreased total protein) were disregarded as incidental, since they were only slight changes or did not occur in a dose-dependent manner. We estimated a candidate ADI of 300 ng/kg-d based on the NOAEL and a total uncertainty factor of 1000 to account for differences between humans and research animals (10), differences among people (10), and the limited availability of information (10).

Key health effects

At this time, we did not find studies to show that PFUnA has caused carcinogenic, mutagenic, teratogenic, or interactive effects in humans, research animals, or culture cells.

Discussion

In research animals, PFUnA has been shown to impact the liver and spleen, decrease the weight of offspring at birth and during early development, and alter a variety of blood and biochemistry parameters, consistent with effects seen by other long-chain PFAS.^{1,2,24} While studies on the effects of PFUnA among people are limited and mixed, data from available studies suggest that PFUnA may impact fetal and newborn growth, the cardiovascular system, immune response, and thyroid function.^{8,11,25-27} Data on half-life are limited, but PFUnA appears to have similarly long half-lives in humans as other long-chain PFAS, potentially longer than a decade, indicating that the body burden will continue to grow if exposure continues.⁷

A number of studies have demonstrated that liver effects caused by PFAS occur primarily through activation of the nuclear hormone receptor, PPAR α (peroxisome proliferator-activated receptor alpha).²⁸⁻³² Nuclear receptors regulate gene expression and PPAR α regulates the expression of genes involved in lipid and cholesterol metabolism.³³ While PPAR α receptors are found in rodents and humans, levels of these receptors are much higher in rats and mice than in monkeys and humans.³³ This means that effects caused by activation of PPAR α are more likely to occur in rodents than they are to occur in people. As such, the effects on the liver observed in the critical study reviewed here are likely not relevant to humans.

Other studies have demonstrated associations between other long-chain PFAS and birth weight and development, both in humans and research animals.¹ The evidence in animals and limited evidence in humans suggests that PFUnA may have consistent effects to these other PFAS. Related developmental effects have been observed in research animals, including delays in hormone and motor development, though these effects have either not been seen or not been studied for PFUnA.¹

Standard Selection

DHS recommends an enforcement standard of 3 $\mu\text{g/L}$ for PFUnA.

There are no federal numbers and no state drinking water standard for PFUnA. Additionally, the EPA has not evaluated the carcinogenicity or established an ADI (oral reference dose) for PFUnA.

However, we found a 2014 study that evaluated the toxicity of PFUnA in rats.² In calculating the ADI as specified in s. 160.13, Wisc. Statute, DHS used the NOAEL based on adverse effects on birth weight and body weight gain in pups, because low birth weight has been demonstrated to increase risk of newborn mortality and increase the risk for other diseases later in life, such as diabetes, cardiovascular diseases, and asthma.³⁴ Further, a human study has demonstrated that there may be an association between PFUnA and birth weight and weight gain.⁸ As described above, we obtained an ADI of 0.0003 mg/kg-d (300 ng/kg-d) as described above. To determine the recommended enforcement standard, DHS used the ADI, and, as required by Ch. 160, Wis. Stats., a body weight of 10 kg, a water consumption rate of 1 L/d, and a relative source contribution of 100%.

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information

DHS recommends a preventive action limit of 0.6 $\mu\text{g/L}$ for PFUnA.

DHS recommends that the preventive action limit for PFUnA be set at 20% of the enforcement standard because PFUnA has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people or research animals.¹

Prepared by Nathan Kloczko, MPH and Sarah Yang, Ph.D.

Wisconsin Department of Health Services

References

1. ATSDR. Toxicological Profile for Perfluoroalkyls - Draft for Public Comment. In: Registry AftSaD, ed. Atlanta, GA2017.
2. Takahashi M, Ishida S, Hirata-Koizumi M, Ono A, Hirose A. Repeated dose and reproductive/developmental toxicity of perfluoroundecanoic acid in rats. *J Toxicol Sci.* 2014;39(1):97-108.
3. (ITRC) ITRC. History and Use of Per- and Polyfluoroalkyl Substances (PFAS). https://pfas-1.itrcweb.org/wp-content/uploads/2017/11/pfas_fact_sheet_history_and_use_11_13_17.pdf. Published 2017. Accessed Oct 2019.
4. Kudo N, Suzuki E, Katakura M, Ohmori K, Noshiro R, Kawashima Y. Comparison of the elimination between perfluorinated fatty acids with different carbon chain length in rats. *Chem Biol Interact.* 2001;134(2):203-216.
5. Kudo N, Suzuki-Nakajima E, Mitsumoto A, Kawashima Y. Responses of the liver to perfluorinated fatty acids with different carbon chain length in male and female mice: in relation to induction of hepatomegaly, peroxisomal beta-oxidation and microsomal 1-acylglycerophosphocholine acyltransferase. *Biol Pharm Bull.* 2006;29(9):1952-1957.
6. Ohmori K, Kudo N, Katayama K, Kawashima Y. Comparison of the toxicokinetics between perfluorocarboxylic acids with different carbon chain length. *Toxicology.* 2003;184(2-3):135-140.
7. Zhang Y, Beesoon S, Zhu L, Martin JW. Biomonitoring of perfluoroalkyl acids in human urine and estimates of biological half-life. *Environ Sci Technol.* 2013;47(18):10619-10627.
8. Lewis M, Kim MH, Liu EJ, Wang N, Chu KH. Biotransformation of 6:2 polyfluoroalkyl phosphates (6:2 PAPs): Effects of degradative bacteria and co-substrates. *J Hazard Mater.* 2016;320:479-486.
9. Huang M, Jiao J, Zhuang P, Chen X, Wang J, Zhang Y. Serum polyfluoroalkyl chemicals are associated with risk of cardiovascular diseases in national US population. *Environ Int.* 2018;119:37-46.
10. Lind PM, Salihovic S, van Bavel B, Lind L. Circulating levels of perfluoroalkyl substances (PFASs) and carotid artery atherosclerosis. *Environ Res.* 2017;152:157-164.
11. Chen MH, Ha EH, Wen TW, et al. Perfluorinated compounds in umbilical cord blood and adverse birth outcomes. *PLoS One.* 2012;7(8):e42474.

12. Impinen A, Nygaard UC, Lodrup Carlsen KC, et al. Prenatal exposure to perfluoroalkyl substances (PFASs) associated with respiratory tract infections but not allergy- and asthma-related health outcomes in childhood. *Environ Res.* 2018;160:518-523.
13. Impinen A, Longnecker MP, Nygaard UC, et al. Maternal levels of perfluoroalkyl substances (PFASs) during pregnancy and childhood allergy and asthma related outcomes and infections in the Norwegian Mother and Child (MoBa) cohort. *Environ Int.* 2019;124:462-472.
14. Bodin J, Groeng EC, Andreassen M, Dirven H, Nygaard UC. Exposure to perfluoroundecanoic acid (PFUnDA) accelerates insulinitis development in a mouse model of type 1 diabetes. *Toxicol Rep.* 2016;3:664-672.
15. USEPA. IRIS Assessments. https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm. Published 2019. Accessed.
16. ITRC. Environmental Fate and Transport for Per- and Polyfluoroalkyl Substances. In: Council ITR, ed2018.
17. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 1402017*.
18. USEPA. National Primary Drinking Water Regulations. <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>. Published 2018. Accessed.
19. USEPA. Drinking Water Contaminant Human Health Effects Information. <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>. Published 2019. Accessed.
20. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 8092018*.
21. IARC. List of Classification, Volumes 1-123. <https://monographs.iarc.fr/list-of-classifications-volumes/>. Published 2018. Accessed May 17, 2019.
22. Ritter L, Totman C, Krishnan K, Carrier R, Vezina A, Morisset V. Deriving uncertainty factors for threshold chemical contaminants in drinking water. *Journal of toxicology and environmental health Part B, Critical reviews.* 2007;10(7):527-557.
23. USEPA. A Review of the Reference Dose and Reference Concentration Processes. 2002(EPA/630/P-02/002F).
24. (NTP) NTP. NTP technical report on the toxicity studies of perfluoroalkyl carboxylates (perfluorohexanoic acid, perfluorooctanoic acid, perfluorononanoic acid, and perfluorodecanoic acid) administered by gavage to Sprague Dawley (Hsd:Sprague Dawley SD) rats. In. Vol Toxicity Report 97. Research Triangle Park, NC: National Toxicology Program2019.
25. Bach CC, Bech BH, Nohr EA, et al. Perfluoroalkyl Acids in Maternal Serum and Indices of Fetal Growth: The Aarhus Birth Cohort. *Environ Health Perspect.* 2016;124(6):848-854.
26. Lee ES, Han S, Oh JE. Association between perfluorinated compound concentrations in cord serum and birth weight using multiple regression models. *Reprod Toxicol.* 2016;59:53-59.

27. Lenters V, Portengen L, Rignell-Hydbom A, et al. Prenatal Phthalate, Perfluoroalkyl Acid, and Organochlorine Exposures and Term Birth Weight in Three Birth Cohorts: Multi-Pollutant Models Based on Elastic Net Regression. *Environ Health Perspect.* 2016;124(3):365-372.
28. Das KP, Wood CR, Lin MT, et al. Perfluoroalkyl acids-induced liver steatosis: Effects on genes controlling lipid homeostasis. *Toxicology.* 2017;378:37-52.
29. Rosen MB, Das KP, Rooney J, Abbott B, Lau C, Corton JC. PPAR alpha-independent transcriptional targets of perfluoroalkyl acids revealed by transcript profiling. *Toxicology.* 2017;387:95-107.
30. Albrecht PP, Torsell NE, Krishnan P, et al. A species difference in the peroxisome proliferator-activated receptor alpha-dependent response to the developmental effects of perfluorooctanoic acid. *Toxicological sciences : an official journal of the Society of Toxicology.* 2013;131(2):568-582.
31. Palkar PS, Anderson CR, Ferry CH, Gonzalez FJ, Peters JM. Effect of prenatal peroxisome proliferator-activated receptor alpha (PPARalpha) agonism on postnatal development. *Toxicology.* 2010;276(1):79-84.
32. Wolf DC, Moore T, Abbott BD, et al. Comparative hepatic effects of perfluorooctanoic acid and WY 14,643 in PPAR-alpha knockout and wild-type mice. *Toxicologic pathology.* 2008;36(4):632-639.
33. Hall AP, Elcombe CR, Foster JR, et al. Liver hypertrophy: a review of adaptive (adverse and non-adverse) changes--conclusions from the 3rd International ESTP Expert Workshop. *Toxicol Pathol.* 2012;40(7):971-994.
34. (WHO) WHO. *WHA Global Nutrition Targets 2025: Low Birth Weight Policy Brief.* 2014.
35. Mobacke I, Lind L, Dunder L, Salihovic S, Lind PM. Circulating levels of perfluoroalkyl substances and left ventricular geometry of the heart in the elderly. *Environ Int.* 2018;115:295-300.
36. Huang R, Chen Q, Zhang L, et al. Prenatal exposure to perfluoroalkyl and polyfluoroalkyl substances and the risk of hypertensive disorders of pregnancy. *Environ Health.* 2019;18(1):5.

Appendix A. Toxicity Data

Table A-I. PFUnA Toxicity Studies from Literature Review

Study Type	Species	Duration	Doses (mg/kg-d)	Route	Endpoints	Toxicity Value (mg/kg-d)	Reference
Short-Term	Sprague-Dawley Rats	42 days	0.1, 0.3, 1.0	Gavage	Liver cell damage	NOAEL: 0.1	Takahashi et al., 2014 ⁽²⁾
Developmental	Sprague-Dawley Rats	14d pre-mating to day 4 of lactation (41-46 days)	0.1, 0.3, 1.0	Gavage	Reduced body weight at birth and weight gain at day four	NOAEL: 0.3	Takahashi et al., 2014 ⁽²⁾

NOAEL: No observed adverse effect level

Table A-2. Critical Study Selection for PFUnA

Reference	Appropriate duration?	Effects consistent with other studies?	Effects relevant to humans?	Number of Doses	Toxicity value identifiable?	Critical study?
Takahashi et al., 2014 ⁽²⁾	✓	✓	✓	3	✓	Yes

To be considered a critical study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.

Table A-3. Recent PFUnA Epidemiological Studies from Literature Review

Study Type	Population	Time period	Exposure	Outcomes	Results	Other PFAS Studied	Reference
Cohort	223 Taiwanese mothers and children	2000-2001	Third trimester maternal serum PFAS concentrations	Fetal and postnatal growth	In females, increased prenatal PFUnA was associated with decreased birth weight (-0.06 kg per ln-unit increase in concentration, 95% CI -0.11, -0.01) and increased odds of small for gestational age (OR 1.83, 95% CI 1.01, 3.32). Likewise, higher PFUnA concentrations were associated with decreased weight and height during childhood in females.	PFOA, PFNA, PFDA, PFDoA	Wang et al., 2016 ⁽⁸⁾
Cross-sectional	1016 Swedish adults > 70 years	2001-2004	Plasma PFAS concentrations	Carotid artery atherosclerosis	PFUnA was significantly associated with the intima-media complex (a marker of lipid infiltration in arteries) and odds of carotid plaque in women (OR 1.59, 95% CI 1.03-2.43)	PFHpA, PFNA, PFDA, PFHxS, PFOS, PFOA, PFOSA	Lind et al., 2017 ⁽¹⁰⁾
Cross-sectional	10,859 individuals from NHANES	1999-2014	Serum PFAS concentrations	Cardiovascular disease, congestive heart failure, coronary heart disease, angina pectoris, heart attack, and stroke.	PFUnA was significantly associated with elevated odds of total cardiovascular disease (Q4 vs Q1 OR 1.47, 95% CI 1.07-2.04; p for trend 0.011) and coronary heart disease (OR 2.02, 95% CI 1.36-3.00).	PFOA, PFOS, PFHxS, PFDA, PFHpA, PFOSA, PFNA, PFDoA	Huang, 2018 ⁽⁹⁾
Cohort	801 Swedish adults > 70 years	2001-2004	Plasma PFAS concentrations	Left ventricular geometry	PFUnA concentrations were negatively related to lower relative wall thickness and higher left ventricular end-diastolic volume.	PFHpA, PFHxS, PFOS, PFDA, PFOSA	Mobacke, 2018 ⁽³⁵⁾
Cohort	641 children from Norway	1992-1993	Cord blood plasma PFAS concentrations	Obstructive airway disease, atopic dermatitis, allergic sensitization,	PFUnA concentrations were associated with more episodes of common cold (0-2 years) and lower respiratory tract infections (0-10 years). PFUnA	PFOS, PFOA, PFOSA, PFHxS, PFNA	Impinen, 2018 ⁽¹²⁾

				respiratory tract infections	concentrations were also associated with higher odds for wheeze before 3 years (OR 1.34, 95% CI 1.01, 1.77), but lower odds of reduced lung function at birth (OR: 0.49, 95% CI: 0.26, 0.92).		
Cohort	792 Norwegian mother-child pairs	1999-2008	Maternal plasma PFAS levels during pregnancy	Asthma, allergic conditions, infections	PFUnA was inversely associated with odds of asthma, wheeze, and atopic eczema.	PFOS, PFOA, PFHxS, PFNA, PFHpS	Impinen, 2019 ⁽¹³⁾
Cross-sectional	674 women giving birth in Shanghai hospitals	2011-2012	Umbilical cord blood plasma PFAS concentrations	Hypertensive disorders of pregnancy (HDP): gestational hypertension and preeclampsia	PFUnA was most strongly negatively associated with preeclampsia in an elastic net regression model.	PFOS, PFNA, PFHxS, PFBS, PFOA, PFDA, PFDoA	Huang, 2019 ⁽³⁶⁾
Cross-sectional	2975 individuals from NHANES	2007-2014	Serum PFAS concentrations	Metabolic syndrome	PFUnA was associated with a decreased risk of metabolic syndrome (OR 0.69, 95% CI 0.54-0.88 for linear model)	PFDA, PFOS, PFOA, PFHxS, PFNA	Christensen, 2019 ⁽³⁶⁾
<p>Epidemiologic terms: OR=odds ratio; AOR=adjusted odds ratio; RR=relative risk; 95% CI=95% confidence interval; r=Spearman correlation coefficient; β=regression coefficient</p> <p>PFAS acronyms: PFOA=perfluorooctanoic acid, PFNA=perfluorononanoic acid, PFDA=perfluorodecanoic acid, PFUnA= perfluoroundecanoic acid, PFDoA=perfluorododecanoic acid, PFTriA=perfluorotetradecanoic acid, PFHxDA=perfluorohexadecanoic acid, PFHxS=perfluorohexane sulfonate, PFOS=perfluorooctane sulfonic acid, PFDS=perfluorodecanesulfonate, PFBS=perfluorobutane sulfonate, PFBA=perfluorobutanoic acid, PFPeA=perfluoropentanoic acid, PFHxA=perfluorohexanoic acid, PFHpA=perfluoroheptanoic acid, 6:2 FTSA=6:2 fluorotelomer sulfonates, NMeFOSAA=N-methyl perfluorooctanesulfonamidoacetate, EPAH=2-(N-ethyl-perfluorooctane sulfonamido)acetate, MPAH=2-(N-methyl-perfluorooctane sulfonamido) acetate, PFSA=perfluorooctane sulfonamide; PFHpS=perfluoroheptane sulfonic acid; PFDS=perfluorodecane sulfonic acid; EtPFOSAA= 2-(Nethyl-perfluorooctane sulfonamido) acetic acid</p>							

PFBS | 2020

Substance Overview

Perfluorobutanesulfonic acid^a (PFBS) is a chemical in a group of contaminants called per- and polyfluoroalkyl substances (PFAS). PFAS are manmade chemicals that have been used in industry and consumer products since the 1940s. Because of their unique physical and chemical properties, PFAS can be found in a variety of commercial products such as paper and textile coatings, food packaging, surfactants, repellants, and fire-fighting foams.^{1,2} Because PFBS has been shown to stay in the human body for significantly less time than other PFAS, it has been widely used as a substitute compound for longer-lasting PFAS.³⁻⁵

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard for PFBS. DHS recommends an enforcement standard of 450 micrograms per liter (µg/L) for PFBS. The recommended standard is based on a study that found that PFBS can increase body weight in female mice and cause kidney damage in male and female mice.⁶

DHS recommends that the preventive action limit for PFBS be set at 20% of the enforcement standard because PFBS has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture studies.

Current Standards

Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

Recommended Standards

Enforcement Standard:	450 µg/L
Preventive Action Limit:	90 µg/L

Health Effects

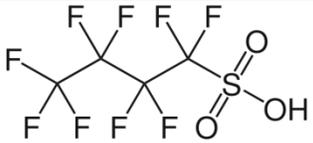
Studies among people exposed to high levels of PFAS have shown that PFBS can increase the risk of total^b cardiovascular disease; infertility and high blood pressure disorders in pregnant women, including preeclampsia; and asthma in male children.⁷⁻¹⁰ Studies in research animals found that high levels of PFBS can cause damage to the liver and kidneys, alter blood chemistry and thyroid hormone levels, and affect development.^{6,11-13} PFBS has not been shown to cause carcinogenic, mutagenic, teratogenic, or

^a This scientific support document and the included groundwater standard recommendations also apply to anion salts of perfluorobutanesulfonic acid.

^b Total cardiovascular disease includes congestive heart failure, coronary artery disease, angina pectoris, heart attack, and stroke.

interactive effects in people, research animals, or cell culture studies.^c The EPA has not evaluated the carcinogenicity of PFBS.¹⁴

Chemical Profile

Structure:	
CAS Number:	375-73-5
Formula:	C ₄ HF ₉ O ₃ S
Molar Mass:	300.09 g/mol
Synonyms:	Nonafluorobutanesulphonic acid 1-Butanesulfonic Acid, nonafluoro- 1-Butanesulfonic acid, 1,1,2,2,3,3,4,4,4-nonafluoro- 1,1,2,2,3,3,4,4,4-Nonafluorobutane-1-sulfonic acid C4 Sulfonate 1-Perfluorobutanesulfonic acid FC-98

Exposure Routes

PFAS, including PFBS, can be released directly into the environment during the manufacture and use of PFAS and can be found in water or soil. PFAS can move between groundwater and surface water. Once in water, PFAS can travel long distances.^{5,15}

People can be exposed to PFAS by drinking water, eating food, and breathing in or accidentally swallowing soil or dust containing PFAS.¹⁴

Current Standard

Wisconsin does not currently have groundwater standards for PFBS.¹⁶

^c Carcinogenic effects means the substance can cause cancer; mutagenic effects means the substance can cause DNA damage; teratogenic means the substance can cause birth defects; and interactive effects mean the substance can affect the toxicity of another substance or its toxicity can be affected by another substance.

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

Draft EPA Oral Reference Doses:	0.04 mg/kg-d	Subchronic
	0.1 mg/kg-d	Subchronic
	0.01 mg/kg-d	Chronic
	0.01 mg/kg-d	Chronic

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

ATSDR Oral Minimum Risk Level:	N/A
--------------------------------	-----

Literature Search

Literature Search Dates:	1900–2019
Total studies evaluated:	Approximately 100
Key studies found?	Yes
Critical studies identified?	Yes

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for PFBS.¹⁷

Health Advisory

The EPA has not established health advisories for PFBS.¹⁸

Drinking Water Concentration (Cancer Risk)

The EPA has not established drinking water concentration based on a cancer risk determination for PFBS.¹⁴

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for PFBS.¹⁹

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Information System (IRIS) program.

Draft EPA Oral Reference Doses

In 2018, the EPA issued draft subchronic and chronic oral reference doses for PFBS (Table 1; note that K⁺PFBS, or potassium perfluorobutane sulfonate, is the related salt of PFBS and is considered the same as PFBS for the purposes of this document)^{20,d}.

Table 1. Summary of draft EPA candidate oral references doses for K⁺PFBS

Oral toxicity value type	Critical effect	Study Type	Study	Reference dose
Subchronic	Thyroid	Gestational exposure	Feng et al., 2017 ¹²	0.04 mg/kg-d
Subchronic	Kidney	Two-generation reproductive	Lieder et al., 2009b ⁶	0.1 mg/kg-d
Chronic	Thyroid	Gestational exposure	Feng et al., 2017 ¹²	0.01 mg/kg-d
Chronic	Kidney	Two-generation reproductive	Lieder et al., 2009b ⁶	0.01 mg/kg-d

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If we determine that something is carcinogenic and there is no federal number or ADI from the EPA, we must recommend an enforcement standard that corresponds to a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

d See the Critical Study section below for more details on these studies.

To evaluate the oncogenic potential of PFBS, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of PFBS. If so, we look to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA and International Agency for Research on Cancer (IARC) have not evaluated the carcinogenicity of PFBS.^{14,21}

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for PFBS.¹⁴

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information not considered when the value was established that indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For PFBS, we searched for values that been published before or during September 2019. We did not find any relevant guidance values for PFBS.

Literature Search

Our literature review focused on relevant scientific literature on the health effects of PFBS published before or during September 2019. We looked for studies related to PFBS toxicity or effects on a disease state in which information on exposure or dose was included as part of the study.^e Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

Approximately 100 toxicity studies were returned by the search engines. We excluded studies on non-mammalian or cell systems, non-oral exposure routes, and those that did not evaluate health risks from further review. After applying these exclusion criteria, we located four key toxicity studies on PFBS

^e We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

Title/abstract: PFBS or "Perfluorobutanesulfonic acid" or "Perfluorobutane sulfonate"

Subject area: toxicology

Language: English

We also searched online for toxicity studies published by national research programs.

(summarized in Table A-1). To be considered a critical toxicity study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.^{f-g} Three studies met the criteria to be considered a critical toxicity study (see Table A-2).

In our search, we also located a handful of epidemiology studies (See Table A-3 for a summary). While multiple potential exposure sources and the ability for other PFAS compounds to cause similar health effects preclude using these data to establish a health-based value, such studies are helpful in identifying the critical effects and ensuring that the animal data used to establish the standard are relevant to people.

Critical Toxicity Studies

To compare between results from recently found studies and the study used to set the current enforcement standard, we calculated an acceptable daily intake (ADI) for each study/effect. The ADI is the estimated amount of PFBS that a person can be exposed to every day and not experience health impacts. As such, we calculated ADI by dividing a toxicity value from either a no-observed adverse effect level (NOAEL), lowest observed adverse effect level (LOAEL), or benchmark dose lower confidence level (BMDL) identified in a study by a factor accounting for various sources of scientific uncertainty.^h Uncertainty factors were included, as appropriate, to account for differences between humans and animals, differences between healthy and sensitive human populations, using data from short-term experiments to protect against effects from long-term exposure, and using data where a health effect was observed to estimate the level that does not cause an effect. To ensure appropriate protection, we have chosen to not use studies that have significant uncertainty as the basis for the recommended enforcement standards.ⁱ This approach is consistent with that taken by EPA when establishing oral reference doses.²²

Feng et al., 2017

Feng et al. exposed female mice to different concentrations of PFBS (0, 50, 200, and 500 milligrams of PFBS per kilogram body weight per day or mg/kg-d) through gavage during pregnancy (gestation days 1

f Appropriate toxicity values include the no observable adverse effect level (NOAEL), lowest observable adverse effect level (LOAEL), and benchmark dose (BMD).

g Due to the limited availability of data for this substance, we considered a study to have a duration appropriate for review if animals were exposed for at least 28 days or if the exposure occurred during pregnancy. We accounted for differences in exposure duration through the use of an uncertainty factor.

h The NOAEL is the highest dose tested that did not cause an adverse effect, the LOAEL is the lowest dose tested that caused an adverse effect, and the BMDL is the lower limit of a one-sided 95% confidence interval established using benchmark dose modeling. Benchmark dose modeling is considered the state of the science for establishing health-based values like an acceptable daily intake. Benchmark dose modeling takes account of all of the data for a particular effect from a particular experiment, allows for increased consistency, and can better account for statistical uncertainties.^{22,24-26}

i DHS considers a study to have significant uncertainty if the total uncertainty factors is greater than 3,000.

through 20).¹² The researchers evaluated the health of the exposed animals and that of their female offspring. They found that PFBS affected thyroid hormone levels in mothers and thyroid hormone levels, perinatal growth, pubertal onset, and reproductive organ development in offspring (Table 2).

Table 2. Statistically Significant Effects Observed in Feng et al., 2017 (12)

Effects observed in dams		Dose (mg/kg-d)		
		50	200	500
Endocrine	Lower serum triiodothyronine (T3)		✓	✓
	Lower serum and free thyroxine (T4)		✓	✓
	Higher serum thyroid-stimulating hormone (THS)		✓	✓
Effects observed in offspring		Dose (mg/kg-d)		
		50	200	500
Growth	Lower body weight		✓	✓
	Development	Delayed eye opening		✓
Delayed vaginal opening			✓	✓
Delayed first estrous			✓	✓
Lower relative ovarian weight			✓	✓
Lower relative uterine weight			✓	✓
Fewer follicles per ovary			✓	✓
Decreased uterine thickness			✓	✓
Prolonged diestrus stage			✓	✓
Endocrine	Lower serum estrogen		✓	✓
	Higher serum luteinizing hormone		✓	✓
	Lower serum progesterone		✓	✓
	Lower serum triiodothyronine (T3)		✓	✓
	Lower serum thyroxine (T4)		✓	✓

From this study, we identified a No Observable Adverse Effect Level (NOAEL) of 50 mg/kg-d based on effects on growth, development, and endocrine parameters at higher doses. We estimated a candidate ADI of 0.050 mg/kg-d PFBS from this study based on the NOAEL and a total uncertainty factor of 1000 to account for differences between people and research animals (10), differences among people (10), and the limited availability of information (10).

Lieder et al., 2009a

Lieder et al. exposed male and female rats to different concentrations of PFBS (0, 60, 200, and 600 mg/kg-d PFBS) through gavage for 90 days.¹³ They found that PFBS caused clinical toxicity, altered organ weights, and affected the blood and liver (Table 3). They also found that PFBS increased the incidence of lesions in the nasal cavity at 200 and 600 mg/kg-d in both sexes and caused hyperplasia of kidney epithelial cells and necrosis of stomach epithelial cells at 600 mg/kg-d in both sexes.

Table 3. Statistically Significant Effects Observed in Lieder et al., 2009a

Effects observed in males		Dose (mg/kg-d)		
		60	200	600

Spleen	Lower spleen weight	*	*	*
Blood	Lower blood hemoglobin		✓	✓
	Lower blood hematocrit		✓	✓
	Higher serum chloride values			✓
	Lower red blood cell counts			✓
Clinical observations	Red perioral substance			✓
	Urine-stained abdominal fur			✓
		Dose (mg/kg-d)		
Effects observed in females		60	200	600
Blood	Higher mean corpuscular hemoglobin concentration	*		
Liver	Lower serum total protein			✓
	Lower serum albumin			✓

* The authors did not consider this finding to be biologically meaningful or related to PFBS-treatment (see Lieder et al., 2009a for more details).¹³

From this study, we identified a NOAEL of 60 mg/kg-d based on clinical effects and effects on the blood. We estimated a candidate ADI of 0.020 mg/kg-d PFBS from this study based on the NOAEL and a total uncertainty factor of 3000 to account for differences between people and rodents (10), differences among people (10), use of a shorter duration study to protect against effects from long-term exposure (3), and the limited availability of information (10).

Lieder et al., 2009b

Lieder et al. also conducted a two-generation study in which they exposed male and female rats to different concentrations of PFBS (0, 30, 100, 300, and 1000 mg/kg-d PFBS) through gavage for 10 weeks.⁶ In this study, second-generation pups were not directly dosed but were studied through three weeks after birth. They found that PFBS exposure affected liver weight in parental generation males and body weight and onset of preputial separation in first generation males (Table 4). In addition to these statistically significant findings, among both parental and first generations, PFBS caused higher incidence of adaptive hepatocellular hypertrophy in males and microscopic findings in the medulla and papilla of the kidney in males and females at 300 and 1000 mg/kg-d.

Table 4. Statistically Significant Effects Observed in Lieder et al., 2009b (6)

		Dose (mg/kg-d)			
Effects observed in parental generation		30	100	300	1000
Liver	Increased absolute and relative liver weight (males)			✓	✓
Brain	Decreased absolute brain weight (males)	*			
	Decreased absolute brain weight (females)				✓
Reproduction	Lower testicular sperm count				*
		Dose (mg/kg-d)			
Effects observed in first generation pups		30	100	300	1000
Growth	Higher terminal body weight (females)	✓	✓	✓	✓
	Higher body weight at GD 21 (females)	✓	✓		

	Delayed preputial separation (males)	*			*
	Lower terminal body weight (males)				✓
Spleen	Increased absolute spleen weight (females)	*	*	*	
Development	Lower pup survival at PND 4 (viability index)	*			
	Lower pup survival at PND 21 (lactation index)		*	*	
Reproduction	Higher proportion of abnormal sperm cells				*
	Lower absolute weight of seminal vesicles with and without fluid				*
	Prolonged diestrus (>6 days)		*		*
Liver	Increased relative liver weight (males)				✓
		Dose (mg/kg-d)			
Effects observed in second generation pups		30	100	300	1000
Development	Lower pup survival at PND 21 (lactation index)				*

GD = gestation day; PND = postnatal day

* The authors did not consider this finding to be biologically meaningful or related to PFBS-treatment (see Lieder et al., 2009b for more details).⁶

In 2018, the Minnesota Department of Health established a benchmark dose lower confidence level (BMDL) of 45 mg/kg-d based on this study.²³ We estimated a candidate ADI of 0.045 mg/kg-d PFBS from this study based on the BMDL established by the Minnesota Department of Health and a total uncertainty factor of 1,000 to account for differences between humans and research animals (10), differences among people (10), and the limited availability of information (10).

Key Health Effects

We did not find any studies indicating that PFBS can cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture studies.

Discussion

While information about the health effects of PFBS exposure is limited, studies in research animals indicate that PFBS can affect health. These studies indicate that high levels of PFBS can affect male and female reproductive systems, impact growth and development (e.g., body weight), affect hormone levels and other components of the blood, cause liver and kidney damage and a variety of clinical effects.^{6,12,13} Studies among people exposed to high levels of PFAS have shown that PFBS can increase the risk of total cardiovascular disease^j; infertility and high blood pressure disorders in pregnant women, including preeclampsia; and asthma in male children.⁷⁻¹⁰

Other studies have demonstrated associations between other long-chain PFAS and birth weight and development, both in humans and research animals.⁵ Evidence in animals suggests that PFBS may have

^j Total cardiovascular disease includes congestive heart failure, coronary artery disease, angina pectoris, heart attack, and stroke.

consistent effects to these other PFAS, including developmental delays and abnormal birth weights and hormone levels.^{6,12} Related developmental effects have been observed in research animals, including delays in motor development, though these effects have not been observed or studied for PFBS.⁵

In the 2009b, Lieder et al., critical study, the researchers found that PFBS increased body weight in female mice.⁶ Other PFAS have been shown to affect body weight.⁵ Newer studies among people suggest that PFAS may cause or contribute to weight gain and obesity.²⁴⁻³⁴ Some of these studies among both sexes showed increases in body weight among females only, or stronger weight gain associations among females when compared to males.^{27,30,31,33}

A number of studies have demonstrated that liver effects caused by PFAS, like PFBA, occur primarily through activation of the nuclear hormone receptor, PPAR α (peroxisome proliferator-activated receptor alpha).³⁵⁻⁴⁴ Nuclear receptors regulate gene expression and PPAR α regulates the expression of genes involved in lipid and cholesterol metabolism.⁴⁵ While PPAR α receptors are found in rodents and humans, levels of these receptors are much higher in rats and mice than in monkeys and humans.⁴⁵ This means that effects caused by activation of PPAR α are more likely to occur in rodents than they are to occur in people. As such, the effects on the liver observed in the critical studies reviewed here are likely not relevant to humans.

Standard Selection

DHS recommends an enforcement standard of 450 $\mu\text{g/L}$ for PFBS.

There are no federal numbers and no state drinking water standard for PFBS. Additionally, the EPA has not evaluated the carcinogenicity or established an ADI (oral reference dose) for PFBS.

However, we found several critical studies evaluating the toxicity of PFBS. To calculate the ADI as specified in s. 160.13, Wisc. Statute, DHS selected the two-generation study by Lieder et al. as the critical study due to its robust two-generational design.⁶ We used the BMDL established by the Minnesota Department of Health and a total uncertainty factor of 1000 to obtain an ADI of 0.045 mg/kg-d as described above. To determine the recommended enforcement standard, DHS used the ADI, and, as required by Ch. 160, Wis. Stats., a body weight of 10 kg, a water consumption rate of 1 L/d, and a relative source contribution of 100%.

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information

DHS recommends a preventive action limit of 90 $\mu\text{g/L}$ for PFBS.

DHS recommends that the preventive action limit for PFBS be set at 20% of the enforcement standard because PFBS has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture studies.

References

1. Kissa E. *Fluorinated Surfactants and Repellants*. New York, NY: Marcel Dekker; 2001.
2. Calafat AM, Wong LY, Kuklennyik Z, Reidy JA, Needham LL. Polyfluoroalkyl chemicals in the U.S. population: data from the National Health and Nutrition Examination Survey (NHANES) 2003-2004 and comparisons with NHANES 1999-2000. *Environ Health Perspect*. 2007;115(11):1596-1602.
3. Olsen GW, Chang SC, Noker PE, et al. A comparison of the pharmacokinetics of perfluorobutanesulfonate (PFBS) in rats, monkeys, and humans. *Toxicology*. 2009;256(1-2):65-74.
4. Persson S, Rotander A, Karrman A, van Bavel B, Magnusson U. Perfluoroalkyl acids in subarctic wild male mink (*Neovison vison*) in relation to age, season and geographical area. *Environ Int*. 2013;59:425-430.
5. ATSDR. Toxicological Profile for Perfluoroalkyls - Draft for Public Comment. In: Registry AfTSD, ed. Atlanta, GA2017.
6. Lieder PH, York RG, Hakes DC, Chang SC, Butenhoff JL. A two-generation oral gavage reproduction study with potassium perfluorobutanesulfonate (K+PFBS) in Sprague Dawley rats. *Toxicology*. 2009;259(1-2):33-45.
7. Huang M, Jiao J, Zhuang P, Chen X, Wang J, Zhang Y. Serum polyfluoroalkyl chemicals are associated with risk of cardiovascular diseases in national US population. *Environ Int*. 2018;119:37-46.
8. Huang R, Chen Q, Zhang L, et al. Prenatal exposure to perfluoroalkyl and polyfluoroalkyl substances and the risk of hypertensive disorders of pregnancy. *Environmental Health*. 2019;18.
9. Wang B, Zhang R, Jin F, et al. Perfluoroalkyl substances and endometriosis-related infertility in Chinese women. *Environ Int*. 2017;102:207-212.
10. Zhu Y, Qin XD, Zeng XW, et al. Associations of serum perfluoroalkyl acid levels with T-helper cell-specific cytokines in children: By gender and asthma status. *Sci Total Environ*. 2016;559:166-173.
11. Bijland S, Rensen PCN, Pieterman EJ, et al. Perfluoroalkyl Sulfonates Cause Alkyl Chain Length-Dependent Hepatic Steatosis and Hypolipidemia Mainly by Impairing Lipoprotein Production in APOE*3-Leiden CETP Mice. *Toxicological Sciences*. 2011;123(1):290-303.
12. Feng XJ, Cao XY, Zhao SS, et al. Exposure of Pregnant Mice to Perfluorobutanesulfonate Causes Hypothyroxinemia and Developmental Abnormalities in Female Offspring. *Toxicological Sciences*. 2017;155(2):409-419.

13. Lieder PH, Chang SC, York RG, Butenhoff JL. Toxicological evaluation of potassium perfluorobutanesulfonate in a 90-day oral gavage study with Sprague-Dawley rats. *Toxicology*. 2009;255(1-2):45-52.
14. USEPA. IRIS Assessments. https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm. Published 2019. Accessed.
15. ITRC. Environmental Fate and Transport for Per- and Polyfluoroalkyl Substances. In: Council ITR, ed2018.
16. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 140*2017.
17. USEPA. National Primary Drinking Water Regulations. <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>. Published 2018. Accessed.
18. USEPA. Drinking Water Contaminant Human Health Effects Information. <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>. Published 2019. Accessed.
19. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 809*2018.
20. USEPA. Human Health Toxicity Values for Perfluorobutane Sulfonic Acid (CASRN 375-73-5) and Related Compound Potassium Perfluorobutane Sulfonate (CASRN 29420-49-3). In:2018.
21. IARC. List of Classification, Volumes 1-123. <https://monographs.iarc.fr/list-of-classifications-volumes/>. Published 2018. Accessed May 17, 2019.
22. USEPA. A Review of the Reference Dose and Reference Concentration Processes. 2002(EPA/630/P-02/002F).
23. Health MDo. Toxicological Summary for: Perfluorobutane sulfonate. In:2017.
24. Braun JM, Chen A, Romano ME, et al. Prenatal perfluoroalkyl substance exposure and child adiposity at 8 years of age: The HOME study. *Obesity (Silver Spring)*. 2016;24(1):231-237.
25. Hoyer BB, Ramlau-Hansen CH, Vrijheid M, et al. Anthropometry in 5- to 9-Year-Old Greenlandic and Ukrainian Children in Relation to Prenatal Exposure to Perfluorinated Alkyl Substances. *Environ Health Perspect*. 2015;123(8):841-846.
26. Maisonet M, Terrell ML, McGeehin MA, et al. Maternal concentrations of polyfluoroalkyl compounds during pregnancy and fetal and postnatal growth in British girls. *Environ Health Perspect*. 2012;120(10):1432-1437.
27. Halldorsson TI, Rytter D, Haug LS, et al. Prenatal exposure to perfluorooctanoate and risk of overweight at 20 years of age: a prospective cohort study. *Environ Health Perspect*. 2012;120(5):668-673.
28. Lauritzen HB, Larose TL, Oien T, et al. Prenatal exposure to persistent organic pollutants and child overweight/obesity at 5-year follow-up: a prospective cohort study. *Environ Health*. 2018;17(1):9.
29. Karlsen M, Grandjean P, Weihe P, Steuerwald U, Oulhote Y, Valvi D. Early-life exposures to persistent organic pollutants in relation to overweight in preschool children. *Reprod Toxicol*. 2017;68:145-153.

30. Tian YP, Zeng XW, Bloom MS, et al. Isomers of perfluoroalkyl substances and overweight status among Chinese by sex status: Isomers of C8 Health Project in China. *Environ Int.* 2019;124:130-138.
31. Liu G, Dhana K, Furtado JD, et al. Perfluoroalkyl substances and changes in body weight and resting metabolic rate in response to weight-loss diets: A prospective study. *PLOS Medicine.* 2018;15(2):e1002502.
32. Cardenas A, Hauser R, Gold DR, et al. Association of Perfluoroalkyl and Polyfluoroalkyl Substances With Adiposity. *JAMA Network Open.* 2018;1(4):e181493-e181493.
33. Mora AM, Oken E, Rifas-Shiman SL, et al. Prenatal Exposure to Perfluoroalkyl Substances and Adiposity in Early and Mid-Childhood. *Environ Health Perspect.* 2017;125(3):467-473.
34. Domazet SL, Grontved A, Timmermann AG, Nielsen F, Jensen TK. Longitudinal Associations of Exposure to Perfluoroalkylated Substances in Childhood and Adolescence and Indicators of Adiposity and Glucose Metabolism 6 and 12 Years Later: The European Youth Heart Study. *Diabetes Care.* 2016;39(10):1745-1751.
35. Das KP, Wood CR, Lin MT, et al. Perfluoroalkyl acids-induced liver steatosis: Effects on genes controlling lipid homeostasis. *Toxicology.* 2017;378:37-52.
36. Rosen MB, Das KP, Rooney J, Abbott B, Lau C, Corton JC. PPAR alpha-independent transcriptional targets of perfluoroalkyl acids revealed by transcript profiling. *Toxicology.* 2017;387:95-107.
37. Albrecht PP, Torsell NE, Krishnan P, et al. A species difference in the peroxisome proliferator-activated receptor alpha-dependent response to the developmental effects of perfluorooctanoic acid. *Toxicological sciences : an official journal of the Society of Toxicology.* 2013;131(2):568-582.
38. Palkar PS, Anderson CR, Ferry CH, Gonzalez FJ, Peters JM. Effect of prenatal peroxisome proliferator-activated receptor alpha (PPARalpha) agonism on postnatal development. *Toxicology.* 2010;276(1):79-84.
39. Wolf DC, Moore T, Abbott BD, et al. Comparative hepatic effects of perfluorooctanoic acid and WY 14,643 in PPAR-alpha knockout and wild-type mice. *Toxicologic pathology.* 2008;36(4):632-639.
40. Rosenmai AK, Ahrens L, le Godec T, Lundqvist J, Oskarsson A. Relationship between peroxisome proliferator-activated receptor alpha activity and cellular concentration of 14 perfluoroalkyl substances in HepG2 cells. *J Appl Toxicol.* 2018;38(2):219-226.
41. Wolf CJ, Takacs ML, Schmid JE, Lau C, Abbott BD. Activation of mouse and human peroxisome proliferator-activated receptor alpha by perfluoroalkyl acids of different functional groups and chain lengths. *Toxicol Sci.* 2008;106(1):162-171.
42. Foreman JE, Chang SC, Ehresman DJ, et al. Differential hepatic effects of perfluorobutyrate mediated by mouse and human PPAR-alpha. *Toxicol Sci.* 2009;110(1):204-211.
43. Mahapatra CT, Damayanti NP, Guffey SC, Serafin JS, Irudayaraj J, Sepulveda MS. Comparative in vitro toxicity assessment of perfluorinated carboxylic acids. *J Appl Toxicol.* 2017;37(6):699-708.

44. Bjork JA, Wallace KB. Structure-activity relationships and human relevance for perfluoroalkyl acid-induced transcriptional activation of peroxisome proliferation in liver cell cultures. *Toxicol Sci.* 2009;111(1):89-99.
45. Hall AP, Elcombe CR, Foster JR, et al. Liver hypertrophy: a review of adaptive (adverse and non-adverse) changes--conclusions from the 3rd International ESTP Expert Workshop. *Toxicol Pathol.* 2012;40(7):971-994.

Appendix A. Toxicity Data

Table A-I. PFBS Toxicity Studies from Literature Review

Study Type	Species	Duration	Chemical Form	Doses (mg/kg-d)	Route	Key Findings	Toxicity Value (mg/kg-d)	Reference
Short-Term	Mouse	28 d	K ⁺ PFBS	0, 30	Diet	PFBS reduced plasma triglycerides, very low density lipoprotein-cholesterol, hepatic lipase activity, liver cholesteryl esters, liver free cholesterol, plasma glycerol, plasma cholesteryl ester transfer protein.	NOAEL: N/A LOAEL: 30	Bijland et al., 2011 ⁽¹¹⁾
Reproduction/development	Mouse	Gestation days 1-20	K ⁺ PFBS	0, 50, 200, 500	Gavage	At 200 and 500 mg/kg-d, PFBS decreased body weight, delayed eye opening, and affected reproductive development in females by delaying vaginal opening, reducing the size of the ovaries and uterus, delaying menstruation time, and altering hormone levels.	NOAEL: 50 LOAEL: 200	Feng et al., 2017 ⁽¹²⁾
Sub-chronic	Rat	90 d	K ⁺ PFBS	0, 60, 200, 600	Gavage	At the highest dose, PFBS caused red perioral substance, urine-stained abdominal fur, increased chloride values, and reduced red blood cells in males; reduced total protein and albumin values in females; and histological changes in the stomach and kidney in both sexes. At 200 and 600 mg/kg-d, PFBS caused lower blood hemoglobin and hematocrit in males.	NOAEL: 60 LOAEL: 200	Lieder et al., 2009a ⁽¹³⁾
Reproduction/development	Rat	2 generations	K ⁺ PFBS	0, 30, 100, 300, 1000	Gavage	At the highest dose, PFBS caused decreased absolute brain weight among females and lower terminal body weight in males. At 300 and 1000 mg/kg-day, PFBS caused increased absolute and relative liver weight in males. At all doses, PFBS caused higher terminal body weight in females.	NOAEL: N/A LOAEL: 30	Lieder et al., 2009b ⁽⁶⁾

K⁺ PFBS = potassium salt of perfluorobutanesulfonate

Table A-2. Critical Study Selection for PFBS

Reference	Appropriate duration?	Effects consistent with other studies?	Effects relevant to humans?	Number of Doses	Toxicity value identifiable?	Critical study?
Bijland et al., 2011 ¹¹	✓	✓	✓	1	✓	No
Feng et al., 2017 ¹²	✓	✓	✓	3	✓	Yes
Lieder et al., 2009a ¹³	✓	✓	✓	3	✓	Yes
Lieder et al., 2009b ⁶	✓	✓	✓	4	✓	Yes

To be considered a critical study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.

Table A-3. PFBS Epidemiological Studies from Literature Review

Study Type	Population	Time period	Exposure	Outcomes	Results	Other PFASs analyzed	Reference
Cross-sectional	U.S. population (NHANES)	1999-2014	PFBS concentrations in blood serum	Total cardiovascular disease (CVD), including congestive heart failure, coronary heart disease, chest pain, heart attack, stroke	Total CVD AOR (core model): 1.34, 95% CI: 1.05-1.72, <i>p</i> =0.0193	PFOA, PFOS, PFHxS, EPAH, MPAH, PFDE, PFHP, PFNA, PFSA, PFUA, PFDO	Huang et al., 2018 (7)
Cross-sectional	Chinese women	2011-2012	PFBS concentrations in umbilical cord plasma	Preeclampsia Overall hypertensive disorders of pregnancy (HDP)	Preeclampsia AOR: 1.81, 95% CI: 1.03-3.17 Overall HDP AOR: 1.64, 95% CI: 1.09-2.47	PFHxS, PFUnA	Huang et al., 2019 (8)
Case-control	Chinese reproductive-age women	2014-2015	PFBS concentrations in blood plasma	Endometriosis-related infertility	Second vs. lowest tertile: OR=3.74, 95% CI: 2.04, 6.84 Highest vs. lowest tertile: OR=3.04, 95% CI: 1.65, 5.57	PFHpA, PFHxS, PFNA	Wang et al., 2017 (9)
Cross-sectional	Taiwanese children	2009-2010	PFBS concentrations in blood serum	Asthma	Asthma AOR among males (highest vs. lowest quartile): 2.59, 95% CI: 1.14, 5.87 <i>No significant associations among females detected</i>	PFOS, PFOA, PFDA, PFHxS, PFNA	Zhu et al., 2016 (10)
<p>Epidemiologic terms: OR=odds ratio; AOR=adjusted odds ratio; RR=relative risk; 95% CI=95% confidence interval; r=Spearman correlation coefficient; β=regression coefficient PFAS acronyms: PFOA=perfluorooctanoic acid, PFNA=perfluorononanoic acid, PFDA=perfluorodecanoic acid, PFUnA= perfluoroundecanoic acid, PFDoA=perfluorododecanoic acid, PFTriA=perfluorotetradecanoic acid, PFHxDA=perfluorohexadecanoic acid, PFHxS=perfluorohexane sulfonate, PFOS=perfluorooctane sulfonic acid, PFDS=perfluorodecanesulfonate, PFBS=perfluorobutane sulfonate, PFBA=perfluorobutanoic acid, PFPeA=perfluoropentanoic acid, PFHxA=perfluorohexanoic acid, PFHpA=perfluoroheptanoic acid, 6:2 FTSA=6:2 fluorotelomer sulfonates, NMeFOSAA=N-methyl perfluorooctanesulfonamidoacetate, EPAH=2-(N-ethyl-perfluorooctane sulfonamido)acetate, MPAH=2-(N-methyl-perfluorooctane sulfonamido) acetate, PFSA=perfluorooctane sulfonamide; PFHpS=perfluoroheptane sulfonic acid; PFDS= perfluorodecane sulfonic acid; EtPFOSAA= 2-(Nethyl-perfluorooctane sulfonamido) acetic acid</p>							

PFHxS | 2020

Substance Overview

Perfluorohexanesulfonic acid (PFHxS) is a chemical in a group of contaminants called per- and polyfluoroalkyl substances (PFAS). PFHxS has been used as a replacement of perfluorooctanesulfonic acid (PFOS) and perfluorooctanoic acid (PFOA) in some industrial and commercial products like firefighting foam, stain repellants, and packaging.¹ PFAS with long carbon chains, like PFHxS, cannot be easily broken down and excreted from the body and they can persist in the environment and in the human body for long periods of time.¹

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard for PFHxS. DHS recommends an enforcement standard of 40 nanograms per liter (ng/L) for PFHxS. The recommended standard is based on a chronic reference dose developed from a study that found that PFHxS can cause reproductive toxicity in research animals.²

DHS recommends that the preventive action limit for PFHxS be set at 10% of the enforcement standard because PFHxS has been shown to cause interactive effects with endocrine disrupting chemicals in research animals.³

Current Standards	
Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

Recommended Standards	
Enforcement Standard:	40 ng/L
Preventive Action Limit:	4 ng/L

Health Effects

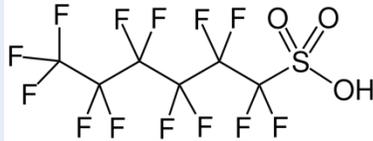
Studies among people have shown that high levels of PFHxS can decrease immune response to vaccines and may increase the risk of osteoarthritis and asthma.¹ Studies in research animals have found that high levels of PFHxS can damage the liver and alter cholesterol levels and may impact fertility and neurological function.³⁻⁹

Limited information is available about the carcinogenic, mutagenic, teratogenic, and interactive effects of PFHxS.⁹ The United States Environmental Protection Agency (EPA) and the International Agency for Research on Cancer (IARC) have not assessed the carcinogenicity of PFHxS. However, a study in human cells found PFHxS caused DNA damage.⁸ Limited studies in animals exposed to high levels of PFHxS

a Carcinogenic effects means the substance can cause cancer; mutagenic effects means the substance can cause DNA damage; teratogenic means the substance can cause birth defects; and interactive effects mean the substance can affect the toxicity of another substance or its toxicity can be affected by another substance.

during pregnancy did not see effects on the development of unborn babies. In a recent developmental study in rats, researchers found that exposure to both PFHxS and a mixture of endocrine disrupting chemicals significantly increased nipple retention in male offspring indicating that PFHxS may cause interactive effects.³

Chemical Profile

PFHxS	
Structure:	
CAS Number:	355-46-4
Formula:	C ₆ HF ₁₃ O ₃ S
Molar Mass:	400.11 g/mol
Synonyms:	Perfluorohexanesulfonic acid Tridecafluorohexane-1-sulfonic acid Perfluorohexane-1-sulphonic acid 1,1,2,2,3,3,4,4,5,5,6,6,6-tridecafluoro-1-Hexanesulfonic acid tridecafluoro-1-Hexanesulfonic acid 1-Perfluorohexanesulfonic acid Tridecafluorohexanesulfonic acid EC 206-587-1

Exposure Routes

People can be exposed to PFHxS by drinking contaminated water, swallowing contaminated soil, eating food that was packaged in material that contains PFHxS, consuming fish from contaminated waters, and breathing in or swallowing dust that contains PFHxS.¹⁰ Babies born to mothers exposed to PFHxS can themselves be exposed to the substance during pregnancy and during breastfeeding.^{1,11,12}

In the environment, PFHxS can be found in water or soil from its manufacturing and use in consumer products and fire-fighting foam.¹³ PFHxS can stay in the environment for a long time (years).¹ PFHxS can move between groundwater and surface water. Once in groundwater, PFHxS can travel long distances.¹

Current Standard

Wisconsin does not currently have groundwater standards for PFHxS.¹⁴

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

EPA Oral Reference Dose:	N/A
--------------------------	-----

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

ATSDR Chronic Oral Minimum Risk Level:	0.00002 mg/kg-d	(2016)
--	-----------------	--------

Literature Search

Literature Search Dates:	2000 – 2019
Key studies found?	Yes
Critical studies identified?	Yes

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for PFHxS.¹⁵

Health Advisory

The EPA has not established health advisories for PFHxS.¹⁶

Drinking Water Concentration (Cancer Risk)

The EPA has not established a drinking water concentration based on a cancer risk level determination for PFHxS.¹⁶

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for PFHxS.¹⁷

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Information System (IRIS) program.

EPA Oral Reference Dose

The EPA does not have an oral reference dose for PFHxS.¹⁸

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If we determine that something is carcinogenic and there is no federal number or ADI from the EPA, then we must set the standard at a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of PFHxS, we looked to see if the EPA, IARC, or another agency has classified the cancer potential of PFHxS. If so, we look to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA and IARC have not evaluated the carcinogenicity of PFHxS.^{18,19}

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for PFHxS.¹⁶

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information that was not considered when the value was established and indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For PFHxS, we searched for any guidance values that had been published on or before the date of the present review (July 2019). We found a relevant guidance value from the Agency for Toxic Substances and Disease Registry (ATSDR).

ATSDR Intermediate Oral Minimum Reference Level (Draft)

In 2018, the ATSDR released a draft Toxicological Profile for Perfluoroalkyls.¹ In this Profile, they recommended an intermediate oral minimum risk level of 20 ng/kg-d for PFHxS.^b

The ATSDR evaluated four studies on PFHxS toxicity in research animals (see Table A-1 for a summary of these studies). They selected a 2009 study by Butenhoff et al. as the critical study.⁵ In this study, male and female rats were exposed to different concentrations of PFHxS (0, 0.3, 1, 3, and 10 milligrams per kilogram per day or mg/kg-d) prior to mating, during mating, pregnancy, and lactation through gavage. The researchers did not observe any effects on reproduction or development nor did they observe any treatment-related effects in dams or offspring. They found that PFHxS reduced serum total cholesterol at all doses. It increased liver-to-body weight and liver-to-brain weight ratios, caused centrilobular hepatocellular hypertrophy, and hyperplasia of thyroid follicular cells at 3 and 10 mg/kg-d. In parental males, PFHxS also reduced prothrombin time at 0.3, 3, and 10 mg/kg-d, reduced hematocrit at 3 and 10 mg/kg-d, reduced triglyceride levels at 10 mg/kg-d, and increased albumin, blood urea nitrogen (BUN), alkaline phosphatase (ALP), calcium, and albumin/globulin ratio at 10 mg/kg-d.

Due to differences between rodents and humans, the ATSDR did not consider the observed liver effects to be relevant to humans.^c ATSDR selected a No Observable Adverse Effect Level (NOAEL) of 1 mg/kg-d based on thyroid follicular cell damage observed in parental males at higher concentrations. To account for differences in the half-life of PFHxS in people versus research animals, the ATSDR estimated human equivalent dose from measured serum concentrations in animals using the trapezoid rule.²⁰ To obtain the MRL, they used a human equivalent dose of 0.0047 mg/kg-d and applied a total uncertainty factor of

b The ATSDR's intermediate minimum risk levels are protective of exposures between 15 and 364 days. The ATSDR did not recommend a chronic oral reference dose for PFOA because they felt that the available data for chronic exposure (more than 1 year) are limited and were uncertain whether the most sensitive endpoint for chronic exposure has been identified in the current research.

c For more details on this determination, see Hall et al., 2012 (19) and section 2.9 of ATSDR's Toxicological Profile (1).

300 to account for differences between people and research animals (3), differences among people (10), and the limited availability of information (10).

Literature Search

Our literature review focused on relevant scientific literature on the health effects of PFHxS published on or before August 2020. We looked for studies related to PFHxS toxicity or effects on a disease state in which information on exposure or dose was included as part of the study or studies related to modeling PFHxS exposure or dose using pharmacokinetics in animals or humans.^d Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

Approximately 500 toxicity studies were returned by the search engines. We excluded studies on non-mammalian or cell systems, non-oral exposure routes, and those that did not evaluate health risks from further review. After applying these exclusion criteria, we located five key toxicity studies on PFHxS that were not evaluated by ATSDR (summarized in Table A-2). To be considered a critical toxicity study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.^{e,f} Three of the key studies that we located meet the criteria to be considered a critical toxicity study (see Table A-3 for more details).

To be considered a critical pharmacokinetics study, the study must model oral exposure in humans or rodents. One of the key studies met the criteria to be considered a critical pharmacokinetic study (the section below has more details on these studies).

In our search, we also located a number of epidemiology studies in our search (See Table A-4 for a summary). While the long half-life of PFHxS in people, multiple potential exposure sources, and the ability for other PFAS compounds to cause similar health effects preclude using these data to establish a health-based value, such studies are helpful in identifying the crucial effects and ensuring that the

^d We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

Title/abstract: PFHxS or "perfluorohexane sulfonate"

Subject area: toxicology

Language: English

We also searched online for toxicity studies published by national research programs.

^e Due to the limited availability of data for this substance, we considered a study to be an appropriate duration if animals were exposed for at least 28 days or if the exposure occurred during pregnancy. We accounted for differences in exposure duration through the use of an uncertainty factor.

^f Appropriate toxicity values include the no observable adverse effect level (NOAEL), lowest observable adverse effect level (LOAEL), and benchmark dose (BMD).

animal data used to establish the standard are relevant to people. In these studies, PFHxS exposure was primarily associated with physical and mental developmental markers, diabetes, and thyroid function, among other effects (Table A-1).^{11,22-30}

Critical Toxicity Studies

To compare between results between the critical studies, we calculated an acceptable daily intake (ADI) for each study/effect. The ADI is the estimated amount of PFHxS that a person can be exposed to every day and not experience health impacts. Because the half-life of PFHxS is significantly longer in people than it is in research animals (years compared to days), we followed the approach used by ATSDR to establish candidate ADIs. In this approach, a human equivalent dose is calculated from either a no-observed adverse effect level (NOAEL), lowest observed adverse effect level (LOAEL), or benchmark dose (BMD) and from measured serum concentrations.^g This approach ensures that the ADI is adequately protective of human health by accounting for slow elimination of PFHxS in people and its ability to accumulate in the body. More details on this calculation are provided in Appendix B.

To obtain the ADI, we divided the human equivalent dose by a factor accounting for various sources of scientific uncertainty. We included uncertainty factors to account for differences between humans and animals, differences between healthy and sensitive human populations, using data from short-term experiments to protect against effects from long-term exposure, and using data where a health effect was observed to estimate the level that does not cause an effect, as appropriate. To ensure appropriate protection, we have chosen to not use studies that have significant uncertainty as the basis for the recommended enforcement standards.^h This approach is consistent with that taken by EPA when establishing oral reference doses.²¹

Chang et al., 2018

Cheng et al. exposed male and female mice to different concentrations of PFHxS (0, 0.3, 1, and 3 mg/kg-d) prior to mating, during mating, pregnancy, and lactation through gavage. PFHxS caused some statistically significant effects in parents and offspring (Table 1).

Table 1. Statistically Significant Effects Observed in Chang et al, 2018 (6)

Effects Observed in Parents		0.3	1	3
Weight	Increased body weight gain	*	*	
Reproduction	Reduced mean live litter size		*	*
Biochemistry	Reduced serum cholesterol (males)			✓

^g The NOAEL is the highest dose tested that did not cause an adverse effect, the LOAEL is the lowest dose tested that caused an adverse effect, and the BMD is an estimation of the dose that would cause a specific level of response (typically 5 or 10%).

^h DHS considers a study to have significant uncertainty if the total uncertainty factors is greater than 3,000.

	Reduced total bilirubin (males)			*
	Increased alkaline phosphatase (males)			*
Liver	Increased mean liver weight (males and females)		*	*
	Increased mean relative liver weight to body weight ratio (males and females)		*	*
	Cytoplasmic alteration, ground-glass (males and females)	*	*	*
	Microvesicular fatty changes (males)			*
	Cytoplasmic vasculature (females)			*
	Centrilobular hepatocellular hypertrophy (males and females)	*	*	*
	Single-cell necrosis (males)			*
Effects Observed in Offspring		0.3	1	3
Development	Increased relative anogenital distance (males)	*	*	*
Liver	Increased mean liver weight at PND 36 (males)			*
	Increased mean relative liver weight to body weight ratio (PND 21 and 36) (males and females)			*
	Cytoplasmic alteration (males and females)			*
	Centrilobular hepatocellular hypertrophy (males and females)			*

* The authors did not consider this finding to not be treatment-related, toxicologically relevant, or adverse (see Chang et al., 2018 for more details).⁶

We identified a NOAEL of 0.3 mg/kg-d based on the reduced mean live litter size at higher concentrations. The authors did not consider the reduced litter size to be toxicologically relevant because similar results were not observed by Butenhoff et al. in rats and by Ramhoj et al. in mice.^{3,5} However, the previous studies tested either a different species, a lower internal dose of PFHxS (likely because PFHxS has a longer half-life in mice compared to rats), or a different exposure period.^{2,3,5} These differences between the studies could have accounted for the differences in observed results. Additionally, exposure to other PFAS have shown a reduction to litter size in research animals and PFHxS is known to have a longer half-life in humans compared to research animals.¹ Therefore, DHS considers this effect on litter size to be valid for use in establishing a proposed ADI as effects on reproduction and litter size can have serious implications for public health. Additional data are limited on the potential impacts of PFHxS to reproduction and development in research animals and the mechanism behind most of the observed effects is still under investigation.¹

From this study, we obtained a human equivalent dose by estimating the time-weighted average serum concentration at the NOAEL and applied a total uncertainty factor of 300 to account for differences between people and research animals (3), differences among people (10), and the limited availability of information (10). We obtained a candidate ADI of 4 ng/kg-d for PFHxS from this study. Additional details

on this calculation are included in Table A-5. In 2019, Ali et al., used the Cheng et al., study to produce a chronic reference dose of 4 ng/kg-d.^{2,6}

Ramhøj et al., 2018

Ramhøj et al. conducted two studies evaluating the effect of PFHxS on development.³ In the first study, they exposed female rats to high concentrations of PFHxS (0, 25, and 45 mg/kg-d) during pregnancy and lactation through gavage with and without the addition of EDmix (32.11 mg/kg-d).ⁱ The second study used lower concentrations of PFHxS (0, 0.05, and 5 mg/kg-d). They found that PFHxS alone did not cause overt toxicity in dams and offspring, but did reduce thyroxine (T4) levels in mothers and offspring at doses at and above 5 mg/kg-d (Table 2). They also found that co-exposure with EDmix increased nipple retention in males at the PFHxS levels at and above 5 mg/kg-d.

Table 2. Statistically Significant Effects Observed in Ramhoj et al., 2018 (3)

Effects of PFHxS alone		0.05	5	25*	45
Thyroid	Reduced serum T4 levels in mothers at GD 15		✓	✓	N/A
	Reduced serum T4 levels in mothers at PND 22		✓	✓	✓
	Reduced serum T4 levels in offspring at PND 16/17		✓	✓	✓
Growth	Slightly Reduced offspring birth weight			✓	
	Reduced body weight in female offspring at PNDs 6 and 14		✓		
Liver	Increased relative liver weight in male offspring at PND 16			✓	
	Increased relative liver weight in females (PND 17)		✓	✓	
Effects of PFHxS enhanced by EDmix		0.05	5	25*	45
Development	Increased nipple retention in males (PND 14)		✓	✓	✓

*Work was conducted as 2 studies: study 1 examined doses of 25 and 45 and study 2 examined doses of 0.05, 5, and 25. Except for Reduced pup birth weight, all effects at 25 mg/kg-d were seen in both Study 1 and Study 2.

We identified a NOAEL of 0.05 mg/kg-d based on reduced serum T4 levels at higher concentrations in mothers and offspring at several time points. Because the researchers did not measure serum concentrations at the NOAEL or LOAEL, we were unable to estimate the time-weight average serum

ⁱ EDmix is a mixture of endocrine disruptors including dibutyl phthalate (DBP), di-2-ethylhexyl phthalate (DEHP), vinclozolin, prochloraz, procymidone, linuron, epoxyconazole, 4-methylbenzylidene camphor (4-MBC), octyl methoxycinnamate (OMC), *p,p'*-dichlorodiphenyldichloroethylene (*p,p'*-DDE), bisphenol A (BPA), and butyl paraben.

concentration. Instead, we assumed that the human equivalent dose equaled the NOAEL. We applied a total uncertainty factor of 1000 directly to account for differences between people and research animals (10), differences among people (10), and the limited availability of information (10). We obtained a candidate ADI of 50 ng/kg-d for PFHxS from this study. Additional details on this calculation are included in Table A-5.

NTP, 2018

In 2019, the National Toxicology Program (NTP) published a report describing a study in which they exposed adult rats to different concentrations of PFHxS (0, 0.625, 1.25, 2.5, 5, and 10 mg/kg-d for males and 0, 3.12, 6.25, 12.5, 25, and 50 mg/kg-d for females) for 28 days through gavage.³¹ PFHxS caused some statistically significant effects in males and females (Table 3).

Table 3. Statistically Significant Effects Observed in NTP, 2018⁽³¹⁾

Effects in males		0.625	1.25	2.5	5	10
Biochemistry	Reduced free T4, total T4 and T3	✓	✓	✓	✓	✓
	Reduced cholesterol		✓	✓	✓	✓
	Reduced total bilirubin			✓	✓	✓
	Reduced triglycerides			✓	✓	✓
	Increased acetyl Co-A				✓	
Adrenal	Reduced relative adrenal weights			✓	✓	✓
Liver	Increased relative liver weight		✓	✓	✓	✓
	Increased absolute liver weight		✓	✓	✓	✓
	Increased incidence and severity of hypertrophy			✓	✓	✓
	Increased relative liver weight					✓
Effects in females		3.12	6.25	12.5	25	50
Biochemistry	Reduced free and total T4		✓	✓	✓	✓
Adrenal	Increased absolute adrenal weight			✓	✓	✓
	Increased relative adrenal weight				✓	✓
Liver	Increased relative liver weight	✓	✓	✓	✓	✓
	Increased absolute liver weight	✓	✓	✓	✓	✓
Olfactory	Increased incidence of olfactory epithelial changes and degeneration					✓

We identified a LOAEL of 0.625 mg/kg-d based on reduced free T4 and total T3 and T4 high males at this concentration and higher. We calculated the human equivalent dose by estimating the time-weighted average serum concentration at the LOAEL and applied an total uncertainty factor of 30,000 to account for differences between people and research animals (3), differences among people (10), using data from short-term experiments to protect against effects from long-term exposure (10), using a LOAEL instead of a NOAEL (10), and the limited availability of information (10). While we obtained a candidate

ADI of 0.07 ng/kg-d for PFHxS from this study, this study was not used to establish a recommended enforcement standard due to significant uncertainty. Additional details on this calculation are included in Table A-5.

Critical Pharmacokinetic Studies

A physiologically-based pharmacokinetic (PBPK) model can be used to relate the amount of a chemical exposure to the amount of a chemical found in different organs at different time points.³² PBPK models are developed using mathematical values and equations that describe characteristics and processes of the body, such as body weight, blood flow rate, and metabolism rate. The PBPK model uses a NOAEL and known rat serum levels to estimate a human point of departure (POD_{Human}). POD_{Human} is defined as the point on a toxicological dose-response curve established from experimental data that corresponds to an estimated no effect level.³² The POD_{Human} and PBPK models can be used to better understand what animal toxicity data means for human health and PBPK models provide a critical link between chemical toxicity and exposure information, as well as an important tool for using animal experiments to inform evaluations of the health effects of chemicals on humans.³²

Kim et al., 2018

In 2018, Kim et al., published a study in which they developed a physiology-based pharmacokinetic (PBPK) model to estimate PFHxS serum levels in male and female rats from exposure to the NOAEL used in Butenhoff et al.^{5,32}

The researchers obtained a point of departure for humans (POD_{Human}) of 71.09 µg/kg-d for males and 15.89 µg/kg-d for females (Table 4). To obtain the candidate ADIs, we applied an uncertainty factor of 1000 to account for differences among people (10), using data from a short-term study to protect from long-term effects (10), and the limited availability of data (10). We established candidate ADIs of 70 ng/kg-d for males and 20 ng/kg-d for females.

Table 4. Toxicity parameters and ADIs from Kim et al. 2018³²

Sex	NOAEL (mg/kg-d)	Endpoint	POD _{Human} (µg/kg-d)	Total Uncertainty Factor	ADI (ng/kg-d)
Female	1	Liver toxicity*	15.89	1000**	20
Male	1	Liver toxicity*	71.09	1000**	70

* Toxicity endpoint from Butenhoff et al.⁵
 ** DHS selected a total uncertainty factor of 1000 to account for differences among people (10), use of a shorter duration study to protect against effects from long-term exposure (10), and the limited availability of data (10).

Key Health Effects

We did not find any studies indicating PFHxS can cause carcinogenic, mutagenic, or teratogenic effects in people, research animals, or cell culture. However, we did find a study that found that PFDA caused interactive effects with endocrine disrupting chemicals in research animals.³ One study exposed male rats to different concentrations of PFNA (0, 0.05, 5, 25 mg/kg-d), to a mixture of endocrine disrupting chemicals (ED_{mix}).³ PFHxS exposure alone decreased T4 levels starting at 5 mg/kg-d. PFHxS and ED_{mix} exposure decreased offspring weight at birth and increased liver weights at birth as compared to PFHxS exposure alone. The authors concluded that PFHxS can interact with endocrine disrupting chemicals on a variety of toxicity endpoints and thus have interactive effects.³

Discussion

PFHxS has been shown to affect the liver, cholesterol levels, thyroid hormones, increase relative liver weight, cause histological changes in the liver, alter biomarkers of liver damage, reduce serum cholesterol levels, and reduce litter size in research animals or people.^{3,5,6,31}

Studies in research animals and people have shown that PFHxS can affect developmental endpoints.^{5,6,11,23,29} The evidence in animals suggests that PFHxS may have consistent effects to other long-chained PFAS including reduced litter size, decreased offspring birth weight, and delays in hormone and motor development. Thyroid hormones are crucial for development, energy balance, and metabolism in all species.³³ Studies in research animals and people have shown that PFHxS, as well as other PFAS, can affect the levels of thyroid hormones.¹ In people, thyroid hormones play an important role in the development of the brain, lungs, and heart.³³ Scientists have learned that certain PFAS, including PFHxS, can bind to transport proteins involved in moving thyroid hormones throughout the body.³⁴ Scientists are still learning how this effect occurs and its impact on health.

Studies in rodents have shown that PFHxS, as well as other PFAS, can reduce cholesterol levels.¹ However, studies in people have shown that PFAS exposure is associated with increased total cholesterol and LDL levels.¹ Scientists are learning more about the way in which PFAS reduce cholesterol levels in rodents and believe that they are associated with activation of nuclear hormone receptors.⁴ Because nuclear receptors, like PPAR α , are important regulators of lipid metabolism, changes in their expression patterns can affect lipid transport and alter cholesterol levels. However, at this time, it is unclear whether PFAS act in the same way in people.

A number of studies have demonstrated that liver effects caused by PFHxS, as well as other PFAS, occur primarily through activation of the nuclear hormone receptor, PPAR α (peroxisome proliferator-activated receptor alpha).^{7,35-38} Nuclear receptors regulate gene expression and PPAR α regulates the expression of genes involved in lipid and cholesterol metabolism.³⁹ While PPAR α receptors are found in rodents and humans, levels of these receptors are much higher in rats and mice than in monkeys and humans.³⁹ This means that effects caused by activation of PPAR α are more likely to occur in rodents than they are to

occur in people. As such, the effects on the liver observed in the critical studies reviewed here are likely not relevant to humans. This conclusion is supported by the lack of associations between PFAS exposure and liver disease in epidemiological studies.⁴⁰⁻⁴⁵

Standard Selection

DHS recommends an enforcement standard of 40 ng/L for PFHxS.

There are no federal numbers and no state drinking water standard for PFHxS. Additionally, the EPA has not established a cancer slope factor or ADI (oral reference dose) for PFHxS. However, we found several critical studies evaluating the toxicity of PFHxS.^{3,5,6,31}

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information

As such, DHS recommends using available scientific information to establish an ADI through the procedures specified in Ch. 160, Wis. Stats. To establish the ADI, we used the 2018 study by Chang et al. as the critical study and effects on litter size as the critical effect.⁶ Toxicity studies in animals continue to show that developmental endpoints are critical effects for PFAS, including PFHxS, with effects occurring in offspring after exposure during pregnancy and lactation.^{1,5,6} Additionally, epidemiology studies continue to associate maternal PFHxS exposure to developmental problems in offspring, as PFHxS has been shown to cross the placenta during pregnancy.^{1,11,23,29} We selected reduced litter size as the critical effect because it indicates maternal and prenatal toxicity during pregnancy and through lactation.⁴⁶

We obtained an ADI of 4 ng/kg-d by estimating the human equivalent dose at a NOAEL from measured serum concentrations and applying a total uncertainty factor of 300. To determine the recommended enforcement standard, we used a body weight of 10 kg, a water consumption rate of 1 L/d, and a relative source contribution of 100% as required by Ch. 160, Wis. Stats.

DHS recommends a preventive action limit of 4 ng/L for PFHxS.

DHS recommends that the preventive action limit for PFHxS be set at 10% of the enforcement standard because a recent study has shown that PFHxS can cause interactive effects in research animals.³ At this time, PFHxS has not been shown to cause carcinogenic, mutagenic, or teratogenic effects in people, research animals, or cell culture studies.

Prepared by Sarah Yang, PhD, Gavin Dehnert, PhD, and Nathan Kloczko, MPH

Wisconsin Department of Health Services

References

1. ATSDR. Toxicological Profile for Perfluoroalkyls - Draft for Public Comment. In: Registry AftSaD, ed. Atlanta, GA2017.
2. Ali JM, Roberts SM, Gordon DS, Stuchal LD. Derivation of a chronic reference dose for perfluorohexane sulfonate (PFHxS) for reproductive toxicity in mice. *Regulatory toxicology and pharmacology : RTP*. 2019;108:104452.
3. Ramhoj L, Hass U, Boberg J, et al. Perfluorohexane Sulfonate (PFHxS) and a Mixture of Endocrine Disruptors Reduce Thyroxine Levels and Cause Antiandrogenic Effects in Rats. *Toxicol Sci*. 2018;163(2):579-591.
4. Bijland S, Rensen PCN, Pieterman EJ, et al. Perfluoroalkyl Sulfonates Cause Alkyl Chain Length-Dependent Hepatic Steatosis and Hypolipidemia Mainly by Impairing Lipoprotein Production in APOE*3-Leiden CETP Mice. *Toxicol Sci*. 2011;123(1):290-303.
5. Butenhoff JL, Chang SC, Ehresman DJ, York RG. Evaluation of potential reproductive and developmental toxicity of potassium perfluorohexanesulfonate in Sprague Dawley rats. *Reprod Toxicol*. 2009;27(3-4):331-341.
6. Chang S, Butenhoff JL, Parker GA, et al. Reproductive and developmental toxicity of potassium perfluorohexanesulfonate in CD-1 mice. *Reprod Toxicol*. 2018;78:150-168.
7. Das KP, Wood CR, Lin MT, et al. Perfluoroalkyl acids-induced liver steatosis: Effects on genes controlling lipid homeostasis. *Toxicology*. 2017;378:37-52.
8. Wielsoe M, Long M, Ghisari M, Bonefeld-Jorgensen EC. Perfluoroalkylated substances (PFAS) affect oxidative stress biomarkers in vitro. *Chemosphere*. 2015;129:239-245.
9. Viberg H, Lee I, Eriksson P. Adult dose-dependent behavioral and cognitive disturbances after a single neonatal PFHxS dose. *Toxicology*. 2013;304:185-191.
10. Hu XDC, Dassuncao C, Zhang XM, et al. Can profiles of poly- and Perfluoroalkyl substances (PFASs) in human serum provide information on major exposure sources? *Environ Health*. 2018;17:15.
11. Cao W, Liu X, Liu X, et al. Perfluoroalkyl substances in umbilical cord serum and gestational and postnatal growth in a Chinese birth cohort. *Environment international*. 2018;116:197-205.
12. Harris MH, Oken E, Rifas-Shiman SL, et al. Prenatal and childhood exposure to per- and polyfluoroalkyl substances (PFASs) and child cognition. *Environment international*. 2018;115:358-369.
13. ITRC. Environmental Fate and Transport for Per- and Polyfluoroalkyl Substances. In: Council ITR, ed2018.
14. WIDNR. Drinking Water and Groundwater Quality Standards/Advisory Levels. 2017.

15. USEPA. 2018 Edition of the Drinking Water Standards and Health Advisories Table 2018.
16. USEPA. Drinking Water Contaminant Human Health Effects Information. <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>. Published 2019. Accessed.
17. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 8092018*.
18. USEPA. IRIS Assessments. https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm. Published 2019. Accessed.
19. IARC. List of Classification, Volumes 1-123. <https://monographs.iarc.fr/list-of-classifications-volumes/>. Published 2018. Accessed May 17, 2019.
20. ATSDR. Calculation of AUC and TWA serum concentrations of perfluoroalkyls from serum concentration data using the trapezoid rule. In: DHS, ed2019.
21. USEPA. A Review of the Reference Dose and Reference Concentration Processes. 2002(EPA/630/P-02/002F).
22. Maisonet M, Terrell ML, McGeehin MA, et al. Maternal concentrations of polyfluoroalkyl compounds during pregnancy and fetal and postnatal growth in British girls. *Environmental health perspectives*. 2012;120(10):1432-1437.
23. Ernst A, Brix N, Lauridsen LLB, et al. Exposure to Perfluoroalkyl Substances during Fetal Life and Pubertal Development in Boys and Girls from the Danish National Birth Cohort. *Environmental health perspectives*. 2019;127(1):17004.
24. Hoffman K, Webster TF, Weisskopf MG, Weinberg J, Vieira VM. Exposure to polyfluoroalkyl chemicals and attention deficit/hyperactivity disorder in U.S. children 12-15 years of age. *Environmental health perspectives*. 2010;118(12):1762-1767.
25. Wang Y, Zhang L, Teng Y, et al. Association of serum levels of perfluoroalkyl substances with gestational diabetes mellitus and postpartum blood glucose. *Journal of environmental sciences (China)*. 2018;69:5-11.
26. Matilla-Santander N, Valvi D, Lopez-Espinosa MJ, et al. Exposure to Perfluoroalkyl Substances and Metabolic Outcomes in Pregnant Women: Evidence from the Spanish INMA Birth Cohorts. *Environmental health perspectives*. 2017;125(11):117004.
27. Shapiro GD, Dodds L, Arbuckle TE, et al. Exposure to organophosphorus and organochlorine pesticides, perfluoroalkyl substances, and polychlorinated biphenyls in pregnancy and the association with impaired glucose tolerance and gestational diabetes mellitus: The MIREC Study. *Environmental research*. 2016;147:71-81.
28. Kim MJ, Moon S, Oh BC, et al. Association between perfluoroalkyl substances exposure and thyroid function in adults: A meta-analysis. *PloS one*. 2018;13(5):e0197244.

29. Preston EV, Webster TF, Oken E, et al. Maternal Plasma per- and Polyfluoroalkyl Substance Concentrations in Early Pregnancy and Maternal and Neonatal Thyroid Function in a Prospective Birth Cohort: Project Viva (USA). *Environmental health perspectives*. 2018;126(2):027013.
30. Christensen KY, Raymond M, Meiman J. Perfluoroalkyl substances and metabolic syndrome. *International journal of hygiene and environmental health*. 2019;222(1):147-153.
31. NTP. NTP Technical Report on the Toxicity Studies of Perfluoroalkyl Sulfonates (Perfluorobutane Sulfonic Acid, Perfluorohexane Sulfonate Potassium Salt, and Perfluorooctane Sulfonic Acid) Administered by Gavage to Sprague Dawley Rats. In: Program NT, ed. Vol Tox 962019.
32. Kim SJ, Shin H, Lee YB, Cho HY. Sex-specific risk assessment of PFHxS using a physiologically based pharmacokinetic model. *Arch Toxicol*. 2018;92(3):1113-1131.
33. Calsolaro V, Pasqualetti G, Niccolai F, Caraccio N, Monzani F. Thyroid Disrupting Chemicals. *Int J Mol Sci*. 2017;18(12):17.
34. Ren XM, Qin WP, Cao LY, et al. Binding interactions of perfluoroalkyl substances with thyroid hormone transport proteins and potential toxicological implications. *Toxicology*. 2016;366-367:32-42.
35. Rosen MB, Das KP, Rooney J, Abbott B, Lau C, Corton JC. PPAR alpha-independent transcriptional targets of perfluoroalkyl acids revealed by transcript profiling. *Toxicology*. 2017;387:95-107.
36. Albrecht PP, Torsell NE, Krishnan P, et al. A species difference in the peroxisome proliferator-activated receptor alpha-dependent response to the developmental effects of perfluorooctanoic acid. *Toxicological sciences : an official journal of the Society of Toxicology*. 2013;131(2):568-582.
37. Palkar PS, Anderson CR, Ferry CH, Gonzalez FJ, Peters JM. Effect of prenatal peroxisome proliferator-activated receptor alpha (PPARalpha) agonism on postnatal development. *Toxicology*. 2010;276(1):79-84.
38. Wolf DC, Moore T, Abbott BD, et al. Comparative hepatic effects of perfluorooctanoic acid and WY 14,643 in PPAR-alpha knockout and wild-type mice. *Toxicologic pathology*. 2008;36(4):632-639.
39. Hall AP, Elcombe CR, Foster JR, et al. Liver hypertrophy: a review of adaptive (adverse and non-adverse) changes--conclusions from the 3rd International ESTP Expert Workshop. *Toxicologic pathology*. 2012;40(7):971-994.
40. Alexander BH, Olsen GW, Burris JM, Mandel JH, Mandel JS. Mortality of employees of a perfluorooctanesulphonyl fluoride manufacturing facility. *Occupational and environmental medicine*. 2003;60(10):722-729.
41. Anderson-Mahoney P, Kotlerman J, Takhar H, Gray D, Dahlgren J. Self-reported health effects among community residents exposed to perfluorooctanoate. *New solutions : a journal of environmental and occupational health policy : NS*. 2008;18(2):129-143.

42. Darrow LA, Groth AC, Winquist A, Shin HM, Bartell SM, Steenland K. Modeled Perfluorooctanoic Acid (PFOA) Exposure and Liver Function in a Mid-Ohio Valley Community. *Environmental health perspectives*. 2016;124(8):1227-1233.
43. Grice MM, Alexander BH, Hoffbeck R, Kampa DM. Self-reported medical conditions in perfluorooctanesulfonyl fluoride manufacturing workers. *Journal of occupational and environmental medicine*. 2007;49(7):722-729.
44. Olsen GW, Church TR, Larson EB, et al. Serum concentrations of perfluorooctanesulfonate and other fluorochemicals in an elderly population from Seattle, Washington. *Chemosphere*. 2004;54(11):1599-1611.
45. Steenland K, Zhao L, Winquist A. A cohort incidence study of workers exposed to perfluorooctanoic acid (PFOA). *Occupational and environmental medicine*. 2015;72(5):373-380.
46. Haschek W, Rousseaux C, Wallig M. *Fundamentals of Toxicologic Pathology*. Vol 2nd Edition 2010.
47. Lee I, Viberg H. A single neonatal exposure to perfluorohexane sulfonate (PFHxS) affects the levels of important neuroproteins in the developing mouse brain. *Neurotoxicology*. 2013;37:190-196.
48. Blake BE, Pinney SM, Hines EP, Fenton SE, Ferguson KK. Associations between longitudinal serum perfluoroalkyl substance (PFAS) levels and measures of thyroid hormone, kidney function, and body mass index in the Fernald Community Cohort. *Environmental pollution (Barking, Essex : 1987)*. 2018;242(Pt A):894-904.
49. Lin PD, Cardenas A, Hauser R, et al. Per- and polyfluoroalkyl substances and blood lipid levels in pre-diabetic adults-longitudinal analysis of the diabetes prevention program outcomes study. *Environment international*. 2019;129:343-353.
50. Vuong AM, Braun JM, Yolton K, et al. Prenatal and childhood exposure to perfluoroalkyl substances (PFAS) and measures of attention, impulse control, and visual spatial abilities. *Environment international*. 2018;119:413-420.
51. Sundstrom M, Chang SC, Noker PE, et al. Comparative pharmacokinetics of perfluorohexanesulfonate (PFHxS) in rats, mice, and monkeys. *Reprod Toxicol*. 2012;33(4):441-451.
52. Kim SJ, Heo SH, Lee DS, Hwang IG, Lee YB, Cho HY. Gender differences in pharmacokinetics and tissue distribution of 3 perfluoroalkyl and polyfluoroalkyl substances in rats. *Food and chemical toxicology : an international journal published for the British Industrial Biological Research Association*. 2016;97:243-255.
53. Fu J, Gao Y, Cui L, et al. Occurrence, temporal trends, and half-lives of perfluoroalkyl acids (PFAAs) in occupational workers in China. *Scientific reports*. 2016;6:38039.

54. Li Y, Fletcher T, Mucs D, et al. Half-lives of PFOS, PFHxS and PFOA after end of exposure to contaminated drinking water. *Occupational and environmental medicine*. 2018;75(1):46-51.
55. MDH. Minnesota Department of Health. Environmental Health & Biomonitoring Advisory Panel June 9, 2015 Meeting Background Materials. In: Health MDo, ed2015.
56. Worley RR, Moore SM, Tierney BC, et al. Per- and polyfluoroalkyl substances in human serum and urine samples from a residentially exposed community. *Environment international*. 2017;106:135-143.

Appendix A. Available Scientific Information

Table A-I. PFHxS Rodent Toxicity Studies Reviewed by ATSDR

Study Type	Species	Duration	Doses (mg/kg-d)	Route	Key Findings	Toxicity Value (mg/kg-d)	Reference
Short-Term	Mouse	28 d	0, 6	Diet	Reduced plasma triglycerides, non HDL-cholesterol, and HDL-cholesterol. Increased liver weight and hepatic triglyceride levels. These effects were a result of PPAR- α and PXR (Pregnane-X Receptor) activation.	NOAEL: N/A LOAEL: 6	Bijland et al., 2011 (⁴)
Reproduction/Development	Rat	Prior to cohabitation, during cohabitation, until sacrifice	0, 0.3, 1, 3, 10	Gavage	No effects on reproduction and development observed. No treatment-related effects in dams or offspring. In parental males, PFHxS reduced serum total cholesterol at all doses, reduced prothrombin time at 0.3, 3, and 10 mg/kg-d. It increased liver-to-body weight and liver-to-brain weight ratios, caused centrilobular hepatocellular hypertrophy, hyperplasia of thyroid follicular cells, reduced hematocrit at 3 and 10 mg/kg-d. PFHxS reduced triglycerides and increased albumin, blood urea nitrogen (BUN), alkaline phosphatase (ALP), calcium, and albumin/globulin ratio at 10 mg/kg-d.	NOAEL: N/A LOAEL: 0.3	Butenhoff et al., 2009* (⁵)
Short-term (Mechanism)	Mouse (wildtype, PPAR- α knockout, PPAR α antagonist)	7 d	0, 10	Gavage	PFHxS increased liver weight and cell size and reduced DNA content per mg of liver. This affect was observed in knockout mice but not antagonist mice. PFHxS increased lipid accumulation in wildtype and knockout mice. PFHxS elevated liver triglycerides in WT.	N/A	Das et al., 2017 (⁷)

Acute	Mouse	Single dose	0, 0.61, 6.1, 9.2	Gavage	Altered adult spontaneous behavior and cognitive function in males and females at all levels.	NOAEL: N/A LOAEL: 0.61	Viberg et al., 2013 (⁹)
<p>* = Study was used by ATSDR to establish their intermediate oral minimum risk level. i. 21 days of lactation or presumed gestation day 25 for females and minimum of 42 days of treatment for males.</p>							

Table A-2. PFHxS Rodent Toxicity Studies Not Reviewed by ATSDR

Study Type	Species	Duration	Doses (mg/kg-d)	Route	Key Findings	Toxicity Value (mg/kg-d)	Reference
Reproduction/Development	Mouse	2 generations ⁱ	0, 0.3, 1, 3	Gavage	PFHxS decreased live litter size at 1 and 3 mg/kg-d. Adaptive hepatocellular hypertrophy. Reduced serum cholesterol and increased alkaline phosphatase in F0 males at 3 mg/kg-d. PFHxS did not affect reproductive parameters, hematology/clinical pathology, thyroid stimulating hormone (TSH), neurobehavioral effects, or histopathology. PFHxS did not affect postnatal survival, development or onset of preputial separation or vaginal openings in F1 mice.	NOAEL: N/A LOAEL: 0.3	Chang et al., 2018 ⁽⁶⁾
Acute	Mouse	Single dose	0, 6.1, 9.2	Gavage	PFHxS altered neuroprotein levels (CaMKII, GAP-43, synaptophysin, tau) in the hippocampus and cerebral cortex of neonatal males and females at both levels.	NOAEL: N/A LOAEL: 6.1	Lee et al., 2013 ⁽⁴⁷⁾
Sub-chronic	Rat	28 d	Males: 0, 0.625, 1.25, 2.5, 5, 10 Females: 0, 3.12, 6.25, 12.5, 25, 50	Gavage	In males, PFHxS reduced free and total T4 (thyroxine) and total T3 (triiodothyronine) at all doses; increased relative and absolute liver weights and reduced cholesterol at 1.25 mg/kg-d and greater. PFHxS also reduced relative adrenal weights, total bilirubin, triglycerides, and increased incidence and severity of hypertrophy at 2.5 mg/kg-d and greater. In females, PFHxS increased relative and absolute liver weight at all doses; reduced free and total T4 at 6.25 mg/kg-d and greater. PFHxS also increased absolute adrenal weight at 12.5 mg/kg-d and greater; increased relative adrenal weight at 25	NOAEL: N/A LOAEL: 0.625	NTP, 2018 ⁽³¹⁾

					mg/kg- and greater, and affected the olfactory epithelium at the highest dose.		
Co-exposure	Rat	Gestation through lactation (GD 7 – PND 22)	Study 1 PFHxS: 0, 25, 45 EDmix: 32.11 Study 2 PFHxS: 0, 0.05, 5 EDmix ⁱⁱ : 32.11	Gavage	PFHxS did not cause overt toxicity in dams and offspring. PFHxS alone reduced T4 levels in dams and offspring at doses at 5 mg/kg-d and greater. PFHxS with EDmix reduced male pup birth weight and slightly increased liver weights at high doses.	NOAEL: 0.05 LOAEL: 0.5	Ramhoj et al., 2018 ⁽³⁾
Short-term (Mechanism)	Mouse (WT; PPARα-null)	7 d	0, 3, 10	Gavage	11-24% of genes in PFAS-treated mice were independent of PPAR-α. PFAS gene expression profiles were similar to those exposed to agonists of CAR (constitutive activated receptor), ER-α (estrogen receptor alpha), and PPAR-γ.	N/A	Rosen et al., 2017 ⁽³⁵⁾
<p>i. F0 treated before mating (for at least 42 days in males) through gestation and lactation for females. F1 pups were treated for 14 days after weaning. ii. EDmix is a fixed dose of 12 environmentally relevant endocrine disrupting chemicals.</p>							

Table A-3. Critical Study Selection for PFHxS

Reference	Appropriate duration?	Effects consistent with other studies?	Effects relevant to humans?	Number of Doses	Toxicity value identifiable?	Critical study?
Chang et al., 2018	✓	✓	✓	3	✓	Yes
Lee et al., 2013	⊘	✓	✓	2	✓	No
NTP, 2018	✓	✓	✓	5	✓	Yes
Ramhoj et al., 2018	✓	✓	✓	4	✓	Yes
Rosen et al., 2017	⊘	✓	✓	2	⊘	No

To be considered a critical study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.

* = Study was reviewed by ATSDR

Table A-4. Recent PFHxS Human Studies from Literature Review

Study Type	Population	Time period	Exposure	Outcomes	Results	Other PFAS Studied	Reference
Cohort	768 Pregnant women and 501 newborns	1999-2002	Maternal plasma PFAS concentrations	Thyroid hormones: thyroxine (T ₄), free T ₄ Index (FT ₄ I) , thyroid stimulating hormone (TSH)	PFHxS concentrations associated with lower maternal FT ₄ I and lower male newborn T ₄ levels.	PFOA, PFOS, PFNA, PFHxS, EtFOSAA, MeFOSAA	Preston et al., 2018 ⁽²⁹⁾
Cohort	210 community members (median age 38)	1990-2008	Drinking water exposure; serum PFAS concentrations	Thyroid disruption, kidney function, and BMI	Intraquartile PFHxS increase associated with 2.06% eGFR decrease. No associations seen with thyroid disruption or BMI.	PFOS, PFOA, PFNA, PFHxS, PFDeA, PFOSA, Me-PFOSA, Et-PFOSA	Blake et al., 2018 ⁽⁴⁸⁾
Clinical Trial/cross-sectional	888 from Diabetes Prevention Program	1996-2014	Blood plasma PFAS concentrations	Hypercholesterolemia, hypertriglyceridemia	PFHxS had slightly higher risk for hypertriglyceridemia in placebo group	PFOS, PFOA, PFHxS, EtFOSAA, MeFOSAA, PFNA	Lin et al., 2019 ⁽⁴⁹⁾
Cohort/cross-sectional	631 children from birth cohort	1999-2002	Blood plasma PFAS concentrations	Neurologic development	Visual-motor scores on the Wide Range Assessment of Visual Motor Abilities were lower among children with higher PFHxS (Q4 vs. Q1: -5.0, 95% CI: -9.1, -0.8).	PFOS, PFOA, PFHxS, PFNA, EtFOSAA, MeFOSAA, PFDeA	Harris et al., 2018 ⁽¹²⁾
Cohort/cross-sectional	218 mother-child dyads from HOME Study	2003-2006	Prenatal and childhood PFAS serum concentrations	Neurologic development	Higher concentrations of PFHxS were associated with shorter times in the Virtual Morris Water Maze	PFOS, PFOA, PFHxS, PFNA	Vuong et al., 2018 ⁽⁵⁰⁾

Nested Cohort	445 Danish children in birth cohort	2012-2017	PFAS in maternal plasma from early gestation	Hormonal developmental outcomes	PFHxS was associated with lower mean age at puberty in both boys and girls.	PFOS, PFOA, PFHxS, PFHpS, PFNA, PFDA	Ernst, 2019 ⁽²³⁾
Cross-sectional	2975 individuals from NHANES	2007-2014	Serum PFAS concentrations	Metabolic syndrome	Higher concentrations of PFHxS were associated with elevated triglycerides.	PFDA, PFOS, PFOA, PFHxS, MPAH, PFNA, PFUnA	Christensen, 2019 ⁽³⁰⁾
Cohort	282 Chinese infants	2013-2015	PFAS concentrations in cord blood	Gestational and postnatal growth	Newborns in the highest exposure group of PFHxs had decreased birth length, but increased head circumference.	PFOA, PFNA, PFDA, PFUnA, PFDoA, PFTrDA, PFTeDA, PFHxDA, PFHxS, PFOS, PFDS	Cao, 2018 ⁽¹¹⁾

**This literature review is not exhaustive as the primary purpose of the search was to identify epidemiological studies that support toxicological findings.*

Epidemiologic terms: OR=odds ratio; AOR=adjusted odds ratio; RR=relative risk; 95% CI=95% confidence interval; r=Spearman correlation coefficient; β =regression coefficient

PFAS acronyms: PFOA=perfluorooctanoic acid, PFNA=perfluorononanoic acid, PFDA=perfluorodecanoic acid, PFUnA= perfluoroundecanoic acid, PFDoA=perfluorododecanoic acid, PFTriA=perfluorotetradecanoic acid, PFHxDA=perfluorohexadecanoic acid, PFHxS=perfluorohexane sulfonate, PFOS=perfluorooctane sulfonic acid, PFDS=perfluorodecanesulfonate, PFBS=perfluorobutane sulfonate, PFBA=perfluorobutanoic acid, PFPeA=perfluoropentanoic acid, PFHxA=perfluorohexanoic acid, PFHpA=perfluoroheptanoic acid, 6:2 FTSA=6:2 fluorotelomer sulfonates, NMeFOSAA=N-methyl perfluorooctanesulfonamidoacetate, EPAH=2-(N-ethyl-perfluorooctane sulfonamido)acetate, MPAH=2-(N-methyl-perfluorooctane sulfonamido) acetate, PFSA=perfluorooctane sulfonamide; PFHpS=perfluoroheptane sulfonic acid; PFDS= perfluorodecane sulfonic acid; EtPFOSAA= 2-(Nethyl-perfluorooctane sulfonamido) acetic acid

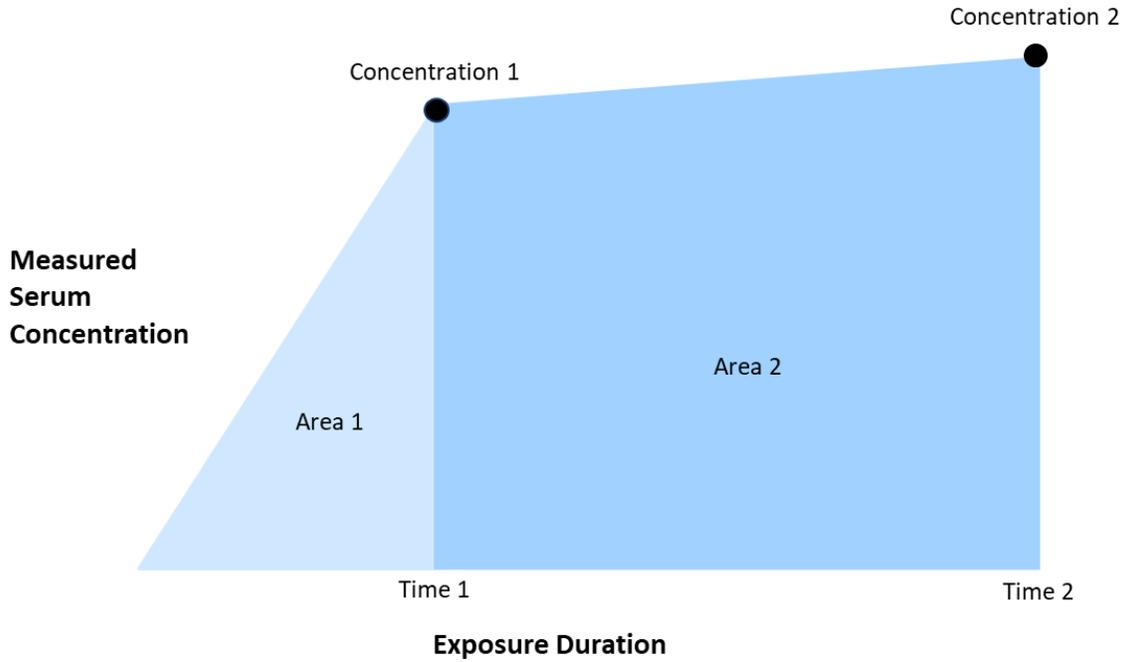
Table A-5. PFHxS Estimated Acceptable Daily Intakes (ADIs) for the Identified Critical Toxicity Studies

Reference	Toxicity Value (mg/kg-d)	Measured serum concentration at toxicity value (µg/L)	Area under the curve	Time-weight average serum concentration (µg/L)	Human equivalent dose (mg/kg-d)	Total Uncertainty Factor ^b	ADI (ng/kg-d)
Chang, 2018	0.3 (NOAEL)	0.0005 at day 0 ^a 20.8 at GD 18 22.1 at PND 4 12.9 at PND 21 19.6 at PND 56	316	23.9	0.0025	300	4
Ramhoj, 2018	0.05 (NOAEL)	N/A	N/A	N/A	N/A	1,000	50 ^c
NTP, 2018	0.625 (LOAEL)	0.102 at day 0 ^a 66.76 at day 28	936	33.4	0.0034	30,000	0.11
NTP, 2018	(BMDL) ^d	0.102 at day 0 32.4 at day 28	33.4	16.3	0.0017	3,000	0.56
<p>GD = gestation day; PND = postnatal day</p> <p>a. Used measured serum concentration in control animals as day 0 value</p> <p>b. See the <i>Critical Toxicity Studies</i> section above for description of factors accounted for in the total uncertainty factor</p> <p>c. Because serum concentrations at the NOAEL and LOAEL were not measured, the ADI was calculated directly from the toxicity value.</p> <p>d. Measured as PFHxS in serum (mg/L) as reported by Minnesota (REFERENCE)</p>							

Appendix B. Calculation of Human Equivalent Dose

To calculate the human equivalent dose for PFHxS, we followed a three-step process.

1. We first calculated the area under the curve at the selected toxicity value using the trapezoid rule.



In this mathematical approach, the area under the curve is divided into one or more trapezoids and area of each trapezoid is calculated (Equation B-1).

$$\text{Equation B-1} \quad \text{Area} = \frac{h}{2}(p+q)$$

Where:

- h = The difference in time between the data points.
- q = Measured serum concentration at first time point
- p = Measured serum concentration at second time point

The areas of all of the trapezoids are summed to give the area under the curve (Equation B-2).

$$\text{Equation B-2} \quad \text{AUC} = \text{Area}_1 + \text{Area}_2 + \dots + \text{Area}_n$$

2. We then calculated the time-weight average serum concentration as a surrogate for the steady-state serum concentration (Equation B-3).

Equation B-3
$$TWA = \frac{AUC}{ED}$$

Where:

AUC = The difference in time between the data points.
 ED = Exposure duration (days)

3. Finally, we calculated the human equivalent dose (HED) by accounting for the long half-life in people.

$$HED = \frac{TWA \times \frac{\ln 2}{t_{1/2}} \times V_d}{AF}$$

Where:

$t_{1/2}$ = half-life in days^j
 Since the early 2000s, a number of studies have been published evaluating the half-life of PFHxS in people (Table B-1). We selected a half-life of 5.3 years because the study was well designed in that it included a wide age range and equal representation of genders. The study is relevant for groundwater exposure in that the PFAS exposure evaluated in the study was from drinking water.

V_d = Volume of distribution^k
 While data on the V_d of PFHxS in people are not available, one study has estimated V_d for nonhuman primates.⁵¹ Because the critical effect that we identified occurred in male animals, we used the V_d for male nonhuman primates (0.287 L/kg).

AF = Gastrointestinal absorption fraction^l
 Data on the AF of PFHxS in people are not available. A few studies have estimated AF in research animals.^{51,52} Because PFHxS has been shown to be completely bioavailable in some of these studies, we used an AF of 1.

These values were also used by ATSDR in establishing their draft intermediate oral minimum risk level.¹

j The half-life is a measure of elimination rate. The longer the half-life, the longer it takes a chemical to be removed from the body.

k The volume of distribution is the theoretical volume needed to contain the amount of the chemical administered at the measured serum concentration.

l The gastrointestinal absorption factor accounts for the bioavailability of the chemical after oral exposure. In other words, it is a measure how much of the chemical is available to cause harm within the body.

Table B-I. Summary of Studies evaluating the half-life of PFHxS in people

Reference	Study type	Location	Exposure source	Population Size	Age	Arithmetic mean half-life (years)	Notes
Fu et al., 2016 ⁽⁵³⁾	Longitudinal	China	Workers	41	14-65	14.7 (daily clearance) 3.6 (annual decline)	Strengths: Occupational exposures with tiered exposures over 5 years, reliable measurements Limitations: change in PFOS production through study duration, high turnover in worker population. Big difference between two reported values. Notes: Values shown are geometric means.
Li et al., 2018 ⁽⁵⁴⁾	Longitudinal	Sweden	Drinking water	106	4 – 86	5.3	Strengths: Longitudinal nature, general population included wide age range and equal representation of genders, drinking water route of exposure, and the well-controlled nature of the environmental “stop-exposure.” Limitations: Inter-individual variation was substantial. Laboratory analysis did not happen in a single batch.
MDH, 2015 ⁽⁵⁵⁾	Longitudinal	Minnesota	Drinking water	149	53 (mean)	8.3	Notes: Percent change was calculated using 2010 and 2014 Geometric Means as well as by averaging the change in participant’s individual PFC levels. The elimination rate for PFHxS was 8.3 years for both approaches. Results published in community report.
Olsen et al, 2007	Longitudinal	Minnesota	Retired workers	26	61 (mean)	8.5	Strengths: Strong “stop-exposure” condition with retirees, longitudinal measurements. Limitations: Small number of participants, the male/female ratio heavily skewed towards males, and the study population was older adults.
Worley et al. 2017 ⁽⁵⁶⁾	Longitudinal	Alabama	Land spreading	45	62.6 (mean)	15.5	Strengths: Considered ongoing exposures to PFAS in order to refine the half-life estimates, well-defined exposure, longitudinal nature.

							Limitations: Only two time points were analyzed (2010 and 2016). Older age of cohort (avg age 52 years in 2010, 63 years in 2016).
Zhang et al., 2013	Cross-sectional	China	General population	86	(22-88)	7.7	Strengths: Wide population age range across genders Limitations: Cross-sectional nature, only partial serum evaluation (primarily urine, which has more variable levels)

NEtFOSE, NEtFOSAA, NEtFOSA, FOSA | 2020

Substance Overview

N-Ethyl perfluorooctane sulfonamideoethanol (NEtFOSE), N-Ethyl perfluorooctane sulfonamidacetic acid (NEtFOSAA), N-Ethyl perfluorooctane sulfonamide (NEtFOSA), and Perfluorooctane sulfonamide (FOSA) are a group of chemicals in a larger group of contaminants called per- and polyfluoroalkyl substances (PFAS). This group of chemicals are found as impurities in stain repellants, food packaging and other packaging, and some fire-fighting foams.¹⁻⁴ NEtFOSE, NEtFOSAA, NEtFOSA, and FOSA are precursors to perfluorooctane sulfonate (PFOS) which means they are known to turn into PFOS in in the body and the environment (Figure 1).^{3,4-10} Exposure to high levels of PFOS is associated with a number of negative health effects in people and research animals.^{4,11-13}

Recommendations

Wisconsin does not currently have NR140 Groundwater Quality Public Health Enforcement Standards for NEtFOSE, NEtFOSAA, NEtFOSA, or FOSA.

DHS recommends a combined enforcement standard of 20 nanograms per liter (ng/L) for NEtFOSE, NEtFOSAA, NEtFOSA, FOSA, PFOS, and perfluorooctanoic acid (PFOA).^b The recommended standard is based on multiple studies that show NEtFOSE, NEtFOSAA, NEtFOSA, and FOSA breakdown into PFOS in both the body and the environment.⁴⁻⁹

DHS recommends that the preventive action limit for the combined standard of NEtFOSE, NEtFOSAA, NEtFOSA, FOSA, PFOA, and PFOS be set at 10% of the enforcement standard because PFOS has been shown to cause carcinogenic, teratogenic and interactive effects in people and research animals.^{7,14}

Health Effects

Studies are limited on the health effects of NEtFOSE, NEtFOSAA, NEtFOSA, and FOSA in people and research animals. However, several studies have shown that NEtFOSE exposure can cause health effects in research animals, such as mice and rats, with the most consistent effects being decreased body

Current Standards

Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

Recommended Standards

Enforcement Standard:	20 ng/L*
Preventive Action Limit:	2 ng/L*

*Applies to the sum of NEtFOSE, NEtFOSAA, NEtFOSA, FOSA, PFOA, and PFOS

a NEtFOSE, NEtFOSAA, NEtFOSA, and FOSA have been shown to breakdown into PFOA in the environment such as soil, aqueous solutions, activated sludge, and marine sediments.

b A combined groundwater standard of 20 ng/L for PFOA and PFOS is currently set in Wisconsin.

weight, decreased body weight gain, decreased food consumption, and decreased offspring weight.^{5,7,14-16}

Limited information is available about the carcinogenic, mutagenic, teratogenic, and interactive effects of NtEtFOSE, NtEtFOSAA, NtEtFOSA, and FOSA.^{c,5-7,17} One developmental study in rabbits found that NtEtFOSE increased the incidence of cleft palates in offspring.⁷ NtEtFOSE, NtEtFOSAA, NtEtFOSA, and FOSA may also cause interactive effects with PFOS and PFOA because they have similar chemical structure and therefore, they could act similarly to PFOS and PFOA in the body.^{14,18}

PFOS has been studied much more extensively.^{2,14,19} Studies among people have shown that PFOS exposure is associated with increased cholesterol, liver damage, pregnancy-induced hypertension, increased risk for thyroid disease, decreased antibody response to vaccines, decreased fertility, and small decreases in birth weight.^{2,14} Additionally, studies in research animals have found that both PFOS and PFOA can decrease offspring survival, weight, motor activity, delay of eye opening, antibody responses to vaccines, spatial learning and memory, as well as lead to kidney and liver damage.²

PFOS and PFOA have been shown to have teratogenic, carcinogenic, and interactive effects.¹⁴ PFOS can cause teratogenic effects such as bone development defects, ribcage defects, and cleft palate in research animals.^{2,14,20,21} The United States Environmental Protection Agency (EPA) have classified PFOS and PFOA as having suggestive evidence of carcinogenic potential.¹⁴

Chemical Breakdown

Studies suggest that NtEtFOSE is metabolized (turned) into NtEtFOSAA, NtEtFOSA, and FOSA and eventually to PFOS in the body (Figure 1).^{d,4-9} More specifically, in human liver cells and rats these studies have shown that transformation of NtEtFOSE to NtEtFOSAA, NtEtFOSA, and PFOS can occur. In fact, Xie et al. detected measureable amounts of NtEtFOSAA, NtEtFOSA, FOSA, and PFOS in the blood and liver of rats that were exposed only to NtEtFOSE.^{5,6} Further studies have shown that cytochrome P450s – a class of enzymes known to metabolize drugs and other contaminants – are involved in this metabolism that occurs within the liver. These enzymes turn NtEtFOSE, NtEtFOSAA, and NtEtFOSA into FOSA, FOSA into perfluorooctane sulfinic acid (PFOSI), and then PFOSI into PFOS.^{4-9,22} While studies show NtEtFOSE, NtEtFOSAA, NtEtFOSA, and FOSA are biotransformed in the body, the percentage of each chemical that is biotransformed into PFOS is unknown.^{6,42} Taken altogether, NtEtFOSE, NtEtFOSAA, NtEtFOSA, and FOSA can be metabolized into PFOS in both research animals and people.⁴⁻⁹

c Carcinogenic effects means the substance can cause cancer; mutagenic effects means the substance can cause DNA damage; teratogenic means the substance can cause birth defects; and interactive effects mean the substance can affect the toxicity of another substance or its toxicity can be affected by another substance.

d Chemical profiles for NtEtFOSE, NtEtFOSAA, NtEtFOSA and FOSA are described in Table A3.

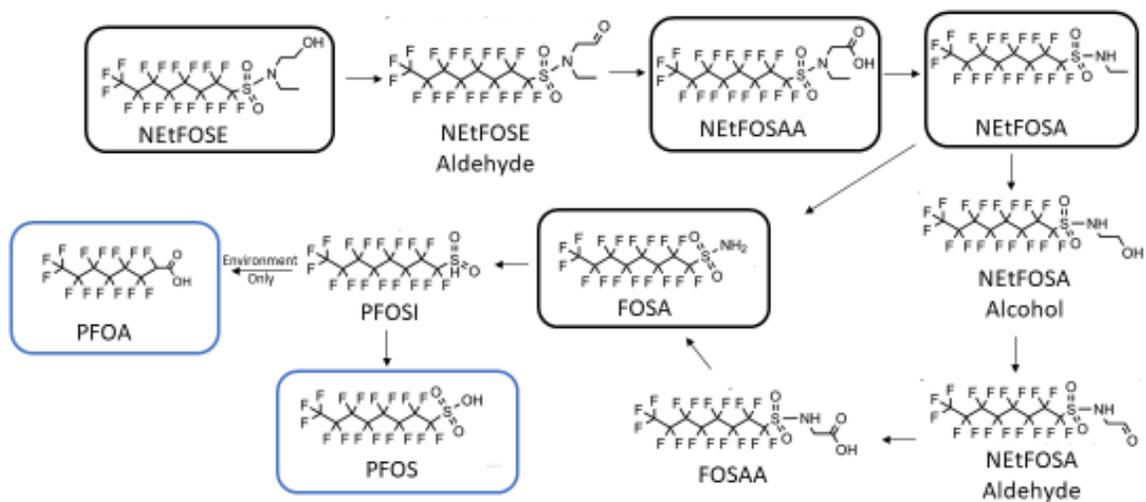


Figure 1. Metabolism process of NETFOSE, NETFOSAA, NETFOSA, and FOSA to PFOS in the body. Adapted from Mejia Avendano and Liu, 2015 and Parsons et al., 2008.^{4,23} Black boxes indicate the chemicals to be added to the combined PFOS and PFOA standard. Blue boxes indicate the chemicals that are already in the combined PFOS and PFOA standard.

The chemical structures of NETFOSE, NETFOSAA, NETFOSA, and FOSA indicate they will have long half-lives in the body. Studies in rat and human cell culture studies have shown that long chain PFAS (those six or more carbons) have a higher affinity for proteins in the blood and molecule transporters in tissues than short chain PFAS (those five or fewer carbons).^{24,25} Because of this, longer carbon chain PFAS are usually excreted out of the body at a slower rate than shorter carbon chained PFAS.^{8,9,18,22,26} For instance, PFOA, PFOA, and PFHxS have been shown to stay in the human body for long periods of time (half-lives of 5-8 years).^{e,14,26,27} Limited studies in rats have determined half-life data for NETFOSE, NETFOSAA, NETFOSA, and FOSA (see Table A1), however, PFAS half-lives in rodents may not accurately determine PFAS half-lives in humans because of different PFAS affinities to proteins in the blood and molecule transporters in tissues. To date, there are no half-lives of NETFOSE, NETFOSAA, NETFOSA, and FOSA in human epidemiology studies, however, because NETFOSE, NETFOSAA, NETFOSA, and FOSA are long chain PFAS, they are expected to act in similar fashion to PFOS and stay in the body for prolonged periods of time.^f While in the body, NETFOSE, NETFOSAA, NETFOSA, or FOSA can be metabolized into PFOS in the body contributing to the overall burden of PFOS observed in people, which is supported by epidemiology studies that have found NETFOSE, NETFOSAA, NETFOSA, FOSA, and PFOS in the same human blood samples.²⁸⁻³⁰

Exposure Routes

e PFOA has a seven carbon chain length. PFOS and PFHxS have six carbon chain lengths.

f NETFOSE, NETFOSAA, NETFOSA, and FOSA have eight carbon chain lengths.

People can be exposed to NtFOSE, NtFOSAA, NtFOSA, and FOSA by drinking contaminated water, swallowing contaminated soil, eating fish from contaminated water, eating food that was packaged in material that these substances, and breathing in or swallowing dust that contain these substances.^{2,31} Mothers exposed to some PFAS, specifically FOSA, can also expose their babies during pregnancy and breastfeeding.^{2,32,33}

In the environment, NtFOSE, NtFOSAA, NtFOSA, and FOSA can be found in water or soil as an impurity from the use of other PFAS in manufacturing and consumer products. They can also get into the water or soil through the use of fire-fighting foam. PFAS, like NtFOSE, NtFOSAA, NtFOSA, and FOSA, can move between groundwater and surface water. Once in groundwater, PFAS, like NtFOSE, NtFOSAA, NtFOSA, and FOSA, can travel long distances.²

NtFOSE, NtFOSAA, NtFOSA, and FOSA are known to degrade (breakdown) into PFOS and PFOA in abiotic (non-living) environments including soil, aqueous solutions, activated sludge, and marine sediments (Figure 1).^{4,10,34-38} Transformation of NtFOSE, NtFOSAA, NtFOSA, and FOSA to PFOS or PFOA can be highly variable in the environment and is dependent on temperature, sunlight, and microbial presence.^{4,34-38} While data on the half-lives of NtFOSE, NtFOSAA, NtFOSA, and FOSA in the environment are limited (Table A2), PFOS and PFOA have been shown to stay in the environment for more than a decade.^{4,22,24,36,38-41,49} Additionally, environmental studies provide evidence that NtFOSE, NtFOSAA, NtFOSA, and FOSA are found with PFOS and PFOA in the same water and soil samples, indicating they co-occur in the environment together.^{4,31,42}

Current Standard

Wisconsin does not currently have groundwater standards for NtFOSE, NtFOSAA, NtFOSA, or FOSA.⁴³

Standard Development

	NtFOSE	NtFOSAA	NtFOSA	FOSA
Federal Numbers				
Maximum Contaminant Level:	N/A	N/A	N/A	N/A
Health Advisory:	N/A	N/A	N/A	N/A
Drinking Water Concentration (Cancer Risk):	N/A	N/A	N/A	N/A
State Drinking Water Standard				
NR 809 Maximum Contaminant Level:	N/A	N/A	N/A	N/A
Acceptable Daily Intake				
EPA Oral Reference Dose:	N/A	N/A	N/A	N/A
Oncogenic Potential				
EPA Cancer Slope Factor:	N/A	N/A	N/A	N/A
Guidance Values				
ATSDR Oral Minimum Risk Level:	N/A	N/A	N/A	N/A
Literature Search				

Literature Search Dates:	Up to 2020	Up to 2020	Up to 2020	Up to 2020
Key toxicity studies?	Yes	No	No	Yes
Critical toxicity identified?	Yes	No	No	No

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for NtFOSE, NtFOSAA, NtFOSA, or FOSA.⁴⁴

Health Advisory

The EPA has not established health advisories for NtFOSE, NtFOSAA, NtFOSA, or FOSA.⁴⁵

Drinking Water Concentration (Cancer Risk)

The EPA has not established drinking water concentration based on a cancer risk level determination for NtFOSE, NtFOSAA, NtFOSA, or FOSA.⁴⁶

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for NtFOSE, NtFOSAA, NtFOSA, or FOSA.⁴⁷

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Information System (IRIS) program.

EPA Oral Reference Dose

The EPA does not have an oral reference dose for NtFOSE, NtFOSAA, NtFOSA, or FOSA.⁴⁶

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If we determine that something is

carcinogenic and there is no federal number or ADI from the EPA, DHS must set the standard at a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of NEtFOSE, NEtFOSAA, NEtFOSA, and FOSA, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of NEtFOSE, NEtFOSAA, NEtFOSA, and FOSA. If so, we look to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA and International Agency for Research on Cancer (IARC) have not evaluated the carcinogenicity of FOSA.^{46,48}

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for NEtFOSE, NEtFOSAA, NEtFOSA, or FOSA.⁴⁶

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information that was not considered when the value was established and indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For NEtFOSE, NEtFOSAA, NEtFOSA, and FOSA, we searched for values that been published on or before September, 2020. While the Agency for Toxic Substances and Disease Registry (ATSDR) reviewed the toxicity of NEtFOSE, NEtFOSAA, NEtFOSA, and FOSA in 2018, they did not establish any guidance values for NEtFOSE, NEtFOSAA, NEtFOSA, or FOSA.^{g,2}

Literature Search

Our literature review focused on relevant scientific literature on the health effects of NEtFOSE, NEtFOSAA, NEtFOSA, and FOSA published on or before September, 2020. We looked for studies related to NEtFOSE, NEtFOSAA, NEtFOSA, and FOSA toxicity or effects on a disease state in which information on exposure or dose was included as part of the study. Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans. For more details on the individual literature searches of NEtFOSE, NEtFOSAA, NEtFOSA, and FOSA, see appendix B below.

^g ATSDR stated that they did not identify any intermediate-duration oral studies for FOSA in their literature review.

We found key toxicity studies for NtFOSE and FOSA, but not for NtFOSAA or NtFOSA.

Critical Toxicity Studies

We did not identify any critical toxicity studies for NtFOSAA, NtFOSA, or FOSA.

We found one critical study for NtFOSE. Case et al. exposed pregnant rats and rabbits with varying concentrations of NtFOSE to determine effects on the pregnant mother and the development of their offspring.⁷ NtFOSE exposure caused maternal weight loss, increased resorptions, and decreased litter size in both research rats and rabbits. At higher concentrations, NtFOSE exposure led to severe maternal toxicity and maternal death in rabbits. In the offspring, NtFOSE exposure decreased offspring weight, delayed bone growth, and increased incidences of cleft palate. For more detail regarding critical studies, see Appendix C below.

Key health effects

We did not identify any studies evaluating carcinogenic, teratogenic, mutagenic, or interactive effects of NtFOSAA, NtFOSA, or FOSA. For NtFOSE, we found one study that suggested teratogenic effects. Case et al. found that NtFOSE exposure (35 mg/kg-d) increased the incidence of cleft palate in offspring of pregnant rats.^{h,7} However, the researchers considered cleft palate at the highest dose level to be incidental and not a teratogenic effect. Despite this, two animal studies have indicated that exposure to PFOS, a major metabolite of NtFOSE, can contribute to cleft palate in the offspring if the mother is exposed during pregnancy.^{20,21} Therefore, we considered the observed cleft palate attributable to the high dose levels of NtFOSE and its ability to breakdown to PFOS.⁷ Furthermore, NtFOSE, NtFOSAA, NtFOSA, and FOSA may cause interactive effects with PFOS and PFOA because they have similar chemical structure and therefore, NtFOSE, NtFOSAA, NtFOSA, and FOSA could act similarly to PFOS and PFOA in the body.^{2,14}

PFOS has been shown to have, teratogenic, and interactive effects in people and research animals. Multiple studies have shown that PFOS can cause teratogenic effects such as bone development defects, ribcage defects, and other birth defects (such as cleft palate described above) in research animals.^{2,14} Additionally, the EPA has classified PFOS as having suggestive evidence of carcinogenic potential.^{2,14}

^h NtFOSE exposure to 35mg/kg-d also caused maternal toxicity such as decreased body weight gain, decreased food consumption, and thin/ emaciated pregnant rats.

Standard Selection

DHS recommends a combined enforcement standard of 20 ng/L for NEtFOSE, NEtFOSAA, NEtFOSA, FOSA, PFOS and PFOA.

There are no federal numbers and no state drinking water standards for NEtFOSE, NEtFOSAA, NEtFOSA, and FOSA. Additionally, the EPA has not evaluated the carcinogenicity or established an acceptable daily intake (oral reference dose) for NEtFOSE, NEtFOSAA, NEtFOSA, and FOSA.

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information

DHS recommends a combined enforcement standard of 20 nanograms per liter (ng/L) for NEtFOSE, NEtFOSAA, NEtFOSA, FOSA, PFOS and PFOA. Multiple studies have shown that NEtFOSE, NEtFOSAA, NEtFOSA, and FOSA metabolize or breakdown into PFOS in the body.^{4,11,34-37,42,49} Additionally, studies have shown that similarly structured long carbon chained PFAS can have long half-lives which would provide sufficient time for NEtFOSE, NEtFOSAA, NEtFOSA, and FOSA to be metabolized into PFOS in people.^{24,25} Moreover, NEtFOSE, NEtFOSAA, NEtFOSA, and FOSA have been found to co-occur with PFOS and PFOA in the environment.^{4,31,42} Therefore, exposure to NEtFOSE, NEtFOSAA, NEtFOSA, and FOSA can contribute to the burden of PFOS in people and exposure to high levels of PFOS is associated with a number of health effects among people.^{10,27} To ensure adequate protection of people from exposure to PFOS resulting from ingestion of NEtFOSE, NEtFOSAA, NEtFOSA, and FOSA, we recommend a combined enforcement standard of 20 ng/L for NEtFOSE, NEtFOSAA, NEtFOSA, FOSA, PFOS and PFOA.

DHS recommends a preventive action limit of 2 ng/L for NEtFOSE, NEtFOSAA, NEtFOSA, FOSA, PFOS and PFOA.

DHS recommends that the preventive action limit for NEtFOSE, NEtFOSAA, NEtFOSA, FOSA, PFOS and PFOA be set at 10% of the enforcement standard because PFOS has been shown to have carcinogenic, teratogenic, and interactive effects in people and research animals. Multiple studies have shown that PFOS can cause teratogenic effects such as bone development defects, ribcage defects, and other birth defects in research animals.^{2,14} Additionally, the EPA has classified PFOS as having suggestive evidence of carcinogenic potential.^{2,14} Last, NEtFOSE, NEtFOSAA, NEtFOSA, and FOSA may cause interactive effects with PFOS and PFOA because they have similar chemical structure and therefore, NEtFOSE, NEtFOSAA, NEtFOSA, and FOSA could act similarly to PFOS and PFOA in the body.^{2,14}

Prepared by Gavin Dehnert, Ph.D., Brita Kilburg-Basnyat, Ph.D, and Sarah Yang, Ph.D.

Wisconsin Department of Health Services

References

1. Chu S, Letcher RJ. In vitro metabolic formation of perfluoroalkyl sulfonamides from copolymer surfactants of pre- and post-2002 scotchgard fabric protector products. *Environmental science & technology*. 2014;48(11):6184-6191.
2. ATSDR. Toxicological Profile for Perfluoroalkyls - Draft for Public Comment. In: Registry AftSaD, ed. Atlanta, GA2017.
3. Anderson RH, Long GC, Porter RC, Anderson JK. Occurrence of select perfluoroalkyl substances at U.S. Air Force aqueous film-forming foam release sites other than fire-training areas: Field-validation of critical fate and transport properties. *Chemosphere*. 2016;150:678-685.
4. Mejia Avendaño S, Liu J. Production of PFOS from aerobic soil biotransformation of two perfluoroalkyl sulfonamide derivatives. *Chemosphere*. 2015;119:1084-1090.
5. Xie W, Wu Q, Kania-Korwel I, et al. Subacute exposure to N-ethyl perfluorooctanesulfonamidoethanol results in the formation of perfluorooctanesulfonate and alters superoxide dismutase activity in female rats. *Archives of toxicology*. 2009;83(10):909-924.
6. Xu L, Krenitsky DM, Seacat AM, Butenhoff JL, Anders M. Biotransformation of N-ethyl-N-(2-hydroxyethyl) perfluorooctanesulfonamide by rat liver microsomes, cytosol, and slices and by expressed rat and human cytochromes P450. *Chemical research in toxicology*. 2004;17(6):767-775.
7. Case MT, York RG, Christian MS. Rat and rabbit oral developmental toxicology studies with two perfluorinated compounds. *International journal of toxicology*. 2001;20(2):101-109.
8. Xie W, Wu Q, Kania-Korwel I, et al. Subacute exposure to N-ethyl perfluorooctanesulfonamidoethanol results in the formation of perfluorooctanesulfonate and alters superoxide dismutase activity in female rats. *Archives of Toxicology*. 2009;83(10):909-924.
9. Manning R, Bruckner J, Mispagel M, Bowen J. Metabolism and disposition of sulfluramid, a unique polyfluorinated insecticide, in the rat. *Drug metabolism and disposition*. 1991;19(1):205-211.
10. Liu J, Mejia Avendaño S. Microbial degradation of polyfluoroalkyl chemicals in the environment: a review. *Environ Int*. 2013;61:98-114.
11. Xu L, Krenitsky DM, Seacat AM, Butenhoff JL, Anders MW. Biotransformation of N-ethyl-N-(2-hydroxyethyl)perfluorooctanesulfonamide by rat liver microsomes, cytosol, and slices and by expressed rat and human cytochromes P450. *Chemical Research in Toxicology*. 2004;17(6):767-775.
12. Xu L, Krenitsky DM, Seacat AM, Butenhoff JL, Tephly TR, Anders MW. N-glucuronidation of perfluorooctanesulfonamide by human, rat, dog, and monkey liver microsomes and by expressed rat and human UDP-glucuronosyltransferases. *Drug metabolism and disposition: the biological fate of chemicals*. 2006;34(8):1406-1410.
13. Xu L, Seacat AM, Butenhoff JL, Anders MW. Biotransformation of N-ethyl-N-(2-hydroxyethyl)perfluorooctanesulfonamide E (N-EtFOSE) by rat liver microsomes, cytosol, and slices. *Toxicological Sciences*. 2003;72:314-314.

14. DHS. *Recommended Public Health Groundwater Quality Standards: Scientific Support Documents for Cycle 10 Substances*. 2019.
15. USEPA. Memorandum: Chlorantraniliprole; Human Health Risk Assessment for Proposed Use on Tobacco. In: Office of Prevention P, and Toxic Substances, ed. Vol DP#3692242010.
16. USEPA. Pesticide Factsheet: Chlorantraniliprole. In: Office of Prevention P, and Toxic Substances, ed2008.
17. Bonefeld-Jørgensen EC, Long M, Fredslund SO, Bossi R, Olsen J. Breast cancer risk after exposure to perfluorinated compounds in Danish women: a case-control study nested in the Danish National Birth Cohort. *Cancer causes & control : CCC*. 2014;25(11):1439-1448.
18. Christensen GY, Sarah. Perfluorooctane sulfonic acid (PFOS); Perfluorooctanoic acid (PFOA). In: Services WDoH, ed. Cycle 10 of Recommended Groundwater Standards2019.
19. Seacat AM, Luebker DJ. *Toxicokinetic Study of Perfluorooctane Sulfonamide (PFOSA; T-7132.2) in Rats*. 3M Medical Department, Corporate Toxicology;2000.
20. Era S, Harada KH, Toyoshima M, et al. Cleft palate caused by perfluorooctane sulfonate is caused mainly by extrinsic factors. *Toxicology*. 2009;256(1-2):42-47.
21. Thibodeaux JR, Hanson RG, Rogers JM, et al. Exposure to perfluorooctane sulfonate during pregnancy in rat and mouse. I: maternal and prenatal evaluations. *Toxicological sciences : an official journal of the Society of Toxicology*. 2003;74(2):369-381.
22. Chang S, Mader BT, Lindstrom KR, et al. Perfluorooctanesulfonate (PFOS) Conversion from N-Ethyl-N-(2-hydroxyethyl)-perfluorooctanesulfonamide (EtFOSE) in male Sprague Dawley rats after inhalation exposure. *Environmental research*. 2017;155:307-313.
23. Parsons JR, Sáez M, Dolfig J, de Voogt P. Biodegradation of perfluorinated compounds. *Reviews of environmental contamination and toxicology*. 2008;196:53-71.
24. Zhao S, Ma X, Fang S, Zhu L. Behaviors of N-ethyl perfluorooctane sulfonamide ethanol (N-EtFOSE) in a soil-earthworm system: transformation and bioaccumulation. *Science of the Total Environment*. 2016;554:186-191.
25. Weaver YM, Ehresman DJ, Butenhoff JL, Hagenbuch B. Roles of rat renal organic anion transporters in transporting perfluorinated carboxylates with different chain lengths. *Toxicological Sciences*. 2010;113(2):305-314.
26. Gebbink WA, Glynn A, Berger U. Temporal changes (1997-2012) of perfluoroalkyl acids and selected precursors (including isomers) in Swedish human serum. *Environmental pollution (Barking, Essex : 1987)*. 2015;199:166-173.
27. USEPA. Health Effects Support Document for Perfluorooctane Sulfonate (PFOS). 2016.
28. Lum KJ, Sundaram R, Barr DB, Louis TA, Buck Louis GM. Perfluoroalkyl Chemicals, Menstrual Cycle Length, and Fecundity: Findings from a Prospective Pregnancy Study. *Epidemiology (Cambridge, Mass)*. 2017;28(1):90-98.
29. Kvaalem HE, Nygaard UC, Lodrup Carlsen KC, Carlsen KH, Haug LS, Granum B. Perfluoroalkyl substances, airways infections, allergy and asthma related health outcomes - implications of gender, exposure period and study design. *Environment international*. 2020;134:105259.

30. Blake BE, Pinney SM, Hines EP, Fenton SE, Ferguson KK. Associations between longitudinal serum perfluoroalkyl substance (PFAS) levels and measures of thyroid hormone, kidney function, and body mass index in the Fernald Community Cohort. *Environmental pollution (Barking, Essex : 1987)*. 2018;242(Pt A):894-904.
31. Sinclair E, Mayack DT, Roblee K, Yamashita N, Kannan K. Occurrence of perfluoroalkyl surfactants in water, fish, and birds from New York State. *Archives of Environmental Contamination and Toxicology*. 2006;50(3):398-410.
32. Impinen A, Nygaard UC, Lodrup Carlsen KC, et al. Prenatal exposure to perfluoroalkyl substances (PFASs) associated with respiratory tract infections but not allergy- and asthma-related health outcomes in childhood. *Environmental research*. 2018;160:518-523.
33. Yao Q, Shi R, Wang C, et al. Cord blood Per- and polyfluoroalkyl substances, placental steroidogenic enzyme, and cord blood reproductive hormone. *Environment international*. 2019;129:573-582.
34. Benskin JP, Li B, Ikonomou MG, Grace JR, Li LY. Per- and polyfluoroalkyl substances in landfill leachate: patterns, time trends, and sources. *Environ Sci Technol*. 2012;46(21):11532-11540.
35. Boulanger B, Vargo JD, Schnoor JL, Hornbuckle KC. Evaluation of perfluorooctane surfactants in a wastewater treatment system and in a commercial surface protection product. *Environ Sci Technol*. 2005;39(15):5524-5530.
36. Rhoads KR, Janssen EM, Luthy RG, Criddle CS. Aerobic biotransformation and fate of N-ethyl perfluorooctane sulfonamidoethanol (N-EtFOSE) in activated sludge. *Environ Sci Technol*. 2008;42(8):2873-2878.
37. Rhoads KR, Rostkowski KH, Kitanidis PK, Criddle CS. Use of on-site bioreactors to estimate the biotransformation rate of N-ethyl perfluorooctane sulfonamidoethanol (N-EtFOSE) during activated sludge treatment. *Chemosphere*. 2013;92(6):702-707.
38. Nguyen TV, Reinhard M, Gin KY. Rate laws and kinetic modeling of N-ethyl perfluorooctane sulfonamidoethanol (N-EtFOSE) transformation by hydroxyl radical in aqueous solution. *Water research*. 2013;47(7):2241-2250.
39. Butenhoff J, Seacat A. Comparative sub-chronic toxicity of perfluorooctanesulfonate (PFOS) and N-ethyl perfluorooctanesulfonamidoethanol (N-EtFOSE) in the rat. *Toxicologist*. 2001;60:348.
40. Lange FTS, C.; Brauch, H.J. *Perfluoroalkylcarboxylates and --sulfonates: emerging contaminants for drinking water supplies?* : Association of River Waterworks--RIWA,;2006.
41. Hao J, Wang P, Kang Y, et al. Degradation of Perfluorooctane Sulfonamide by Acinetobacter Sp. M and Its Extracellular Enzymes. *Chemistry--An Asian Journal*. 2019;14(16):2780-2784.
42. Benskin JP, Holt A, Martin JW. Isomer-specific biotransformation rates of a perfluorooctane sulfonate (PFOS)-precursor by cytochrome P450 isozymes and human liver microsomes. *Environ Sci Technol*. 2009;43(22):8566-8572.
43. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 140*2017.
44. USEPA. National Primary Drinking Water Regulations. <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>. Published 2018. Accessed.

45. USEPA. Drinking Water Contaminant Human Health Effects Information. <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>. Published 2019. Accessed.
46. USEPA. Secondary Drinking Water Standards: Guidance for Nuisance Chemicals. <https://www.epa.gov/dwstandardsregulations/secondary-drinking-water-standards-guidance-nuisance-chemicals>. Published 2019. Accessed.
47. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 8092018*.
48. IARC. List of Classification, Volumes 1-123. <https://monographs.iarc.fr/list-of-classifications-volumes/>. Published 2018. Accessed May 17, 2019.
49. Ross MS, Wong CS, Martin JW. Isomer-specific biotransformation of perfluorooctane sulfonamide in Sprague-Dawley rats. *Environ Sci Technol*. 2012;46(6):3196-3203.
50. Grossman MR, Mispagel ME, Bowen JM. Distribution and tissue elimination in rats during and after prolonged dietary exposure to a highly fluorinated sulfonamide pesticide. *Journal of agricultural and food chemistry*. 1992;40(12):2505-2509.
51. Vitayavirasuk B, Bowen JM. Pharmacokinetics of sulfluramid and its metabolite desethylsulfluramid after intravenous and intraruminal administration of sulfluramid to sheep. *Pesticide science*. 1999;55(7):719-725.
52. Lange C. The aerobic biodegradation of N-EtFOSE alcohol by the microbial activity present in municipal wastewater treatment sludge. *For 3M Company*. 2000.
53. Ritter L, Totman C, Krishnan K, Carrier R, Vezina A, Morisset V. Deriving uncertainty factors for threshold chemical contaminants in drinking water. *Journal of toxicology and environmental health Part B, Critical reviews*. 2007;10(7):527-557.
54. USEPA. A Review of the Reference Dose and Reference Concentration Processes. 2002(EPA/630/P-02/002F).
55. ITRC. PFAS Fact Sheets: PFAS Water and Soil Values Table Excel File. In: Council ITR, ed2020.
56. Albrecht PP, Torsell NE, Krishnan P, et al. A species difference in the peroxisome proliferator-activated receptor alpha-dependent response to the developmental effects of perfluorooctanoic acid. *Toxicological sciences : an official journal of the Society of Toxicology*. 2013;131(2):568-582.
57. Louis GM, Sapra KJ, Barr DB, Lu Z, Sundaram R. Preconception perfluoroalkyl and polyfluoroalkyl substances and incident pregnancy loss, LIFE Study. *Reproductive toxicology (Elmsford, NY)*. 2016;65:11-17.
58. Louis GM, Chen Z, Schisterman EF, et al. Perfluorochemicals and human semen quality: the LIFE study. *Environ Health Perspect*. 2015;123(1):57-63.
59. Halldorsson TI, Rytter D, Haug LS, et al. Prenatal Exposure to Perfluorooctanoate and Risk of Overweight at 20 Years of Age: A Prospective Cohort Study. *Environmental Health Perspectives*. 2012;120(5):668-673.

Appendix A: Biotransformation of PFAS Precursors in the Body and Environment

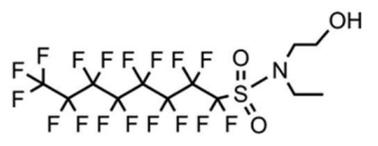
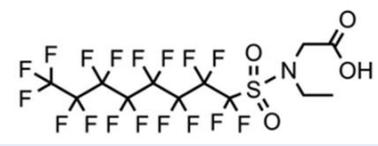
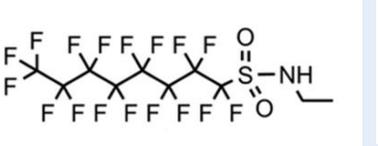
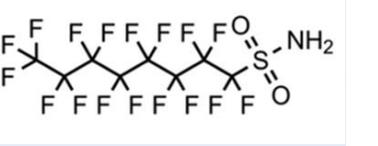
Table A I. NEtFOSE, NEtFOSA, NEtFOSAA, and FOSA biotransformation studies in the human cell cultures and research animals.

Compound	Microcosm	Summary	Reference
NEtFOSE	Rats	Rats were exposed to NEtFOSE. NEtFOSE was biotransformed into FOSE, NEtFOSAA, FOSA, and PFOS. These metabolites were found in both the liver and the blood stream.	Xie et al., 2009 ⁵
NEtFOSE	Rat liver slices and microsome cell cultures	NEtFOSE was exposed to rat liver slices, microsomes, and cytosol. NEtFOSE was metabolized by rat liver slices and microsomes into FOSE, NEtFOSAA and FOSA by cytochrome P450s. FOSA was further metabolized by liver slices into PFOS by cytochrome P450s.	Xu et al., 2004 ⁶
NEtFOSA	Human microsome cell cultures	NEtFOSA was exposed to human microsomes with recombinant human cytochrome P450s. NEtFOSA was metabolized into FOSA by cytochrome P450s. The estimated arithmetic mean half-life of NEtFOSA was 0.7 to 20 minutes.	Benskin et al., 2009 ⁴²
NEtFOSA	Rats	Rats were fed a diet that contained NEtFOSA. NEtFOSA was metabolized into FOSA. The estimated arithmetic mean half-life was approximately 11 days in the blood, 7 days in the liver, and 1 day in the fat.	Grossman et al., 1992 ⁵⁰
FOSA	Rats	Rats were given a single dose of FOSA. The estimated arithmetic mean half-life of FOSA was approximately 5.2 days in the liver and less than 4 days in the blood.	Seacat 2000 ¹⁹
FOSA	Rats	Rats were exposed to FOSA. FOSA was biotransformed into PFOS isomers and was detected in the blood, liver, and other tissues. The estimated arithmetic mean half-life of FOSA was 2.5 – 6 days	Ross et al., 2012 ⁴⁹
NEtFOSA	Rats	Rats were given a single dose of NEtFOSA. NEtFOSA was biotransformed into FOSA. The estimated arithmetic mean half-life of NEtFOSA was 4.2 days.	Manning et al. 1991 ⁹
NEtFOSA	Sheep	Sheep were administered an intravenous or intraruminal bolus dose of NEtFOSA. NEtFOSA was biotransformed into FOSA. The estimated arithmetic mean half-life of NEtFOSA was 2.1 – 3.1 days.	Vitayavirasuk and Bowen, 1999 ⁵¹

Table A2. Half-live studies in the environment.

Compound	Microcosm	Length of study	Estimated Arithmetic mean half-life	Notes	Reference
NETFOSE	Activated sludge	35 days	Less than 5 days (low dose) 2-3 days (high dose)	Produced both PFOA (0.6%) and PFOS (7%).	Lange, 2000 ⁵²
NETFOSE	Activated sludge	4 days	4.2 days	Produced PFOS (0.6%)	Boulanger et al., 2005 ³⁵
NETFOSE	Activated sludge	10 days	0.7 days	N/A	Rhoads et al., 2008 ³⁶
NETFOSE	Marine sediment	120 days	44 days (25 Degrees Celsius) 160 days (4 Degrees Celsius)	Produced PFOS (12% at 25 Degrees Celsius). Produced PFOS (0.44% at 4 Degrees Celsius).	Benskin et al. 2012 ³⁴
NETFOSE	Rate laws and kinetic modeling of aqueous solutions	N/A	1 day	Produced FOSA (18%). Produced PFOA (39%).	Nguyen et al., 2013 ³⁸
NETFOSA	Aerobic soil	182 days	13.9 days	Produced PFOS (4%).	Mejia Avendano and Liu, 2015 ⁴

Appendix B: Chemical Profiles for NEtFOSE, NEtFOSAA, NEtFOSA, and FOSA

	NEtFOSE	NEtFOSAA	NEtFOSA	FOSA
Structure:				
CAS Number:	1691-99-2	1336-61-4	4151-50-2	754-91-6
Formula:	C ₁₂ H ₁₀ F ₁₇ NO ₃ S	C ₁₂ H ₈ F ₁₇ NO ₄ S	C ₁₀ H ₆ F ₁₇ NO ₂ S	C ₈ H ₂ F ₁₇ NO ₂ S
Molar Mass:	571.25 g/mol	585.23 g/mol	527.2 g/mol	499.15 g/mol
Synonyms:	N-Ethyl-N-(2-hydroxyethyl)perfluorooctanesulfonamide N-EtFOSE	2-(N-ethylperfluorooctanesulfonyl)glycine EtFOSAA Glycine, N-ethyl-N-((heptadecafluorooctyl)sulfonyl)- N-Ethyl-N-(1,1,2,2,3,3,4,4,5,5,6,6,7,7,8,8-heptadecafluorooctane-1-sulfonyl)glycine	Sulfluramid N-Ethylperfluorooctanesulfonamide N-ethyl-1,1,2,2,3,3,4,4,5,5,6,6,7,7,8,8-heptadecafluorooctane-1-sulfonamide EtFOSA	1,1,2,2,3,3,4,4,5,5,6,6,7,7,8,8-heptadecafluorooctane-1-sulfonamide PFOSA Perfluorooctane sulfonamide Perfluorooctylsulfonamide Heptadecafluorooctanesulfonamide Perfluorooctanesulfonic acid amide

Appendix C. Available Toxicity Data

Literature Search

Our literature review focused on relevant scientific literature on the health effects of NtFOSE, NtFOSAA, NtFOSA, and FOSA published on or before September, 2020. We looked for studies related to NtFOSE, NtFOSAA, NtFOSA, and FOSA toxicity or effects on a disease state in which information on exposure or dose was included as part of the study.ⁱ Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

Approximately 100 toxicity studies were returned by the search engines for NtFOSE, approximately 50 studies for NtFOSAA, approximately 40 for NtFOSA and approximately 200 studies for FOSA. We excluded studies on non-mammalian or cell systems, non-oral exposure routes, and those that did not evaluate health risks from further review. After applying these exclusion criteria, we located two key studies on NtFOSE, one key toxicity study on FOSA, and no key studies for NtFOSAA and NtFOSA. The key studies are summarized in Table B-1.

To be considered a critical toxicity study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.^{j-k} For NtFOSE, one of the key studies met the criteria to be considered a critical toxicity study, but the key FOSA study did not meet the criteria to be considered a critical toxicity study (see Table B-2).

In our search, we located a handful of epidemiology studies for FOSA in our search (See Table B-3 for a summary). While multiple potential exposure sources and the ability for other PFAS compounds to cause similar health effects preclude using these data to establish a health-based value, such studies are helpful in identifying the crucial effects and ensuring that the animal data used to establish the standard is relevant to people.

ⁱ We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

Title/abstract: NtFOSE or EtFOSE or "N-Ethyl-N-(2-hydroxyethyl) perfluorooctanesulfonamide" for NtFOSE

NtFOSAA or EtFOSAA or "N-Ethyl – Perfluorooctane sulfonamidaacetic acid" for NtFOSAA

NtFOSA or EtFOSA or "N-Ethyl – Perfluorooctane sulfonamide" for NtFOSA

PFOSA or "Perfluorooctane sulfonamide" for FOSA

Language: English

We also searched online for toxicity studies published by national research programs.

^j Appropriate toxicity values include the no observable adverse effect level (NOAEL), lowest observable adverse effect level (LOAEL), and benchmark dose (BMD).

^k Due to the limited availability of data for this substance, we considered a study to be of an appropriate duration if animals were exposed for at least 28 days or if the exposure occurred during pregnancy. We accounted for differences in exposure duration through the use of an uncertainty factor.

NEtFOSE Critical Toxicity Studies

To compare results from studies identified in the literature review, we calculated an acceptable daily intake (ADI) for each study/effect. The ADI is the estimated amount of NEtFOSE that a person can be exposed to every day and not experience health impacts. As such, we calculated ADI by dividing a toxicity value from either a no-observed adverse effect level (NOAEL), lowest observed adverse effect level (LOAEL), or benchmark dose (BMD) identified in a study by a factor accounting for various sources of scientific uncertainty.^l We included uncertainty factors to account for differences between humans and animals, differences between healthy and sensitive human populations, using data from short-term experiments to protect against effects from long-term exposure, and using data where a health effect was observed to estimate the level that does not cause an effect, as appropriate. To ensure appropriate protection, we have chosen to not use studies that have significant uncertainty as the basis for the recommended enforcement standards.^m This approach is consistent with that taken by EPA when establishing oral reference doses.⁵⁴

Case et al., 2001

Case et al. exposed pregnant rats and rabbits with varying concentrations of NEtFOSE to determine the effects of NEtFOSE on the pregnant mother and the development of their offspring.⁷

Rats

Case et al. exposed pregnant female rats to different concentrations of NEtFOSE (0, 5, 10, 20, 25, and 35 milligrams of NEtFOSE per kilogram body weight per day or mg/kg-d) by gavage for gestational days 6 through 17. NEtFOSE caused some significant effects including decreased body weight gain and decreased offspring weights in pregnant females and their offspring (Table C1).

Table C1. Significant Effects Observed in Pregnant Female Rats and Offspring in Case et al., 2001⁷

Effects observed in pregnant females		Dose (mg/kg-d)					
		1	5	10	20	25	35
Growth	Decreased body weight gain during pregnancy			✓	✓	✓	✓
	Observed as thin/emaciated						✓
	Reduced food consumption					✓	✓
Reproduction	Increased reabsorption						*
Effects observed in offspring							
Growth	Decreased fetal weights				✓	✓	✓
Development	Increased incidence of cleft palate						*
	Increased incident of anasarca (swelling of the entire body)						*

^l The NOAEL is the highest dose tested that did not cause an adverse effect, the LOAEL is the lowest dose tested that caused an adverse effect, and the BMD is an estimation of the dose that would cause a specific level of response (typically 5 or 10%).

^m DHS considers a study to have significant uncertainty if the total uncertainty factors is greater than 3,000.

*The authors considered this an incidental finding and not NtEtFOSE related. Increased reabsorption was defined by the authors as “possibly” increased and cleft palate in an entire litter and whole body swelling in two offspring in the same litter was considered by the authors to be incidental.

From this dose-ranging experiment, we identified a no observable adverse effect level (NOAEL) of 5 mg/kg-d based on decreased body weight gain in pregnancy and decreased fetal weights which are consistent with the effects of other PFAS.⁵⁵ We determined the candidate ADI using this NOAEL and a total uncertainty factor of 1,000 to account for differences between humans and research animals (10), differences among people (10), and the limited availability of information (10). We obtained a candidate ADI of 5,000 ng/kg-d for NtEtFOSE from this study.

In a follow-up developmental study, the researchers exposed pregnant female rats to different concentrations of NtEtFOSE (0, 1, 5, 10, and 20 mg/kg-d NtEtFOSE) by gavage for gestational days 6 through 17. NtEtFOSE caused some significant effects including decreased body weight gain and decreased offspring weights in pregnant females and their offspring (Table C2).

Table C2. Significant Effects Observed in Pregnant Female Rats and Offspring in Case et al., 2001

Effects observed in pregnant females		Dose (mg/kg-d)			
		1	5	10	20
Body Weight	Decreased body weight gain during pregnancy			✓	✓
Food consumption	Reduced maternal food consumption			✓	✓
Effects observed in offspring					
Body Weight	Decreased offspring weights			✓	✓
Ossification	Increase in reversible delays in ossification				*

*The authors did not consider this finding to be toxicologically meaningful and attributed the delay in ossification to small fetus size that was corrected after birth (see Case et al., 2009) for more details⁵⁶

From this developmental experiment, the researchers identified a maternal and developmental No Observed Effect Level (NOEL) of 5 mg/kg-d based on decreased body weight and reduced food consumption in pregnant rats and decreased body weights in offspring. We determined the candidate ADI using this NOEL and a total uncertainty factor of 1,000 to account for differences between humans and research animals (10), differences among people (10), and the limited availability of information (10). We obtained a candidate ADI of 5,000 ng/kg-d for NtEtFOSE in pregnant females.

Rabbits

Case et al. also exposed pregnant female rabbits to different concentrations of NtEtFOSE (0, 1, 5, 10, 25, 50, and 75 mg/kg-d NtEtFOSE) by gavage for gestational days 6 through 20. NtEtFOSE caused some significant effects including decreased body weight gain and decreased offspring weights in pregnant females and their offspring (Table C3). Females dosed ≥25 mg/kg-d were terminated early on Day 15 due to severe toxicity including early maternal death and lethargy.

Table C3. Significant Effects Observed in Pregnant Female Rabbits and Offspring in Case et al., 2001

Effects observed in pregnant females		Dose (mg/kg-d)					
		1	5	10	25	50	75

Growth	Weight loss	✓	✓	✓	✓	✓
	Reduced maternal food consumption			✓	✓	✓
Clinical toxicity	Ungroomed coats (indicates stress)			✓	✓	✓
	No or scant feces			✓	✓	✓
	Became lethargic			✓	✓	✓
Death	Early maternal death *				✓	✓
	Sacrificed early due to severe toxicity			✓	✓	✓
Reproduction	Increased abortions	✓	✓ ^a	X	X	X
	Increased resorptions	✓	X	X	X	X
Effects observed in offspring						
	Reduced litter size	✓	X	X	X	X
Development	Decreased offspring weight	✓	X	X	X	X

*The authors did not consider the death of one animal in the 1mg/kg-d group this finding to be toxicologically meaningful (see Case et al., 2009 for more details).⁵⁶

X= sacrificed in moribund condition on Gestational Day 15 due to severe toxicity

^a=all 5 pregnant females aborted their litters leading to no offspring available to determine litter size, resorptions, or fetal weight in the 10 mg/kg-d dose group,

From this experiment, we identified a NOAEL of 1 mg/kg-d based on weight loss, reduced food consumption in pregnant females, early maternal death, reduced litter size, and decreased offspring weights. We determined the candidate ADI using this NOAEL and a total uncertainty factor of 1,000 to account for differences between humans and research animals (10), differences among people (10), and the limited availability of information (10). We obtained a candidate ADI of 1,000 ng/kg-d for NETFOSE for pregnant females.

In a follow-up study, Case et al. exposed pregnant female rabbits to different concentrations of NETFOSE (0.1, 1.0, 2.5, and 3.75 mg/kg-d NETFOSE) by gavage for gestational days 7 through 20. NETFOSE caused some significant effects, including decreased maternal body weight gain and decreased offspring weights, in pregnant females and their offspring (Table C4).

Table C4. Significant Effects Observed in Pregnant Female Rabbits and Offspring in Case et al., 2001

Effects observed in pregnant females		Dose (mg/kg-d)			
		0.1	1.0	2.5	3.75
Growth	Reduced body weight gains		✓	✓	✓
	Reduced maternal food consumption		✓	✓	✓
Clinical toxicity	No or scant feces			✓	✓
Reproduction	Increased abortions			✓	✓
	Increased resorptions				✓

From this experiment, the researchers identified a maternal NOEL of 0.1 mg/kg-d based on reduced body weights and food consumption. The developmental NOEL of 1.0 mg/kg-d was based on increased abortions and reabsorptions. We determined the candidate ADI using the maternal NOEL and a total uncertainty factor of 1,000 to account for differences between humans and research animals (10),

differences among people (10), and the limited availability of information (10). We obtained a candidate ADI of 100 ng/kg-d for NEtFOSE for pregnant females.

Table C-5. Key toxicity Studies from Literature Review

Compound	Study Type	Species	Duration	Doses (mg/kg-d)	Route	Endpoints	Toxicity Value (mg/kg-d)	Reference
NETFOSE	Developmental	Rabbit	Days 6 to 20	0, 1, 5, 10, 25, 50, 75	Gavage	Weight loss/decreased body weight gain at ≥ 5 mg/kg-d; decreased food consumption, ungroomed coats, no or scant feces and lethargic in ≥ 25 mg/kg-d; rabbits administered ≥ 25 mg/kg-d were sacrificed on gestational day 15 due to severity of symptoms.	NOAEL: 1	Case et al., 2001 ⁷
NETFOSE	Developmental	Rabbit	Days 7 to 20	0, 0.1, 1.0, 2.5, 3.75	Gavage	Increased abortions at ≥ 2.5 mg/kg-d; Decreased body weight gains at doses ≥ 2.5 mg/kg-d; Reduced body weight gains in doses on gestational days 7 to 13 for 1.0 mg/kg-d; weight reductions correlated with decreased food consumption; Increased number of late resorptions in 3.75 mg/kg-d	NOAEL: 0.1	Case et al., 2001 ⁷
NETFOSE	Developmental	Sprague-Dawley Female Rats	Day 6 to 17	0, 1, 5, 10, 20, 25, 35	Gavage	Emaciation and thin observation in high dose group; Reduced body weight gains > 10 mg/kg-d and reduced feed consumption for > 25 mg/kg-d. Reduced fetal weights > 20 mg/kg-d. One dam administered 35 mg/kg-d had 13 fetuses with cleft palate and 2 with anasarca (swelling).	NOAEL: 5	Case et al., 2001 ⁷
NETFOSE	Developmental	Sprague-Dawley Female Rats	Day 6 to 17	0, 1, 5, 10, 20	Gavage	Reduced body weight gain in mothers > 10 mg/kg-d (20 mg/kg-d was significant); significantly reduced fetal weights in > 10 mg/kg-d groups.	NOEL: 5	Case et al., 2001 ⁷

NEtFOSE	Short-Term	Sprague-Dawley Female Rats	21 days (5 days/week, no administration on Days 6 and 7)	5	Gavage	Decreased growth rate, increased relative liver weight, and increased activity of SOD in the liver and uterus	N/A	Xie et al., 2009 ⁸
FOSA	Single exposure	Rat	^a 29 Days	0, 5	Gavage in oil vehicle	No impacts on liver weight	NOAEL: N/A LOAEL: N/A	Seacat and Luebker 2000 ¹⁹

^a Only a single dose was administered and animals were evaluated for 29 days

Table C-6. Critical Study Selection

Compound	Reference	Appropriate duration?	Effects consistent with other studies?	Effects relevant to humans?	Number of Doses	Toxicity value identifiable?	Critical study?
NEtFOSE	Xie et al., 2009 ⁵	⊘	✓	✓	1	⊘	No
NEtFOSE	Case et al., 2001 ⁷ (females-Dose range finder)	✓	✓	✓	6	✓	Yes
NEtFOSE	Case et al., 2001 ⁷ (females-rabbits)	✓	✓	✓	4	✓	Yes
NEtFOSE	Case et al., 2001 ⁷ (females-Dose range finder rats)	✓	✓	✓	6	✓	Yes
NEtFOSE	Case et al., 2001 ⁷ (females-rats)	✓	✓	✓	4	✓	Yes
FOSA	Seacat and Leubker 2000 ¹⁹	⊘	⊘	✓	1	⊘	No

To be considered a critical study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.

Table C-7. Epidemiological Studies from Literature Review

Compound	Study Type	Population	Time period	Exposure	Outcomes	Results	Other PFAS Studied	Reference
FOSA	Cohort	210 community members (median age 38)	1990-2008	Drinking water exposure; serum PFAS concentrations	Thyroid disruption, kidney function, and BMI	FOSA had no associations with thyroid disruption, estimated glomerular filtration rate, or BMI.	PFOS, PFOA, PFNA, PFHxS, PFOSA, Me-PFDA, Et-PFOA	Blake et al., 2018 ³⁰
FOSA	Cohort	501 mothers from Michigan and Texas	2005-2009	Maternal PFAS serum concentrations	Pregnancy loss	FOSA was not associated with an increase in pregnancy loss.	PFNA, PFOS, PFOA, PFDA, Et-PFOSE-AcOH, Me-PFOA-AcOH	Buck Louis et al., 2016 ⁵⁷
FOSA	Cohort	501 fathers from Michigan and Texas	2005-2009	Paternal PFAS serum concentrations	Semen parameters	An increase in FOSA (1-unit ln transformed ng/mL) was associated with a decrease in sperm head area (μm^2); $\beta = -2.295$ (95% CI: -4.052, -0.538) and perimeter (μm); $\beta = -1.252$ (95% CI: -2.276, -0.228). An increase in FOSA (1-unit ln transformed ng/mL) was associated with a decrease in the percentage of DNA stability within the sperm (%); $\beta = -15.153$ (95% CI: -26.559, -3.747) and an increase in the percentage of immature sperm (%); $\beta = -90.881$ (95% CI: 51.226, 130.496).	PFNA, PFOS, PFOA, PFDA, Me-PFOA-AcOH	Buck Louis et al., 2015 ⁵⁸
FOSA	Case-Cohort	483 women (250 women with breast cancer) recruited from the Danish National Birth Cohort	1996-2002	PFAS serum concentrations	Breast cancer	An increase in FOSA concentrations (ng/ml) was associated with an increase in relative risk for breast cancer RR = 1.89 (95%CI 1.01,3.54) and an increase in relative risk from breast cancer under 40 years old RR = 2.45 (95% CI 1.00,6.00).	PFOS, PFOA, PFNA, PFHxs	Bonefeld-Jorgensen et al., 2014 ¹⁷
FOSA	Cohort	665 mother-offspring pairs from the Danish Birth Cohort	1988 - 2009	Maternal PFAS serum concentrations	Anthropometry outcomes (Measurements and proportions)	Maternal FOSA concentrations were not associated with any offspring anthropometry outcomes tested 20 years later.	PFOA, PFOS, PFNA	Halldorsson et al., 2012 ⁵⁹

					of the human body)			
FOSA	Cohort	641 infants in the Environment and Childhood Asthma (ECA) prospective birth cohort study	1992-2003	Cord PFAS blood concentrations	Health outcomes Reduced lung function at birth, asthma, allergic rhinitis, AD nor allergic sensitization	An increase in cord blood FOSA concentrations (log2 transformed) was associated with an increase in the number of lower respiratory tract infections between 0 and 10 years old $\beta = 0.10$ (95% CI: 0.06, 0.14). Cord blood FOSA concentrations were not associated with offspring lung function at birth, asthma, allergic rhinitis, atopic dermatitis, nor allergic sensitization	PFOS, PFOA, PFHxS, PFNA, PFUnA	Impinen et al., 2018 ³²
FOSA	Cross sectional study	378 ten-year-old's form Oslo, Norway.	1992-2009	Serum PFAS concentrations	Health outcomes	FOSA concentrations were not associated with atopic dermatitis, lower respiratory tract infections, common colds, or allergy outcomes. FOSA concentrations were inversely associated with rhinitis in boys RR = 0.49 [95%CI: 0.26;0.94.	PFBA, PFPeA, PFHxA, PFDoDA, PFTrDA, PFDS, NMeFOSA, NEtFOSA, PFOA, PFNA, PFUnA, PFHxS, PFHpS	Kvalem et al., 2020 ²⁹
FOSA	Cohort	501 couples from Michigan and Texas	2005-2009	Serum PFAS concentrations	Menstrual cycle and fecundity	FOSA concentrations were not associated to any menstrual cycle or fecundity outcomes.	PFNA, PFOA, PFOS, FOSA, Et-PFOSA-AcOH, Me-PFOSA-AcOH	Lum et al., 2017 ²⁸
FOSA	Cohort	351 mother-child pairs from Laizhou, China	2010-2013	Cord serum PFAS concentrations	Reproductive hormones, steroidogenic enzymes,	FOSA concentrations were not associated with any reproductive hormones or steroidogenic concentrations.	PFOS, PFOA, PFBS, PFDoA, PFHpA, PFHxS, PFNA, PFDA, PFUA	Yao et al., 2019 ³³
<p>*This literature review is not exhaustive as the primary purpose of the search was to identify epidemiological studies that support toxicological findings.</p> <p>Epidemiologic terms: OR=odds ratio; AOR=adjusted odds ratio; RR=relative risk; 95% CI=95% confidence interval; r=Spearman correlation coefficient; β=regression coefficient</p> <p>PFAS acronyms: PFOA=perfluorooctanoic acid, PFNA=perfluorononanoic acid, PFDA=perfluorodecanoic acid, PFUnA= perfluoroundecanoic acid, PFTrDA=perfluorotridecanoic acid, PFTeDA=perfluorotetradecanoic acid, PFHxDA=perfluorohexadecanoic acid, PFHxS=perfluorohexane sulfonate, PFOS=perfluorooctane sulfonic acid, PFDS=perfluorodecanesulfonate, PFBS=perfluorobutane sulfonate, PFBA=perfluorobutanoic acid, PFPeA=perfluoropentanoic acid, PFHxA=perfluorohexanoic acid, PFHpA=perfluoroheptanoic acid, 6:2 FTSA=6:2 fluorotelomer sulfonates, NMeFOSAA=N-methyl perfluorooctanesulfonamidoacetate, EPAH=2-(N-ethyl-perfluorooctane sulfonamido)acetate, MPAH=2-(N-methyl-perfluorooctane sulfonamido) acetate, PFSA=perfluorooctane sulfonamide; PFHpS=perfluoroheptane sulfonic acid; PFDS= perfluorodecane sulfonic acid; EtPFOSAA= 2-(Nethyl-perfluorooctane sulfonamido) acetic acid</p>								

PFD_oA | 2020

Substance Overview

Perfluorododecanoic acid (PFD_oA) is a chemical in a group of contaminants called per- and polyfluoroalkyl substances (PFAS). Because of its unique chemical properties, PFD_oA is used in textile manufacturing and commercial and industrial products such as paper wrapping, food packaging, and fire-fighting foams.¹ PFAS can persist in the environment and in the human body for long periods of time.² PFAS with carbon chains consisting of seven or more carbon atoms, like PFD_oA, cannot be easily broken down and excreted from the body; these compounds have been shown to build up more and persist longer in the body than shorter chain PFAS.³⁻⁵

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard for PFD_oA. DHS recommends an enforcement standard of 500 nanograms per liter (ng/L) for PFD_oA. The recommended standard is based on studies which found that PFD_oA reduced body weight and levels of testosterone and progesterone in male rats.^{6,7}

DHS recommends that the preventive action limit for PFD_oA be set at 20% of the enforcement standard because PFD_oA has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell cultures.

Current Standards

Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

Recommended Standards

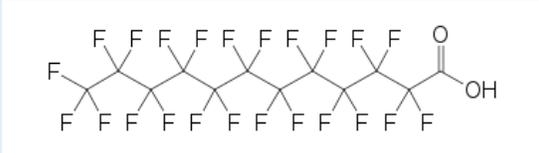
Enforcement Standard:	500 ng/L
Preventive Action Limit:	100 ng/L

Health Effects

Studies among people exposed to high levels of PFAS have shown that PFD_oA is associated with altered thyroid hormone levels in mothers and their offspring, physical growth and psychological development in infants and children, development and severity of asthma and allergies in children, risk of cardiovascular disease and immune response in adults, risk of gestational diabetes and polycystic ovarian syndrome related infertility in women, and may contribute to insulin resistance and prevalence of diabetes in adults.⁸⁻²² Studies in research animals have found that high levels of PFD_oA can lower body weight, affect several blood and metabolic parameters, alter reproductive steroid levels and fertility, and can cause liver damage.^{6,7,23-30}

PFDaA has not been shown to cause carcinogenic (cancer), mutagenic (DNA damage), teratogenic (birth defects), or interactive effects in people, research animals, or cell cultures.² The EPA has not evaluated the carcinogenicity of PFDaA.

Chemical Profile

PFDaA	
Structure:	
CAS Number:	307-55-1
Formula:	C ₁₂ HF ₂₃ O ₂
Molar Mass:	514.086 g/mol
Synonyms:	Tricosafuorododecanoic acid Dodecanoic acid, 2,2,3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,11,11,12,12 ,12-tricosafuoro- 2,2,3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,11,11,12,12 ,12-Tricosafuorododecanoic acid Perfluorolauric acid Dodecanoic acid, tricosafuoro- tricosafuorododecanoic acid

Exposure Routes

PFAS, including PFDaA, can be released directly into the environment during the manufacture and use of PFAS and can be found in water or soil. PFAS can move between groundwater and surface water. Once in water, PFAS can travel long distances.^{2,31}

People can be exposed to PFAS by drinking water, eating food, and breathing in or accidentally swallowing soil or dust containing PFAS.²

Current Standard

Wisconsin does not currently have groundwater standards for PFDaA.³²

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

EPA Oral Reference Dose:	N/A
--------------------------	-----

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

ATSDR Oral Minimum Risk Level:	N/A
--------------------------------	-----

Literature Search

Literature Search Dates:	1900–2019
Key studies found?	Yes
Critical studies identified?	Yes

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for PFDa.³³

Health Advisory

The EPA has not established health advisories for PFDa.³⁴

Drinking Water Concentration (Cancer Risk)

The EPA has not established a drinking water concentration based on a cancer risk level determination for PFDa.³⁵

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for PFDoA.³⁶

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides or for use by the Integrated Risk Information System (IRIS) program.

EPA Oral Reference Dose

The EPA does not have an oral reference dose for PFDoA.³⁵

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If we determine that something is carcinogenic and there is no federal number or ADI from the EPA, DHS must set the standard at a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of PFDoA, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of PFDoA. If so, we look to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA and International Agency for Research on Cancer (IARC) have not evaluated the carcinogenicity of PFDoA.

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for PFDoA.³⁵

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information that was not considered when the value was established and that indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For PFDoA, we searched for values that had been published on or before November 2019. While the Agency for Toxic Substances and Disease Registry (ATSDR) reviewed the toxicity of PFDoA in 2018, they did not establish any guidance values for PFDoA.^{a 2}

Literature Search

Our literature review focused on relevant scientific literature on the health effects of PFDoA published on or before November 2019. We looked for studies related to PFDoA toxicity or effects on a disease state in which information on exposure or dose was included as part of the study.^b Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

Approximately 45 toxicity studies were returned by the search engines. We excluded studies on non-mammalian or cell systems, non-oral exposure routes, and those that did not evaluate health risks from further review. After applying these exclusion criteria, we located 12 key toxicity studies on PFDoA (summarized in Table A-1). To be considered a critical toxicity study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable

a ATSDR stated that they did not identify any chronic-duration oral studies for PFDoA in their literature review. While they located one intermediate-duration study, they did not establish a minimum risk level (MRL) for PFDoA.²² Given the limited number of endpoints examined in this study, including the lack of histopathological examination, ATSDR did not consider the database adequate for identifying the critical target of toxicity and thus for derivation of an MRL.

b We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

Title/abstract: "Perfluorododecanoic acid" or "PFDoA"

Subject area: toxicology

Language: English

We also searched online for toxicity studies published by national research programs.

toxicity value.^{c,d} Five of these studies met the criteria to be considered a critical toxicity study (see Table A-2).

In our search, we also located several epidemiology studies in our search (See Table A-3 for a summary). While multiple potential exposure sources and the ability for other PFAS compounds to cause similar health effects preclude using these data to establish a health-based value, such studies are helpful in identifying the critical effects and ensuring that the animal data used to establish the standard are relevant to people.

Critical Toxicity Studies

To compare results from studies identified in the literature review, we calculated an acceptable daily intake (ADI) for each study/effect. The ADI is the estimated amount of PFDoA that a person can be exposed to every day and not experience health impacts. Since we could not find studies with information about the half-life of PFDoA in people or research animals, As such, we calculated ADI by dividing a toxicity value from either a no-observed adverse effect level (NOAEL), lowest observed adverse effect level (LOAEL), or benchmark dose (BMD) identified in a study by a factor accounting for various sources of scientific uncertainty.^e Uncertainty factors were included, as appropriate, to account for differences between humans and animals, differences between healthy and sensitive human populations, using data from short-term experiments to protect against effects from long-term exposure, and using data where a health effect was observed to estimate the level that does not cause an effect. To ensure appropriate protection, we have chosen to not use studies that have significant uncertainty as the basis for the recommended enforcement standards.^f This approach is consistent with that taken by EPA when establishing oral reference doses.³⁸

Kato et al., 2014

Kato et al. exposed male and female rats to different concentrations of PFDoA (0, 0.1, 0.5, and 2.5 milligrams of PFDoA per kilogram body weight per day or mg/kg-d) through oral gavage for 42 days (males) or for 14 days prior to and through gestation day 25 or postnatal day 5 (females).²⁵ A subset of males and females in the 0 and 2.5 mg/kg-d dose groups were withheld treatment for an additional 14

c Appropriate toxicity values include the no observable adverse effect level (NOAEL), lowest observable adverse effect level (LOAEL), and benchmark dose (BMD).

d Due to the limited availability of data for this substance, we considered a study to be of an appropriate duration if animals were exposed for at least 28 days or if the exposure occurred during pregnancy. We accounted for differences in exposure duration through the use of an uncertainty factor.

e The NOAEL is the highest dose tested that did not cause an adverse effect, the LOAEL is the lowest dose tested that caused an adverse effect, and the BMD is an estimation of the dose that would cause a specific level of response (typically 5 or 10%).³⁴

f DHS considers a study to have significant uncertainty if the total uncertainty factors is greater than 3,000.

days (see Kato et al. for more information on recovery group effects). In the study, the authors found that PFDoA lowered body weight, increased relative liver and kidney weights, affected several blood, metabolic, and endocrine parameters, and produced abnormalities in the liver, pancreas, and adrenal gland (Table 1).

Table 1. Statistically Significant Effects Observed in Male Rats in Kato et al., 2014

Effects observed		Dose (mg/kg-d)		
		0.1	0.5	2.5
Growth	Lower body weight			✓
Blood	Higher mean cell hemoglobin			✓
	Higher reticulocyte level			✓
Metabolism	Lower plasma glucose			✓
	Lower serum calcium			✓
Liver	Higher relative liver weight		✓	✓
	Higher serum alkaline phosphatase (ALP)		✓	✓
	Lower serum total protein			✓
	Elevated serum total bilirubin			✓
	Lower serum total albumin			✓
	Higher serum albumin fraction			✓
	Higher serum albumin to globulin ratio			✓
	Lower serum globulin α_1 fraction			✓
	Lower serum globulin α_2 fraction		✓	✓
	Higher serum globulin γ fraction			✓
	Lower serum total cholesterol	✓	✓	†
	Spleen	Lower absolute spleen weight		
Heart	Lower absolute heart weight			✓
Brain	Higher relative brain weight			✓
Pituitary gland	Lower absolute pituitary gland weight			✓
Thymus	Lower absolute and relative thymus weight			✓
Thyroid	Lower absolute thyroid weight			✓
Adrenal gland	Lower absolute adrenal gland weight			✓
	Deterioration of adrenal gland cortex			✓
Reproduction	Lower absolute epididymis weight			✓
Kidney	Lower creatinine			✓
	Higher serum blood urea nitrogen (BUN)			✓
	Lower absolute and relative kidney weight			✓
Pancreas	Decrease in specialized pancreatic cell organelles			✓

†The authors believe that at the highest dose, impaired cholesterol excretion due to a decrease in bile flow could have accounted for a lack of statistical association with lower total cholesterol.

From this repeated dose experiment in males, the authors identified a No Observable Adverse Effect Level (NOAEL) of 0.1 mg/kg-d based on hypertrophy, necrosis, and inflammatory cholestasis in the liver. We estimated a candidate ADI of 10 nanograms per kilogram-day (ng/kg-d) PFD_oA based on this NOAEL and an uncertainty factor of 10,000 to account for differences between humans and research animals (10), differences among people (10), using a short-term study to extrapolate to long-term exposures (10), and the limited availability of information (10). While we obtained a candidate ADI for males from this study, this study was not used to establish to recommended enforcement standard due to significant uncertainty.

In females, the authors found that PFD_oA also lowered body weight and absolute weights of the spleen, heart, pituitary gland, and thymus, increased blood platelet concentrations, and affected several liver parameters (Table 2).

Table 2. Statistically Significant Effects Observed in Female Rats in Kato et al., 2014

Effects observed		Dose (mg/kg-d)		
		0.1	0.5	2.5
Growth	Lower body weight			✓
Blood	Higher platelet concentration	✓		N/A
Spleen	Lower absolute spleen weight		✓	N/A
Heart	Lower absolute heart weight		✓	N/A
Pituitary gland	Lower absolute pituitary gland weight		✓	N/A
Thymus	Lower absolute thymus weight		✓	N/A
	Deterioration of thymus cortex			✓
Pancreas	Swelling within the pancreas			✓
Liver	Higher serum total albumin	✓		N/A
	Higher serum albumin to globulin ratio		✓	N/A
	Higher serum albumin fraction	✓	✓	N/A
	Lower serum globulin α_2 fraction	✓		N/A
	Liver cell death			✓
	Liver cell enlargement			✓
	Higher relative liver weight		✓	N/A
Uterus	Uterine bleeding at the implantation site			✓
Reproduction	Lower proportion of pregnancies yielding live litters			✓

N/A: Authors could not statistically analyze effects of a 2.5 mg/kg-d dose among main group females as data from only one animal were available. In this group, other females did not deliver pups normally or survive to the end of the study.

From this reproductive and developmental experiment in females, we identified a NOAEL of 0.1 mg/kg-d based upon effects on lower absolute weights of the spleen, heart, pituitary gland, and thymus seen at

0.5 mg/kg-d in females. It should be noted that authors were not able to evaluate these effects at the highest dose due to the low number of surviving animals. We selected this value instead of the higher NOAEL identified by the authors because lower organ weights are consistent with lower overall body weight findings in other PFDoA toxicity studies in research animals.^{6,6,23-27,39} Additionally, effects on the pituitary gland are consistent with findings from other toxicity studies in research animals which show that PFDoA can affect levels of luteinizing hormone, follicle-stimulating hormone, testosterone, progesterone, and estradiol.^{6,7,23,26,27} Finally, several epidemiologic studies among people support a link between PFDoA and thyroid effects, demonstrating an association between PFDoA and altered thyroid hormone levels.^{8,14,22,40}

We estimated a candidate ADI of 100 ng/kg-d for females in this study based on this NOAEL and a total uncertainty factor of 1000 to account for differences between people and research animals (10), differences among people (10), and the limited availability of information (10).

Ding et al., 2014

Ding et al. exposed male rats to different concentrations of PFDoA (0, 0.02, 0.05, 0.2, and 0.5 mg/kg-d of PFDoA) through oral gavage for 110 days.²⁴ The authors found that PFDoA decreased body weight at the highest dose and increased glucose and absolute and relative liver weights at all doses (Table 3). In addition to these statistically significant findings, the study observed liver tissue abnormalities in rats treated at 0.05, 0.2, and 0.5 mg/kg-d PFDoA, as well as a decrease in liver triglycerides.

Table 3. Statistically Significant Effects Observed in Ding et al., 2014

Effects observed		Dose (mg/kg-d)			
		0.02	0.05	0.2	0.5
Growth	Lower body weight				✓
Metabolism	Higher serum glucose	✓	✓	✓	✓
Liver	Higher absolute and relative liver weight	✓	✓	✓	✓
	Lower serum triglycerides		✓	✓	✓
	Lower serum low density lipid-cholesterol	✓	✓		
	Higher serum creatinine			✓	✓
	Higher serum total bilirubin				✓
	Higher serum total bile acids			✓	✓
	Higher serum alkaline phosphatase (ALP)				✓
	Higher serum albumin	✓	✓	✓	✓
	Higher serum creatinine kinase	✓	✓		✓
	Kidney	Higher serum urea nitrogen			✓

⁶ Kato et al. identified a reproductive/developmental NOAEL of 0.5 mg/kg-d due to continuous estrous in females at the 2.5 mg/kg-d dose.

From this study, we identified a Lowest Observable Adverse Effect Level (LOAEL) of 0.02 mg/kg-d based on effects on glucose levels and the liver seen in rats at all treatment doses at the end of the exposure period. We estimated a candidate ADI of 2 ng/kg-d PFDoA from this study based on the LOAEL and a total uncertainty factor of 10,000 to account for differences between people and research animals (10), differences among people (10), the use of a LOAEL instead of a NOAEL (10), and the limited availability of information (10). While we obtained a candidate ADI for PFDoA from this study, this study was not used to establish to recommended enforcement standard due to significant uncertainty.

Shi et al., 2009a

Shi et al. exposed pubertal female rats to different concentrations of PFDoA (0, 0.5, 1.5, and 3 mg/kg-d of PFDoA) through oral gavage for 28 days.²⁶ The authors found that PFDoA lowered body weight and levels of estradiol, and raised total cholesterol at the highest dose (Table 4). In addition to these findings, the study observed statistically significant changes in the expression of key genes responsible for estrogen synthesis at various treatment doses.

Table 4. Statistically Significant Effects Observed in Shi et al., 2009a

Effects observed		Dose (mg/kg-d)		
		0.5	1.5	3
Growth	Lower body weight			✓
Liver	Higher total cholesterol			✓
Endocrine	Lower estradiol			✓

From this study, we identified a NOAEL of 1.5 mg/kg-d based on effects on body weight and levels of estradiol and total cholesterol at the highest dose. We estimated a candidate ADI of 150 ng/kg-d PFDoA from this study based on the NOAEL and a total uncertainty factor of 10,000 to account for differences between people and research animals (10), differences among people (10), use of a shorter duration study to protect against effects from long-term exposure (10), and the limited availability of information (10). While we obtained a candidate ADI for PFDoA from this study, this study was not used to establish to recommended enforcement standard due to significant uncertainty.

Shi et al., 2009b

Shi et al. exposed male rats to different concentrations of PFDoA (0, 0.02, 0.05, and 0.2, and 0.5 mg/kg-d of PFDoA) through oral gavage for 110 days.⁶ The authors found that PFDoA decreased body weight at the highest dose and testosterone levels at 0.2 and 0.5 mg/kg-d (Table 5). In addition to these findings, the study observed changes in the structure of the testes at the highest dose and statistically significant changes in the expression of key genes responsible for male reproductive steroid synthesis at various doses.

Table 5. Statistically Significant Effects Observed in Shi et al., 2009b

Effects observed		Dose (mg/kg-d)			
		0.02	0.05	0.2	0.5
Growth	Lower body weight				✓
Endocrine	Lower testosterone			✓	✓

From this study, we identified a NOAEL of 0.05 mg/kg-d based on effects on testosterone levels at higher doses. We estimated a candidate ADI of 50 ng/kg-d PFDa from this study based on the NOAEL and a total uncertainty factor of 1000 to account for differences between people and research animals (10), differences among people (10), and the limited availability of information (10).

Shi et al., 2010

Shi et al. exposed male rats to different concentrations of PFDa (0, 0.02, 0.2, and 0.5 mg/kg-d of PFDa) through oral gavage for 110 days.⁷ The authors found that PFDa decreased progesterone levels at 0.2 and 0.5 mg/kg-d (Table 6). Additionally, the study observed statistically significant changes in the expression of key genes responsible for male reproductive steroid synthesis at various doses.

Table 6. Statistically Significant Effects Observed in Shi et al., 2010

Effects observed		Dose (mg/kg-d)			
		0.02	0.05	0.2	0.5
Endocrine	Lower progesterone			✓	✓

From this study, we identified a NOAEL of 0.05 mg/kg-d based on effects on progesterone levels at higher doses. We estimated a candidate ADI of 50 ng/kg-d PFDa from this study based on the NOAEL and a total uncertainty factor of 1000 to account for differences between people and research animals (10), differences among people (10), and the limited availability of information (10).

Key Health Effects

In our literature search, we did not find any studies to indicate that PFDa caused carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell cultures.

Discussion

While information about the health effects of PFDa exposure is limited, studies in research animals indicate that PFDa can affect health. These studies have found that high levels of PFDa can lower body weight, affect several blood and metabolic parameters, alter reproductive steroid levels and fertility, and can cause liver damage.^{6,7,23-30} Studies among people exposed to high levels of PFAS have shown that PFDa may affect thyroid hormone levels in mothers and their offspring, physical growth and psychological development in infants and children, development and severity of asthma and allergies in children, cardiovascular disease and immune response in adults, risk of gestational diabetes

and polycystic ovarian syndrome related infertility in women, and may contribute to insulin resistance and prevalence of diabetes in adults.⁸⁻²²

Lower birth weight is a common finding among PFDoA toxicity studies in research animals and in other studies of long-chain PFAS.^{2,6,23-27,39} In limited studies among human infants, the relationship between PFDoA and birthweight is mixed; some studies found an association with lower birthweight while others found no association with birthweight.^{9,20,41,42}

Other common findings among PFDoA toxicity studies in research animals are effects on the pituitary gland or the hormones it creates or controls, including luteinizing hormone, follicle-stimulating hormone, testosterone, progesterone, and estradiol.^{6,7,23,26,27} Several epidemiologic studies among people support a link between PFDoA and thyroid effects, demonstrating an association between PFDoA and altered thyroid hormone levels.^{8,14,22,40}

A number of studies have demonstrated that liver effects caused by PFAS occur primarily through activation of the nuclear hormone receptor, PPAR α (peroxisome proliferator-activated receptor alpha).⁴³⁻⁴⁷ Nuclear receptors regulate gene expression and PPAR α regulates the expression of genes involved in lipid and cholesterol metabolism.⁴⁸ While PPAR α receptors are found in rodents and humans, levels of these receptors are much higher in rats and mice than in monkeys and humans.⁴⁸ This means that effects caused by activation of PPAR α are more likely to occur in rodents than they are to occur in people. As such, the effects on the liver observed in the critical studies reviewed here are likely not relevant to humans.

Standard Selection

DHS recommends an enforcement standard of 500 ng/L for PFDoA.

There are no federal numbers and no state drinking water standard for PFDoA. Additionally, the EPA has not evaluated the carcinogenicity or established an ADI (oral reference dose) for PFDoA.

However, we found several studies evaluating the toxicity of PFDoA in research animals. To calculate the ADI as specified in s. 160.13, Wisc. Statute, DHS selected a 2009 study by Shi et al. as the critical study.⁶ We selected this study because it is a long-term study which found that PFDoA can affect reproductive function by lowering testosterone levels in rats, a finding consistent with other studies in people and research animals that have shown associations between PFDoA and altered hormone levels, including thyroid and sex hormones.

As described above, we obtained an ADI of 50 ng/kg-d from a NOAEL of 0.05 mg/kg-d and a total uncertainty factor of 1000. To determine the recommended enforcement standard, DHS used the ADI,

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information

and, as required by Ch. 160, Wis. Stats., a body weight of 10 kg, a water consumption rate of 1 L/d, and a relative source contribution of 100%.

DHS recommends a preventive action limit of 100 ng/L for PFDoA.

DHS recommends that the preventive action limit for PFDoA be set at 20% of the enforcement standard because PFDoA has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell cultures.²

Prepared by Amanda Koch, MPH and Sarah Yang, Ph.D.

Wisconsin Department of Health Services

References

1. Kissa E. *Fluorinated Surfactants and Repellants*. New York, NY: Marcel Dekker; 2001.
2. ATSDR. Toxicological Profile for Perfluoroalkyls - Draft for Public Comment. In: Registry AftSaD, ed. Atlanta, GA2017.
3. Kudo N, Suzuki E, Katakura M, Ohmori K, Noshiro R, Kawashima Y. Comparison of the elimination between perfluorinated fatty acids with different carbon chain length in rats. *Chem Biol Interact*. 2001;134(2):203-216.
4. Kudo N, Suzuki-Nakajima E, Mitsumoto A, Kawashima Y. Responses of the liver to perfluorinated fatty acids with different carbon chain length in male and female mice:in relation to induction of hepatomegaly, peroxisomal beta-oxidation and microsomal 1-acylglycerophosphocholine acyltransferase. *Biol Pharm Bull*. 2006;29(9):1952-1957.
5. Ohmori K, Kudo N, Katayama K, Kawashima Y. Comparison of the toxicokinetics between perfluorocarboxylic acids with different carbon chain length. *Toxicology*. 2003;184(2-3):135-140.
6. Shi Z, Ding L, Zhang H, Feng Y, Xu M, Dai J. Chronic exposure to perfluorododecanoic acid disrupts testicular steroidogenesis and the expression of related genes in male rats. *Toxicol Lett*. 2009;188(3):192-200.
7. Shi Z, Zhang H, Ding L, Feng Y, Wang J, Dai J. Proteomic analysis for testis of rats chronically exposed to perfluorododecanoic acid. *Toxicol Lett*. 2010;192(2):179-188.
8. Aimuzi R, Luo K, Chen Q, et al. Perfluoroalkyl and polyfluoroalkyl substances and fetal thyroid hormone levels in umbilical cord blood among newborns by prelabor caesarean delivery. *Environ Int*. 2019;130:104929.

9. Cao W, Liu X, Liu X, et al. Perfluoroalkyl substances in umbilical cord serum and gestational and postnatal growth in a Chinese birth cohort. *Environ Int.* 2018;116:197-205.
10. Chen Q, Huang R, Hua L, et al. Prenatal exposure to perfluoroalkyl and polyfluoroalkyl substances and childhood atopic dermatitis: a prospective birth cohort study. *Environ Health.* 2018;17(1):8.
11. Dong GH, Tung KY, Tsai CH, et al. Serum polyfluoroalkyl concentrations, asthma outcomes, and immunological markers in a case-control study of Taiwanese children. *Environ Health Perspect.* 2013;121(4):507-513.
12. Goudarzi H, Miyashita C, Okada E, et al. Effects of prenatal exposure to perfluoroalkyl acids on prevalence of allergic diseases among 4-year-old children. *Environ Int.* 2016;94:124-132.
13. Huang M, Jiao J, Zhuang P, Chen X, Wang J, Zhang Y. Serum polyfluoroalkyl chemicals are associated with risk of cardiovascular diseases in national US population. *Environ Int.* 2018;119:37-46.
14. Itoh S, Araki A, Miyashita C, et al. Association between perfluoroalkyl substance exposure and thyroid hormone/thyroid antibody levels in maternal and cord blood: The Hokkaido Study. *Environ Int.* 2019;133(Pt A):105139.
15. Kim JH, Park HY, Jeon JD, et al. The modifying effect of vitamin C on the association between perfluorinated compounds and insulin resistance in the Korean elderly: a double-blind, randomized, placebo-controlled crossover trial. *Eur J Nutr.* 2016;55(3):1011-1020.
16. Niu J, Liang H, Tian Y, et al. Prenatal plasma concentrations of Perfluoroalkyl and polyfluoroalkyl substances and neuropsychological development in children at four years of age. *Environ Health.* 2019;18(1):53.
17. Rahman ML, Zhang C, Smarr MM, et al. Persistent organic pollutants and gestational diabetes: A multi-center prospective cohort study of healthy US women. *Environ Int.* 2019;124:249-258.
18. Seo SH, Son MH, Choi SD, Lee DH, Chang YS. Influence of exposure to perfluoroalkyl substances (PFASs) on the Korean general population: 10-year trend and health effects. *Environ Int.* 2018;113:149-161.
19. Wang W, Zhou W, Wu S, et al. Perfluoroalkyl substances exposure and risk of polycystic ovarian syndrome related infertility in Chinese women. *Environ Pollut.* 2019;247:824-831.
20. Wang Y, Adgent M, Su PH, et al. Prenatal Exposure to Perfluorocarboxylic Acids (PFCAs) and Fetal and Postnatal Growth in the Taiwan Maternal and Infant Cohort Study. *Environ Health Perspect.* 2016;124(11):1794-1800.
21. Wang Y, Rogan WJ, Chen PC, et al. Association between maternal serum perfluoroalkyl substances during pregnancy and maternal and cord thyroid hormones: Taiwan maternal and infant cohort study. *Environ Health Perspect.* 2014;122(5):529-534.

22. Yang L, Li J, Lai J, et al. Placental Transfer of Perfluoroalkyl Substances and Associations with Thyroid Hormones: Beijing Prenatal Exposure Study. *Sci Rep.* 2016;6:21699.
23. Chen Y, Li H, Mo J, et al. Perfluorododecanoic Acid Blocks Rat Leydig Cell Development during Prepuberty. *Chem Res Toxicol.* 2019;32(1):146-155.
24. Ding L, Hao F, Shi Z, et al. Systems biological responses to chronic perfluorododecanoic acid exposure by integrated metabolomic and transcriptomic studies. *J Proteome Res.* 2009;8(6):2882-2891.
25. Kato H, Fujii S, Takahashi M, et al. Repeated dose and reproductive/developmental toxicity of perfluorododecanoic acid in rats. *Environ Toxicol.* 2015;30(11):1244-1263.
26. Shi Z, Zhang H, Ding L, Feng Y, Xu M, Dai J. The effect of perfluorododecanoic acid on endocrine status, sex hormones and expression of steroidogenic genes in pubertal female rats. *Reprod Toxicol.* 2009;27(3-4):352-359.
27. Shi Z, Zhang H, Liu Y, Xu M, Dai J. Alterations in gene expression and testosterone synthesis in the testes of male rats exposed to perfluorododecanoic acid. *Toxicol Sci.* 2007;98(1):206-215.
28. Zhang H, Ding L, Fang X, et al. Biological responses to perfluorododecanoic acid exposure in rat kidneys as determined by integrated proteomic and metabolomic studies. *PLoS One.* 2011;6(6):e20862.
29. Zhang H, Shi Z, Liu Y, Wei Y, Dai J. Lipid homeostasis and oxidative stress in the liver of male rats exposed to perfluorododecanoic acid. *Toxicol Appl Pharmacol.* 2008;227(1):16-25.
30. Kielsen K, Shamim Z, Ryder LP, et al. Antibody response to booster vaccination with tetanus and diphtheria in adults exposed to perfluorinated alkylates. *J Immunotoxicol.* 2016;13(2):270-273.
31. ITRC. Environmental Fate and Transport for Per- and Polyfluoroalkyl Substances. In: Council ITR, ed2018.
32. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 140*2017.
33. USEPA. National Primary Drinking Water Regulations. <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>. Published 2018. Accessed.
34. USEPA. Drinking Water Contaminant Human Health Effects Information. <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>. Published 2019. Accessed.
35. USEPA. IRIS Assessments. https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm. Published 2019. Accessed.
36. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 809*2018.
37. Ritter L, Totman C, Krishnan K, Carrier R, Vezina A, Morisset V. Deriving uncertainty factors for threshold chemical contaminants in drinking water. *Journal of toxicology and environmental health Part B, Critical reviews.* 2007;10(7):527-557.

38. USEPA. A Review of the Reference Dose and Reference Concentration Processes. 2002(EPA/630/P-02/002F).
39. Kawabata K, Matsuzaki H, Nukui S, et al. Perfluorododecanoic Acid Induces Cognitive Deficit in Adult Rats. *Toxicol Sci.* 2017;157(2):421-428.
40. Wang Y, Rogan WJ, Chen PC, et al. Association between Maternal Serum Perfluoroalkyl Substances during Pregnancy and Maternal and Cord Thyroid Hormones: Taiwan Maternal and Infant Cohort Study. *Environmental Health Perspectives.* 2014;122(5):529-534.
41. Lee ES, Han S, Oh JE. Association between perfluorinated compound concentrations in cord serum and birth weight using multiple regression models. *Reprod Toxicol.* 2016;59:53-59.
42. Lenters V, Portengen L, Rignell-Hydbom A, et al. Prenatal Phthalate, Perfluoroalkyl Acid, and Organochlorine Exposures and Term Birth Weight in Three Birth Cohorts: Multi-Pollutant Models Based on Elastic Net Regression. *Environ Health Perspect.* 2016;124(3):365-372.
43. Das KP, Wood CR, Lin MT, et al. Perfluoroalkyl acids-induced liver steatosis: Effects on genes controlling lipid homeostasis. *Toxicology.* 2017;378:37-52.
44. Rosen MB, Das KP, Rooney J, Abbott B, Lau C, Corton JC. PPAR alpha-independent transcriptional targets of perfluoroalkyl acids revealed by transcript profiling. *Toxicology.* 2017;387:95-107.
45. Albrecht PP, Torsell NE, Krishnan P, et al. A species difference in the peroxisome proliferator-activated receptor alpha-dependent response to the developmental effects of perfluorooctanoic acid. *Toxicological sciences : an official journal of the Society of Toxicology.* 2013;131(2):568-582.
46. Palkar PS, Anderson CR, Ferry CH, Gonzalez FJ, Peters JM. Effect of prenatal peroxisome proliferator-activated receptor alpha (PPARalpha) agonism on postnatal development. *Toxicology.* 2010;276(1):79-84.
47. Wolf DC, Moore T, Abbott BD, et al. Comparative hepatic effects of perfluorooctanoic acid and WY 14,643 in PPAR-alpha knockout and wild-type mice. *Toxicologic pathology.* 2008;36(4):632-639.
48. Hall AP, Elcombe CR, Foster JR, et al. Liver hypertrophy: a review of adaptive (adverse and non-adverse) changes--conclusions from the 3rd International ESTP Expert Workshop. *Toxicol Pathol.* 2012;40(7):971-994.
49. Liu H, Zhang H, Cui R, Guo X, Wang D, Dai J. Activation of peroxisome proliferator-activated receptor alpha ameliorates perfluorododecanoic acid-induced production of reactive oxygen species in rat liver. *Arch Toxicol.* 2016;90(6):1383-1397.
50. Shi ZM, Hou JJ, Guo XJ, Zhang HX, Yang FQ, Dai JY. Testicular phosphoproteome in perfluorododecanoic acid-exposed rats. *Toxicology Letters.* 2013;221(2):91-101.

51. Ji K, Kim S, Kho Y, et al. Serum concentrations of major perfluorinated compounds among the general population in Korea: dietary sources and potential impact on thyroid hormones. *Environ Int.* 2012;45:78-85.
52. Mattsson K, Rignell-Hydbom A, Holmberg S, et al. Levels of perfluoroalkyl substances and risk of coronary heart disease: Findings from a population-based longitudinal study. *Environ Res.* 2015;142:148-154.
53. Okada E, Sasaki S, Kashino I, et al. Prenatal exposure to perfluoroalkyl acids and allergic diseases in early childhood. *Environ Int.* 2014;65:127-134.
54. Qin XD, Qian Z, Vaughn MG, et al. Positive associations of serum perfluoroalkyl substances with uric acid and hyperuricemia in children from Taiwan. *Environ Pollut.* 2016;212:519-524.
55. Zeng XW, Qian Z, Emo B, et al. Association of polyfluoroalkyl chemical exposure with serum lipids in children. *Sci Total Environ.* 2015;512-513:364-370.

Appendix A. Toxicity Data

Table A-I. PFDoA Toxicity Studies from Literature Review

Study Type	Species	Duration	Doses (mg/kg-d)	Route	Key Findings	Toxicity Value (mg/kg-d)	Reference
Short-Term	Rat (male)	PND 21–35	0, 5, 10	Gavage	<p>Lower testosterone, serum luteinizing hormone, and follicle-stimulating hormone at 5 and 10 mg/kg-d</p> <p>Lower body weight and testis weight at 10 mg/kg-d</p> <p>Down-regulation of Leydig cell gene expression at 5 and/or 10 mg/kg-d</p>	NOAEL: N/A LOAEL: 5	Chen et al., 2019 ²³
Chronic	Rat (male)	110 days	0, 0.02, 0.05, 0.2, 0.5	Gavage	<p>Higher absolute liver weight and relative liver weight, higher glucose and albumin concentrations at all treatment doses</p> <p>Observations of liver tissue abnormalities and higher liver triglycerides, and lower blood triglycerides at 0.05, 0.2, and 0.5 mg/kg-d</p> <p>Higher creatinine, urea nitrogen, total bile acids, alkaline phosphatase at 0.2 and 0.5 mg/kg-d</p> <p>Lower body weight and higher total bilirubin at 0.5 mg/kg-d</p>	NOAEL: N/A LOAEL: 0.02	Ding et al., 2009 ²⁴
Subchronic	Rat (males)	42 days	0, 0.1, 0.5, 2.5	Gavage	<p>Lower body weight, higher relative brain weight, lower relative thymus and kidney weights, lower absolute</p>	NOAEL: 0.1 LOAEL: 0.5 (reported)	Kato et al., 2014 ²⁵

					thyroid and pituitary gland weights, and altered blood and biochemistry parameters at 2.5 mg/kg-d		
					Altered liver parameters at 0.5 and 2.5 mg/kg-d		
					Lower cholesterol at 0.1 and 0.5 mg/kg-d		
					Observed abnormalities in the liver, pancreas, and adrenal gland		
Reproduction/development	Rat (females)	14 days prior to mating through GD 1–25 or PND 5	0, 0.1, 0.5, 2.5	Gavage	Lower body weight, swelling within the pancreas, liver cell enlargement and death, uterine bleeding at the implantation site, and lower proportion of pregnancies yielding live litters at 2.5 mg/kg-d	NOAEL: 0.5 LOAEL: 2.5 (reported)	Kato et al., 2014 ²⁵
					Lower absolute weights of the spleen, heart, pituitary gland, and thymus at 0.5 mg/kg-d		
					Altered liver parameters at various doses		
Acute	Rat	Single dose	0, 50	Gavage	Lower body weight, decrease in weight gain, higher absolute liver weight and relative liver weight, higher relative brain weight, cognitive deficit	NOAEL: N/A LOAEL: 50	Kawabata et al., 2017 ³⁹
Chronic	Rat	110 days	0, 0.05, 0.2, 0.5	Gavage	Observed changes in protein levels in the liver at various doses of PFDoA	N/A	Liu et al., 2015 ⁴⁹
Short-Term	Rat (male)	14 days	0, 1, 5, 10	Gavage	Lower body weight, higher relative testis weight, lower testosterone, observations of apoptosis in Leydig, Sertoli, and spermatogenic cells, and	NOAEL: 1 LOAEL: 5	Shi et al., 2007 ²⁷

					declines in mRNA expression of genes involved in cholesterol transport and steroid biosynthesis at 5 and 10 mg/kg-d		
					Lower testis weight, higher total cholesterol, lower luteinizing hormone at 10 mg/kg-d		
Short-Term	Rat (female)	28 days	0, 0.5, 1.5, 3	Gavage	Decreased body weight, increased total cholesterol and decreased estradiol at 3 mg/kg-d	NOAEL: 1.5 LOAEL: 3	Shi et al., 2009a ²⁶
					Changes in expression of genes responsible for estrogen synthesis at various treatment doses		
Chronic	Rat (male)	110 days	0, 0.02, 0.05, 0.2, 0.5	Gavage	Lower testosterone at 0.2 and 0.5 mg/kg-d	NOAEL: 0.05 LOAEL: 0.2	Shi et al., 2009b ⁶
					Lower body weight at 0.5 mg/kg-d		
					Changes in expression of genes responsible for male reproductive steroid synthesis at various treatment doses		
Chronic	Rat (male)	110 days	0, 0.02, 0.2, 0.5	Gavage	Lower serum progesterone at 0.2 and 0.5 mg/kg-d	NOAEL: 0.02 LOAEL: 0.2	Shi et al., 2010 ⁷
					Changes in levels of proteins involved in male reproductive steroid synthesis at various treatment doses		
Chronic	Rat (male)	110 days	0, 0.02, 0.2, 0.5	Gavage	Increase in the number of casein kinase 2kinase-modified peptides and other protein expression levels in the testes at various doses of PFDoA	N/A	Shi et al., 2013 ⁵⁰

Short-Term	Rat (male)	14 days	0, 1, 5, 10	Gavage	Decrease in absolute liver weight, increase in relative liver weight, observed liver cell damage at 5 and 10 mg/kg-d Increase in blood and liver triglycerides, increase in liver cholesterol at 10 mg/kg-d	NOAEL: 1 LOAEL: 5	Zhang et al., 2008 ²⁹
Chronic	Rat	110 days	0, 0.05, 0.2, 0.5	Gavage	Increase in pyruvate, lactate, acetate, choline, and a variety of amino acids in the highest dose group	NOAEL: N/A LOAEL: 0.05	Zhang et al., 2011 ²⁸

GD=gestation day, PND=postnatal day

Table A-2. Critical Study Selection for PFD_oA

Reference	Appropriate duration?	Effects consistent with other studies?	Effects relevant to humans?	Number of Doses	Toxicity value identifiable?	Critical study?
Chen et al., 2019 ²³	⊗	✓	✓	2	✓	No
Ding et al., 2009 ²⁴	✓	✓	✓	4	✓	Yes
Kato et al., 2015 ²⁵	✓	✓	✓	3	✓	Yes
Liu et al., 2015	✓	✓	✓	3	⊗	No
Shi et al., 2007 ²⁷	⊗	✓	✓	3	✓	No
Shi et al., 2009a ²⁶	✓	✓	✓	3	✓	Yes
Shi et al., 2009b ²⁷	✓	✓	✓	4	✓	Yes
Shi et al., 2010 ⁷	✓	✓	✓	3	✓	Yes
Zhang et al., 2008 ²⁹	⊗	✓	⊗	3	✓	No
Zhang et al., 2011 ²⁸	✓	✓	⊗	3	✓	No

To be considered a critical study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.

Table A-3. PFDoA Epidemiological Studies from Literature Review*

Study Type	Population	Time period	Exposure	Outcomes	Results	Other PFAS evaluated	Reference
Cross-sectional	Prelabor caesarian deliveries	2012–2013	PFAS concentrations in cord blood	Thyroid hormone levels	<p>A standardized unit increase in PFDoA was significantly associated with a 0.062 pmol/L (95% CI: -0.108, -0.017) decrease in thyroid stimulating hormone (TSH) levels <i>PFDoA was not associated with TSH in girls</i></p> <p>A standardized unit increase in PFDoA was significantly associated with a 0.054 pmol/L (95% CI: 0.019, 0.119) increase in free thyroxine (FT4) levels in boys and a 0.174 pmol/L (95% CI: 0.019, 0.331) increase in girls</p> <p>A standardized unit increase in PFDoA was significantly associated with a 0.251 pmol/L (95% CI: -0.361, -0.144) decrease in free tri-iodothyronine (FT3) levels in boys and a 0.124 pmol/L (95% CI: -0.185, -0.056) decrease in girls</p>	PFOA, PFOS, PFNA, PFDA, PFUnA, PFHxS, PFBS, PFHpA, PFSA	Aimuzi et al., 2019 ⁸
Prospective cohort	Chinese infants	2013–2015	PFAS concentrations in umbilical cord blood	Gestational and postnatal growth	<p>Lower birthweight among infants with 0.02-0.04 ng/mL PFDoA vs. those with <0.02 ng/mL ($p<0.05$)</p> <p>Higher postnatal length among infants with >0.04 ng/mL PFDoA vs. those with 0.02-0.04 or <0.02 ng/mL ($p<0.05$)</p>	PFOA, PFNA, PFDA, PFUnA, PFTTrDA, PFTTeDA, PFHxDA, PFHxS, PFOS, PFDS	Cao et al., 2018 ⁹
Prospective cohort	Chinese infants up to 24 months	2012–2015	PFAS concentrations in cord blood plasma	Prevalence of atopic dermatitis (AD)	<p>Among females: Positive association between PFDoA and risk for childhood AD (second vs. lowest quartile): OR=3.24, 95% CI: 1.44, 7.27; overall $p=0.74$</p> <p><i>No association found between other quartile comparisons</i></p>	PFOS, PFOA, PFNA, PFDA, PFUnA, PFHxS, PFBS	Chen et al., 2018 ¹⁰
Cross-sectional	Taiwanese children	2009–2010	PFAS concentrations in serum	Prevalence of asthma	<p>Among all children:</p>	PFOS, PFOA, PFBS, PFDA, PFHxA, PFHxS, PFNA, PFTTeDA	Dong et al., 2013 ¹¹

					Positive association between PFD _o A and asthma (highest vs. lowest quartile): AOR=1.81, 95% CI: 1.02, 3.23		
					Among asthmatic children: Positive association between PFD _o A and asthma severity score (overall $p=0.001$)		
Prospective cohort	4-year-old Japanese children	2003–2013	PFAS concentrations in maternal plasma	Prevalence of total allergic diseases (TAD)* <i>*TAD includes wheezing, eczema, and rhinoconjunctivitis symptoms</i>	Negative association between PFD _o A and total allergic diseases (highest vs. lowest quartile) among boys: AOR=0.492, 95% CI: 0.314, 0.766; $p=0.001$ <i>PFD_oA was not associated with TAD in girls</i>	PFHxS, PFOS, PFHxA, PFHpA, PFOA, PFNA, PFDA, PFUnA, PFTrDA, PFTeDA	Gourdazi et al., 2016 ¹²
Cross-sectional	U.S. adults	1999–2014	PFAS concentrations in serum	Prevalence of total* and individual cardiovascular diseases (CVD) <i>*Total CVD includes congestive heart failure, coronary heart disease, angina pectoris, heart attack, and stroke</i>	Positive association between PFD _o A and presence of total CVD (highest vs. lowest quartile): AOR=1.53, 95% CI: 1.14, 2.04; $p=0.0075$ Positive association between PFD _o A and congestive heart failure (highest vs. lowest quartile): AOR=1.60, 95% CI: 1.01, 2.53; $p=0.0162$ Positive association between PFD _o A and angina pectoris (highest vs. lowest quartile): AOR=1.64, 95% CI: 1.06, 2.54; $p=0.0138$	PFOA, PFOS, PFHxS, EPAH, MPAH, PFDA, PFBS, PFHpA, PFNA, PFSA, PFUnA	Huang et al., 2018 ¹³

Prospective cohort	Japanese mothers and their infants	2002–2005	PFAS concentrations in maternal and cord blood	Expression of thyroid hormones and thyroid antibodies (TA)	Positive association between maternal PFDoA (in TA-positive mothers) and lower free thyroxine among girls ($\beta = -0.077$, 95% CI: -0.148, -0.006; $p=0.037$)	PFHxA, PFHxS, PFHpA, PFOS, PFOA, PFNA, PFDA, PFUnA, PFTrDA, PFTeDA	Itoh et al., 2019 ¹⁴
Cross-sectional	Korean adults	2008	PFAS concentrations in serum	Thyroid hormone concentrations	No associations between serum PFDoA and TSH or T ₄ were identified.	PFHxS, PFHpS, PFOS, PFDS, PFOA, PFNA, PFDA, PFUnA, PFTrDA, PFTeDA, NMeFOSAA, EtPFOSAA	Ji et al., 2012 ⁵¹
Prospective cohort	Korean elderly	2011–2012	PFAS concentrations in serum	Insulin resistance	Insulin resistance among the >90 th percentile PFDoA exposures vs. those with lower PFDoA exposures ($p<0.0001$)	PFBS, PFHxS, PFOS, PFDS, PFBA, PFPeA, PFHxA, PFHpA, PFOA, PFNA, PFDA, PFUnA, PFTrDA, PFTeDA	Kim et al., 2016 ¹⁵
Prospective cohort	Danish adults	Not reported	PFAS concentrations in serum	Antibody response	Negative association between PFDoA and diphtheria antibody levels (Percent change in exponential change of antibody concentrations between days 4 and 10 post-vaccination: -15.64, 96% CI: -0.98, -29.14; $p=0.038$) Negative association between PFDoA and tetanus antibody levels (Percent change in exponential change of antibody concentrations between days 4 and 10 post-vaccination: -10.78, 96% CI: -0.64, -19.90; $p=0.038$)	PFHxS, PHHpA, PFOS, PFOA, PFNA, PFDA, PFUnA	Kielsen et al., 2016 ³⁰
Cross-sectional	South Korean newborns	2008	PFAS concentrations in cord serum	Birth weight	No associations between cord blood PFDoA levels and birth weight observed.	PFBS, PFHxS, PFHpS, PFOS, PFOA, PFNA, PFDA, PFUnA	Lee et al., 2016 ⁴¹
Prospective cohort	Greenlander, Polish, and Ukrainian	2002–2004	PFAS concentrations in maternal serum	Birth weight	No associations between maternal PFDoA levels and birth weight observed.	PFHpA, PFHxS, PFOS, PFOA, PFNA, PFDA, PFUnA	Lenters et al., 2016 ⁴²

	mothers and their infants						
Case-control	Swedish adult males	1990-2009	PFAS concentrations in serum	Coronary heart disease (CHD)	No association between PFDoA and risk for developing CHD was identified.	PFOA, PFOS, PFNA, PFDA, PFHpA, PFHxS, PFUnA	Mattsson et al., 2015 ⁵²
Prospective cohort	4-year-old Chinese children	2012	PFAS concentrations in maternal plasma	Neuro-psychological development	Positive association between increasing PFDoA concentrations and risk of problems in personal-social skills among girls (RR=1.62, 95% CI: 1.04, 2.54)	PFOS, PFOA, PFHxS, PFNA, PFDA, PFUnA, PFTTrDA	Niu et al., 2019 ¹⁶
Prospective cohort	Japanese mothers and their infants	2003–2009	PFAS concentrations in maternal plasma	Infant allergic diseases (eczema, wheezing, and allergic rhino-conjunctivitis symptoms)	No associations between maternal serum PFDoA levels and the risk of allergic disease or eczema in infants were identified.	PFHxA, PFHpA, PFOA, PFNA, PFDA, PFUnA, PFTTrDA, PFTeDA, PFHxS, PFOS	Okada et al., 2014 ⁵³
Cross-sectional	Taiwanese children ages 12–15 years	2009–2010	PFAS concentrations in serum	Serum uric acid levels	No associations between serum PFDoA levels and serum uric acid levels or the risk of hyperuricemia were observed.	PFBS, PFHxS, PFOS, PFHxA, PFHpA, PFNA, PFOA, PFDA, PFTeDA	Qin et al., 2016 ⁵⁴
Prospective cohort	U.S. women	2009–2013	PFAS concentrations in plasma	Gestational diabetes (GDM)	Positive association between PFDoA in women with family history of tyoe-2 diabetes and risk of GDM (RR=3.18, 95% CI: 1.35, 7.52; $p<0.05$)	NMeFOSAA, PFDA, PFHpA, PFHxS, PFNA, PFOA, PFOS, PFUnA	Rahman et al., 2019 ¹⁷
Prospective cohort	Korean adults	2006–2015	PFAS concentrations in serum	Various adverse health effects (increased cholesterol, elevated uric acid levels, hypothyroidism, diabetes mellitus)	Higher PFDoA levels among participants with diabetes compared to those without diabetes ($p<0.05$)	PFBS, PFHxA, PFHpA, PFHxS, PFOA, PFNA, PFOS, PFDA, PFUnA, PFDS, PFTTrDA, PFTeDA	Seo et al., 2018 ¹⁸
Prospective cohort	Greenlander and Ukrainian mothers and their children	2002-2012	PFAS concentrations in maternal serum	Asthma, eczema, and wheezing	No associations between maternal serum PFDoA levels and risk of asthma diagnosis, eczema, or wheezing were identified.	PFHpA, PFHxS, PFOS, PFOA, PFNA, PFDA, PFUnA	Smit et al., 2015

Cross-sectional	Taiwanese mothers and their infants	2000–2001	PFAS concentrations in maternal and cord serum	Thyroid hormone levels	<p>A standardized unit increase (1 ng/mL) in maternal PFDoA was significantly associated with a 0.132 ng/dL (95% CI: -0.204, -0.059) decrease in maternal free T₄ ($p < 0.001$)</p> <p>A standardized unit increase in maternal PFDoA was significantly associated with a 1.742 µg/dL (95% CI: -2.785, -0.700) decrease in maternal total T₄ ($p < 0.01$)</p> <p>A standardized unit increase in maternal PFDoA was significantly associated with a 1.920 µg/dL (95% CI: -3.345, -0.495) decrease in fetal total T₄ ($p < 0.01$)</p> <p>A standardized unit increase in maternal PFDoA was significantly associated with a 0.022 ng/dL (95% CI: -0.035, -0.009) decrease in fetal total T₃ ($p < 0.01$)</p>	PFHxS, PFOA, PFOS, PFNA, PFDA, PFUnA	Wang et al., 2014 ²¹
Prospective cohort	Taiwanese mothers and their infants	2000–2001	PFAS concentrations in maternal serum	Fetal and postnatal growth	<p>Among females exposed to maternal PFDoA:</p> <p>A 1-ln-unit increase in prenatal PFDoA concentration (ng/mL) was significantly associated with a 0.12 kg (95% CI: -0.21, -0.02) decrease in birth weight ($p < 0.05$)</p> <p>A 1-ln-unit increase in prenatal PFDoA concentration (ng/mL) was significantly associated with a 0.38 cm (95% CI: -0.74, -0.02) decrease in birth head circumference ($p < 0.05$)</p> <p>A 1-ln-unit increase in prenatal PFDoA concentration (ng/mL) was significantly associated with a 0.30 (95% CI: -0.55, -0.06) decrease in childhood weight z-score ($p < 0.05$)</p>	PFOA, PFNA, PFDA, PFUnA	Wang et al., 2016 ²⁰

					A 1-ln-unit increase in prenatal PFDoA concentration (ng/mL) was significantly associated with a 0.25 (95% CI: -0.49, 0.00) decrease in childhood height z-score ($p<0.05$)			
Case-control	Chinese women	2014	PFAS concentrations in plasma	Polycystic ovarian syndrome (PCOS) related infertility	Positive association between PFDoA and risk of PCOS-related infertility (medium vs. low tertile: OR=2.36, 95% CI: 1.12, 4.99, $p=0.02$; high vs. low tertile: OR=3.04, 95% CI: 1.19, 7.67, $p=0.02$; overall $p=0.01$)	PFBS, PFHpA, PFHxS, PFOA, PFOS, PFNA, PFDA, PFUnA	Wang et al., 2019 ¹⁹	
Cross-sectional	Chinese mothers and their infants	2013	PFAS concentrations in maternal and cord serum	Thyroid hormone (TSH, T3, T4, FT3, FT4) levels	Negative correlations between maternal PFDoA and all five maternal thyroid hormones ($r = -0.301$ to -0.160 , $p<0.05$ for all crude and adjusted models)	PFOA, PFNA, PFDA, PFUnA, PFHxS, PFOS, 6:2 FTSA, NMeFOSAA	Yang et al., 2016 ²²	
Cross-sectional	Taiwanese children	2009–2015	PFAS concentrations in serum	Serum lipid concentrations	Negative correlations between fetal PFDoA and all five maternal thyroid hormones ($r = -0.268$ to -0.047 , $p<0.05$ for all crude and some adjusted models)	No associations between serum PFDoA and total cholesterol, LDL cholesterol, HDL cholesterol, or triglyceride levels identified.	PFOS, PFOA, PFBS, PFDA, PFHxA, PFHxS, PFHpA, PFNA, PFTeDA	Zeng et al., 2015 ⁵⁵
<p>*This literature review is not exhaustive as the primary purpose of the search was to identify epidemiological studies that support toxicological findings.</p> <p>Epidemiologic terms: OR=odds ratio; AOR=adjusted odds ratio; RR=relative risk; 95% CI=95% confidence interval; r=Spearman correlation coefficient; β=regression coefficient</p> <p>PFAS acronyms: PFOA=perfluorooctanoic acid, PFNA=perfluorononanoic acid, PFDA=perfluorodecanoic acid, PFUnA= perfluoroundecanoic acid, PFTrDA=perfluorotridecanoic acid, PFTeDA=perfluorotetradecanoic acid, PFHxDA=perfluorohexadecanoic acid, PFHxS=perfluorohexane sulfonate, PFOS=perfluorooctane sulfonic acid, PFDS=perfluorodecanesulfonate, PFBS=perfluorobutane sulfonate, PFBA=perfluorobutanoic acid, PFPeA=perfluoropentanoic acid, PFHxA=perfluorohexanoic acid, PFHpA=perfluoroheptanoic acid, 6:2 FTSA=6:2 fluorotelomer sulfonates, NMeFOSAA=N-methyl perfluorooctanesulfonamidoacetate, EPAH=2-(N-ethyl-perfluorooctane sulfonamido)acetate, MPAH=2-(N-methyl-perfluorooctane sulfonamido) acetate, PFSA=perfluorooctane sulfonamide; PFHpS=perfluoroheptane sulfonic acid; PFDS=perfluorodecane sulfonic acid; EtPFOSAA= 2-(Nethyl-perfluorooctane sulfonamido) acetic acid</p>								

6:2 FTSA | 2020

Substance Overview

6:2 fluorotelomer sulfonic acid (6:2 FTSA) is a chemical in a group of contaminants called per- and polyfluoroalkyl substances (PFAS). PFAS are manmade chemicals that have been used in industry and consumer products since the 1940s. Because of their unique physical and chemical properties, PFAS can be found in a variety of commercial products such as paper and textile coatings, food packaging, surfactants, repellants, and fire-fighting foams.^{1,2} 6:2 FTSA is commonly used as a substitute for perfluorooctane sulfonate (PFOS), a longer-chain PFAS that can last for long periods of time in the environment and has been shown to affect human health.³⁻⁷

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard for 6:2 FTSA. DHS cannot recommend an enforcement standard for 6:2 FTSA due to insufficient technical information.

Current Standards	
Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

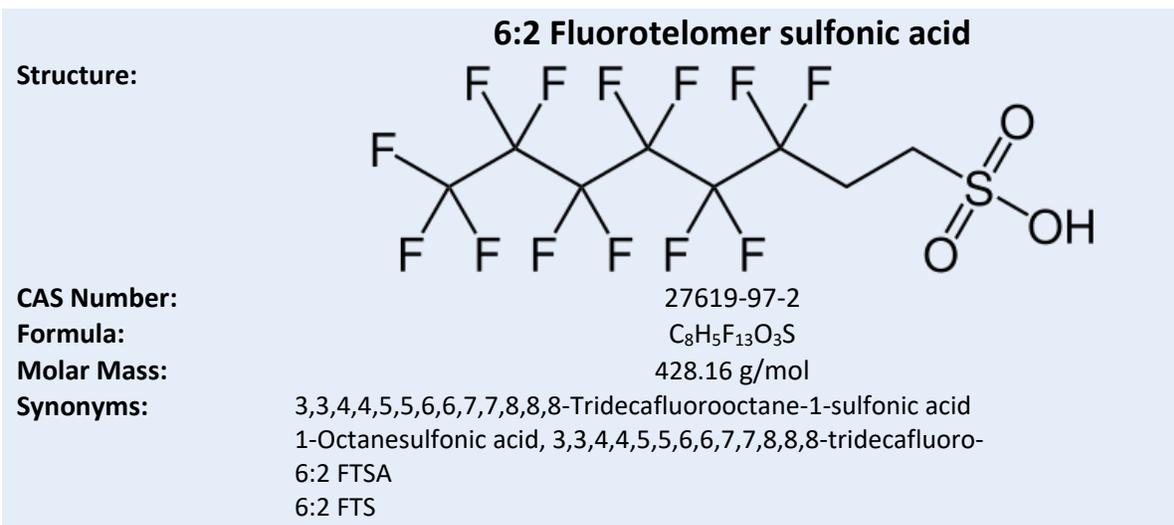
Health Effects

There is limited published information on the health effects of exposure to 6:2 FTSA among people or in research animals. One study in research animals shows that a high level of 6:2 FTSA can cause liver effects, including increased absolute and relative liver weights, increased serum aspartate aminotransferase (AST) and albumin, swelling, and cell damage (necrosis) in the liver of male mice.⁷ However, this study did not evaluate more than one oral dose. One *in vitro* study supporting liver effects found that 6:2 FTSA was more toxic in human liver cells than PFOS and perfluorooctanoic acid (PFOA).⁸ Additional supporting evidence for the association between 6:2 FTSA and liver effects has not been observed in the published literature. 6:2 FTSA has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture studies.⁹ The EPA has not evaluated the carcinogenicity of 6:2 FTSA.⁹

Recommended Standards	
Enforcement Standard:	N/A
Preventive Action Limit:	N/A

a Carcinogenic effects means the substance can cause cancer; mutagenic effects means the substance can cause DNA damage; teratogenic means the substance can cause birth defects; and interactive effects mean the substance can affect the toxicity of another substance or its toxicity can be affected by another substance.

Chemical Profile



Exposure Routes

PFAS, including 6:2 FTSA, can be released directly into the environment during the manufacture and use of PFAS and can be found in water or soil. PFAS can move between groundwater and surface water. Once in water, PFAS can travel long distances.^{10,11} Limited environmental data suggest that 6:2 FTSA can break down in the environment to form shorter-chain compounds, including other PFAS such as perfluoroheptanoic acid (PFHpA), 5:3 fluorotelomer carboxylic acid (5:3 FTCA), perfluoropentanoic acid (PFPeA), and perfluorohexanoic acid (PFHxA).^{12,13}

People can be exposed to PFAS by drinking water, eating food, and breathing in or accidentally swallowing soil or dust containing PFAS.¹⁰

Current Standard

Wisconsin does not currently have a groundwater standard for 6:2 FTSA.¹⁴

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

EPA Oral Reference Dose:	N/A
--------------------------	-----

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

ATSDR Oral Minimum Risk Level:	N/A
--------------------------------	-----

Literature Search

Literature Search Dates:	1900–2019
Key studies found?	Yes
Critical studies identified?	No

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for 6:2 FTSA.¹⁵

Health Advisory

The EPA has not established health advisories for 6:2 FTSA.¹⁶

Drinking Water Concentration (Cancer Risk)

The EPA has not established drinking water concentration based on a cancer risk determination for 6:2 FTSA.⁹

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for 6:2 FTSA.¹⁷

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS uses an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Information System (IRIS) program.

EPA Oral Reference Dose

The EPA does not have an oral reference dose for 6:2 FTSA.⁹

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If we determine that something is carcinogenic and there is no federal number or ADI from the EPA, we must recommend an enforcement standard that corresponds to a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of 6:2 FTSA, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of 6:2 FTSA. If so, we look to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA and International Agency for Research on Cancer (IARC) have not evaluated the carcinogenicity of 6:2 FTSA.^{9,18}

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for 6:2 FTSA.⁹

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information not considered when the value was established that indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For 6:2 FTSA, we searched for values that have been published before or during December 2019. We did not find any relevant guidance values for 6:2 FTSA. While the Agency for Toxic Substances and Disease Registry (ATSDR) reviewed the toxicity of 14 PFAS compounds in 2018, they did not review nor establish any guidance values for 6:2 FTSA.¹⁰

Literature Search

Our literature review focused on relevant scientific literature on the health effects of 6:2 FTSA published before or during December 2019. We looked for studies related to 6:2 FTSA toxicity or effects on a disease state in which information on exposure or dose was included as part of the study.^b Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

In our search, we also looked for epidemiology studies studying the health effects of exposure to 6:2 FTSA among humans. No epidemiologic studies were identified in our search.

Approximately 75 studies on 6:2 FTSA were returned by the search engines. We excluded studies on non-mammalian or cell systems, non-oral exposure routes, and those that did not evaluate health risks from further review. After applying these exclusion criteria, we located one key toxicity study on 6:2 FTSA (summarized in Table A-1). To be considered a critical toxicity study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.^{c,d} Because the key toxicity study did not evaluate more than one dose, it did not meet the criteria to be considered a critical toxicity study (see Table A-2).

b We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

Title/abstract: "6:2 fluorotelomer sulfonate"

Subject area: N/A

Language: English

We also searched online for toxicity studies published by national research programs.

c Due to the limited availability of data for this substance, we considered a study to have a duration appropriate for review if animals were exposed for at least 28 days or if the exposure occurred during pregnancy. We accounted for differences in exposure duration through the use of an uncertainty factor.

d Appropriate toxicity values include the no observable adverse effect level (NOAEL), lowest observable adverse effect level (LOAEL), and benchmark dose (BMD). The NOAEL is the highest dose tested that did not cause an adverse effect, the LOAEL is the lowest dose tested that caused an adverse effect, and the BMD is an estimation of the dose that would cause a specific level of response (typically 5 or 10%).¹⁹ USEPA. A Review of the Reference Dose and Reference Concentration Processes. 2002(EPA/630/P-02/002F).

Key health effects

In the literature search, we did not find studies that show 6:2 FTSA can cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture studies.

Standard Selection

DHS does not recommend an enforcement standard for 6:2 FTSA.

The available health information on 6:2 FTSA is very limited. There are no federal numbers nor a state drinking water standard for 6:2 FTSA. Additionally, the EPA has not evaluated the carcinogenicity or established an ADI (oral reference dose) for 6:2 FTSA. Furthermore, we were unable to identify any critical toxicity studies. Therefore, we have concluded that there is insufficient evidence to establish an enforcement standard for 6:2 FTSA at this time.

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information

DHS does not recommend a preventive action limit for 6:2 FTSA.

There is insufficient evidence to establish a preventive action limit for 6:2 FTSA.

Prepared by Amanda Koch, MPH and Sarah Yang, Ph.D.

Wisconsin Department of Health Services

References

1. Kissa E. *Fluorinated Surfactants and Repellants*. New York, NY: Marcel Dekker; 2001.
2. Calafat AM, Wong LY, Kuklennyik Z, Reidy JA, Needham LL. Polyfluoroalkyl chemicals in the U.S. population: data from the National Health and Nutrition Examination Survey (NHANES) 2003-2004 and comparisons with NHANES 1999-2000. *Environ Health Perspect*. 2007;115(11):1596-1602.
3. Jin L, Jiang C, Zhang P. Photochemical decomposition of 1H,1H,2H,2H-perfluorooctane sulfonate (6:2FTS) induced by ferric ions. *J Environ Sci (China)*. 2017;51:120-127.
4. Shi G, Xie Y, Guo Y, Dai J. 6:2 fluorotelomer sulfonamide alkylbetaine (6:2 FTAB), a novel perfluorooctane sulfonate alternative, induced developmental toxicity in zebrafish embryos. *Aquat Toxicol*. 2018;195:24-32.
5. Urtiaga A, Soriano A, Carrillo-Abad J. BDD anodic treatment of 6:2 fluorotelomer sulfonate (6:2 FTSA). Evaluation of operating variables and by-product formation. *Chemosphere*. 2018;201:571-577.

6. Yang XL, Huang J, Zhang KL, Yu G, Deng SB, Wang B. Stability of 6:2 fluorotelomer sulfonate in advanced oxidation processes: degradation kinetics and pathway. *Environmental Science and Pollution Research*. 2014;21(6):4634-4642.
7. Sheng N, Zhou XJ, Zheng F, et al. Comparative hepatotoxicity of 6: 2 fluorotelomer carboxylic acid and 6: 2 fluorotelomer sulfonic acid, two fluorinated alternatives to long-chain perfluoroalkyl acids, on adult male mice. *Archives of Toxicology*. 2017;91(8):2909-2919.
8. Sheng N, Cui RN, Wang JH, Guo Y, Wang JS, Dai JY. Cytotoxicity of novel fluorinated alternatives to long-chain perfluoroalkyl substances to human liver cell line and their binding capacity to human liver fatty acid binding protein. *Archives of Toxicology*. 2018;92(1):359-369.
9. USEPA. IRIS Assessments. 2019; https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm.
10. ATSDR. Toxicological Profile for Perfluoroalkyls - Draft for Public Comment. In: Registry AfTSA, ed. Atlanta, GA2017.
11. ITRC. Environmental Fate and Transport for Per- and Polyfluoroalkyl Substances. In: Council ITR, ed2018.
12. Eberle M, Edelman M, Denly E, Rabah N. Evaluation of the effects of PFAS soil adsorption and transformation in the presence of divalent cations under ambient conditions. *Remediation-the Journal of Environmental Cleanup Costs Technologies & Techniques*. 2019;30(1):15-25.
13. Zhang S, Lu X, Wang N, Buck RC. Biotransformation potential of 6:2 fluorotelomer sulfonate (6:2 FTSA) in aerobic and anaerobic sediment. *Chemosphere*. 2016;154:224-230.
14. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 140*2017.
15. USEPA. National Primary Drinking Water Regulations. 2018; <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>.
16. USEPA. Drinking Water Contaminant Human Health Effects Information. 2019; <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>.
17. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 809*2018.
18. IARC. List of Classification, Volumes 1-123. 2018; <https://monographs.iarc.fr/list-of-classifications-volumes/>. Accessed May 17, 2019.
19. USEPA. A Review of the Reference Dose and Reference Concentration Processes. 2002(EPA/630/P-02/002F).

Appendix A. Toxicity Data

Table A-1. 6:2 FTSA Toxicity Studies from Literature Review

Study Type	Species	Duration	Chemical Form	Doses (mg/kg-d)	Route	Key Findings	Toxicity Value (mg/kg-d)	Reference
Short-Term	Mouse (male)	28 d	6:2 FTSA	0, 5	Oral gavage	At 5 mg/kg-d, 6:2 FTSA increased absolute and relative liver weights, increased serum AST and albumin, caused cell damage (necrosis) and swelling in the liver, and caused an increase in PPAR γ * and related proteins	NOAEL: N/A LOAEL: 5	Sheng et al., 2017 ⁷
*PPAR γ is a gene that plays an important role in maintaining healthy levels of glucose and fat cells, and is also associated with immune response.								

Table A-2. Critical Study Selection for 6:2-FTSA

Reference	Appropriate duration?	Effects consistent with other studies?	Effects relevant to humans?	Number of Doses	Toxicity value identifiable?	Critical study?*
Sheng et al., 2017 ⁷	✓	✓	✓	1	✓	No

**To be considered a critical study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.*

8:2 FTSA | 2020

Substance Overview

8:2 fluorotelomer sulfonic acid (8:2 FTSA) is a chemical in a group of contaminants called per- and polyfluoroalkyl substances (PFAS). PFAS are manmade chemicals that have been used in industry and consumer products since the 1940s. Because of their unique physical and chemical properties, PFAS can be found in a variety of commercial products such as paper and textile coatings, food packaging, surfactants, repellants, and fire-fighting foams.^{1,2}

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard for 8:2 FTSA. DHS cannot recommend an enforcement standard for 8:2 FTSA due to insufficient technical information currently available.

Current Standards	
Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

Health Effects

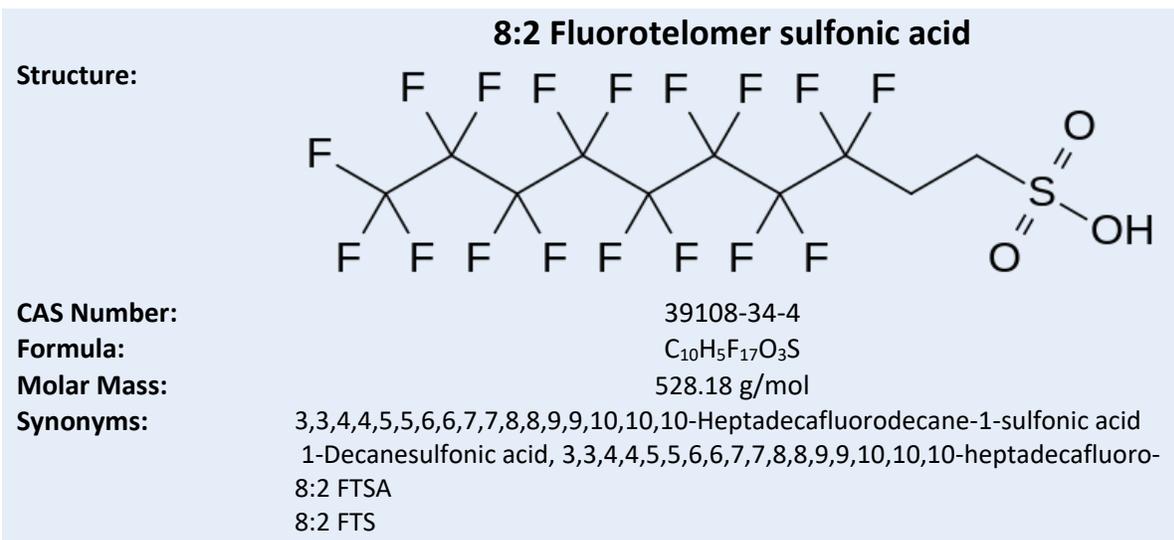
There is limited published information on the health effects of exposure to 8:2 FTSA among people or in research

Recommended Standards	
Enforcement Standard:	N/A
Preventive Action Limit:	N/A

animals. No studies among people or in research animals were identified. 8:2 FTSA has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture studies.³ The EPA has not evaluated the carcinogenicity of 8:2 FTSA.³

a Carcinogenic effects means the substance can cause cancer; mutagenic effects means the substance can cause DNA damage; teratogenic means the substance can cause birth defects; and interactive effects mean the substance can affect the toxicity of another substance or its toxicity can be affected by another substance.

Chemical Profile



Exposure Routes

PFAS, including 8:2 FTSA, can be released directly into the environment during the manufacture and use of PFAS and can be found in water or soil. PFAS can move between groundwater and surface water. Once in water, PFAS can travel long distances.^{4,5}

People can be exposed to PFAS by drinking water, eating food, and breathing in or accidentally swallowing soil or dust containing PFAS.⁴

Current Standard

Wisconsin does not currently have a groundwater standard for 8:2 FTSA.⁶

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

EPA Oral Reference Dose:	N/A
--------------------------	-----

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

ATSDR Oral Minimum Risk Level:	N/A
--------------------------------	-----

Literature Search

Literature Search Dates:	1900–2019
Key studies found?	No
Critical studies identified?	No

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for 8:2 FTSA.⁷

Health Advisory

The EPA has not established health advisories for 8:2 FTSA.⁸

Drinking Water Concentration (Cancer Risk)

The EPA has not established drinking water concentration based on a cancer risk determination for 8:2 FTSA.³

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for 8:2 FTSA.⁹

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS uses an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Information System (IRIS) program.

EPA Oral Reference Dose

The EPA does not have an oral reference dose for 8:2 FTSA.³

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If we determine that something is carcinogenic and there is no federal number or ADI from the EPA, we must recommend an enforcement standard that corresponds to a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of 8:2 FTSA, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of 8:2 FTSA. If so, we look to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA and International Agency for Research on Cancer (IARC) have not evaluated the carcinogenicity of 8:2 FTSA.^{3,10}

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for 8:2 FTSA.³

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information not considered when the value was established that indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we searched for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For 8:2 FTSA, we searched for values that have been published before or during December 2019. We did not find any relevant guidance values for 8:2 FTSA. While the Agency for Toxic Substances and Disease Registry (ATSDR) reviewed the toxicity of 14 PFAS compounds in 2018, they did not review nor establish any guidance values for 8:2 FTSA.⁴

Literature Search

Our literature review focused on relevant scientific literature on the health effects of 8:2 FTSA published before or during December 2019. We looked for studies related to 8:2 FTSA toxicity or effects on a disease state in which information on exposure or dose was included as part of the study.^b Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

In our search, we also looked for epidemiology studies studying the health effects of exposure to 8:2 FTSA among humans. While multiple potential exposure sources and the ability for other PFAS compounds to cause similar health effects preclude using these data to establish a health-based value, such studies are helpful in identifying the critical effects and ensuring that the animal data used to establish the standard are relevant to people.

We excluded studies on non-mammalian or cell systems, non-oral exposure routes, and those that did not evaluate health risks from further review. After applying these exclusion criteria, no key toxicity studies were found.

Key health effects

We did not find studies that show 8-2 FTSA can cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture studies.

^b We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

Title/abstract: "8:2 fluorotelomer sulfonate"

Subject area: N/A

Language: English

We also searched online for toxicity studies published by national research programs.

Standard Selection

DHS does not recommend an enforcement standard for 8:2 FTSA.

The available health information on 8:2 FTSA is very limited. There are no federal numbers nor a state drinking water standard for 8:2 FTSA. Additionally, the EPA has not evaluated the carcinogenicity or established an ADI (oral reference dose) for 8:2 FTSA.

Furthermore, we were unable to identify any critical toxicity studies. Therefore, we have concluded that there is insufficient evidence to establish an enforcement standard for 8:2 FTSA at this time.

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information

DHS does not recommend a preventive action limit for 8:2 FTSA.

There is insufficient evidence to establish a preventive action limit for 8:2 FTSA.

Prepared by Amanda Koch, MPH and Sarah Yang, Ph.D.

Wisconsin Department of Health Services

References

1. Kissa E. *Fluorinated Surfactants and Repellants*. New York, NY: Marcel Dekker; 2001.
2. Calafat AM, Wong LY, Kuklennyik Z, Reidy JA, Needham LL. Polyfluoroalkyl chemicals in the U.S. population: data from the National Health and Nutrition Examination Survey (NHANES) 2003-2004 and comparisons with NHANES 1999-2000. *Environ Health Perspect*. 2007;115(11):1596-1602.
3. USEPA. IRIS Assessments. https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm. Published 2019. Accessed.
4. ATSDR. Toxicological Profile for Perfluoroalkyls - Draft for Public Comment. In: Registry AftSaD, ed. Atlanta, GA2017.
5. ITRC. Environmental Fate and Transport for Per- and Polyfluoroalkyl Substances. In: Council ITR, ed2018.
6. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 140*2017.
7. USEPA. National Primary Drinking Water Regulations. <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>. Published 2018. Accessed.

8. USEPA. Drinking Water Contaminant Human Health Effects Information. <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>. Published 2019. Accessed.
9. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 809*2018.
10. IARC. List of Classification, Volumes 1-123. <https://monographs.iarc.fr/list-of-classifications-volumes/>. Published 2018. Accessed May 17, 2019.

PFDS | 2020

Substance Overview

Perfluorodecane sulfonic acid^a (PFDS) is a chemical in a group of contaminants called per- and polyfluoroalkyl substances (PFAS). Because of its chemical properties, PFDS can be found as an impurity in stain repellants in commercial products like carpet and fabric, as a coating for packaging, and in some fire-fighting foams.^{1,2} PFAS, like PFDS, can persist in the environment for decades.¹

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard or Preventive Action Limit for PFDS.

DHS did not identify sufficient technical information to recommend an enforcement standard or preventive action limit for PFDS.

Health Effects

Studies investigating the health effects of PFDS are limited.

In our literature search, we did not find any studies that looked at the toxicity of PFDS using research animals or cell culture assays. One epidemiology study suggests PFDS exposure is associated with higher cholesterol levels and lower triglyceride levels, but supporting evidence for those associations are not available.³

PFDS has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell cultures.^b The EPA has not evaluated the carcinogenicity of PFDS.⁴

Current Standards

Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

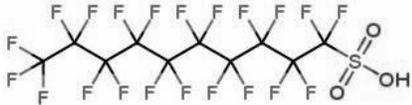
Recommended Standards

Enforcement Standard:	N/A
Preventive Action Limit:	N/A

a This scientific support document also applies to anion salts of perfluorodecane sulfonic acid.

b Carcinogenic effects means the substance can cause cancer; mutagenic effects means the substance can cause DNA damage; teratogenic means the substance can cause birth defects; and interactive effects mean the substance can affect the toxicity of another substance or its toxicity can be affected by another substance.

Chemical Profile

PFDS	
Structure:	
CAS Number:	335-77-3
Formula:	C ₁₀ HF ₂₁ O ₃ S
Molar Mass:	600.15 g/mol
Synonyms:	1,1,2,2,3,3,4,4,5,5,6,6,7,7,8,8,9,9,10,10,10-henicosafluorodecane-1-sulfonic acid Heneicosafluoro-1-decanesulfonic acid Perfluordecansulfonsaure

Exposure Routes

People can be exposed to PFDS by drinking contaminated water, swallowing contaminated soil, eating contaminated fish, eating food that was packaged in material that contains PFDS, and breathing in or swallowing contaminated dust.^{1,2,5,6}

In the environment, PFDS can be found in water or soil as an impurity from the use of other PFAS in manufacturing and consumer products.^{1,2} It can also get into the water or soil from the use of fire-fighting foam. PFAS, like PFDS, can move between groundwater and surface water.² Once in groundwater, PFDS can travel long distances.²

Current Standard

Wisconsin does not currently have groundwater standards for PFDS.⁷

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

EPA Oral Reference Dose:	N/A
--------------------------	-----

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

None available	N/A
----------------	-----

Literature Search

Literature Search Dates:	1900-2020
Total studies evaluated:	14
Key studies found?	No
Critical studies identified?	No

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for PFDS.⁸

Health Advisory

The EPA has not established health advisories for PFDS.⁹

Drinking Water Concentration (Cancer Risk)

The EPA has not established drinking water concentration based on a cancer risk level determination for PFDS.⁴

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for PFDS.¹⁰

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Information System (IRIS) program.

EPA Oral Reference Dose

The EPA does not have an oral reference dose for PFDS.⁴

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If we determine that something is carcinogenic and there is no federal number or ADI from the EPA, DHS must set the standard at a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of PFDS, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of PFDS. If so, we look to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA and International Agency for Research on Cancer (IARC) have not evaluated the carcinogenicity of PFDS.^{4,11}

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for PFDS.⁴

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information that was not considered when the value was established and indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For PFDS, we searched for values that have been published on or before March 2020. The Agency for Toxic Substances and Disease Registry (ATSDR) did not review the toxicity of PFDS in 2018.¹

Literature Search

Our literature review focused on relevant scientific literature on the health effects of PFDS published on or before March 2020. We looked for studies related to PFDS toxicity or effects on a disease state in which information on exposure or dose was included as part of the study.^c Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

Fourteen toxicity studies were returned by the search engines. We excluded studies on non-mammalian or cell systems, non-oral exposure routes, and those that did not evaluate health risks from further review. After applying these exclusion criteria, we did not locate any key toxicity studies on PFDS.

In our search, we located one epidemiology study (See Table A-1 for a summary). While multiple potential exposure sources and the ability for other PFAS to cause similar health effects preclude using these data to establish a health-based value, such studies are helpful in identifying the crucial effects and ensuring that the animal data used to establish the standard is relevant to people.

Critical toxicity studies

We did not identify any critical toxicity studies.

Key health effects

^c We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

Title/abstract: PFDS or "Perfluorodecane sulfonic acid"

Subject area: N/A

Language: English

We also searched online for toxicity studies published by national research programs.

We did not find studies that suggest PFDS has caused carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell cultures.

Standard Selection

DHS did not identify sufficient technical information to recommend an enforcement standard for PFDS.

The available health information on PFDS is limited. There are no federal numbers and no state drinking water standard for PFDS. Additionally, the EPA has not evaluated the carcinogenicity or established an ADI (oral reference dose) for PFDS.

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information

One epidemiology study did find associations between elevated cholesterol levels and decreased triglyceride levels and cord blood levels.³ Due to limited scientific information available, we have concluded that there is insufficient evidence to establish an enforcement standard for PFDS at this time.

DHS does not recommend a preventive action limit for PFDS.

There is insufficient evidence to establish a preventive action limit for PFDS at this time.

Prepared by Gavin Dehnert, Ph.D. and Sarah Yang, Ph.D.

Wisconsin Department of Health Services

References

1. ATSDR. Toxicological Profile for Perfluoroalkyls - Draft for Public Comment. In: Registry AftSaD, ed. Atlanta, GA2017.
2. ITRC. Environmental Fate and Transport for Per- and Polyfluoroalkyl Substances. In: Council ITR, ed2018.
3. Spratlen MJ, Perera FP, Lederman SA, et al. The Association Between Perfluoroalkyl Substances and Lipids in Cord Blood. *Journal of Clinical Endocrinology & Metabolism*. 2020;105(1):43-54.
4. USEPA. IRIS Assessments. 2019; https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm.
5. Domingo JL, Jogsten IE, Eriksson U, et al. Human dietary exposure to perfluoroalkyl substances in Catalonia, Spain. Temporal trend. *Food chemistry*. 2012;135(3):1575-1582.

6. Wu N, Cai DM, Guo MJ, Li M, Li X. Per- and polyfluorinated compounds in saleswomen's urine linked to indoor dust in clothing shops. *Science of the Total Environment*. 2019;667:594-600.
7. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 140*2017.
8. USEPA. National Primary Drinking Water Regulations. 2018; <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>.
9. USEPA. Drinking Water Contaminant Human Health Effects Information. 2019; <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>.
10. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 809*2018.
11. IARC. List of Classification, Volumes 1-123. 2018; <https://monographs.iarc.fr/list-of-classifications-volumes/>. Accessed May 17, 2019.

Appendix A. Toxicity Data

Table A-I. PFDS Epidemiological Studies from Literature Review

Study Type	Population	Time period	Exposure	Outcomes	Results	Other PFAS Evaluated	Reference
Cohort	222 infants born in the Columbia University birth cohort New York, New York	2001-2002	Maternal cord blood PFAS concentrations	Total lipids levels, total cholesterol levels, and triglyceride levels	An increase in one percent PFDS concentration was associated with 0.91 percent increase in total cholesterol (95% CI: 0.006%, 0.177%). An increase in PFDS concentrations was negatively associated with triglyceride levels (p-trend = 0.04).	PFOS, PFOA, PFHxS, PFNA	Spratlen, 2020 (reference)

Epidemiologic terms: OR=odds ratio; AOR=adjusted odds ratio; RR=relative risk; 95% CI=95% confidence interval; r=Spearman correlation coefficient; β =regression coefficient

PFAS acronyms: PFOA=perfluorooctanoic acid, PFNA=perfluorononanoic acid, PFDA=perfluorodecanoic acid, PFUnA= perfluoroundecanoic acid, PFDoA=perfluorododecanoic acid, PFTriA=perfluorotetradecanoic acid, PFHxDA=perfluorohexadecanoic acid, PFHxS=perfluorohexane sulfonate, PFOS=perfluorooctane sulfonic acid, PFDS=perfluorodecanesulfonate, PFBS=perfluorobutane sulfonate, PFBA=perfluorobutanoic acid, PFPeA=perfluoropentanoic acid, PFHxA=perfluorohexanoic acid, PFHpA=perfluoroheptanoic acid, 6:2 FTSA=6:2 fluorotelomer sulfonates, NMeFOSAA=N-methyl perfluorooctanesulfonamidoacetate, EPAH=2-(N-ethyl-perfluorooctane sulfonamido)acetate, MPAH=2-(N-methyl-perfluorooctane sulfonamido) acetate, PFSA=perfluorooctane sulfonamide; PFHpS=perfluoroheptane sulfonic acid; PFDS=perfluorodecane sulfonic acid; EtPFOSAA= 2-(Nethyl-perfluorooctane sulfonamido) acetic acid

HFPO-DA | 2020

Substance Overview

Hexafluoropropylene oxide dimer acid (HFPO-DA) is a chemical in a group of contaminants called per- and polyfluoroalkyl substances (PFAS).¹ HFPO-DA is commonly referred to as its trade name GenX and was developed to replace longer chained PFAS such as perfluorooctanoic acid (PFOA) and perfluorooctane sulfonate (PFOS), chemicals with known human health impacts.^{2,3} Because of its chemical properties, HFPO-DA can be found in stain repellants in commercial products like carpet and fabric, as a coating for packaging, and in some fire-fighting foams.³⁻⁵

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard for HFPO-DA. DHS recommends an enforcement standard of 300 nanograms per liter (ng/L) for HFPO-DA. The recommended standard is based on a study that found HFPO-DA exposure can increase liver and kidney toxicity and increase liver cell death.^{3,6}

DHS recommends that the preventive action limit for HFPO-DA be set at 10% of the enforcement standard because HFPO-DA has been shown to cause carcinogenic and mutagenic effects in research animals and cell culture studies.^{3,4}

Current Standards	
Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

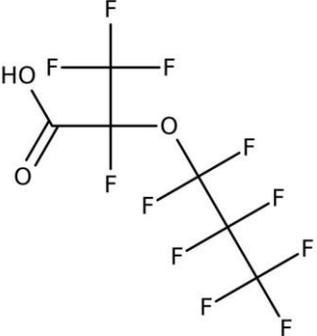
Recommended Standards	
Enforcement Standard:	300 ng/L
Preventive Action Limit:	30 ng/L

Health Effects

Data on the health effects of HFPO-DA exposure are limited.^{3,4,6-17} There are no studies that have evaluated the effects of HFPO-DA among people. High levels of HFPO-DA can decrease red blood cell numbers, hemoglobin, and offspring weight, and increase kidney and liver damage in mice and rats.^{3,4,6-17}

Limited studies suggest HFPO-DA has been shown to cause mutagenic and carcinogenic (cancer) effects in research animals and cell cultures.^{3,4,15} One study found that HFPO-DA can promote DNA damage in rat thyroid cells and disrupt DNA transcription-factors in genes.⁴ A long-term study in rats found that high levels of HFPO-DA increased the incidence of liver and pancreatic tumors.^{3,8,18} The EPA determined that there is suggestive evidence of carcinogenic potential for HFPO-DA in humans.³

Chemical Profile

HFPO-DA	
Structure:	
CAS Number:	13252-13-6
Formula:	C ₆ HF ₁₁ O ₃
Molar Mass:	330.05 g/mol
Synonyms:	GenX 2,3,3,3-tetrafluoro-2-(heptafluoropropoxy)propanoic acid Perfluoro(2-methyl-3-oxahexanoic) acid 2,3,3,3-tetrafluoro-2-(1,1,2,2,3,3,3-heptafluoropropoxy)propanoic acid

Exposure Routes

People can be exposed to HFPO-DA by drinking contaminated water, swallowing contaminated soil, eating fish from contaminated lakes, eating food that was packaged in material that contains HFPO-DA, and breathing in or swallowing dust that contains HFPO-DA.^{3,5,14,19-22} Babies born to mothers exposed to PFAS, like HFPO-DA, can be exposed to PFAS during pregnancy and breastfeeding.^{1,3}

In the environment, HFPO-DA can be found in water as it is highly water soluble and HFPO-DA can move between groundwater and surface water. Once in groundwater, HFPO-DA can travel long distances.³

Current Standard

Wisconsin does not currently have groundwater standards for HFPO-DA.²³

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

Draft EPA Subchronic Oral Reference Dose:	200 ng/kg-d	(2018)
Draft EPA Chronic Oral Reference Dose:	80 ng/kg-d	(2018)

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

None available

Literature Search

Literature Search Dates:	2019-2020
Key studies found?	Yes
Critical studies identified?	Yes

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for HFPO-DA.²⁴

Health Advisory

The EPA has not established health advisories for HFPO-DA.²⁵

Drinking Water Concentration (Cancer Risk)

The EPA has not established drinking water concentration based on a cancer risk level determination for HFPO-DA.²⁶

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for HFPO-DA.²⁷

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Information System (IRIS) program.

Draft EPA Oral Reference Dose

In 2018, the EPA established a draft sub-chronic reference dose of 200 nanograms per kilogram per day (ng/kg-d) and draft chronic reference dose of 80 ng/kg-d for HFPO-DA.³ The EPA evaluated several studies including those that observed effects on development, reproduction, the immune system, and liver and kidney toxicity in research animals. They selected a DuPont study from 2010 (18405-1037) that observed liver cell death in male parental mice after being exposed for 84-85 days as the critical study.³ The EPA identified a No Observable Adverse Effect Level (NOAEL) of 0.1 milligrams per kilogram per day (mg/kg-d) from this study.³

The EPA used benchmark dose modeling to estimate a human equal dose.³ The EPA estimated a human equivalent dose of 0.023 mg/kg-d. For the sub-chronic reference dose, the EPA applied a total uncertainty factor of 100 to account for differences between people and research animals (3), differences among people (10), and limited availability of information (3). This resulted in a sub-chronic oral reference dose of 200 ng/kg-d.³

For the chronic reference dose, the EPA applied a total uncertainty factor of 300 to the benchmark dose to account for differences between people and research animals (3), using short term data to predict long term health effects (3), differences among people (10), and limited availability of information (3). This resulted in a chronic oral reference dose of 80 ng/kg-d.³

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If we determine that something is carcinogenic and there is no federal number or ADI from the EPA, DHS must set the standard at a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of HFPO-DA, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of HFPO-DA. If so, we look to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA determined there is suggestive evidence of carcinogenic potential of oral exposure to HFPO-DA in humans which is based on an increased incidence of cancer and tumors in female rats and increased incidence of combined pancreatic cancer and tumors in male rats.^{3,8,18}

The International Agency for Research on Cancer (IARC) has not evaluated the carcinogenicity of HFPO-DA.²⁶

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for HFPO-DA.²⁸

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information that was not considered when the value was established and indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For HFPO-DA, we searched for values that been published on or before September 2020. We did not find any additional guidance values.

Literature Search

Our literature review focused on relevant scientific literature on the health effects of HFPO-DA published on or before September 2020. We looked for studies related to HFPO-DA toxicity or effects on a disease state in which information on exposure or dose was included as part of the study.^a Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

Approximately 100 toxicity studies were returned by the search engines. We excluded studies on non-mammalian or cell systems, non-oral exposure routes, those that did not evaluate health risks, and those already evaluated by the EPA *Human Health Toxicity Values* from further review. After applying these exclusion criteria, we located multiple key toxicity studies on HFPO-DA (summarized in Table A-1). To be considered a critical toxicity study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.^{b-c} All of these studies met the criteria to be considered a critical toxicity study (see Table A-2).

Critical Toxicity Studies

To compare between results from recently found studies and the study used to set the current enforcement standard, we calculated an acceptable daily intake (ADI) for each study/effect. The ADI is the estimated amount of HFPO-DA that a person can be exposed to every day and not experience health impacts. As such, we calculated ADI by dividing a toxicity value from either a no-observed adverse effect level (NOAEL), lowest observed adverse effect level (LOAEL), or benchmark dose (BMD) identified in a study by a factor accounting for various sources of scientific uncertainty.^d Uncertainty factors were included, as appropriate, to account for differences between humans and animals, differences between healthy and sensitive human populations, using data from short-term experiments to protect against

a We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

Title/abstract: GenX or HFPO-DA or HFPO dimer acid ammonium salt or "Hexafluoropropylene Oxide Dimer Acid"

Language: English

We also searched online for toxicity studies published by national research programs.

b Appropriate toxicity values include the no observable adverse effect level (NOAEL), lowest observable adverse effect level (LOAEL), and benchmark dose (BMD).

c Due to the limited availability of data for this substance, we considered a study to be of an appropriate duration if animals were exposed for at least 28 days or if the exposure occurred during pregnancy. We accounted for differences in exposure duration through the use of an uncertainty factor.

d The NOAEL is the highest dose tested that did not cause an adverse effect, the LOAEL is the lowest dose tested that caused an adverse effect, and the BMD is an estimation of the dose that would cause a specific level of response (typically 5 or 10%).

effects from long-term exposure, and using data where a health effect was observed to estimate the level that does not cause an effect.

Blake et al., 2020

In 2020, Blake et al. exposed pregnant female mice to different concentrations of HFPO-DA (0, 2 or 10 milligrams of HFPO-DA per kilogram body weight per day or mg/kg-d) through gavage for gestational days 1-17.¹⁴ They found that HFPO-DA increased maternal weight gain, maternal liver and kidney weight, the incidence of maternal placental lesions, and placental thyroid hormone T4 levels (Table 1).

Table 1. Statistically Significant Effects Observed in Blake et al, 2020¹⁴

Effects observed in pregnant females		Dose (mg/kg-d)	
		2	10
Growth	Increased maternal weight gain		✓
Liver	Increased liver weight	✓	✓
		✓	✓
Kidney	Increased kidney weight		✓
Reproduction	Increased placental weight	✓	✓
	Increased placental lesions	✓	✓
	Increased placental T4 concentrations		✓

For this study, we identified a LOAEL of 2 mg/kg-d based on effects on placental and liver toxicity. We estimated a candidate ADI of 0.0002 mg/kg-d HFPO-DA or 200 nanograms of HFPO-DA per kilogram body weight per day or ng/kg-d from this study based on a LOAEL of 2 mg/kg-d and a total uncertainty factor of 10,000 to account for differences between people and research animals (10), differences among people (10), using a LOAEL instead of a NOAEL (10), and the limited availability of information (10). While we obtained a candidate ADI for HFPO-DA from this study, this study was not used to establish a recommended enforcement standard due to significant uncertainty.

Conley et al., 2019

In 2019, Conley et al. exposed pregnant female rats to different concentrations of HFPO-DA (0, 1, 3, 10, 30, 62.5, 125, 250, or 500 mg/kg-d) through gavage for gestational days 14-18.¹⁷ They found that HFPO-DA decreased maternal thyroid hormone levels, maternal lipid levels, offspring birth weight, and offspring reproductive organ weight (Table 2).

Table 2. Statistically Significant Effects Observed in Conley et al, 2019¹⁷

Effects observed in pregnant females		Dose (mg/kg-d)							
		1	3	10	30	62.5	125	250	500
Growth	Decreased body weight gain							✓	✓

Liver	Increased liver weight	✓	✓	✓	✓
	Decreased serum triglyceride levels				✓
	Decreased total cholesterol levels			✓	✓
	Decreased high-density lipoprotein			✓	✓
	Decreased low-density lipoprotein		✓	✓	✓
Thyroid	Decreased T3 concentrations	✓	✓	✓	✓
General biochemistry	Lower calcium levels			✓	✓

For this study, we identified a NOAEL of 10 mg/kg-d based on effects on thyroid hormones at higher concentrations. We estimated a candidate ADI of 0.003 mg/kg-d or 3000 ng/kg-d HFPO-DA from this study based on a NOAEL of 10 mg/kg-d and a total uncertainty factor of 3000 to account for differences between people and research animals (10), differences among people (10), use of a shorter duration study to protect against effects from long-term exposure (3), and the limited availability of information (10).

DuPont, 2008a (study number: 24447)

In 2008, DuPont exposed male and female rats to different concentrations of HFPO-DA (males: 0, 0.3, 3, and 30 mg/kg-d; females: 0, 3, 30, 300 mg/kg-d) through gavage for 28 days (study number: 24447).⁹ HFPO-DA decreased red blood cells, hemoglobin, hematocrit, and immature red blood cell, cholesterol, and triglycerides levels and increased liver weights, liver cell size, albumin and glucose levels, and oxidation and enzyme activity in the liver (Table 3).

Table 3. Statistically significant effects observed in DuPont, 2008a (study number: 24447)⁹

Effects observed in males		Dose (mg/kg-d)		
		0.3	3	30
Blood	Decreased red blood cells		*	*
	Decreased hemoglobin		*	*
	Decreased hematocrit		*	*
	Increased immature red blood cells		*	*
Liver	Increased liver weight		*	*
	Increased albumin levels		*	*
	Decreased cholesterol levels	*	*	*

	Decreased triglyceride levels	*	*	*
	Increased glucose levels			*
	Increase in liver cell size		*	*
	Increase in liver oxidation activity	*	*	*
	Increase in liver p450 enzyme activity			*
		Dose (mg/kg-d)		
Effects observed in females		3	30	300
Liver	Increased liver weight			*
	Increased liver cell size			*
	Increased liver oxidation activity		*	*
*The authors did not consider this effect to be adverse (see DuPont, 2008a for more details).				

For this study, we identified a LOAEL of 0.3 mg/kg-d based on effects on cholesterol. We selected this value instead of the NOAEL identified by the researchers because this effect was observed in males at all doses and impacts on cholesterol have been observed for other PFAS. We estimated a candidate ADI of 3 nanograms per kilogram per day (ng/kg-d) from this study based the LOAEL and a total uncertainty factor of 100,000 to account for differences between people and research animals (10), differences among people (10), use of a LOAEL instead of a NOAEL (10), use of a shorter duration study to protect against effects from long-term exposure (10), and the limited availability of information (10). While we obtained a candidate ADI for HFPO-DA from this study, this study was not used to establish a recommended enforcement standard due to significant uncertainty.

DuPont, 2008b (study number: 24459)

In 2008, DuPont exposed male and female mice to different concentrations of HFPO-DA (0, 0.1, 3, and 30 mg/kg-d) through gavage for 28 days (study number: 24459).¹⁰ HFPO-DA increased body weight and food consumption, decreased red blood cells, hemoglobin, and hematocrit levels, increased liver cell death, and increased liver and kidney weight in rats (Table 4).

Table 4. Statistically significant effects observed in DuPont, 2008b¹⁰

		Dose (mg/kg-d)		
Effects observed in males		0.1	3	30
Body weight	Increased body weight			✓
	Increased food consumption			✓
Blood	Decreased red blood cells		*	*
	Decreased hemoglobin levels		*	*
	Decreased hematocrit levels		*	*
Liver	Increased liver weight		*	*
	Increased in liver cell size		*	*
	Increased liver cell death		✓	✓

	Decrease in cholesterol levels		*	
	Increase in alanine aminotransferase		*	*
	Increase in alkaline phosphatase		*	*
	Increase in sorbitol dehydrogenase		*	*
	Increase in liver oxidation activity	*	*	*
	Increase in liver p450 enzyme activity		*	*
Kidney	Increased kidney weight			✓
	Increased kidney cell size			✓
		Dose (mg/kg-d)		
Effects observed in females		0.1	3	30
Body weight	Increased body weight			✓
	Increased food consumption			✓
Liver	Increased liver weights		*	*
	Increased liver cell size		*	*
	Increased liver cell death			✓
	Increase in alanine aminotransferase			*
	Increase in alkaline phosphatase			*
	Increase in sorbitol dehydrogenase			*
	Increase in liver oxidation activity		*	*
Uterus	Decreased uterus weight			✓
	Increased number of animals in the diestrus stage			✓
*The authors did not consider this effect to be adverse (see DuPont, 2008b for more details).				

For this study, we identified a NOAEL of 0.1 mg/kg-d based on liver cell death and toxicity effects of the blood. We did not consider the effects on liver oxidation activity to be adverse because this effect is considered non-adverse for humans when it is observed in rats (see summary below).³⁰ Therefore, we selected the same NOAEL value that was identified by the authors. We estimated a candidate ADI of 0.00001 mg/kg-d or 10 ng/kg-d from this study based the NOAEL and a total uncertainty factor of 10,000 to account for differences between people and research animals (10), differences among people (10), use of a shorter duration study to protect against effects from long-term exposure (10), and the limited availability of information (10). While we obtained a candidate ADI for HFPO-DA from this study, this study was not used to establish a recommended enforcement standard due to significant uncertainty.

DuPont, 2009 (study number: 17751-1026)

In 2009, DuPont exposed male and female rats to different concentrations of HFPO-DA (males: 0, 0.1, 1, and 100 mg/kg-d; females: 0, 10, 100, 1000 mg/kg-d) through gavage for 90 days (study number: 17751-

1026).¹¹ HFPO-DA decreased red blood cells, hemoglobin, and hematocrit levels, increased liver and kidney weight, and caused death (Table 5).

Table 5. Statistically significant effects observed in DuPont, 2009¹¹

Effects observed in males		Dose (mg/kg-d)		
		0.1	10	100
Blood	Decreased red blood cells		*	✓
	Decreased hemoglobin		*	✓
	Decreased hematocrit		*	✓
Liver	Increased liver weight		*	*
	Decreased cholesterol levels		*	*
	Increase liver cell size		*	*
Kidney	Increased kidney weight		✓	✓
Effects observed in females		Dose (mg/kg-d)		
		10	100	1000
Body	Decreased survival			✓
	Increased food consumption			✓
Liver	Increased liver weight			*
	Increased liver cell size			*
	Decreased alkaline phosphatase			*
	Decreased total bilirubin levels		*	
	Decreased cholesterol levels		*	*
Blood	Decreased red blood cells			✓
	Decreased hemoglobin			✓
	Decreased hematocrit			✓
Kidney	Increased kidney weight	*	*	*
	Lower pH in urine			*
	Lower osmolality in urine			*
	Higher urine amounts			*
Survival	Increase in death			✓
*The authors did not consider this effect to be adverse (see DuPont, 2009 for more details).				

For this study, we identified a NOAEL of 0.1 mg/kg-d based on toxicity effects observed in the blood and kidney at higher concentrations in male rats. We selected this value instead of the NOAEL identified by the authors because these effects were also observed in males at the middle dose (10 mg/kg-d) and these impacts have been observed for HFPO-DA and other PFAS.¹¹ We estimated a candidate ADI of 0.00003 mg/kg-d or 30 ng/kg-d from this study based the NOAEL and a total uncertainty factor of 3,000 to account for differences between people and research animals (10), differences among people (10),

use of a shorter duration study to protect against effects from long-term exposure (3), and the limited availability of information (10).

DuPont, 2010a (study number: 18405-1307)

In 2010, DuPont exposed male and female mice to different concentrations of HFPO-DA (0, 0.1, 0.5, and 5 mg/kg-d) through gavage for 96 or 97 days (study number: 18405-1307).¹² HFPO-DA increased body weight and caused liver damage in mice (Table 6).

Table 6. Statistically significant effects observed in DuPont, 2010a¹²

Effects observed in males		Dose (mg/kg-d)		
		0.1	0.5	5
Body	Increase in body weight			*
Kidney	Increased kidney weight			*
	Increased kidney cell size			*
Liver	Increased liver weight			*
	Increased liver cell size		*	*
	Increased relative liver weight		*	*
	Increase in cell death			✓
	Decreased cholesterol levels			*
	Increase in albumin levels			*
	Increased glucose levels			*
	Increased aspartate aminotransferase			*
	Increased alanine aminotransferase			*
	Increased sorbitol dehydrogenase			*
	Increased alkaline phosphatase			*
	Increased total bile acids			*
	Effects observed in females		Dose (mg/kg-d)	
0.1			0.5	5
Liver	Increased liver weight			*
	Increased liver cell size			*
	Increased cell death			✓
	Increased total bile acids			*
	Increase in in albumin levels			*
	Increased aspartate aminotransferase			*
	Increased alanine aminotransferase			*
	Increased sorbitol dehydrogenase			*
	Increased alkaline phosphatase			*

*The authors did not consider this effect to be adverse (see DuPont, 2010a for more details).

For this study, the researchers identified a NOAEL of 0.5 mg/kg-d based on liver cell death in male and female rats at high concentrations. We estimated a candidate ADI of 500 nanograms per kilogram per day (ng/kg-d) from this study based the NOAEL and a total uncertainty factor of 1,000 to account for differences between people and research animals (10), differences among people (10), and the limited availability of information (10).

DuPont, 2010b (study number: 18405-1037)

In 2010, DuPont exposed males and females mice to different concentrations of HFPO-DA (0, 0.1, 0.5, and 5 mg/kg-d) by gavage for 84 to 85 days for males and for 14 days before impregnation and through gestational day 20 by gavage for females (study number: 18405-1037.⁶ HFPO-DA increased body weight, liver weight and liver cell death, and kidney weight and decreased offspring body weight and body weight gain (Table 7).

Table 3. Statistically significant effects observed in DuPont, 2010b⁶

		Dose (mg/kg-d)		
Effects observed in females		0.1	0.5	5
Growth	Increased body weight		✓	✓
	Increased food consumption		✓	✓
Liver	Increased liver weight		*	*
	Increased absolute liver weight		*	*
	Increase in liver cell size			*
	Increase in liver cell death		✓	✓
Kidney	Increased kidney weight			*
		Dose (mg/kg-d)		
Effects observed in offspring		0.1	0.5	5
Growth	Decreased body weight			✓
	Decreased body weight gains			✓
		Dose (mg/kg-d)		
Effects observed in males		0.1	0.5	5
Growth	Increase in body weight			✓
Liver	Increased liver weight			*
	Increased liver cell size		*	*
	Increased relative liver weight		*	*
	Increase in liver cell size		*	*
	Increased liver cell death		✓	✓
Kidney	Increased kidney weight			*
	Increased kidney cell size		*	*
*The authors did not consider this effect to be adverse (see DuPont, 2010b for more details).				

For this study, the researchers identified a NOAEL of 0.1 mg/kg-d based on liver cell death at higher concentrations in male mice. We estimated a candidate ADI of 0.00003 mg/kg-d or 30 ng/kg-d from this study based the NOAEL and a total uncertainty factor of 3,000 to account for differences between people and research animals (10), differences among people (10), use of a shorter duration study to protect against effects from long-term exposure (3), and the limited availability of information (10).

DuPont, 2010c (study number: 18405-841)

In 2010, DuPont exposed female rats to different concentrations of HFPO-DA (0, 10, 100, and 1000 mg/kg-d) through gavage for gestational days 6 to 20 (study number: 18405-841).¹³ HFPO-DA decreased liver, kidney, and uterine weights in pregnant females and offspring birthweights and caused early delivery of the offspring (Table 8).

Table 8. Statistically significant effects observed in DuPont, 2010c¹³

		Dose (mg/kg-d)		
Effects observed in females		10	100	1000
Reproduction	Early delivery time		✓	✓
	Decreased uterine weight		✓	✓
Liver	Increased liver weight		✓	✓
	Increased liver cell death		✓	✓
Kidney	Increased kidney weight			✓
		Dose (mg/kg-d)		
Effects observed in offspring		10	100	1000
Growth	Decreased birth weights		✓	✓

For this study, the researchers identified a NOAEL of 10 mg/kg-d based on liver cell death and reproduction toxicity effects at higher concentrations in female rats, and decreased birth weights at high concentrations in the offspring. We estimated a candidate ADI of 0.01 mg/kg-d or 10,000 ng/kg-d from this study based the NOAEL and a total uncertainty factor of 1,000 to account for differences between people and research animals (10), differences among people (10), and the limited availability of information (10).

Rae et al. 2015 (DuPont, 2013)

In 2013, Rae et al. gave different concentrations of HFPO-DA to male rats (0, 0.1, 1, and 50 mg/kg-d) and female rats (0, 1, 50, and 500 mg/kg-d) through gavage for 2 years.^{8,18} HFPO-DA decreased body weight, hemoglobin and hematocrit levels, caused liver damage, and increased the incidence of pancreatic and liver cancer in rats (Table 9). In addition to these statistically significant findings, the study observed an increase in liver tumors, lesions in the liver, and liver cell death at the highest concentration of HFPO-DA exposure in female rats and an increased incidence of pancreatic tumors and cancer combined at the highest concentration of HFPO-DA exposure in male rats. Furthermore, the study also observed an

increased incidence of pathology severity in the kidney such as tubular dilatation, lesions, tubular mineralization, renal papillary necrosis, and chronic progressive nephropathy (CPN) at the highest concentration of HFPO-DA exposure in females.

Table 9. Statistically significant effects observed in Rae et al., 2015^{8,18}

		Dose (mg/kg-d)		
Effects observed in males		0.1	1	50
Growth	Decreased body weight			✓
Blood	Decreased red cell mass			✓
Liver	Increased alanine aminotransferase			✓
	Increased sorbitol dehydrogenase			✓
	Increased liver cell death			✓
	Increased albumin levels			✓
Pancreases	Increased incidence of pancreatic cancer			✓
		Dose (mg/kg-d)		
Effects observed in females		1	50	500
Growth	Decreased body weight			✓
	Decreased body weight gain			✓
	Decreased food efficiency			✓
Blood	Decrease hemoglobin			✓
	Decrease in hematocrit			✓
	Increased incidence of liver cancer			✓

For this study, the researchers identified a NOAEL of 1 mg/kg-d based on toxicity effects on decreased body weight, decreased red blood cell mass, and the observed increased incidence of pancreatic cancer at higher concentrations in male rats. We estimated a candidate ADI of 0.001 mg/kg-d or 1,000 ng/kg-d from this study based the NOAEL and a total uncertainty factor of 1,000 to account for differences between people and research animals (10), differences among people (10), and the limited availability of information (10).

Rushing et al., 2017

In 2017, Rushing et al. exposed male and female rats to different concentrations of HFPO-DA (0, 1, 10, and 100 mg/kg-d) through gavage for 28 days.⁷ HFPO-DA exposure increased liver weights and decreased antibody response in mice.

Table 1. Statistically significant effects observed in Rushing et al. 2017⁷

		Dose (mg/kg-d)		
Effects observed in males		1	10	100
Liver	Increased relative liver weight		✓	✓

Immune	Decreased T lymphocyte numbers			✓
		Dose (mg/kg-d)		
Effects observed in females		1	10	100
Liver	Increased relative liver weight			✓
Immune	Decreased antibody response			✓

For this study, we identified a NOAEL of 10 mg/kg-d based on the immune toxicity effects at higher concentrations in both male and female rats. We did not consider the effects on liver weight to be adverse because this effect is considered non-adverse for humans when it is observed in rats (see summary below).³⁰ We estimated a candidate ADI of 0.001 mg/kg-d or 1000 ng/kg-d HFPO-DA from this study based on the NOAEL of 10 mg/kg-d and a total uncertainty factor of 10,000 to account for differences between people and research animals (10), differences among people (10), use of a shorter duration study to protect against effects from long-term exposure (10), and the limited availability of information (10). While we obtained a candidate ADI for HFPO-DA from this study, this study was not used to establish a recommended enforcement standard due to significant uncertainty.

Key Health Effects

We found studies that indicate HFPO-DA may cause carcinogenic and mutagenic effects in research animals and cell cultures.^{3,4,15} In their draft of Human Health Toxicity Values for HFPO-DA risk assessment, the EPA has concluded that there is suggestive evidence of carcinogenic potential of oral exposure to HFPO-DA chemicals in humans based on the Rae et al., 2015 study that saw an increase in liver and pancreatic tumors and carcinomas in rats when exposed to high levels of HFPO-DA for two years (Table 9).^{8,18} HFPO-DA has also been shown to increase DNA damage in rat thyroid cells.⁴

We did not find any studies indicating HFPO-DA can cause teratogenic or interactive effects in people, research animals, or cell culture.

Discussion

While studies on the effects of HFPO-DA among research animals are limited, the available data indicate that Gen X has been associated with decreased red blood cell numbers, decreased hemoglobin, increased kidney damage, altered cholesterol levels, decreased offspring weight, and delayed offspring development in mice and rats.^{3,14,17}

Studies in research animals and people have shown that PFAS can affect the levels of thyroid hormones; for HFPO-DA in particular, this has been observed in research animals.^{1,3} Thyroid hormones are crucial for development, energy balance, and metabolism in all species.³¹ In people, thyroid hormones play an important role in the development of the brain, lungs, and heart.³¹ Scientists have learned that certain PFAS, including HFPO-DA, can bind to transport proteins involved in moving thyroid hormones throughout the body.³⁰ Scientists are still learning how this effect occurs and its impact on health.

A number of studies have demonstrated associations between long-chain PFAS and birth weight and development in people and research animals.¹ Multiple studies using research animals have noted a decrease in fetal body weight and a decrease in body weight gain when exposed to HFPO-DA.^{1,3,14,17} Scientists have associated multiple health issues in adulthood with lower weight at birth such as adult obesity, unfavorable metabolic outcomes, and increased risk of cardiovascular disease.³¹⁻³³ Scientists are still continuing to learn how lower birth weight can affect adult human health.

Studies in research animals have shown that PFAS, including HFPO-DA, can affect the blood (e.g., red blood cell concentrations, hemoglobin level, hematocrit level).¹ Testing whether these components fall within a normal range can help determine the health of a human or an animal and whether disease is present or there are issues with normal organ function.³⁴

A number of studies have demonstrated that liver effects caused by PFAS, like HFPO-DA, occur primarily through activation of the nuclear hormone receptor, PPAR α (peroxisome proliferator-activated receptor alpha).³⁵⁻³⁷ Nuclear receptors regulate gene expression and PPAR α regulates the expression of genes involved in lipid and cholesterol metabolism.³⁰ While PPAR α receptors are found in rodents and humans, levels of these receptors are much higher in rats and mice than in monkeys and humans.³⁵ This means that effects caused by activation of PPAR α are more likely to occur in rodents than they are to occur in people. Therefore, we have adapted criteria from Hall et al. (2012) similarly to the Agency for Toxic Substances and Disease Registry (ATSDR) for determining the adversity of the liver effects reported in the rodent PFAS studies to adverse effects in humans.^{1,30} Concentrations associated with an increase in liver weight or liver cell growth were not considered adverse effect levels unless liver cell death, liver cell degenerative biliary/oval cell growth, degeneration, scarring, or bile build up, or other liver cell damage were also present.^{1,30}

Standard Selection

DHS recommends an enforcement standard of 300 ng/L for HFPO-DA.

There are no federal numbers and no state drinking water standard for HFPO-DA. Additionally, the EPA has not established a cancer slope factor for HFPO-DA. However, the EPA currently has draft subchronic and chronic oral reference doses for HFPO-DA.³

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information**

We identified several studies evaluating the toxicity of HFPO-DA. To calculate the ADI as specified in s. 160.13, Wisc. Statute, we selected a 2010 study by DuPont (18405-1037) as the critical study.⁶ We choose this study because it showed liver cell death that was similar to other studies, but at lower concentrations of HFPO-DA. Additionally, this study is used by the EPA to establish their draft oral

reference doses.³ We selected a NOAEL of 0.1 mg/kg-d based on increased liver cell death at higher concentrations. These effects on liver cell death from HFPO-DA exposure are also consistent with those caused by other PFAS in research studies and met to criteria to be considered adverse health effects.^{1,2,30}

As described above, we selected an ADI of 30 ng/kg-d from this NOAEL and a total uncertainty factor of 3000. To determine the recommended enforcement standard, DHS used the ADI, and, as required by Ch. 160, Wis. Stats., a body weight of 10 kg, a water consumption rate of 1 L/d, and a relative source contribution of 100%.

DHS recommends a preventive action limit of 30 ng/L for HFPO-DA.

DHS recommends that the preventive action limit for HFPO-DA be set at 10% of the enforcement standard because HFPO-DA has been shown to cause carcinogenic and mutagenic effects in research animals and cell culture studies.^{3,4,15} HFPO-DA has not been shown to cause teratogenic or interactive effects in people, research animals, or cell culture studies.

Prepared by Gavin Dehnert, Ph.D. and Sarah Yang, Ph.D.

Wisconsin Department of Health Services

References

1. ATSDR. Toxicological Profile for Perfluoroalkyls - Draft for Public Comment. In: Registry AftSaD, ed. Atlanta, GA2017.
2. DHS. *Recommended Public Health Groundwater Quality Standards: Scientific Support Documents for Cycle 10 Substances*. 2019.
3. USEPA. Human Health Toxicity Values for Hexafluoropropylene Oxide (HFPO) Dimer Acid and Its Ammonium Salt (CASRN 13252-13-6 and CASRN 62037-80-3). 2018.
4. Coperchini F, Croce L, Denegri M, et al. Adverse effects of in vitro GenX exposure on rat thyroid cell viability, DNA integrity and thyroid-related genes expression. *Environmental pollution (Barking, Essex : 1987)*. 2020;264:114778.
5. Cahoon LB. GenX Contamination of the Cape Fear River, North Carolina: Analytical Environmental Chemistry Uncovers Multiple System Failures. In: Ahuja S, ed. *Evaluating Water Quality to Prevent Future Disasters, Vol 11*. Vol 11.2019:341-354.
6. DuPont. *An Oral (Gavage) Reproduction/Developmental Toxicity Screening Study of H-28548 in Mice*. 2010. 18405-1037.

7. Rushing BR, Hu Q, Franklin JN, et al. Evaluation of the immunomodulatory effects of 2,3,3,3-tetrafluoro-2-(heptafluoropropoxy)-propanoate in C57BL/6 mice. *Toxicological sciences : an official journal of the Society of Toxicology*. 2017.
8. Caverly Rae JM, Craig L, Slone TW, Frame SR, Buxton LW, Kennedy GL. Evaluation of chronic toxicity and carcinogenicity of ammonium 2,3,3,3-tetrafluoro-2-(heptafluoropropoxy)-propanoate in Sprague-Dawley rats. *Toxicology reports*. 2015;2:939-949.
9. DuPont. A 28-DAY ORAL (GAVAGE) TOXICITY STUDY OF H-28397 IN RATS WITH A 28-DAY RECOVERY. 2008. DuPont-24447.
10. DuPont. A 28-DAY ORAL (GAVAGE) TOXICITY STUDY OF H-28397 IN MICE WITH A 28-DAY RECOVERY. 2008. DuPont-24459.
11. DuPont. A 90-DAY ORAL (GAVAGE) TOXICITY STUDY OF H-28548 IN RATS WITH A 28-DAY RECOVERY. 2009. DuPont-17751-1026.
12. DuPont. H-28548: Subchronic Toxicity 90-Day Gavage Study in Mice. 2010. DuPont-18405-1307.
13. DuPont. AN ORAL (GAVAGE) PRENATAL DEVELOPMENTAL TOXICITY STUDY OF H-28548 IN RATS. 2010. 18405-841.
14. Blake BE, Cope HA, Hall SM, et al. Evaluation of Maternal, Embryo, and Placental Effects in CD-1 Mice following Gestational Exposure to Perfluorooctanoic Acid (PFOA) or Hexafluoropropylene Oxide Dimer Acid (HFPO-DA or GenX). *Environmental health perspectives*. 2020;128(2):27006.
15. Cannon RE, Richards AC, Trexler AW, et al. Effect of GenX on P-Glycoprotein, Breast Cancer Resistance Protein, and Multidrug Resistance-Associated Protein 2 at the Blood-Brain Barrier. *Environmental health perspectives*. 2020;128(3):37002.
16. Chappell GA, Thompson CM, Wolf JC, Cullen JM, Klaunig JE, Haws LC. Assessment of the Mode of Action Underlying the Effects of GenX in Mouse Liver and Implications for Assessing Human Health Risks. *Toxicologic pathology*. 2020;48(3):494-508.
17. Conley JM, Lambright CS, Evans N, et al. Adverse Maternal, Fetal, and Postnatal Effects of Hexafluoropropylene Oxide Dimer Acid (GenX) from Oral Gestational Exposure in Sprague-Dawley Rats. *Environmental health perspectives*. 2019;127(3).
18. DuPont. H-28548: COMBINED CHRONIC TOXICITY/ONCOGENICITY STUDY 2-YEAR ORAL GAVAGE STUDY IN RATS. 2013. 18405-1238.
19. Berendsen BJA, Lakraoui F, Leenders L, van Leeuwen SPJ. The analysis of perfluoroalkyl substances at ppt level in milk and egg using UHPLC-MS/MS. *Food additives & contaminants Part A, Chemistry, analysis, control, exposure & risk assessment*. 2020:1-12.
20. Brandsma SH, Koekkoek JC, van Velzen MJM, de Boer J. The PFOA substitute GenX detected in the environment near a fluoropolymer manufacturing plant in the Netherlands. *Chemosphere*. 2019;220:493-500.

21. Guillette TC, McCord J, Guillette M, et al. Elevated levels of per- and polyfluoroalkyl substances in Cape Fear River Striped Bass (*Morone saxatilis*) are associated with biomarkers of altered immune and liver function. *Environment international*. 2020;136:105358.
22. Munoz G, Liu JX, Duy SV, Sauve S. Analysis of F-53B, Gen-X, ADONA, and emerging fluoroalkylether substances in environmental and biomonitoring samples: A review. *Trends in Environmental Analytical Chemistry*. 2019;23.
23. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 140*2017.
24. USEPA. National Primary Drinking Water Regulations. <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>. Published 2018. Accessed.
25. USEPA. Drinking Water Contaminant Human Health Effects Information. <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>. Published 2019. Accessed.
26. USEPA. IRIS Assessments. https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm. Published 2019. Accessed.
27. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 809*2018.
28. IARC. List of Classification, Volumes 1-123. <https://monographs.iarc.fr/list-of-classifications-volumes/>. Published 2018. Accessed May 17, 2019.
29. Ritter L, Totman C, Krishnan K, Carrier R, Vezina A, Morisset V. Deriving uncertainty factors for threshold chemical contaminants in drinking water. *Journal of toxicology and environmental health Part B, Critical reviews*. 2007;10(7):527-557.
30. Hall AP, Elcombe CR, Foster JR, et al. Liver hypertrophy: a review of adaptive (adverse and non-adverse) changes--conclusions from the 3rd International ESTP Expert Workshop. *Toxicologic pathology*. 2012;40(7):971-994.
31. Andersen LG, Holst C, Michaelsen KF, Baker JL, Sorensen TI. Weight and weight gain during early infancy predict childhood obesity: a case-cohort study. *International journal of obesity (2005)*. 2012;36(10):1306-1311.
32. Sharma D, Shastri S, Sharma P. Intrauterine Growth Restriction: Antenatal and Postnatal Aspects. *Clinical medicine insights Pediatrics*. 2016;10:67-83.
33. Barker DJ. The developmental origins of adult disease. *Journal of the American College of Nutrition*. 2004;23(6 Suppl):588s-595s.
34. National Heart L, and Blood Institute. Blood Tests. <https://www.nhlbi.nih.gov/health-topics/blood-tests>. Accessed 4/9/2020.
35. Wolf CJ, Takacs ML, Schmid JE, Lau C, Abbott BD. Activation of mouse and human peroxisome proliferator-activated receptor alpha by perfluoroalkyl acids of different functional groups and

chain lengths. *Toxicological sciences : an official journal of the Society of Toxicology*. 2008;106(1):162-171.

36. Cheng X, Klaassen CD. Critical role of PPAR-alpha in perfluorooctanoic acid- and perfluorodecanoic acid-induced downregulation of Oatp uptake transporters in mouse livers. *Toxicological sciences : an official journal of the Society of Toxicology*. 2008;106(1):37-45.
37. Cheng X, Klaassen CD. Perfluorocarboxylic acids induce cytochrome P450 enzymes in mouse liver through activation of PPAR-alpha and CAR transcription factors. *Toxicological sciences : an official journal of the Society of Toxicology*. 2008;106(1):29-36.

Appendix A. Toxicity Data

Table A-I. HFPO-DA Toxicity Studies from Literature Review

Study Type	Species	Duration	Doses (mg/kg-d)	Route	Endpoints	Toxicity Value (mg/kg-d)	Reference
Development	Mice	Day 1 to 17 of gestation	0, 2, 10	Gavage	HFPO-DA increased liver weight in a dose-dependent manner starting at 2 mg/kg-d in offspring. HFPO increased kidney weight in a dose-dependent manner starting at 10 mg/kg-d in offspring. HFPO-DA increased cholesterol at 2 mg/kg-d in offspring. HFPO-DA reduced embryo/placental weight ratios at 10 mg/kg-d in female mice.	NOAEL: N/A LOAEL: 2.0	Blake et al, 2020 ¹⁴
Maternal exposure throughout development	Rat	Day 14-18 of gestation	0, 1, 3, 10, 30, 62.5, 125, 250, 500	Gavage	HFPO-DA decreased maternal weight gain in a dose-dependent manner starting at 250 mg/kg-d and increased maternal liver weight in a dose-dependent manner starting at 62.5 mg/kg-d. HFPO-DA decreased liver weight in a dose-dependent manner starting at 62.5 mg/kg-d in the offspring. HFPO-DA decreased total T3 in a dose-dependent manner starting at 30 mg/kg/d and decreased total T4 in a dose-dependent manner starting at 125 mg/kg-d in the offspring. HFPO-DA decreased cholesterol in a dose-dependent manner starting at 250 mg/kg-d. HFPO-DA decreased ano-genital distance at 125 mg/kg-d for female offspring. HFPO-DA decreased testis weight at 125 mg/kg-d for male offspring.	NOAEL: 10 LOAEL: 30	Conley et al., 2020 ¹⁷

Short term	Rats	28 Days	0.3, 3, 30	Gavage	HFPO-DA decreased red blood cells, hemoglobin, hematocrit, and immature red blood cell levels. HFPO-DA increased liver weights and liver cell size HFPO-DA increased albumin and glucose levels and decreased cholesterol and triglycerides levels. HFPO-DA increased oxidation and enzyme activity.	NOAEL: N/A LOAEL: 0.3	DuPont, 2008a (study number - 24447) ⁹
Short Term	Mice	28 Days	0.1, 3, 30	Gavage	HFPO-DA increased body weight and food consumption. HFPO-DA decreased red blood cells, hemoglobin, and hematocrit levels. HFPO-DA increased liver cell death. HFPO-DA increased liver and kidney weight.	NOAEL: 0.1 LOAEL: 3	DuPont, 2008b (study number- 24459) ¹⁰
Sub-chronic	Rats	90 Days	0.1, 10, 100 males 10, 100, 1000 females	Gavage	HFPO-DA decreased red blood cells, hemoglobin, and hematocrit levels HFPO-DA increased liver and kidney weight. At high concentrations HFPO-DA exposure lead to death.	Males: NOAEL: 0.1 LOAEL: 10 Females: NOAEL: N/A LOAEL: 10	DuPont, 2009 (study number- 17751- 1026) ¹¹
Sub-chronic	Mice	95-96 Days	0.1, 0.5 5	Gavage	Exposure to HFPO-DA increased body weight. HFPO-DA increased liver toxicity markers. HFPO-DA increased liver weight.	Males: NOAEL: 0.1 LOAEL: 0.5 Females: NOAEL: 0.5 LOAEL: 5	DuPont, 2010a (study number - 18405- 1307) ¹²
Developmental	Rats	84 to 85 days for males 14 days before impregnation and through gestational day 20 by gavage for females	0.1, 1, 5	Gavage	HFPO-DA can increase body weight, increase liver and kidney weight, increase liver cell death, and decrease offspring body weight and body weight gain	Males: NOAEL: 0.1 LOAEL: 1 Females: NOAEL: 0.1 LOAEL: 1 Offspring: NOAEL: 1 LOAEL: 5	DuPont, 2010b (study number - 18405- 1037) ⁶

Developmental	Rats	Gestational day 6 through 20	10,100, 1000	Gavage	HFPO-DA decreased liver, kidney, and uterine weights in pregnant females. HFPO-DA decreased offspring birthweights. HFPO-DA caused delivery of the offspring early.	Females: NOAEL: 10 LOAEL: 100 Offspring: NOAEL: 10 LOAEL: 100	DuPont, 2010c (study number - 18405-841) ¹³
Chronic	Rats	2 years	0.1, 1, 50 for males 1, 50, 500 females	Gavage	HFPO-DA decreased body weight. HFPO-DA decreased hemoglobin and hematocrit levels. HFPO-DA increased liver toxicity indicators. HFPO-DA increased the incidence of pancreatic tumors and cancer and increased the incidence of liver cancer.	Males: NOAEL: 1 LOAEL: 50 Females: NOAEL: 50 LOAEL: 500	Rae et al. 2015 ^{8,18}
Short term	Mice	28 days	1, 10, 100	Gavage	HFPO-DA decreased relative liver weight. Gen X decreased T cell-dependent antibody responses (TDAR). HFPO-DA increased T lymphocyte numbers.	NOAEL: 1 LOAEL: 10	Rushing et al., 2017 ⁷

Table A-2. Critical Study Selection for HFPO-DA

Reference	Appropriate duration?	Effects consistent with other studies?	Effects relevant to humans?	Number of Doses	Toxicity value identifiable?	Critical study?
Blake et al., 2020 ¹⁴	✓	✓	✓	2	✓	Yes
Conley et al., 2020 ¹⁷	✓	✓	✓	8	✓	Yes
DuPont, 2008a ⁹	✓	✓	✓	3	✓	Yes
DuPont, 2008b ¹⁰	✓	✓	✓	3	✓	Yes
DuPont, 2009 ¹¹	✓	✓	✓	4	✓	Yes
DuPont, 2010a ¹²	✓	✓	✓	3	✓	Yes
DuPont, 2010b ⁶	✓	✓	✓	3	✓	Yes
DuPont, 2010c ¹³	✓	✓	✓	3	✓	Yes
Rae et al., 2015 ^{8,18}	✓	✓	✓	4	✓	Yes
Rushing et al., 2017 ⁷	✓	✓	✓	3	✓	Yes

To be considered a critical study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.

PFODA | 2020

Substance Overview

Perfluorooctadecanoic acid (PFODA) is a chemical in a group of contaminants called per- and polyfluoroalkyl substances (PFAS). PFODA can be found as an impurity in stain repellants in commercial products like carpet and fabric, as a coating for packaging, or as an ingredient in fire-fighting foam.^{1,2} PFAS, like PFODA, can persist in the environment and in the human body for long periods of time.¹

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard for PFODA. DHS recommends an enforcement standard of 400 micrograms per liter (µg/L) for PFODA. The recommended standard is based on a study that found that PFODA can significantly decrease body weight gain in pregnant rats and their offspring.³

DHS recommends that the preventive action limit for PFODA be set at 20% of the enforcement standard because PFODA has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture.

Current Standards

Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

Recommended Standards

Enforcement Standard:	400 µg/L
Preventive Action Limit:	80 µg/L

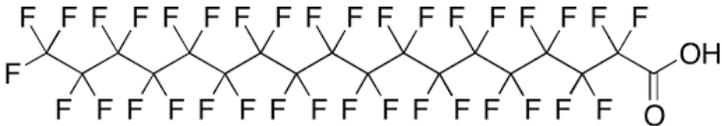
Health Effects

At this time, no studies have evaluated the effects of PFODA among people and data on the effects of PFODA on research animals are limited. A study in rats has shown that high levels of PFODA can affect the blood, cause damage to the liver, pancreas, and spleen, reduce body weight gain in offspring, and alter the weights of the liver, heart, and brain.³

PFODA has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture.^{1,4,a} The EPA has not evaluated the carcinogenicity of PFODA.⁵

a Carcinogenic effects means the substance can cause cancer; mutagenic effects means the substance can cause DNA damage; teratogenic means the substance can cause birth defects; and interactive effects mean the substance can affect the toxicity of another substance or its toxicity can be affected by another substance.

Chemical Profile

PFODA	
Structure:	
CAS Number:	16517-11-6
Formula:	C ₁₈ HF ₃₅ O ₂
Molar Mass:	914.1 g/mol
Synonyms:	Pentanoic acid, 2,2,3,3,4,4,5,5,5-nonafluoro-; Undecafluorohexanoic acid; Perfluorovaleric acid; PFOcDA

Exposure Routes

PFAS, including PFODA, can be released directly into the environment during the manufacture and use of PFAS and can be found in water or soil. PFAS can move between groundwater and surface water. Once in water, PFAS can travel long distances.^{1,2}

People can be exposed to PFAS by drinking water, eating food, and breathing in or accidentally swallowing soil or dust containing PFAS.¹

Current Standard

Wisconsin does not currently have groundwater standards for PFODA.⁶

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

EPA Oral Reference Dose:	N/A
--------------------------	-----

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

None Available

Literature Search

Literature Search Dates:	Up to 2020
Key studies found?	Yes
Critical studies identified?	Yes

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for PFODA.⁷

Health Advisory

The EPA has not established health advisories for PFODA.⁸

Drinking Water Concentration (Cancer Risk)

The EPA has not established drinking water concentrations based on a cancer risk level determination for PFODA.⁵

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for PFODA.⁹

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Information System (IRIS) program.

EPA Oral Reference Dose

The EPA does not have an oral reference dose for PFODA.⁵

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If we determine that something is carcinogenic and there is no federal number or ADI from the EPA, DHS must set the standard at a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of PFODA, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of PFODA. If so, we look to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA and International Agency for Research on Cancer (IARC) have not evaluated the carcinogenicity of PFODA.^{5,10}

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for PFODA.⁵

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information that was not considered when the value was established and that indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For PFODA, we searched for values that had been published on or before August 2020. While the Agency for Toxic Substances and Disease Registry (ATSDR) reviewed the toxicity of 14 PFAS compounds in 2018, they did not review nor establish any guidance values for PFODA.¹

Literature Search

Our literature review focused on relevant scientific literature on the health effects of PFODA published on or before August 2020. We looked for studies related to PFODA toxicity or effects on a disease state in which information on exposure or dose was included as part of the study.^b Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

Fifty-three studies were returned by search engines. We excluded studies on non-mammalian or cell systems, non-oral exposure routes, and those that did not evaluate health risks from further review. After applying these exclusion criteria, we located one key toxicity study on PFODA (summarized in Table A-1). To be considered a critical toxicity study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.^{c-d}

^b We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

- Title/abstract: PFODA OR 2706-90-3 OR "Perfluorooctadecanoic acid"
Subject area: toxicology
Language: English

We also searched online for toxicity studies published by national research programs.

^c Appropriate toxicity values include the no observable adverse effect level (NOAEL), lowest observable adverse effect level (LOAEL), and benchmark dose (BMD).

^d Due to the limited availability of data for this substance, we considered a study to be of an appropriate duration if animals were exposed for at least 28 days or if the exposure occurred during pregnancy. We accounted for differences in exposure duration through the use of an uncertainty factor.

After reviewing the key study, we determined that the key study met the criteria to be considered a critical toxicity study (see Table A-2).

Critical Toxicity Studies

To compare between results in the critical studies, we calculated an acceptable daily intake (ADI) for each study/effect. The ADI is the estimated amount of PFODA that a person can be exposed to every day and not experience health effects. Since there are no studies investigating the half-life in humans we calculated ADI by dividing a toxicity value from either a no-observed adverse effect level (NOAEL), lowest observed adverse effect level (LOAEL), or benchmark dose (BMD) identified in a study by a factor accounting for various sources of scientific uncertainty.^{e,12} Uncertainty factors were included, as appropriate, to account for differences between humans and animals, differences between healthy and sensitive human populations, using data from short-term experiments to protect against effects from long-term exposure, and using data where a health effect was observed to estimate the level that does not cause an effect. To ensure appropriate protection, we have chosen to not use studies that have significant uncertainty as the basis for the recommended enforcement standards.^f This approach is consistent with that taken by EPA when establishing oral reference doses.¹³

Hirata-Koizumi et al., 2012³

Hirata-Koizumi et al. exposed male and female rats to different concentrations of PFODA (0, 40, 200, and 1,000 milligrams of PFODA per kilogram body weight per day or mg/kg-d) by gavage for 42 days in males and from 14 days pre-mating through 5 days after lactation in females.³ A subset of males and females (without mating—dosed 42 days) in the 0 and 1,000 mg/kg-d dose groups were withheld treatment for an additional 14 day recovery period (see Hirata-Koizumi et al. for information on recovery group effects).

In males, PFODA affected body weight, food consumption, blood and biochemistry parameters, organ weights, and increased liver injury and cell death at the mid- and/or highest dose levels (Table 1).

e The NOAEL is the highest dose tested that did not cause an adverse effect, the LOAEL is the lowest dose tested that caused an adverse effect, and the BMD is an estimation of the dose that would cause a specific level of response (typically 5 or 10%).

f DHS considers a study to have significant uncertainty if the total uncertainty factors is equal to or greater than 3,000.

Table 1. Statistically Significant Effects Observed in Males in Hirata-Koizumi et al., 2012

Effects observed in males		Dose (mg/kg-d)		
		40	200	1000
Growth	Decreased body weight			✓
	Decreased food consumption			✓
Blood	Decreased red blood cell count		✓	✓
	Decreased hemoglobin level		✓	✓
	Decreased hematocrit		✓	✓
	Decreased reticulocyte ratio			✓
Biochemistry	Decreased total protein			✓
	Decreased in α_1 -globulin ratio			✓
	Increased γ -globulin ratio			✓
Heart	Decreased absolute heart weight			✓
Spleen	Decreased absolute spleen weight			✓
Thymus	Decreased absolute thymus weight			✓
Brain	Increased relative brain weights			✓
Kidney	Increased relative kidney weights			✓
	Increased BUN			✓
Testis	Increased relative testis weights			✓
Liver	Increased albumin ratio		✓	✓
	Increased ALT			✓
	Increased ALP			✓
	Increased total bilirubin			✓
	Increased absolute liver weight		✓	✓
	Increased relative liver weight		✓	✓
	Increased cellular size		✓	✓
	Increased cellular injury and death			✓
	Increased focal area of cell death		✓	✓
	Hemosiderin (excess iron) deposits			✓

From this repeated dose experiment in males, the researchers identified a no observable adverse effect level (NOAEL) of 40 mg/kg-d based on the effects on the liver. We determined a candidate ADI of 0.01 mg/k-d using this NOAEL and a total uncertainty factor of 3,000 to account for differences between humans and research animals (10), differences among people (10), using a short-term study to extrapolate to long-term exposures (3), and the limited availability of information (10).

In females, PFODA affected forelimb grip strength, body weight gain, food consumption, blood and blood chemistry, organ weights, increased cell injury and death in the liver, and increased cell injury in the pancreas and spleen at the mid- and/or high dose levels (Table 2).

Table 2. Statistically Significant Effects Observed in Females in Hirata-Koizumi et al., 2012

Effects observed in females		Dose (mg/kg-d)		
		40	200	1000
Strength	Decrease in forelimb grip strength			✓
Growth	Decreased body weight			✓
	Decreased food consumption		✓	✓
Blood	Increase in basophils (immune cells)			✓
	Reduced prothrombin time		✓	✓
	Prolongation of activated partial thromboplastin time			✓
Blood Chemistry	Decreased in α_1 -globulin ratio		✓	✓
	Increased γ -globulin ratio			✓
Liver	Increased absolute liver weight			✓
	Increased relative liver weights		✓	✓
	Increased centrilobular hepatocyte hypertrophy (liver			✓
	Increased focal necrosis (liver			✓
	Increased microgranuloma (liver			✓
	Increased ALP			✓
	Decrease in Total Cholesterol			✓
Heart	Decreased absolute heart weight			✓
Brain	Increased relative brain weight			✓
Kidney	Increased BUN			✓
Pancreas	Increased number of decreased pancreatic zymogen granules			✓
Spleen	Increased severity of hemosiderin deposit (spleen)			✓

From this repeated dose study in females, the researchers identified a NOAEL of 40 mg/kg-d based on effects on growth and damage to the pancreas and liver. We determined a candidate ADI of 0.04 mg/kg-d based on the NOAEL and a total uncertainty factor of 1,000 to account for differences between humans and research animals (10), differences among people (10), short term exposure in mating and pregnant rats (1), and the limited availability of information (10).

PFODA affected reproduction and development of offspring with decreases in growth, decreases in the number of endocrine glands (corpora lutea) that develop when eggs mature, decreased implantation, and a decrease in the number of live offspring (Table 3).

Table 3. Statistically Significant Effects Observed in Offspring in Hirata-Koizumi et al., 2012

Effects observed in offspring	Dose (mg/kg-d)		
	40	200	1000
Growth	Decreased body weight at birth		✓
	Decreased body weight gain of offspring		✓
Viability	Decreased corpora lutea		✓
	Decreased implantation sites		✓
Survivability	Decreased total number of offspring born		✓
	Decreased number of live offspring		✓
	Stillborn		✓

From this reproductive/development study, the researchers identified a NOAEL of 200 mg/kg-d based on decreased growth rates, decreased viability, and decreased survivability of offspring. We determined a candidate ADI of 0.2 mg/kg-d based on the NOAEL and a total uncertainty factor of 1,000 to account for differences between humans and research animals (10), differences among people (10), and the limited availability of information (10).

Key health effects

In our literature search, we did not find studies that suggest that PFODA can cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell cultures. The EPA has not evaluated the carcinogenicity of PFODA.

Discussion

PFODA has been shown to impact the liver, spleen, and pancreas, decrease maternal body weight and the body weight of offspring during development, and alter a variety of blood and biochemistry parameters in research animals. These findings are consistent with those seen in other long-chain PFAS.^{1,14,15 16,17}

Decreased body weight gain in pregnant females and in offspring is consistent with findings in other PFAS toxicity studies in research animals. The evidence in animals suggests that PFODA may have consistent effects to other long-chained PFAS, such as PFHxS, PFOA, and PFOS, including reduced litter size and decreased offspring birth weight.¹

A number of studies have demonstrated that liver effects caused by PFAS occur primarily through activation of the nuclear hormone receptor, PPAR α (peroxisome proliferator-activated receptor

alpha).¹⁸⁻²² Nuclear receptors regulate gene expression and PPAR α regulates the expression of genes involved in lipid and cholesterol metabolism.²³ While PPAR α receptors are found in rodents and humans, levels of these receptors are much higher in rats and mice than in monkeys and humans.²³ This means that effects caused by activation of PPAR α are more likely to occur in rodents than they are to occur in people. As such, the effects on the liver observed in the critical study reviewed here are likely not relevant to humans.

Standard Selection

DHS recommends an enforcement standard of 400 $\mu\text{g/L}$ PFODA.

There are no federal numbers and no state drinking water standard for PFODA. Additionally, the EPA has not established a cancer slope factor or ADI (oral reference dose) for PFODA.

However, we found one critical study evaluating the toxicity of PFODA. To calculate the ADI as specified by Ch. 160.13, Wisc, Statute, DHS selected the 2012 study by Hirata-Koizumi et al. as the critical study.³ We selected this study because it evaluated the effects of PFODA exposure during reproductive and development phase in maternal and offspring endpoints.^{24,25} Toxicity studies in animals continue to show that developmental endpoints are critical effects for PFAS, including PFOSA. We chose the critical effects as decreased maternal body weight and food consumption and damage to the pancreas and liver.³

As described above, we obtained an ADI of 0.04 mg/kg-d from a NOAEL of 40 mg/kg-d and a total uncertainty factor of 1000 for pregnant females. This ADI has a lower uncertainty factor compared to males (3000) and uses a lower NOAEL compared to offspring (200 mg/kg-d) based on the lower body weight gain, pancreas, and liver effects observed in pregnant females.³ To determine the recommended enforcement standard, DHS used the ADI, and, as required by Ch. 160, Wis. Stats., a body weight of 10 kg, a water consumption rate of 1 L/d, and a relative source contribution of 100%.

DHS recommends a preventive action limit of 80 $\mu\text{g/L}$ PFODA.

DHS recommends that the preventive action limit for PFODA be set at 20% of the enforcement standard because PFODA has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture.

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information

Prepared by Brita Kilburg-Basnyat, Ph.D. and Sarah Yang, Ph.D.

Wisconsin Department of Health Services

References

1. ATSDR. Toxicological Profile for Perfluoroalkyls - Draft for Public Comment. In: Registry AfTsaD, ed. Atlanta, GA2017.
2. ITRC. Environmental Fate and Transport for Per- and Polyfluoroalkyl Substances. In: Council ITR, ed2018.
3. Hirata-Koizumi M, Fujii S, Furukawa M, Ono A, Hirose A. Repeated dose and reproductive/developmental toxicity of perfluorooctadecanoic acid in rats. *J Toxicol Sci*. 2012;37(1):63-79.
4. Buhrike T, Kibellus A, Lampen A. In vitro toxicological characterization of perfluorinated carboxylic acids with different carbon chain lengths. *Toxicol Lett*. 2013;218(2):97-104.
5. USEPA. IRIS Assessments. https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm. Published 2019. Accessed.
6. WIDNR. Groundwater Quality. In: Resources WDoN, ed. *Chapter NR 140*2017.
7. USEPA. National Primary Drinking Water Regulations. <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>. Published 2018. Accessed.
8. USEPA. Drinking Water Contaminant Human Health Effects Information. <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>. Published 2019. Accessed.
9. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 809*2018.
10. IARC. List of Classification, Volumes 1-123. <https://monographs.iarc.fr/list-of-classifications-volumes/>. Published 2018. Accessed May 17, 2019.
11. Ritter L, Totman C, Krishnan K, Carrier R, Vezina A, Morisset V. Deriving uncertainty factors for threshold chemical contaminants in drinking water. *Journal of toxicology and environmental health Part B, Critical reviews*. 2007;10(7):527-557.
12. Zhang Y, Beeson S, Zhu L, Martin JW. Biomonitoring of perfluoroalkyl acids in human urine and estimates of biological half-life. *Environ Sci Technol*. 2013;47(18):10619-10627.
13. USEPA. A Review of the Reference Dose and Reference Concentration Processes. 2002(EPA/630/P-02/002F).

14. Takahashi M, Ishida S, Hirata-Koizumi M, Ono A, Hirose A. Repeated dose and reproductive/developmental toxicity of perfluoroundecanoic acid in rats. *J Toxicol Sci.* 2014;39(1):97-108.
15. (NTP) NTP. NTP technical report on the toxicity studies of perfluoroalkyl carboxylates (perfluorohexanoic acid, perfluorooctanoic acid, perfluorononanoic acid, and perfluorodecanoic acid) administered by gavage to Sprague Dawley (Hsd:Sprague Dawley SD) rats. In. Vol Toxicity Report 97. Research Triangle Park, NC: National Toxicology Program 2019.
16. ITRC. PFAS Fact Sheets: PFAS Water and Soil Values Table Excel File. In: Council ITR, ed2020.
17. ITRC. Human and Ecological Health Effects of select PFAS. In: Council ITR, ed2020.
18. Das KP, Wood CR, Lin MT, et al. Perfluoroalkyl acids-induced liver steatosis: Effects on genes controlling lipid homeostasis. *Toxicology.* 2017;378:37-52.
19. Rosen MB, Das KP, Rooney J, Abbott B, Lau C, Corton JC. PPAR alpha-independent transcriptional targets of perfluoroalkyl acids revealed by transcript profiling. *Toxicology.* 2017;387:95-107.
20. Albrecht PP, Torsell NE, Krishnan P, et al. A species difference in the peroxisome proliferator-activated receptor alpha-dependent response to the developmental effects of perfluorooctanoic acid. *Toxicol Sci.* 2013;131(2):568-582.
21. Palkar PS, Anderson CR, Ferry CH, Gonzalez FJ, Peters JM. Effect of prenatal peroxisome proliferator-activated receptor alpha (PPARalpha) agonism on postnatal development. *Toxicology.* 2010;276(1):79-84.
22. Wolf DC, Moore T, Abbott BD, et al. Comparative hepatic effects of perfluorooctanoic acid and WY 14,643 in PPAR-alpha knockout and wild-type mice. *Toxicol Pathol.* 2008;36(4):632-639.
23. Hall AP, Elcombe CR, Foster JR, et al. Liver hypertrophy: a review of adaptive (adverse and non-adverse) changes--conclusions from the 3rd International ESTP Expert Workshop. *Toxicol Pathol.* 2012;40(7):971-994.
24. Harris MW, Birnbaum LS. Developmental toxicity of perfluorodecanoic acid in C57BL/6N mice. *Fundamental and applied toxicology : official journal of the Society of Toxicology.* 1989;12(3):442-448.
25. Harris MW, Uraih LC, Birnbaum LS. Acute toxicity of perfluorodecanoic acid in C57BL/6 mice differs from 2,3,7,8-tetrachlorodibenzo-p-dioxin. *Fundamental and applied toxicology : official journal of the Society of Toxicology.* 1989;13(4):723-736.

Appendix A. Toxicity Data

Table A-I. PFODA Toxicity Studies from Literature Review

Study Type	Species	Duration	Doses (mg/kg-d)	Route	Endpoints	Toxicity Value (mg/kg-d)	Reference
Long-term	Male rats	42 days	0, 40, 200, 1000	Oral gavage	Decreased body weight and food consumption, decreased blood and blood biochemistry parameters (including liver indicators ALT, ALP, total bilirubin), organ weight changes, and increased cell injury and cell death in the liver.	NOAEL: 40	Hirata-Koizumi et al., 2012 ³
Long-term	Pregnant rats	~42 days	0, 40, 200, 1000	Oral gavage	Decreased forelimb grip strength, decreased body weight and food consumption, decreased blood and blood chemistry parameters, increased incidence of moribund condition, increased cell injury/cell death in the liver, increased organ weights, increased cell damage in the pancreas and spleen.	NOAEL:40	Hirata-Koizumi et al., 2012 ³
Reproductive/developmental	Female Rats and Offspring	~ 42 days	0, 40, 200, 1000	Oral Gavage	Decreased body weight at birth, decreased body weight gain, decreased corpora lutea and implantation site, decreased total number of offspring born and live offspring.	NOAEL: 200	Hirata-Koizumi et al., 2012 ³
NOAEL: No observed adverse effect level							

Table A-3. Critical Study Selection for PFODA

Reference	Appropriate duration?	Effects consistent with other studies?	Effects relevant to humans?	Number of Doses	Toxicity value identifiable?	Critical study?
Hirata-Koizumi et al, 2012 (Males)	✓	✓	✓	3	✓	Yes
Hirata-Koizumi et al, 2012 (Pregnant rats)	✓	✓	✓	3	✓	Yes
Hirata-Koizumi et al, 2012 (Females and offspring)	✓	✓	✓	3	✓	Yes

To be considered a critical study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.

DONA | 2020

Substance Overview

4,8-Dioxa-3H-perfluorononanoic acid^a (DONA) is a chemical in a group of contaminants called per- and polyfluoroalkyl substances (PFAS). DONA is an emulsifier (stabilizer) used to create high performance fluoropolymer chemicals.¹ DONA was developed to replace longer chained PFAS such as perfluorooctanoic acid (PFOA), a chemical with known human health impacts.¹⁻⁴ Because of its chemical properties, DONA can be found as an impurity in stain repellants, as a coating for packaging, and in non-stick coatings on cookware and other food contact surfaces.^{2,4}

Recommendations

Wisconsin does not currently have an NR140 Groundwater Quality Public Health Enforcement Standard for DONA. DHS recommends an enforcement standard of 3 micrograms per liter (µg/L) for DONA. The recommended standard is based on a study that found DONA exposure can decrease red blood cell, hemoglobin, and hematocrit levels and cause liver toxicity in male rats.³

DHS recommends that the preventive action limit for DONA be set at 20% of the enforcement standard because DONA has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people or research animals.

Current Standards	
Enforcement Standard:	N/A
Preventive Action Limit:	N/A
Year:	N/A

Recommended Standards	
Enforcement Standard:	3 µg/L
Preventive Action Limit:	0.6 µg/L

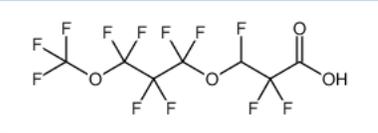
Health Effects

Data on the health effects of DONA exposure are limited. There are no studies that have evaluated the effects of DONA among people. The studies that have evaluated the effects of DONA in research animals have shown that DONA can decrease body weight, red blood cell counts, and cholesterol levels and increase glucose levels and liver and kidney weight.^{1,3}

DONA has not been shown to cause carcinogenic (cancer), mutagenic (DNA damage), teratogenic (birth defects), or interactive effects in people, research animals, or cell culture studies. Additionally, the EPA has not evaluated the carcinogenicity of DONA.

^a This scientific support document and the included groundwater standard recommendations also apply to anion salts of 4,8-Dioxa-3H-perfluorononanoic acid.

Chemical Profile

DONA	
Structure:	
CAS Number:	919005-14-4
Formula:	C ₇ H ₂ F ₁₂ O ₄
Molar Mass:	378.07 g/mol
Synonyms:	Propanoic acid, 2,2,3-trifluoro-3- [1,1,2,2,3,3-hexafluoro-3- (trifluoromethoxy)propoxy]- SCHEMBL1681505 DTXSID40881350

Exposure Routes

People can be exposed to DONA by drinking contaminated water, swallowing contaminated soil, eating fish that come from contaminated waterways, eating food that was packaged in material that contains DONA, and breathing in or swallowing dust that contains DONA.^{1,5} Babies born to mothers exposed to PFAS, like DONA, can themselves be exposed to PFAS during pregnancy and breastfeeding and one recent study has detected DONA in human breast milk.^{2,6}

In the environment, DONA can be found in water or soil as an impurity from the use of other PFAS in manufacturing and consumer products.¹ PFAS, like DONA, can move between groundwater and surface water. Once in groundwater, PFAS, like DONA, can travel long distances.^{2,7,8}

Current Standard

Wisconsin does not currently have groundwater standards for DONA.⁹

Standard Development

Federal Numbers

Maximum Contaminant Level:	N/A
Health Advisory:	N/A
Drinking Water Concentration (Cancer Risk):	N/A

State Drinking Water Standard

NR 809 Maximum Contaminant Level:	N/A
-----------------------------------	-----

Acceptable Daily Intake

EPA Oral Reference Dose:	N/A
--------------------------	-----

Oncogenic Potential

EPA Cancer Slope Factor:	N/A
--------------------------	-----

Guidance Values

ATSDR Oral Minimum Risk Level:	N/A
--------------------------------	-----

Literature Search

Literature Search Dates:	Up to 2020
Key studies found?	Yes
Critical studies identified?	Yes

Federal Numbers

Chapter 160, Wis. Stats., requires that DHS use the most recent federal number as the recommended enforcement standard unless one does not exist or there is significant technical information that was not considered when the federal number was established and that indicates a different number should be used.

Maximum Contaminant Level

The EPA does not have a maximum contaminant level for DONA.¹⁰

Health Advisory

The EPA has not established health advisories for DONA.¹¹

Drinking Water Concentration (Cancer Risk)

The EPA has not established drinking water concentration based on a cancer risk level determination for DONA.¹²

State Drinking Water Standard

Chapter 160, Wis. Stats., requires that DHS use a state drinking water standard as the recommended enforcement standard if there are no federal numbers and a state drinking water standard is available.

NR 809 Maximum Contaminant Level

Wisconsin does not have a drinking water standard for DONA.⁹

Acceptable Daily Intake

If a federal number and a state drinking water standard are not available, ch. 160, Wis. Stats., requires that DHS use an acceptable daily intake (ADI) from the EPA to develop the recommendation. Statute allows DHS to recommend a different value if an ADI from the EPA does not exist or if there is significant technical information that is scientifically valid, was not considered when the federal ADI was set, and indicates a different number should be used. The EPA provides ADIs, termed oral reference doses, as part of a health advisory, human health risk assessment for pesticides, or for use by the Integrated Risk Information System (IRIS) program.

EPA Oral Reference Dose

The EPA does not have an oral reference dose for DONA.¹²

Oncogenic Potential

Chapter 160, Wis. Stats., requires that DHS evaluate the oncogenic (cancer-causing; carcinogenic) potential of a substance when establishing the groundwater standard. If we determine that something is carcinogenic and there is no federal number or ADI from the EPA, DHS must set the standard at a level that would result in a cancer risk equivalent to 1 case of cancer in 1,000,000 people. DHS must also set the standard at this level if the EPA has an ADI but using it to set the groundwater standard would result in a cancer risk that is greater than 1 in 1,000,000.

To evaluate the oncogenic potential of DONA, we looked to see if the EPA, the International Agency for Research on Cancer (IARC), or another agency has classified the cancer potential of DONA. If so, we look to see if EPA or another agency has established a cancer slope factor.

Cancer Classification

The EPA and International Agency for Research on Cancer (IARC) have not evaluated the carcinogenicity of DONA.^{12,13}

EPA Cancer Slope Factor

The EPA has not established a cancer slope factor for DONA.¹²

Additional Technical Information

Chapter 160, Wis. Stats., allows DHS to recommend a value other than a federal number or ADI from the EPA if there is significant technical information that was not considered when the value was established and indicates a different value is more appropriate.

To ensure the recommended groundwater standards are based on the most appropriate scientific information, we search for relevant health-based guidance values from national and international agencies and for relevant data from the scientific literature.

Guidance Values

For DONA, we searched for values that been published on or before September 2020. We did not find any additional guidance values.

Literature Search

Our literature review focused on relevant scientific literature on the health effects of DONA published on or before September 2020. Our literature search also focused on health effects of the anion salts of 4,8-Dioxa-3H-perfluorononanoic acid, such as ammonium 4,8-dioxa-3H-perfluorononanoate or ADONA. We looked for studies related to DONA toxicity or effects on a disease state in which information on exposure or dose was included as part of the study.^b Ideally, relevant studies used *in vivo* (whole animal) models and provided data for multiple doses over an exposure duration proportional to the lifetime of humans.

Approximately ten toxicity studies were returned by the search engines. We excluded studies on non-mammalian or cell systems, non-oral exposure routes, and those that did not evaluate health risks from further review. After applying these exclusion criteria, we located one key toxicity study on DONA (summarized in Table A-1). To be considered a critical toxicity study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable

^b We used the National Institutes of Health's PubMed resource and Clarivate Analytics' Web of Science resource for this search. We used the following search terms in the literature review:

Title/abstract: DONA or "4,8-Dioxa-3H-perfluorononanoic acid" or ADONA or "ammonium 4,8-dioxa-3H-perfluorononanoate" or "4,8-dioxa-3H-perfluorononanoate"

Language: English

We also searched online for toxicity studies published by national research programs.

toxicity value.^{c-d} The one key study met the criteria to be considered a critical toxicity study (see Table A-2).

Critical Toxicity Studies

To compare between results from recently found studies and the study used to set the current enforcement standard, we calculated an acceptable daily intake (ADI) for each study/effect. The ADI is the estimated amount of DONA that a person can be exposed to every day and not experience health impacts. As such, we calculated ADI by dividing a toxicity value from either a no-observed adverse effect level (NOAEL), lowest observed adverse effect level (LOAEL), or benchmark dose (BMD) identified in a study by a factor accounting for various sources of scientific uncertainty.^e Uncertainty factors were included, as appropriate, to account for differences between humans and animals, differences between healthy and sensitive human populations, using data from short-term experiments to protect against effects from long-term exposure, and using data where a health effect was observed to estimate the level that does not cause an effect. To ensure appropriate protection, we have chosen to not use studies that have significant uncertainty as the basis for the recommended enforcement standards.^f This approach is consistent with that taken by EPA when establishing oral reference doses.

Gordon et al., 2011

Gordon et al. exposed male and female mice in a series of experiments that varied in concentrations of DONA exposure and varied in length of exposure to DONA to determine the toxicity effects of DONA.³

28 day study

Gordon et al. exposed male and female rats to different concentrations of DONA (0, 10, 30, and 100 milligrams of DONA per kilogram body weight per day or mg/kg-d) for 28 days through gavage. DONA decreased serum calcium levels and increased serum creatinine levels and kidney weights in females (Table 1). In males, DONA decreased hemoglobin, red blood cells bilirubin levels and increased glucose, alkaline phosphatase (ALP), and liver weights (Table 1).

c Appropriate toxicity values include the no observable adverse effect level (NOAEL), lowest observable adverse effect level (LOAEL), and benchmark dose (BMD).

d Due to the limited availability of data for this substance, we considered a study to be of an appropriate duration if animals were exposed for at least 28 days or if the exposure occurred during pregnancy. We accounted for differences in exposure duration through the use of an uncertainty factor.

e The NOAEL is the highest dose tested that did not cause an adverse effect, the LOAEL is the lowest dose tested that caused an adverse effect, and the BMD is an estimation of the dose that would cause a specific level of response (typically 5 or 10%).

f DHS considers a study to have significant uncertainty if the total uncertainty factors is equal to or greater than 3,000.

Table 1. Statistically Significant Effects Observed in Gordon et al, 2011³

Effects observed in females		Dose (mg/kg-d)		
		10	30	100
Serum chemistry	Decreased calcium levels			✓
	Increased creatinine levels			✓
Kidney	Increased kidney weights			✓
Effects observed in males		Dose (mg/kg-d)		
		10	30	100
Blood	Decreased red blood cells			✓
	Decreased hemoglobin levels			✓
Serum chemistry	Increased glucose levels		✓	✓
	Increase potassium levels		✓	✓
Liver	Increased liver weight		✓	✓
	Increased relative liver weight	✓	✓	✓
	Increased alkaline phosphatase levels			✓
	Decreased bilirubin levels	✓	✓	✓
	Increased urea levels			✓
	Increased liver cell size	✓	✓	✓
	Thyroid	Increased thyroid cell size		
	Increased thyroid cell numbers			✓

We identified a LOAEL of 10 mg/kg-d based on toxicity effects observed in the liver. We estimated a candidate ADI of 100 nanograms per kilogram per day (ng/kg-d) from this study based the LOAEL and a total uncertainty factor of 100,000 to account for differences between people and research animals (10), differences among people (10), use of a LOAEL instead of a NOAEL (10), use of a shorter duration study to protect against effects from long-term exposure (10), and the limited availability of information (10). While we obtained a candidate ADI for DONA from this study, this study was not used to establish a recommended enforcement standard due to significant uncertainty.

90 day study

Gordon et al. exposed male rats to different concentrations of DONA (0, 1, 3, and 10 mg/kg-d) and exposed females rats to different concentrations of DONA (0, 10, 30, and 100 mg/kg-d) for 90 days through gavage. DONA decreased serum calcium levels in the urine (an indication of thyroid or kidney problems) and increased white blood cells in females (Table 2). In males, DONA decreased hemoglobin, red blood cell, and hematocrit levels and increased liver cell size (Table 2).³

Table 2. Statistically Significant Effects Observed in Gordon et al, 2011³

Effects observed in females		Dose (mg/kg-d)		
		10	30	100
Urinalysis	Decreased calcium in urine			✓
Blood	Increased white blood cells			✓
Effects observed in males		Dose (mg/kg-d)		
		1	3	10
Blood	Decreased red blood cells			✓
	Decreased hemoglobin levels		✓	✓
	Decreased hematocrit levels		✓	✓
	Decreased white blood cells*	✓	✓	
Liver	Aspartate aminotransferase			✓
	Decreased total cholesterol*	✓	✓	
	Increased liver cells size			✓
* Effects were seen at lower concentrations but remediated at higher concentrations.				

We identified a NOAEL of 1 mg/kg-d based on decreased hemoglobin and hematocrit levels. We estimated a candidate ADI of 300 ng/kg-d from this study based on this NOAEL and a total uncertainty factor of 3,000 to account for differences between people and research animals (10), differences among people (10), use of a shorter duration study to protect against effects from long-term exposure (3), and the limited availability of information (10).

Developmental toxicity study

Gordon et al. exposed pregnant female rats to different concentrations of DONA (10, 30, 90, 270, and 500 mg/kg-d) from gestational day zero through gestational day 24 (or birth) through gavage. DONA exposure decreased body weight gain and food consumption in females and decreased litter size (Table 3).³

Table 3. Statistically Significant Effects Observed in Gordon et al, 2011³

Effects observed in females		Dose (mg/kg-d)			
		10	30	90	270
Body weight	Decreased weight gain				✓
	Decreased food consumption				✓
Liter Size	Decreased litter size			✓	✓

We identified a NOAEL of 30 mg/kg-d based on decreased litter size at higher doses. We estimated a candidate ADI of 0.1 micrograms per kilogram per day ($\mu\text{g}/\text{kg-d}$) from this study based on this NOAEL and a total uncertainty factor of 3,000 to account for differences between people and research animals

(10), differences among people (10), use of a shorter duration study to protect against effects from long-term exposure (3), and the limited availability of information (10).

Key Health Effects

We did not find any studies indicating that DONA can cause carcinogenic, mutagenic, teratogenic, or interactive effects in people, research animals, or cell culture studies.

Discussion

Available data show that DONA can affect the blood, cholesterol levels, the liver and kidney, and at high concentrations can decrease litter size and at high levels lead to death in rats.³

Studies in research animals have shown that DONA can affect the blood (e.g., red blood cell concentrations, hemoglobin level, hematocrit level), which is consistent with other more studied PFAS.² A decrease in red blood cells, hemoglobin levels, and hematocrit levels can lead to anemia and other negative health impacts.¹⁵⁻¹⁷ Testing whether these components fall within a normal range can help determine the health of a human or an animal and whether disease is present or there are issues with normal organ function.

Similar to other PFAS, limited studies in rodents have shown that DONA can reduce cholesterol levels.² However, studies in people have shown that PFAS exposure is associated with increased total cholesterol and LDL levels.^{2,18} Scientists are learning more about the ways in which PFAS reduce cholesterol levels in rodents and believe that they are associated with activation of nuclear hormone receptors.¹⁸ Because nuclear receptors, like PPAR α , are important regulators of lipid metabolism, changes in their expression patterns can affect lipid transport and alter cholesterol levels. However, at this time, it is unclear whether PFAS might act the same way in people as they do in animals.

A number of studies have demonstrated that liver effects caused by PFAS occur primarily through activation of the nuclear hormone receptor, PPAR α (peroxisome proliferator-activated receptor alpha).¹⁹⁻²³ Nuclear receptors regulate gene expression and PPAR α regulates the expression of genes involved in lipid and cholesterol metabolism.²⁴ While PPAR α receptors are found in rodents and humans, levels of these receptors are much higher in rats and mice than in monkeys and humans.²⁴ This means that effects caused by activation of PPAR α are more likely to occur in rodents than they are to occur in people. As such, the effects on the liver observed in the critical studies reviewed here are likely not relevant to humans.

Standard Selection

DHS recommends an enforcement standard of 3 µg/L for DONA.

There are no federal numbers and no state drinking water standard for DONA. Additionally, the EPA has not evaluated the carcinogenicity or established an ADI (oral reference dose) for DONA.

However, we found one study evaluating the toxicity of

DONA. To calculate the ADI as specified in s. 160.13, Wisc. Statute, DHS selected the 2011 study by Gordon et al. as the critical study.³ We selected the 90 day study because it evaluated longer-term DONA exposure to male and female rats which can better estimate the effects after long-term exposure. We choose effect on blood chemistry as the critical effect because decreases in red blood cells, hemoglobin, and hematocrit levels can lead to anemia, and potentially heart problems and pregnancy complications.¹⁵⁻¹⁷ These effects of DONA are also consistent with those caused by other PFAS in research studies.²

As described above, we selected an ADI of 0.300 µg/kg-d from a NOAEL of 1 mg/kg-d and a total uncertainty factor of 3000. To determine the recommended enforcement standard, DHS used the ADI, and, as required by Ch. 160, Wis. Stats., a body weight of 10 kg, a water consumption rate of 1 L/d, and a relative source contribution of 100%.

DHS recommends a preventive action limit of 0.6 µg/L for DONA.

DHS recommends that the preventive action limit for DONA be set at 20% of the enforcement standard because DONA has not been shown to cause carcinogenic, mutagenic, teratogenic, or interactive effects in people or research animals.

Prepared by Gavin Dehnert, Ph.D. and Sarah Yang, Ph.D.

Wisconsin Department of Health Services

Basis for Enforcement Standard

- Federal Number
- Cancer Potential
- EPA Acceptable Daily Intake
- Technical information

References

1. Fromme H, Wöckner M, Roscher E, Völkel W. ADONA and perfluoroalkylated substances in plasma samples of German blood donors living in South Germany. *International journal of hygiene and environmental health*. 2017;220(2 Pt B):455-460.
2. ATSDR. Toxicological Profile for Perfluoroalkyls - Draft for Public Comment. In: Registry AftSaD, ed. Atlanta, GA2017.
3. Gordon SC. Toxicological evaluation of ammonium 4,8-dioxa-3H-perfluorononanoate, a new emulsifier to replace ammonium perfluorooctanoate in fluoropolymer manufacturing. *Regulatory toxicology and pharmacology : RTP*. 2011;59(1):64-80.
4. DHS. *Recommended Public Health Groundwater Quality Standards: Scientific Support Documents for Cycle 10 Substances*. 2019.
5. Zhang B, He Y, Huang Y, et al. Novel and legacy poly- and perfluoroalkyl substances (PFASs) in indoor dust from urban, industrial, and e-waste dismantling areas: The emergence of PFAS alternatives in China. *Environmental pollution (Barking, Essex : 1987)*. 2020;263(Pt A):114461.
6. Awad R, Zhou Y, Nyberg E, et al. Emerging per- and polyfluoroalkyl substances (PFAS) in human milk from Sweden and China. *Environmental science Processes & impacts*. 2020.
7. Li QL, Cheng XH, Zhao Z, et al. [Distribution and Fluxes of Perfluoroalkyl and Polyfluoroalkyl Substances in the Middle Reaches of the Yellow River (Weinan-Zhengzhou Section)]. *Huan jing ke xue= Huanjing kexue*. 2019;40(1):228-238.
8. Pan Y, Zhang H, Cui Q, et al. Worldwide Distribution of Novel Perfluoroether Carboxylic and Sulfonic Acids in Surface Water. *Environmental science & technology*. 2018;52(14):7621-7629.
9. WIDNR. Safe Drinking Water In: Resources WDoN, ed. *Chapter NR 8092018*.
10. USEPA. National Primary Drinking Water Regulations. <https://www.epa.gov/ground-water-and-drinking-water/national-primary-drinking-water-regulations>. Published 2018. Accessed.
11. USEPA. Drinking Water Contaminant Human Health Effects Information. <https://www.epa.gov/dwstandardsregulations/drinking-water-contaminant-human-health-effects-information#hh1>. Published 2019. Accessed.
12. USEPA. IRIS Assessments. https://cfpub.epa.gov/ncea/iris_drafts/AtoZ.cfm. Published 2019. Accessed.
13. IARC. List of Classification, Volumes 1-123. <https://monographs.iarc.fr/list-of-classifications-volumes/>. Published 2018. Accessed May 17, 2019.
14. Ritter L, Totman C, Krishnan K, Carrier R, Vezina A, Morisset V. Deriving uncertainty factors for threshold chemical contaminants in drinking water. *Journal of toxicology and environmental health Part B, Critical reviews*. 2007;10(7):527-557.

15. De Andrade Cairo RC, Rodrigues Silva L, Carneiro Bustani N, Ferreira Marques CD. Iron deficiency anemia in adolescents; a literature review. *Nutricion hospitalaria*. 2014;29(6):1240-1249.
16. Lanier JB, Park JJ, Callahan RC. Anemia in Older Adults. *American family physician*. 2018;98(7):437-442.
17. Lopez A, Cacoub P, Macdougall IC, Peyrin-Biroulet L. Iron deficiency anaemia. *Lancet (London, England)*. 2016;387(10021):907-916.
18. Dong Z, Wang H, Yu YY, Li YB, Naidu R, Liu Y. Using 2003-2014 U.S. NHANES data to determine the associations between per- and polyfluoroalkyl substances and cholesterol: Trend and implications. *Ecotoxicol Environ Saf*. 2019;173:461-468.
19. Das KP, Grey BE, Zehr RD, et al. Effects of perfluorobutyrate exposure during pregnancy in the mouse. *Toxicol Sci*. 2008;105(1):173-181.
20. Wolf CJ, Takacs ML, Schmid JE, Lau C, Abbott BD. Activation of mouse and human peroxisome proliferator-activated receptor alpha by perfluoroalkyl acids of different functional groups and chain lengths. *Toxicological sciences : an official journal of the Society of Toxicology*. 2008;106(1):162-171.
21. Bjork JA, Wallace KB. Structure-activity relationships and human relevance for perfluoroalkyl acid-induced transcriptional activation of peroxisome proliferation in liver cell cultures. *Toxicol Sci*. 2009;111(1):89-99.
22. Cheng X, Klaassen CD. Critical role of PPAR-alpha in perfluorooctanoic acid- and perfluorodecanoic acid-induced downregulation of Oatp uptake transporters in mouse livers. *Toxicological sciences : an official journal of the Society of Toxicology*. 2008;106(1):37-45.
23. Rosenmai AK, Ahrens L, le Godec T, Lundqvist J, Oskarsson A. Relationship between peroxisome proliferator-activated receptor alpha activity and cellular concentration of 14 perfluoroalkyl substances in HepG2 cells. *J Appl Toxicol*. 2018;38(2):219-226.
24. Hall AP, Elcombe CR, Foster JR, et al. Liver hypertrophy: a review of adaptive (adverse and non-adverse) changes--conclusions from the 3rd International ESTP Expert Workshop. *Toxicologic pathology*. 2012;40(7):971-994.

Appendix A. Toxicity Data

Table A-I. DONA Toxicity Studies from Literature Review

Study Type	Species	Duration	Doses (mg/kg-d)	Route	Endpoints	Toxicity Value (mg/kg-d)	Reference
Acute	Rat	5 days	28, 104, 298	Gavage	Decreased body weight, reduced food consumption, reduced blood cells, increased glucose levels, increased kidney weight, and increased liver weight.	LOAEL: 28	Gordon et al., 2011 ³
Short-term	Rat	28 days	10, 30, 100	Gavage	Decreased hemoglobin, decreased hematocrit, increased alkaline phosphatase, increased glucose, decreased total bilirubin, increased urea, increased liver weight, increased adrenal weight, and increase liver and thyroid cell hypertrophy and hyperplasia.	LOAEL: 10	Gordon et al., 2011 ³
Sub-chronic	Rat	90 day	1, 3, 10 male 10, 30, 100 female	Gavage	Decreased red blood cells, decreased hemoglobin, decreased hematocrit, and increased liver weights	Males: NOAEL: 1 LOAEL: 3 Females: NOAEL: 30 LOAEL: 100	Gordon et al., 2011 ³
Development	Rat	Development GD 1-24	10,30, 90, 270	Gavage	Decreased food consumption, decreased weight gain, and decreased liter size.	NOAEL: 30 LOAEL: 90	Gordon et al., 2011 ³

Table A-2. Critical Study Selection for DONA

Reference	Appropriate duration?	Effects consistent with other studies?	Effects relevant to humans?	Number of Doses	Toxicity value identifiable?	Critical study?
Gordon et al., 2011 ³ Acute 5-Day	⊖	✓	✓	3	✓	No
Gordon et al., 2011 ³ Short Term 28-Day	✓	✓	✓	3	✓	Yes
Gordon et al., 2011 ³ Subchronic 90-Day	✓	✓	✓	5	✓	Yes
Gordon et al., 2011 ³ Developmental	✓	✓	✓	4	✓	Yes

To be considered a critical study, the study must be of an appropriate duration (at least 28 days or exposure during gestation), have identified effects that are consistent with other studies and relevant for humans, have evaluated more than one dose, and have an identifiable toxicity value.