United States Environmental Protection Agency
Office of Water Mail Code 4304T
EPA 822-R-16-005 May 2016

Drinking Water Health Advisory for Perfluorooctanoic Acid (PFOA)

ED_005024_00000285-00001

Drinking Water Health Advisory for Perfluorooctanoic Acid (PFOA)

Prepared by:

U.S. Environmental Protection Agency
Office of Water (4304T)
Health and Ecological Criteria Division
Washington, DC 20460

EPA Document Number: 822-R-16-005
May 2016

ED_005024_00000285-00002

ACKNOWLEDGMENTS

This document was prepared by the Health and Ecological Criteria Division, Office of Science and Technology, Office of Water of the U.S. Environmental Protection Agency (EPA). The Agency gratefully acknowledges the valuable contributions of EPA scientists Glinda Cooper, Ph.D.; Barbara Glenn, Ph.D.; Erin Hines, Ph.D.; Michael Wright, Sc.D.; John Wambaugh, Ph.D.; Thomas Speth, Ph.D.; and Daniel Hautman.

This Health Advisory was provided for review by and comments were received from staff in the following EPA program Offices:

Office of Chemical Safety and Pollution Prevention
Office of Children's Health Protection
Office of General Counsel
Office of Land and Emergency Response
Office of Policy
Office of Research and Development
Office of Water

ED_005024_00000285-00003

CONTENTS

ACKNOWLEDGMENTS 3
ABBREVIATIONS AND ACRONYMS 7
EXECUTIVE SUMMARY 9
1.0 INTRODUCTION AND BACKGROUND 11
1.1 Safe Drinking Water Act 11
1.2 Current Advisories and Guidelines 12
1.3 Uses of PFOA 14
2.0 NATURE OF THE STRESSOR 15
2.1 Physical and Chemical Properties 15
2.2 Occurrence and Sources of Exposure 17
2.2.1 Surface Water and Ground Water 17
2.2.2 Drinking Water 18
2.2.3 Food 18
2.2.4 Ambient Air 21
2.2.5 Indoor Dust 22
2.2.6 Soils 22
2.2.7 Biosolids 23
2.2.8 Consumer Products 24
2.3 Environmental Fate 24
2.3.1 Mobility 24
2.3.2 Persistence 24
2.3.3 Bioaccumulation 25
2.4 Toxicokinetics 25
2.5 Human Biomonitoring Data 27
3.0 PROBLEM FORMULATION 28
3.1 Conceptual Model 28
3.1.1 Conceptual Model Diagram for Exposure via finished Drinking Water 29
3.1.2 Factors Considered in the Conceptual Model for PFOA 29
3.2 Analysis Plan 31
3.2.1 Health Advisory Guidelines 31
3.2.2 Establishing the Data Set 31
3.2.3 Approach for HA Calculation 32
3.2.4 Measures of Effect 33
3.2.5 Relative Source Contribution 34

ED_005024_00000285-00004

4.0 EFFECTS ASSESSMENT 35
4.1 Noncancer Health Effects 35
4.1.1 Animal Toxicity Studies 35
4.1.2 Human Epidemiology Studies 39
4.1.3 Noncancer Mode of Action 42
4.2 Cancer 44
4.2.1 Animal Cancer Bioassays 44
4.2.2 Human Epidemiology Studies 45
4.2.3 Cancer Mode of Action 46
4.2.4 Weight of Evidence Classification 47
5.0 DOSE-RESPONSE ASSESSMENT 48
5.1 Uncertainty Factors 50
5.2 RfD Determination 51
6.0 HEALTH ADVISORY VALUES 53
6.1 Relative Source Contribution 53
6.2 Lifetime Health Advisory 54
7.0 QUANTIFICATION OF CANCER RISK 56
8.0 EFFECTS CHARACTERIZATION 57
8.1 Uncertainty and Variability 57
8.2 Use of Human Epidemiology Data 58
8.3 Consideration of Immunotoxicity 58
8.4 Effects on Mammary Gland Development 60
8.5 Alternative Exposure Scenarios 61
8.6 Relative Source Contribution Considerations 61
8.7 Sensitive Populations: Gender Differences 63
8.8 Sensitive Populations: Developmental Effects 63
9.0 ANALYTICAL METHODS 64
10.0 TREATMENT TECHNOLOGIES 65
11.0 REFERENCES 69
12.0 APPENDIX A-QUANTITATIVE CANCER ASSESSMENT MODELING 99

ED_005024_00000285-00005

TABLES

Table 1-1. State Guideline Values for PFOA 13

Table 1-2. International Guideline Values for PFOA 13

Table 2-1. Chemical and Physical Properties of PFOA 16

Table 5-1. Human Equivalent Doses Derived from the Modeled Animal Average Serum Values 49

Table 5-2. Candidate RfDs Derived from the HEDs from the Pharmacokinetic Model Average Serum Values 52

FIGURES

Figure 2-1. Chemical Structures of PFOA and APFO 15

Figure 3-1. Conceptual Model for PFOA in Finished Drinking Water 28

ED_005024_00000285-00006

ABBREVIATIONS AND ACRONYMS

ALT	alanine aminotransferase
ALP	alkaline phosphatase
APFO	ammonium perfluorooctanoate
AST	aspartate aminotransferase
AUC	area under the curve
BAF	bioaccumulation factor
BCF	bioconcentration factor
BMDL	benchmark dose level
BMF	biomagnification factor
bw	body weight
CAR	constitutive androstane receptor
CCK	cholecystokinin
CCL	Contaminant Candidate List
COPD	chronic obstructive airways disease
CWA	Clean Water Act
DWEL	drinking water equivalent level
DWI	drinking water intake
EPA	U.S. Environmental Protection Agency
FXR	farnesoid X receptor
GFR	glomerular filtration rate
GGT	gamma-glutamyl transferase
HA	Health Advisory
HDL	high-density lipoprotein
HED	human equivalent dose
HESD	Health Effects Support Document
IgM	immunoglobulin M
IRIS	Integrated Risk Information System
K_{oc}	organic carbon-water partitioning coefficient
K_{ow}	octanol-water partition coefficient
LCT	Leydig cell tumor
LC/MS/MS	liquid chromatography/tandem mass spectrometry
LDL	low-density lipoprotein
LOAEL	lowest observed adverse effect level
MOA	mode of action
MRL	minimum reporting level
ng/L	nanograms per liter
NHANES	National Health and Nutrition Examination Survey
NOAEL	no observed adverse effect level
PAC	powdered activated carbon
PACT	pancreatic acinar cell tumors
PBPK	physiologically based pharmacokinetic model
PFAS	perfluoroalkyl substance
PFC	perfluoroinated compounds
PFOA	perfluorooctanoic acid
PFOS	perfluorooctanesulfonic acid

ED_005024_00000285-00007

PTFE	polytetrafluoroethylene
pg/L	picograms per liter
PND	post-natal day
POD	point of departure
POE	point-of-entry
POU	point-of-use
PPARα	peroxisome proliferator activated receptor alpha
PWS	public water system
PXR	pregnane X receptor
REACH	Registration, Evaluation, Authorization, and Restriction of Chemicals
RfD	reference dose
RSC	relative source contribution
SDWA	Safe Drinking Water Act
SNUR	Significant New Use Rule
SRBC	sheep red blood cell
TMF	trophic magnification factor
TNSSS	Total National Sewage Sludge Survey
UCMR 3	third Unregulated Contaminant Monitoring Rule
UF	uncertainty factor
UV	ultraviolet

ED_005024_00000285-00008

EXECUTIVE SUMMARY

Perfluorooctanoic acid (PFOA) is a synthetic, fully fluorinated organic acid; it used in a variety of consumer products and in the production of fluoropolymers, and it is generated as a degradation product of other perfluorinated compounds. Because of strong carbon-fluorine bonds, PFOA is stable to metabolic and environmental degradation. PFOA is one of a large group of perfluoroalkyl substances (PFASs) that are used to make products more resistant to stains, grease, and water. These compounds have been widely found in consumer and industrial products, as well as in food items. Major U.S. manufacturers voluntarily agreed to phase out production of PFOA by the end of 2015. Exposure to PFOA in the United States remains possible due to its legacy uses, existing and legacy uses on imported goods, degradation of precursors, and extremely high persistence in the environment and the human body. PFOA was detected in blood serum in 99% of the U.S. general population between 1999 and 2012; however, the levels of PFOA in blood have been decreasing since U.S. companies began to phase out production. Water resources contaminated by PFOA have been associated with releases from manufacturing sites, industrial sites, fire/crash training areas, and industrial or municipal waste sites where products are disposed of or applied.

The U.S. Environmental Protection Agency (EPA) is issuing a lifetime drinking water Health Advisory (HA) for PFOA of 0.07 micrograms per liter (µg/L) based on a reference dose (RfD) derived from a developmental toxicity study in mice; the critical effects included reduced ossification in proximal phalanges and accelerated puberty in male pups following exposure during gestation and lactation. PFOA is known to be transmitted to the fetus in cord blood and to the newborn in breast milk. This lifetime HA is based on the latest health effects information for noncancer and cancer effects for PFOA as described in EPA's 2016 *Health Effects Support Document for Perfluorooctanoic Acid (PFOA)*, which was revised following external peer review. Because the developing fetus and newborn are particularly sensitive to PFOA-induced toxicity, the RfD based on developmental effects also is protective of adverse effects in adults (e.g., liver and kidney toxicity). The lifetime HA is therefore protective of the population at large.

For PFOA, oral animal studies of short-term, subchronic, and chronic duration are available in multiple species including monkeys, rats and mice. These studies report developmental effects (survival, body weight changes, reduced ossification, delays in eye opening, altered puberty, and retarded mammary gland development), liver toxicity (hypertrophy, necrosis, and effects on the metabolism and deposition of dietary lipids), kidney toxicity (weight), immune effects, and cancer (liver, testicular, and pancreatic). Overall, the toxicity studies available for PFOA demonstrate that the developing fetus is particularly sensitive to PFOA-induced toxicity. Human epidemiology data report associations between PFOA exposure and high cholesterol, increased liver enzymes, decreased vaccination response, thyroid disorders, pregnancy-induced hypertension and preeclampsia, and cancer (testicular and kidney).

To derive candidate RfDs, EPA used a peer-reviewed pharmacokinetic model to calculate the average serum concentrations associated with candidate no observed adverse effect levels (NOAELs) and lowest observed adverse effect levels (LOAELs) from six studies for multiple effects. Consistent with EPA's guidance *A Review of the Reference Dose and Reference*

ED_005024_00000285-00009

Concentration Processes (USEPA 2002), EPA applied protective uncertainty factors to address intraspecies variability, interspecies variability, and LOAEL to NOAEL extrapolation.

From a national perspective, the dominant source of human exposure to PFOA is expected to be from the diet; indoor dust from carpets and other sources also is an important source of exposure, especially for children. The HA was calculated using a relative source contribution (RSC) of 20%, which allows for other PFOA exposure sources (e.g., dust, diet, air) to make up 80% of the RfD.

EPA's risk assessment guidelines reflect that, as a general matter, a single exposure to a developmental toxin at a critical time in development can produce an adverse effect (USEPA 1991). In addition, short-term exposure to PFASs can result in a body burden that persists for years and can increase with additional exposures. Thus, EPA recommends that the lifetime HA for PFOA of 0.07 μg/L apply to both short-term (i.e., weeks to months) scenarios during pregnancy and lactation, as well as to lifetime-exposure scenarios.

Adverse effects observed following exposures to PFOA and PFOS are the same or similar and include effects in humans on serum lipids, birth weight, and serum antibodies. Some of the animal studies show common effects on the liver, neonate development, and responses to immunological challenges. Both compounds were also associated with tumors in long-term animal studies. The RfDs for both PFOA and PFOS are based on similar developmental effects and are numerically identical; when these two chemicals co-occur at the same time and location in a drinking water source, a conservative and health-protective approach that EPA recommends would be to compare the sum of the concentrations ([PFOA] + [PFOS]) to the HA (0.07 μg/L).

Under EPA's *Guidelines for Carcinogen Risk Assessment* (USEPA 2005), there is Suggestive Evidence of Carcinogenic Potential for PFOA. Epidemiology studies demonstrate an association of serum PFOA with kidney and testicular tumors among highly exposed members of the general population. Two chronic bioassays of PFOA support a positive finding for the ability of PFOA to be tumorigenic in one or more organs of rats, including the liver, testes, and pancreas. EPA estimated a cancer slope factor of 0.07 per milligram per kilogram-day $(mg/kg\text{-}day)^{-1}$ based on testicular tumors, and confirmed that the lifetime HA based on noncancer effects is protective of the cancer endpoint.

ED_005024_00000285-00010

1.0 INTRODUCTION AND BACKGROUND

The U.S. Environmental Protection Agency (EPA) developed the nonregulatory Health Advisory (HA) Program in 1978 to provide information for public health officials or other interested groups on pollutants associated with short-term contamination incidents or spills that can affect drinking water quality, but are not regulated under the Safe Drinking Water Act (SDWA). At present, EPA lists HAs for more than 200 contaminants.[1]

HAs identify the concentration of a contaminant in drinking water at which adverse health effects are not anticipated to occur over specific exposure durations (e.g., 1 day, 10 days, a lifetime). HAs serve as informal technical guidance to assist federal, state, and local officials, and managers of public or community water systems in protecting public health when emergency spills or other contamination situations occur. An HA document provides information on the environmental properties, health effects, analytical methodology, and treatment technologies for removing drinking water contaminants.

Perfluorooctanoic acid (PFOA) is a manmade chemical in a large family of chemicals called perfluoroalkyl substances (PFASs) (Buck et al. 2011). PFOA has been used in a variety of consumer products and in the production of fluoropolymers, and is generated as a degradation product of other perfluorinated compounds. PFOA is very persistent in the environment and the human body; it has been detected in water, wildlife, and humans worldwide. This document, EPA's 2016 *Drinking Water Health Advisory for Perfluorooctanoic Acid (PFOA)*, presents a guideline concentration for PFOA in drinking water at which adverse health effects are not anticipated to occur over a human lifetime. This lifetime HA is based on the latest health effects information for noncancer and cancer effects for PFOA as described in EPA's *Health Effects Support Document for Perfluorooctanoic Acid (PFOA)* (USEPA 2016a). The HA value is not a legally enforceable federal standard and is subject to change as new information becomes available. The structure, principles, and approach of this document are consistent with EPA's *Framework for Human Health Risk Assessment to Inform Decision Making* (USEPA 2014a).

1.1 Safe Drinking Water Act

SDWA, as amended in 1996, requires EPA to publish a list of unregulated contaminants every 5 years that are not subject to any proposed or promulgated national primary drinking water regulations, are known or anticipated to occur in public water systems (PWSs), and might require regulation under SDWA. This list is known as the Contaminant Candidate List (CCL). PFOA is included on the third CCL (USEPA 2009a) and on the draft fourth CCL (USEPA 2015a).

As part of its responsibilities under SDWA, EPA is required to implement a monitoring program for unregulated contaminants. SDWA requires, among other things, that once every 5 years, EPA issue a list of no more than 30 unregulated contaminants to be monitored by PWSs. In 2012, EPA included PFOA in its third Unregulated Contaminant Monitoring Rule (UCMR 3), which required all large systems serving > 10,000 people, plus a statistically selected group of 800 small systems to monitor for a 1-year period between 2013 and 2015. The last of the

[1] For more information see http://water.epa.gov/drink/standards/hascience.cfm.

ED_005024_00000285-00011

monitoring data are still being compiled, but results to-date indicate that PFOA has been measured at or above the minimum reporting level (0.02 micrograms per liter [µg/L]) by approximately 2% of PWSs nationwide. To-date, PFOA has been measured above the new lifetime HA level of 0.07 µg/L by approximately 0.3% of PWSs. Approximately 1% of PWSs have reported data for which combined PFOA and PFOS results are above 0.07 µg/L. For the latest UCMR 3 results, please refer to https://www.epa.gov/dwucmr/occurrence-data-unregulated-contaminant-monitoring-rule#3.

SDWA requires EPA to make regulatory determinations for at least five CCL contaminants every 5 years. EPA must begin developing a national primary drinking water regulation when the Agency makes a determination to regulate based on three criteria:

- The contaminant may have an adverse effect on the health of persons.
- The contaminant is known to occur or there is substantial likelihood the contaminant will occur in public water systems with a frequency and at levels of public health concern.
- In the sole judgment of the Administrator, regulating the contaminant presents a meaningful opportunity for health risk reductions.

To make these determinations, the Agency uses data to analyze occurrence of these compounds in finished drinking water and data on health effects. If EPA determines the contaminant does not meet any one of the three statutory criteria, the Agency's determination is not to regulate. EPA continues to gather information to inform future regulatory determinations for PFOA under the SDWA.

EPA developed a *Health Effects Support Document for Perfluorooctanoic Acid (PFOA)* and one for another PFAS, perfluorooctane sulfonate (also known as perfluorooctanesulfonic acid or PFOS), to assist federal, state, tribal and local officials, and managers of drinking water systems in protecting public health when these chemicals are present in drinking water (USEPA 2016a, 2016b). The health effects support documents (HESDs) were peer-reviewed in 2014 and were revised as recommended by the peer reviewers with consideration of public comments and inclusion of additional studies published through December 2015. The revised HESD for PFOA (USEPA 2016a) provides an RfD and cancer assessment that serve as the basis for this HA.

The SDWA provides the authority for EPA to publish nonregulatory HAs or take other appropriate actions for contaminants not subject to any national primary drinking water regulation. EPA is providing this HA for PFOA to assist federal, state, and local officials evaluate risks from this contaminant in drinking water. The HA values consider variability in human response across all life stages and population groups while making allowance for contributions from other exposure media.

1.2 Current Advisories and Guidelines

Currently there are no federal regulations under the SDWA or national recommended ambient water quality criteria under the Clean Water Act (CWA) for PFOA. In January 2009, EPA developed a provisional HA for PFOA in drinking water of 0.4 micrograms per liter (µg/L). The provisional HA was developed to reflect an amount of PFOA that could cause adverse health effects in the short term (weeks to months). The provisional HA was intended as a

ED_005024_00000285-00012

guideline for PWSs while allowing time for EPA to develop a lifetime HA. Table 1-1 provides drinking water guideline values that were developed by states.

Table 1-1. State Guideline Values for PFOA

State	Guideline Value (µg/ L)	Source
Delaware Department of Resources and Environmental Control	0.4	DNREC (2016)
Maine Department of Health and Human Services	0.1	Maine DHHS (2014)
Michigan Department of Environmental Quality	0.42	Michigan DEQ (2013)
Minnesota Department of Health	0.3	MDH (2009)
New Jersey Department of Environmental Protection	0.04	NJDEP (2014)
North Carolina Division of Water Quality	2	NCDEQ (2013)
Vermont Agency of Natural Resources	0.02	Vermont ANR (2016)

In 2013 the European Chemicals Agency adopted an agreement that identified PFOA as a "Substance of Very High Concern" because of its persistent, bioaccumulative, and toxic characteristics and placed it onto the Candidate List for Registration, Evaluation, Authorization and Restriction of Chemicals (REACH) (Vierke et al. 2012). Once on the Candidate List, PFOA could be included in Annex XIV of the REACH regulation, which would effectively ban use in manufacturing and in the market.

PFOA also is being considered for listing under The Stockholm Convention on Persistent Organic Pollutants (Convention), a global treaty to protect human health and the environment from persistent organic pollutants. In October 2015, the Persistent Organic Pollutants Review Committee agreed that PFOA meets the screening criteria in Annex D of the Convention, the first of several steps toward listing of chemicals. Listing in various Annexes of the Convention obligates parties to abide by provisions set forth to prohibit, eliminate, or restrict production and use, as well as the import and export of persistent organic pollutants, except as allowed for by specific exemptions. Several international agencies have established guideline values for PFOA (Table 1-2).

Table 1-2. International Guideline Values for PFOA

Country/Agency	Guideline Value (µg/ L)		Source
	Health-based	Administrative	
German Ministry of Health	0.3	Composite precautionary guidance value for PFOA+PFOS is 0.1	German Ministry of Health (2006)
United Kingdom (UK) Drinking Water Inspectorate	5.0	Action levels: Tier 1: potential hazard Tier 2: > 0.3 Tier 3: > 5.0 Tier 4: > 45	UK Drinking Water Inspectorate (2009)
Danish Ministry of the Environment	0.3	Composite drinking water criteria are based on relative toxicity of PFOS, PFOA, and PFOSA	Danish Ministry of the Environment (2015)

ED_005024_00000285-00013

Country/Agency	Guideline Value (µg/ L)		Source
	Health-based	Administrative	
Swedish National Food Agency	--	Also 0.09 for the mixture of: PFOS, PFOA, PFHxS; PFBS; PFHpA, PFHsA, PFPeA (total PFASs) 0.9: Pregnant women, women trying to get pregnant, and infants should not consume if total PFASs exceeds	Livsmedelsverket (2014), cited in Danish Ministry of the Environment (2015)

Notes:
PFOA = perfluorooctanoic acid; PFOS = perfluorooctane sulfonate; PFBS = perfluorobutane sulfonate; PFHpA = perfluoroheptanoic acid; PFHsA = perfluorohexanoic acid; PFHxS = perfluorohexane sulfonic acid; PFOSA = perfluorosulfonamide; PFPeA = perfluoropentanoic acid

1.3 Uses of PFOA

Perfluorinated substances, such as PFOA and its derivatives, are water- and lipid-resistant because of their chemical properties. Therefore, they are commonly used as surface-active agents that alter the surface tension of a mixture. Historically, PFOA was used in the United States in carpets, leathers, textiles, upholstering, paper packaging, and coating additives as a waterproofing or stain-resistant agent. Fire resistance of aviation fluid is increased by adding PFOA, PFOS, and other PFASs to the mixture.

In 2006, EPA initiated the 2010/2015 PFOA Stewardship Program in which eight major companies committed to reduce facility emissions and product contents of PFOA and related chemicals on a global basis by 95% no later than 2010, and to work toward eliminating emissions and product content of these chemicals by 2015 (USEPA 2006). Although the 2010/2015 PFOA Stewardship Program has worked toward eliminating emissions and product content, there are still some ongoing uses that EPA is evaluating. Shorter-chain perfluoroalkyl-based products have been developed to replace these chemicals.

To complement the Stewardship Program, EPA developed Significant New Use Rules[2] (SNURs) to allow EPA to review any significant new uses of PFOA and many PFOA-related chemicals before they are commercialized in the United States. On October 22, 2013, EPA issued a final SNUR (published in the *Federal Register* [FR]; 78 FR 62443) requiring companies to provide notice of any new manufacturing or processing of long-chain perfluoroalkyl carboxylates for use in or on carpets (i.e., to impart soil, water, and stain resistance). Companies must now provide EPA with notice of their intent to manufacture (including import) any of these chemicals if they are used in carpets or to treat carpets. They must also notify EPA for these chemical substances if they intend to import carpets already containing these chemical substances. EPA subsequently proposed another SNUR on January 21, 2015, for PFOA and also for PFOA-related chemicals that have not yet been commercialized (80 FR 2885).

[2] For more information on EPA's SNURs visit http://www.epa.gov/assessing-and-managing-chemicals-under-tsca/long-chain-perfluorinated-chemicals-pfcs.

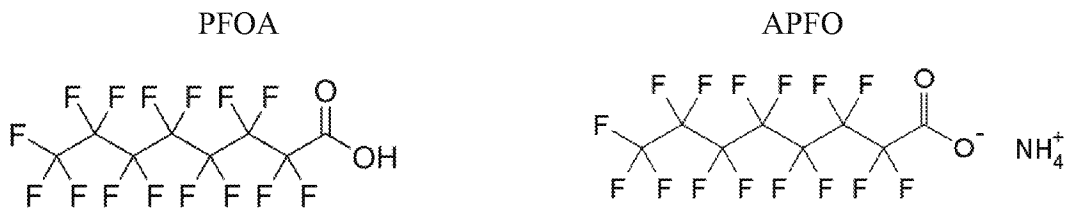
ED_005024_00000285-00014

Given the limited ongoing uses of PFOA-related chemicals, releases to surface water and ground water from PFOA are expected to decline. Exposure to PFOA in the United States remains possible, however, because of its legacy uses, existing and legacy uses on imported goods, degradation of precursors, and extremely high persistence in the environment and in the human body.

2.0 NATURE OF THE STRESSOR

2.1 Physical and Chemical Properties

PFOA and its salts are fluorinated organic compounds and are part of the group of PFASs. PFOA is a completely fluorinated organic synthetic acid that was used in the United States primarily as an aqueous dispersion agent in the manufacture of fluoropolymers and in a variety of water-, oil-, and stain-repellant products. Ammonium perfluorooctanoate (APFO) is the ammonium salt of PFOA (Figure 2-1) which was a processing aid in the manufacture of certain fluoropolymers, especially as an emulsifier during the polymerization of tetrafluoroethylene to make polytetrafluoroethylene (e.g., Teflon™). Most of these primary uses have been voluntarily phased out in the United States as of 2015 (see section 1.3 above); however, limited U.S. uses and imports continue. Some sources of PFOA in the environment result from the atmospheric degradation or transformation and/or surface deposition of precursors, including related fluorinated chemicals (perfluorotelomer alcohols) (Wallington et al. 2006).



Source: SIAR 2008

Figure 2-1. Chemical Structures of PFOA and APFO

The structure of PFOA varies with the manufacturing process. PFOA can be either a linear or branched eight-carbon carboxylic acid with a partial negative charge on each fluorine and an acidic carboxylate functional group. Low concentrations of other perfluorocarboxylate chain lengths can also be present. It will tend to form micelles in aqueous solution and be attracted to surfaces that are characterized by positive charge.

In the environment, the acidic form ionizes in water to a PFOA anion, and the ammonium salt of PFOA rapidly dissociates. Physical and chemical properties and other reference information for PFOA are provided in Table 2-1. These properties help to define the behavior of PFOA in living systems and the environment. PFOA is a highly stable compound. It is a solid at room temperature with a low vapor pressure. The melting point for PFOA is identified as 50 to 60 degrees Celsius (°C); vapor pressures increase at temperatures near the melting point.

ED_005024_00000285-00015

Table 2-1. Chemical and Physical Properties of PFOA

Property	Perfluorooctanoic Acid	Source
Chemical Abstracts Service Registry No. (CASRN[a])	335-67-1	
Chemical Abstracts Index Name	2,2,3,3,4,4,5,5,6,6,7,7,8,8,8-pentadecafluorooctanoic acid	
Synonyms	PFOA; Pentadecafluoro-1-octanoic acid; Pentadecafluoro-n-octanoic acid; Octanoic acid, pentadecafluoro-; Perfluorocaprylic acid; Pentadecafluorooctanoic acid; Perfluoroheptanecarboxylic acid;	
Chemical Formula	$C_8HF_{15}O_2$	
Molecular Weight (g/mol)	414.09	HSDB (2012); Lide (2007); SRC (2016)
Color/Physical State	White powder (ammonia salt)	HSDB (2012); Lewis (2004)
Boiling Point	192.4°C; Stable when bound	HSDB (2012); Lide (2007); SRC (2016)
Melting Point	54.3 °C	HSDB (2012); Lide (2007); SRC (2016)
Vapor Pressure	0.525 mm Hg at 25 °C (measured) 0.962 mm Hg at 59.25 °C (measured)	Hekster et al. (2003); HSDB (2012); SRC (2016) ATSDR (2015); Kaiser et al. (2005)
Henry's Law Constant	Not measureable	ATSDR (2015)
pKa	2.80	SRC (2016)
K_{oc}	2.06	Higgins and Luthy (2006)
K_{ow}	Not measurable	ATSDR (2015); EFSA (2008)
Solubility in Water	9.50×10^3 mg/L at 25 °C (estimated)	ATSDR (2015); Hekster et al. (2003); HSDB (2012); Kauck and Diesslin (1951); SRC (2016)
Half-life in Water (25°C)	Stable	UNEP (2015)
Half-life in Air	Stable when bound	UNEP (2015)

Notes:
K_{ow} = octanol-water partition co-efficient; K_{oc} = organic carbon-water partitioning coefficient; g/mol = grams per mole
[a]The CASRN given is for linear PFOA, but the toxicity studies are based on a mixture of linear and branched; thus, the RfD applies to the total linear and branched.

PFOA is a strong acid that is generally present in solution as perfluorooctanoate anion. It is water soluble and mobile in water, with an estimated log Koc of 2.06. PFOA is stable in environmental media because it is resistant to environmental degradation processes, such as biodegradation, photolysis, and hydrolysis. In water, no natural degradation has been demonstrated, and dissipation is by advection, dispersion, and sorption to particulate matter. PFOA has low volatility in ionized form, but can adsorb to particles and be deposited on the ground and into water bodies. Because of its persistence, it can be transported long distances in air or water, as evidenced by detections of PFOA in the arctic media and biota, including in polar bears, ocean-going birds, and fish found in remote areas (Lindstrom et al. 2011a; Smithwick et al. 2006). PFOA is present in ambient air and seawater globally (Ahrens et al. 2011; McMurdo et al. 2008; Yamashita et al. 2005; Young et al. 2007).

ED_005024_00000285-00016

2.2 Occurrence and Sources of Exposure

PFOA and other PFASs have been discharged into the environment during use as processing aids for fluoropolymers, by degradation of precursors, including fluorotelomer-based polymers, and throughout the life cycle of products containing these compounds (i.e., from the point of product manufacture through its use and disposal) (Washington et al. 2009, 2015a, 2015b). PFOA and other PFASs are man-made chemicals, but because of their widespread use and chemical and physical properties (persistence and mobility) they have been transported into ground water, surface waters (fresh, estuarine, and marine), and soils in the vicinity of their original source and at great distances. Point sources can result in significant exposure to people in some areas. Major sources of PFOA are described below.

2.2.1 Surface Water and Ground Water

Water resources (i.e., surface water and ground water) are susceptible to contamination by PFOA released from manufacturing sites, industrial use, fire/crash training areas, and industrial or municipal waste sites where products are disposed of or applied. PFOA and other PFASs have been reported in wastewater and biosolids as a result of manufacturing activities, disposal of coated paper and other consumer products, and from washing stain-repellant fabrics (Renner 2009). Historically, land application of biosolids has been a source of PFOA and other PFASs in surface water or ground water (Lindstrom et al. 2011b; Washington et al. 2010a, 2010b). The phase-out of the use of these compounds in the United States is expected to reduce PFASs in biosolids.

Some aqueous film forming foams used to combat aviation (or other hydrocarbon) fires release PFOA to the environment (Seow 2013; USEPA 2014b). Surface and ground water resources in close proximity to airports or other areas where these foams have been used can be contaminated (see Moody et al. 2002). PFOA was reported at concentrations as high as 105 μg/L in ground water near a concrete pad formerly used for military fire-training operations in Michigan (ATSDR 2005; Moody et al. 2003). Surface water concentrations as a result of a release of approximately 22,000 L of AFFF at L.B. Pearson International Airport in Toronto, Canada, resulted in peak PFOA concentrations of 11.3 μg/L at the confluence of Etobicoke Creek and Lake Ontario (Moody et al. 2002).

PFOA is not included as an analyte in the U.S. Geological Survey (USGS) National Water Quality Assessment Program, and it is not monitored in water as part of EPA's National Aquatic Resource Surveys. PFOA has been reported in U.S. water bodies, including the Tennessee River (< 25–598 nanograms per liter [ng/L]), Mississippi River (< 1.0–125 ng/L), Lake Erie (21–47 ng/L), Lake Ontario (15–70 ng/L), and the Conasuaga River (253–1,150 ng/L) and Altahama River (3.0–3.1 ng/L) watersheds in Georgia (Boulanger et al. 2004; Hansen et al. 2002; Konwick et al. 2008; Nakayama et al. 2010). In another study, the USGS collaborated with the University of Maryland and sampled three rivers and streams receiving effluent from 11 wastewater treatment facilities in the Chesapeake Bay watershed (USGS 2011); samples were collected in July and August 2010 from the Potomac River, the Patuxent River, and Saint Mary's Run. PFOA concentrations ranged from 3.6 to 20 ng/L in the Patuxent River; from 7.5 to 12 ng/L in the Potomac River; and from <2.0 to 47 ng/L in Saint Mary's Run.

ED_005024_00000285-00017

Studies show that PFOA occurs in marine waters. Yamashita et al. (2005) analyzed samples from the Pacific Ocean, South China Sea, and Mid-Atlantic Ocean, as well as samples from coastal waters of several Asian countries. PFOA was found at levels ranging from several thousand picograms per liter (pg/L) in water samples collected from coastal areas in Japan to tens of pg/L in the central Pacific Ocean. Yamashita et al. (2005) reported that PFOA was the predominant PFAS detected in oceanic waters, followed by PFOS.

2.2.2 Drinking Water

Under EPA's UCMR 3, PFOA was monitored by approximately 5,000 PWSs (all PWSs serving > 10,000 people, and a representative sample of 800 small PWSs) from 2013 through December 2015. The minimum reporting level (MRL) for PFOA in this survey was 0.02 µg/L. To-date, results for more than 36,000 samples have been reported by more than 4,800 PWSs for PFOA. The remainder of the results are expected to be reported by mid-2016. PFOA was measured at or above the MRL by approximately 2% of the PWSs. PFOA was reported above 0.07 µg/L by approximately 0.3% of PWSs that have reported results. Approximately 1% of PWSs have reported data for which combined PFOA and PFOS results are above 0.07 µg/L.

The Environmental Working Group's (EWG[3]) *National Drinking Water Database* includes PFOA analysis at 24 systems between 2004 and 2009 (EWG 2015). EWG obtained data primarily from state drinking water offices; the database includes data from 47,677 water systems in 45 states and the District of Columbia. The database showed that 24 systems reported analyzing for PFOA; of these, five systems in Minnesota reported finding detectable levels. Four of the systems had average concentrations below 0.01 µg/L. One system had an average concentration of 0.09 µg/L and a maximum reported concentration of 0.25 µg/L.

PFOA detections in source water and drinking water are reported in several published studies. These studies frequently involve targeted local sampling; thus, the findings are not representative of national occurrence. For example, in New Jersey, monitoring of raw and finished water between 2006 and 2008 revealed concentrations as high as 0.14 µg/L in finished drinking water (NJDEP 2007; Post et al. 2009). In another study, PFOA concentrations in Little Hocking, Ohio, ranged from 1.5 and 7.2 µg/L in the municipal water distribution system and up to 14 µg/L in private wells between 2002 and 2005 (Emmett et al. 2006). A study in Minnesota reported PFOA concentrations up to 0.9 µg/L in municipal, noncommunity, and private wells between 2004 and 2008 (Goeden and Kelly 2006).

2.2.3 Food

PFOA ingestion from food is an important exposure source. PFOA was detected in a variety of food products including snack foods, vegetables, meat, dairy products, human breast milk, and fish, using data from Europe and North America as reported by Trudel et al. (2008). In North America, snack foods, fish and shellfish, and potatoes were the food items estimated to contribute the most to PFOA exposure, under intermediate and high-exposure conditions. In a survey that included multiple food types, PFOA was the second-most frequently detected PFAS

[3] For more information see http://www.ewg.org.

ED_005024_00000285-00018

and was present at high concentrations relative to other related compounds (Hlouskova et al. 2013). In a 2011 assessment of exposure to Americans, Lorber and Egeghy (2011) concluded that food ingestion appears to be the primary route of exposure in adults, and dust and dietary ingestion is the major contributor for young children, under typical exposure conditions. Recent evidence shows that PFOA levels in food have been declining (Johansson et al. 2014).

Schecter et al. (2010) collected 10 samples of 31 commonly consumed foods from five grocery stores in Dallas, Texas, in 2008 and analyzed them for PFOA. Equal weights of each sample were combined and composited for analysis. Dietary intakes were estimated using data from the 2007 U.S. Department of Agriculture food availability data set. For concentrations below the limit of detection, a value of zero was assigned. The estimated per capita daily estimate for exposure to PFOA was 60 nanograms per day (ng/day), or about 0.75 ng/day for an 80 kilogram (kg) adult. Based on a graphic presentation in the published paper, meat products (n = 8) accounted for about 40 ng/day with the remaining 30% equally distributed between fish (n = 7), vegetables (n = 7: three fat [olive oil, canola, margarine], one cereal, one apple, one potato, and one peanut butter sample), and dairy and egg products (n = 9).

Tittlemier et al. (2007) conducted a Canadian total diet study that collected and analyzed 54 composite food samples. Samples were collected from 1992 to 2004 and represented fish and seafood, meat, poultry, frozen entrées, fast food, and microwave popcorn. PFASs were detected in nine composites (four meat, three fish and shellfish, one fast food, and one microwave popcorn). PFOA and PFOS were most frequently found. The authors concluded that diet represented approximately 60% of total PFASs exposure. PFOA was detected in roast beef, pizza, and microwave popcorn at 0.74 to 3.6 ng/g, wet weight. The average daily PFOA exposure was estimated at 70 ng.

Several studies are available from countries in Western Europe with diets that are comparable to those in the United States. Fromme et al. (2007) collected duplicate diets for 15 male and 16 female healthy subjects (16 to 45 years old) in Germany. The median daily dietary intake for PFOA was 2.9 nanograms per kilogram of body weight (ng/kg) (232 ng/day for an 80 kg adult), with a 90th percentile intake of 672 ng/day. Haug et al. (2010) estimated exposures in Norway using a market basket approach comprised of 21 foods, three drinking water samples, one milk sample, and one tea sample. Total PFOA intake was estimated as 31 ng/day for the general Norwegian population. The highest levels were found in coffee, tea and cocoa (2.1 ng/day), root vegetables/potatoes (0.66 ng/day), tap water (0.54 ng/day), soft drinks (0.45 ng/day), and eggs (0.49 ng/day). Noorlander et al. (2011) estimated mean long-term daily intakes of 0.2 ng/kg (16 ng/day for an 80 kg adult) in the Netherlands using a pooled composite from foods purchased in retail chains with nationwide coverage; the 99th percentile value was 0.5 ng/kg (40 ng/day). Important sources were vegetables, fruits, and flour.

Human studies have shown that PFOA is transferred from mother to infant via cord blood and breast milk. A recent study showed that breast milk contributed > 83% of the PFOA exposure in 6-month-old infants (Haug et al. 2011). Additional information on concentrations of PFOA in breast milk is provided in section 2.5.

PFOA has been detected in beef as a result of cattle ingesting contaminated feed. When cattle were exposed to feed contaminated with 13 ng PFOA/kg wet weight, PFOA accumulated in the liver (9 ng/kg) and in muscle (7 ng/kg) (Vestergren et al. 2013). The study also detected PFOA

ED_005024_00000285-00019

in cow's milk at 6.7 ng/L. In addition, evidence suggests that livestock accumulate PFOA by grazing in fields where biosolids were applied (Renner 2009; Vestergren et al. 2013).

Bioaccumulation in fish and other edible aquatic organisms is another route for potential dietary exposures (Bhavsar et al. 2014; Renzi et al. 2013; Stahl et al. 2014). EPA analyzed fish fillet tissue samples from U.S. rivers and from the Great Lakes as part of EPA's National Aquatic Resource Surveys. These analyses included characterizing perfluorinated compounds (PFCs) in freshwater fish on a national scale during EPA's 2008–2009 National Rivers and Streams Assessment, and on a regional scale during the Great Lakes Human Health Fish Tissue Study component of the EPA 2010 National Coastal Condition Assessment. Fish were collected from randomly selected locations, including 162 urban river sites and 157 nearshore Great Lake sites, and analyzed for 13 PFASs. Results showed that 80% of urban river fish samples and 100% of Great Lakes fish samples contained some detectable PFASs. PFOS was the most frequently detected chemical (in 73% of river fish samples and 100% of Great Lakes fish samples). PFOA was not detected in river fish fillet samples, but it was detected in 12% of the Great Lakes samples. In the 2010 Great Lakes sampling, PFOA was detected in 19 out of 157 samples at a maximum concentration of 0.97 ng/g. The differences in PFOA detections between river and Great Lakes fish samples could be due to the availability of a more sensitive PFAS analytical method with lower detection limits when the Great Lakes study was initiated. Cooking of fish does not reduce the levels of PFOA in the fish (or the consumer's dietary exposure) (Bhavsar et al. 2014).

PFOA has been detected in wild caught and farmed fish, presumably because of bioaccumulation and/or trophic transfer. Bhavsar et al. (2014) found that PFOA concentrations were higher in wild-caught fish than farmed fish, and suggested that fish caught near contaminated sites could represent an important exposure source among recreational and subsistence fishers.

In a survey of French adult freshwater anglers, PFOA was a major contributor of total PFAS exposure from fish. Although some individuals had higher exposures, overall values for this population were close to those for the general population (Denys et al. 2014). In a study of French adults who consumed large amounts of seafood (n = 993), mean lower bound exposure to PFOA was 1.16 ng/kg/day (92.8 ng/day for an 80 kg adult) compared to a lower bound of none in the general population (n = 1918). The mean upper bound values were 2.06 and 0.74 ng/kg/day (164.5 to 59.2 ng/day), respectively, for the same highly exposed and general population groups (Yamada et al. 2014). In a sub-study that was restricted to 106 pregnant women, the upper bound mean was 1.52 ng/kg/day (121.6 ng/day) and the 95th percentile upper bound was 2.41 ng/kg/bw/day (192.8 ng/day).

PFOA can occur in plants grown in soils containing PFOA. For example, PFOA was taken up by corn when grown in biosolid-amended soil; however, the chemical remained in the roots and did not accumulate in edible parts of the plant (Krippner et al. 2014). PFOA accumulation in fruit crops tends to be lower than in shoot or root crops, presumably because there are more compartments through which PFOA would have to pass to reach the edible portion of the plant (Blaine et al. 2014).

ED_005024_00000285-00020

PFOA was previously used in the manufacturing of several types of food packaging; in January 2016, the U.S. Food and Drug Administration (FDA) amended its food additive regulations to no longer allow for the use of perfluoroalkyl ethyl-containing food-contact substances as oil and water repellants for paper and paperboard that comes in contact with aqueous and fatty foods (81 FR 5). PFOA is a breakdown product of the perfluorooctylethanol telomer alcohol used to make coatings for or additives in food contact paper where it adds a moisture or oil barrier to paper-type packaging, including microwave popcorn bags, fast food wrappers, candy wrappers, and pizza box liners (Begley et al. 2005). When used in this way, PFOA can migrate into foods from the packaging material. In a study conducted by FDA, Begley et al. (2005) was able to extract (4 micrograms per square decimeter [$\mu g/dm^2$] paper) of PFOA into food oil before cooking and another 7 $\mu g/dm^2$ from paper after cooking. Based on these results, Begley et al. (2005) concluded that paper with treated coatings had a high potential for migration of fluorochemical to food.

Food can become contaminated with PFOA from preparation in nonstick cookware coated with polytetrafluoroethylene (PTFE) (Teflon™). PFOA is a processing aid in the manufacture of PTFE. Begley et al. (2005) also evaluated migration of PFOA to foods from cooking in Teflon™-lined cookware and found it to be much lower (0.03 $\mu g/dm^2$ polymer) than migration from coated paper. In this study, new pans leached more compared to those that had been used before.

2.2.4 Ambient Air

A number of PFASs are precursors of PFOA and degrade to PFOA in the environment via biotic and abiotic degradation. Some of these precursors are volatile and contribute to the formation of airborne PFOA. Indoor air sampling reportedly contains higher concentrations of these precursors than outdoor air (Vierke et al. 2012). Langer et al. (2010) reported detections of PFOA and precursors in indoor air samples from home residences and at stores that sold outdoor equipment, furniture, and carpet. Fraser et al. (2013) found that PFOA in serum was significantly correlated with air levels collected in offices, likely associated with carpeting, furniture, and paint.

PFOA can be emitted from nonstick cookware coated with PTFE. Schlummer et al. (2015) found that at typical cooking temperatures (< 230°C), perfluoroalkylcarboxylic acids (C4 to C12) dominated (4.75 ng per hour) by PFOA and perfluorobutanoic acid (PFBA) were released to the atmosphere; when pans were overheated PFBA and perfluoro-n-pentanoic acid (PFPeA) were dominant (> 260°C). Emissions were far greater at higher temperatures (12,190 ng per hour at 370 °C; Schlummer et al. 2015). Emissions are expected to decline with use of the product. The authors hypothesized that most of the emissions would end up in household dusts.

Based on its environmental fate properties, PFOA has low volatility. However, PFOA has been reported in ambient air, largely bound to particulate matter. It can be transported long distances via the atmosphere and has been detected at low concentrations in areas as remote as the Arctic (Shoeib et al. 2006) and Antarctic (Del Vento et al. 2012). PFOA levels in outdoor air were measured in a variety of locations, most of which are countries outside the United States. Fromme et al. (2009) reported mean levels of 2 picograms per cubic meter (pg/m^3) in particulate matter for eight samples collected in the summer in Albany, New York with a mean of 3.2 pg/m^3

ED_005024_00000285-00021

present in the gas phase. Mean air concentrations in Spain and England were 6.1 pg/m^3 and 3.5 pg/m^3, respectively (Beser et al. 2011; Goosey and Harrad 2012). In a study conducted in China, airborne PFOA concentrations were similar (Liu et al. 2015). Areas near wastewater treatment plants, waste incinerators, and landfills can be point sources for PFOA in outdoor air (Ahrens et al. 2011). PFOA-derived telomer alcohols can also be present in air (Jogsten et al. 2012).

2.2.5 Indoor Dust

Because of its widespread use in carpets, upholstered furniture, and other textiles, PFOA has been detected in indoor dust from homes, offices, vehicles, and other indoor spaces. Although some of these uses have been phased out, exposure could continue in legacy products and imported goods. As reported by Fraser et al. (2013), particulate matter from fabrics and carpeting are believed to be the source for the PFOA-containing dusts found in homes, offices, and automobiles.

A 2013 survey (Fraser et al. 2013) detected PFOA in samples of house dust (23.7 ng/g), office dust (32.0 ng/g), and vehicles (11.4 ng/g) collected at sites by 31 participants in Boston, Massachusetts. The Wisconsin Department of Health and Human Services collected vacuum cleaner contents from 39 homes as a means of evaluating the concentration of PFOA and 15 other PFASs in dust (Knobeloch et al. 2012). The median PFOA concentration was 44 ng/g. PFOA, PFOS and perfluorohexane sulfonate (PFHxS) accounted for about 70% of the total PFASs present in the dust. Lorber and Egeghy (2011) assessed Americans' PFOA exposure and concluded that ingestion of household dust and food are primary routes of PFOA exposure for 2-year-old children. For median exposed children, exposures were estimated to be 13 and 8 ng/d from dust and food, respectively. For highly exposed children (at the 95th percentile), PFOA exposure from dust was estimated to be three times that from food.

Jogsten et al. (2012) collected dust samples from 10 selected homes in Catalonia, Spain, and analyzed them for 20 PFASs. All samples contained PFOA; the levels ranged from 1.5 to 13.9 ng/g. An important outcome of this study was the identification of PFOA volatile telomer alcohol derivatives in the dust samples at concentrations of up to 1.3 ng/g. The 8:2 telomer alcohols degrade metabolically to PFOA once ingested. A study conducted in Belgium also found that PFOA was present in home (median: 0.7 ng/g dry weight) and office dust (median: 2.2 ng/g dry weight) (D'Hollander et al. 2010). The highest of the indoor dust concentrations of those sampled (114 ng/g) were found in homes in Germany (Xu et al. 2013).

2.2.6 Soils

PFOA persists in soils near manufacturing facilities and disposal sites (Xiao et al. 2015) and in areas, such as military bases, where firefighting foams containing PFOA were heavily used (Filipovic et al. 2015). Measured concentrations of PFOA in surface soils range from 8.0 ng/g (Xiao et al. 2015) to 287 ng/g (Filipovic et al. 2015). These studies focused on two sites, the first in the Minneapolis–St. Paul, Minnesota, metropolitan area where PFASs were manufactured and disposed of, and the second on a former military airport in Sweden abandoned in 1994, where firefighting foams containing PFOA had been used. In both cases, there was ground water contamination. Xiao et al. (2015) determined that levels of PFOA in soils increased with depth,

ED_005024_00000285-00022

providing evidence for migration into ground water (see also section 2.2.1). Filipovic et al. (2015) found that PFOA concentrations in soil cores remained high more than 30 years after usage was discontinued.

Incidental ingestion of soils represents a potential exposure route for PFOA. Regional and geographic differences in soil characteristics can influence PFOA concentrations. Soil contamination tends to occur at manufacturing sites of producers and users, where disposal of treated products has occurred (i.e., landfills), and potentially where biosolids containing PFASs are applied. Calculated residence time in soils suggests that persistence in the environment will extend well beyond the time that PFOA manufacturing ends (Zareitalabad et al. 2013). Contaminated soils also can be transported offsite via water and wind.

2.2.7 Biosolids

Biosolids are sometimes applied as an amendment to soils as fertilizers; in some cases, the biosolids can contain PFOA. For example, in May 2007, a Decatur, Alabama, manufacturer that used PFASs notified the Decatur Utilities Dry Creek Waste Water Treatment plant that it had unknowingly discharged large amounts of perfluorocarboxylic acid precursors (PFOA and perfluorododecanoic acid [PFDA]) to the utility (USEPA 2011a). The Decatur treatment plant also received wastewater from several other industries in the area that manufactured or used a variety of PFAS-containing materials. The incident was reported to EPA and other government agencies because biosolids from the wastewater plant had been applied to 5,000 acres of privately owned agricultural fields for the previous 12 years (1996 to 2008).

Testing revealed that the biosolids from the Decatur plant contained PFOA, PFOS and other PFASs. Concentrations in nine soil samples from the area ranged from 589 to 1,296 parts per billion (ppb) PFOA and 55 to 2,531 ppb PFOS. Subsequently, private wells, ponds, and other surface waters near the biosolids application sites were sampled and found to contain PFOS and PFOA, in some cases at levels greater than EPA's provisional HA values. Several additional rounds of sample collection from the impacted areas confirmed the presence of PFASs, including PFOA and PFOS in the media tested (Lindstrom et al. 2011b; USEPA 2011a; Washington et al. 2010a, 2010b).

PFASs were not analyzed in the 2004 EPA Total National Sewage Sludge Survey (TNSSS), as analytical methods were not available when analytes were selected. Venkatesan and Halden (2013) re-analyzed archived samples for PFASs from the TNSSS in five composite samples, which represented 94 wastewater treatment facilities from 32 U.S. states and the District of Columbia in 2001. PFOS was the most abundant PFC identified (mean 403± 127 μg/kg dry weight), followed by PFOA (mean 34 ± 22 μg/kg dry weight). Armstrong et al. (2016) collected biosolid samples every 2 months from a large municipal water recovery facility between 2005 and 2013. The highest mean PFOA concentration reported was 23.5 μg/kg dry weight. Yoo et al. (2011) found PFOA and PFOS in plants (fescue, barley, bluegrass, and Bermuda grass) grown in soils amended with biosolids. Concentrations of PFOA ranged from 9.9 to 202.7 μg/kg. Concentrations in biosolids are expected to decline because of the phase-out of the use of PFOS and PFOA in manufacturing and industrial processes.

ED_005024_00000285-00023

2.2.8 Consumer Products

Other materials that result in potential human exposure include legacy use and imported goods or continuing uses. Some examples of these uses are listed below.

- Stain/water repellants on clothing, bedding materials, upholstered furniture, carpets, and automobile interiors (e.g., Stainmaster™, Zonyl™, Nuva™, Unidyne™, Baygard™) (Walters and Santillo 2006); these materials can be a particularly important exposure route for infants and children because of their hand-to-mouth behaviors.
- Cooking surfaces (e.g., Teflon™)
- Toothpaste, shampoos, cosmetics
- Polishes and waxes
- Electronics
- Flame repellants
- Paints, varnishes, sealants
- Lubricants/surfactants/emulsifiers (continuing use)
- Food containers and contact paper[4]
- Pesticide
- Aqueous film forming foams (continuing use; used for firefighting)
- Electronics
- Textiles (e.g., Gore-Tex™) and leather
- Plumbing tape
- Cleaning products

2.3 Environmental Fate

2.3.1 Mobility

PFOA is water soluble and has been found in surface water, ground water, and drinking water. It has low volatility in ionized form, but can adsorb to particles in air; because of its persistence, it can be transported long distances to the Arctic (Shoeib et al. 2006) and Antarctic (Del Vento et al. 2012). PFOA has a log K_{oc} of 2.06 and does not easily adsorb to sediments or aquifer materials; therefore, it tends to stay in the water column.

2.3.2 Persistence

PFOA is stable in the environment and resistant to hydrolysis, photolysis, volatilization, and biodegradation (see Table 2-1). No biodegradation or abiotic degradation processes have been found; the only dissipation mechanisms in water are dilution, advection, and sorption. Yamada et

[4] PFOA was used in some grease-proofing paper coatings or additives that can contribute to its presence in foods (Begley et al. 2005). However, in January 2016, FDA amended their food additive regulations to no longer allow for the use of perfluoroalkyl ethyl containing food-contact substances as oil and water repellants for paper and paperboard for use in contact with aqueous and fatty foods (81 FR 5).

ED_005024_00000285-00024

al. (2005) determined that typical municipal waste incinerators destroy PFOA on textiles and paper and do not release it into the atmosphere.

2.3.3 Bioaccumulation

Several criteria can be used to assess bioaccumulation, including octanol-water partition coefficient (K_{ow}), bioconcentration factors (BCF), bioaccumulation factors (BAFs), and biomagnification or trophic magnification factors (BMFs or TMFs, respectively) (Gobas et al. 2009). The K_{ow} and BCF metrics are typically based on partitioning of organic chemicals into octanol or lipids of biota. For PFOA, partitioning appears to be more related to protein binding properties than its lipid partitioning. Thus, the K_{ow} is not a reliable measure of bioaccumulation potential for PFOA (EFSA 2008; UNEP 2015). Information from field studies, BCFs, BMFs, and TMFs provide the most conclusive evidence of accumulation of chemicals in food webs (Gobas et al. 2009) and are the more appropriate metrics for gauging the potential for accumulation of PFOA in fish, wildlife, and humans.

Because of the physical-chemical properties of PFOA, K_{ow} cannot be reliably measured (Table 2-1; UNEP 2015; USEPA 2014b). Model estimates of K_{ow} have been reported; however, verification that these chemicals are within the domain of the models is often not provided. Therefore, the validity of the use of such models is questionable (EFSA 2008; UNEP 2015), Available BCFs determined from lab studies have been reported and generally fall below traditional criteria used to assess bioaccumulation (e.g., Martin et al. 2003c). It is recognized, however, that BCFs determined by existing standard methods derived from lipid-partitioning are not an appropriate metric for assessing bioconcentration of PFOA (EFSA 2008; UNEP 2015). Although evidence of PFOA accumulation in many organisms has been documented, reported BAFs and BCFs for the chemical also fall below traditional criteria used to assess bioaccumulation potential (Loi et al. 2011; Martin et al. 2003a, 2003b; Morikawa et al. 2005; Quinete et al. 2009).

Field evidence of PFOA biomagnification, considered to be the preferable metric for assessing bioaccumulation potential (Gobas et al. 2009), has been documented in many organisms from many locations worldwide (UNEP 2015). Trophic magnification has also been evaluated (Environment Canada and Health Canada, 2012; Houde et al. 2006; Kelly et al. 2009; Loi et al. 2011; Martin et al. 2004). Some field trophic studies revealed TMFs greater than 1, which indicates that PFOA accumulated and increased in concentration with increasing trophic level; other studies reported TMFs less than 1 for some food webs. The weight of evidence for trophic magnification was deemed sufficient to consider PFOA to be bioaccumulative by the Stockholm Convention Persistent Organic Pollutants Review Committee (UNEP 2015).

2.4 Toxicokinetics

Uptake and egress of PFOA from cells is largely regulated by transporters in cell membranes (Anzai et al. 2006; Cheng et al. 2006; Klaassen and Aleksunes 2010; Nakagawa et al. 2007, 2009; Weaver et al. 2009, 2010; Yang et al. 2010). PFOA is absorbed from the gastrointestinal tract as indicated by the serum measurements in humans and treated animals. In serum, it is electrostatically bound to albumin, occupying nine to 12 sites, and sometimes displaces other substances such as nutrients and pharmaceuticals that normally would occupy a site (MacManus-

ED_005024_00000285-00025

Spencer et al. 2009; Salvalaglio et al. 2010; Wu et al. 2009a). Linear PFOA chains display stronger binding than branched chains (Beesoon and Martin 2015). Binding causes a change in the conformation of serum albumin, thereby changing its affinity for the endogenous compounds it normally transports.

PFOA is distributed to tissues by a process requiring transporters. Accordingly, the tissue levels vary from organ to organ as demonstrated by Kemper (2003). The highest tissue concentrations are usually those in the liver. Liver accumulation in males is greater than that in females. Other tissues with a tendency to accumulate PFOA are the kidneys, lungs, heart, and muscle, plus the testes in males and uterus in females (Kemper, 2003). PFOA is not metabolized, thus any effects observed in laboratory toxicological studies are the result of parent compound, not metabolites.

Electrostatic interactions with proteins are an important toxicokinetic feature of PFOA. Studies demonstrate binding or interactions with receptors (e.g., peroxisome proliferator activated receptor alpha [PPARα], triiodothyronine [T3]), transport proteins and enzymes (Luebker et al. 2002; Weiss et al. 2009; L. Zhang et al. 2013). Saturable renal resorption of PFOA from the glomerular filtrate via transporters in the kidney tubules is a major contributor to the long half-life of this compound in humans (Nakagawa et al. 2007, 2009; Weaver et al. 2010; Yang et al. 2009, 2010). Branched-chain PFOAs are less likely to be resorbed than the linear molecules based on half-life information in humans (Y. Zhang et al. 2013). All toxicokinetic models for PFOA are built on the concept of saturable renal resorption first proposed by Andersen et al. (2006). Some PFOA is removed from the body with bile (Genuis et al. 2010), a process that also is transporter-dependent. Accordingly, the levels in fecal matter represent both unabsorbed material and material that is discharged to the intestines with bile.

During pregnancy, PFOA is present in the placenta and amniotic fluid in both animals (Fenton et al. 2009; Hinderliter et al. 2005) and humans (T. Zhang et al. 2013). Post-delivery, PFOA is transferred to offspring through lactation in a dose-related manner (Hinderliter et al. 2005, Fenton et al. 2009). Maternal serum levels decline as those in the pups increase. This also occurs in humans as demonstrated in the study by Mondal et al. (2014) of breastfeeding women and their infants in West Virginia and Ohio.

The half-life in humans for occupationally exposed workers (Olsen et al. 2007) was 3.8 years (95% CI [1.5, 9.1]). Bartell et al. (2010) determined an average half-life of 2.3 years based on a study of the decreases in human serum levels after treatment of drinking water for PFOA removal was instituted by the Lubeck Public Services District in Wqest Virginia and the Little Hocking Water Association in Ohio. This is the value used for humans in this assessment because it applies to the general population and reflects humans whose exposure came primarily from their PWS. Half-lives are reported to be shorter in animals than for humans: 21 days (females) and 30 days (males) for monkeys (Butenhoff et al. 2004b); 11.5 days (males) and 3.4 hours (females) for Sprague-Dawley rats (Kemper 2003); 27.1 days (male) and 15.6 days (female) for CD-1 mice (Lau et al. 2006). Although the animal half-lives are shorter than humans, so, too, are their average lifetimes. In early life, the half-lives are nearly the same for both genders, but once the animals reach sexual maturity resorption increases in male rats, prolonging the half-lives (Hinderliter et al. 2006; Hundley et al. 2006). This change appears to be under the control of hormones in both males and females (Cheng et al. 2006; Kudo et al. 2002).

ED_005024_00000285-00026

2.5 Human Biomonitoring Data

The Fourth National Report on Human Exposure to Environmental Chemicals from the Centers for Disease Control and Prevention (CDC 2009) included exposure data for PFOA from 2003 to 2004 collected by the National Health and Nutrition Examination Survey (NHANES). PFOA was detected in 99.7% of the general U.S. population. Since that time, CDC has issued several updates, the most recent of which was released in 2015 (CDC 2015). Taken together, the data suggest that PFOA concentrations in human serum in the U.S. generally declined between 1999 and 2012. The geometric mean PFOA concentration in human serum decreased from 5.2 to 2.1 μg/L, and the 95th percentile concentration decreased from 11.9 to 5.7 μg/L. During this time, there has been a major reduction in environmental emissions by the manufacturers as well as a phase-out of production of C-8 compounds in the United States. Analysis of the NHANES 2003–2004 subsample demonstrated higher levels of PFOA and PFOS in males and a slight increase in levels of PFOS with age (Calafat et al. 2007a, 2007b).

Precursors might also form PFOA in the body; this represent an important uncertainty in characterizing exposure as measured by blood serum. For example, Lorber and Egeghy (2011), indicated that the precursor fluorotelomer alcohols (FTOHs) and polyfluoroalkyl phosphoric acids (PAPs) would add to exposure but there is uncertainty as to the magnitude of the effect. The authors concluded that precursors "could very well contribute half or more of what is eventually measured as PFOA in the blood."

Evidence shows that PFOA is distributed within the body and can be transferred from pregnant women to their unborn children and offspring. T. Zhang et al. (2013) collected serum and cord blood samples from 30 pregnant women in China. The maternal blood contained variable levels of 10 PFASs, eight acids, and two sulfonates. The mean maternal blood concentration for PFOA was 3.35 nanograms per milliliter (ng/mL). The mean was greater than the median, indicating a distribution skewed toward the higher concentrations. Compared to the mean PFOA blood levels in the pregnant women, the mean levels in cord blood (1.95 milligrams per milliliter [mg/mL]) was 47% of that in the mother's blood.

PFOA has been detected in breast milk (Tao et al. 2008; Völkel et al. 2008) and cord blood (Apelberg et al. 2007; Monroy et al. 2008) at concentrations above the limit of quantification. Mondal et al. (2014) evaluated serum samples from breastfeeding women and their infants in West Virginia and Ohio. For each month of breastfeeding, maternal serum levels of PFOA were reduced by 3% (95% CI: 2%-5%) and infant serum levels increased by 6% (95% CI: 1%-10%]). A publication from the French total diet study (Cariou et al. 2015) also examined human breast milk as an exposure route for infants using 61 mother-infant pairs. PFOA was detected in 77% of the breast milk samples, with a mean concentration of 0.041 ng/mL and a maximum concentration of 0.308 ng/mL. The regression coefficient for the association between the maternal serum concentration and the detected breast milk concentrations was 0.72 (n = 10).

ED_005024_00000285-00027

3.0 PROBLEM FORMULATION

3.1 Conceptual Model

The conceptual model provides useful information to characterize and communicate the potential health risks related to PFOA exposure from drinking water. The sources of PFOA, the routes of exposure for biological receptors of concern (e.g., various human activities related to ingested tap water such as drinking, food preparation, and consumption), and the potential assessment endpoints (e.g., effects such as liver toxicity and developmental effects), and adverse health effects in the populations at risk due to exposure to PFOA are depicted in the conceptual diagram below (Figure 3-1).

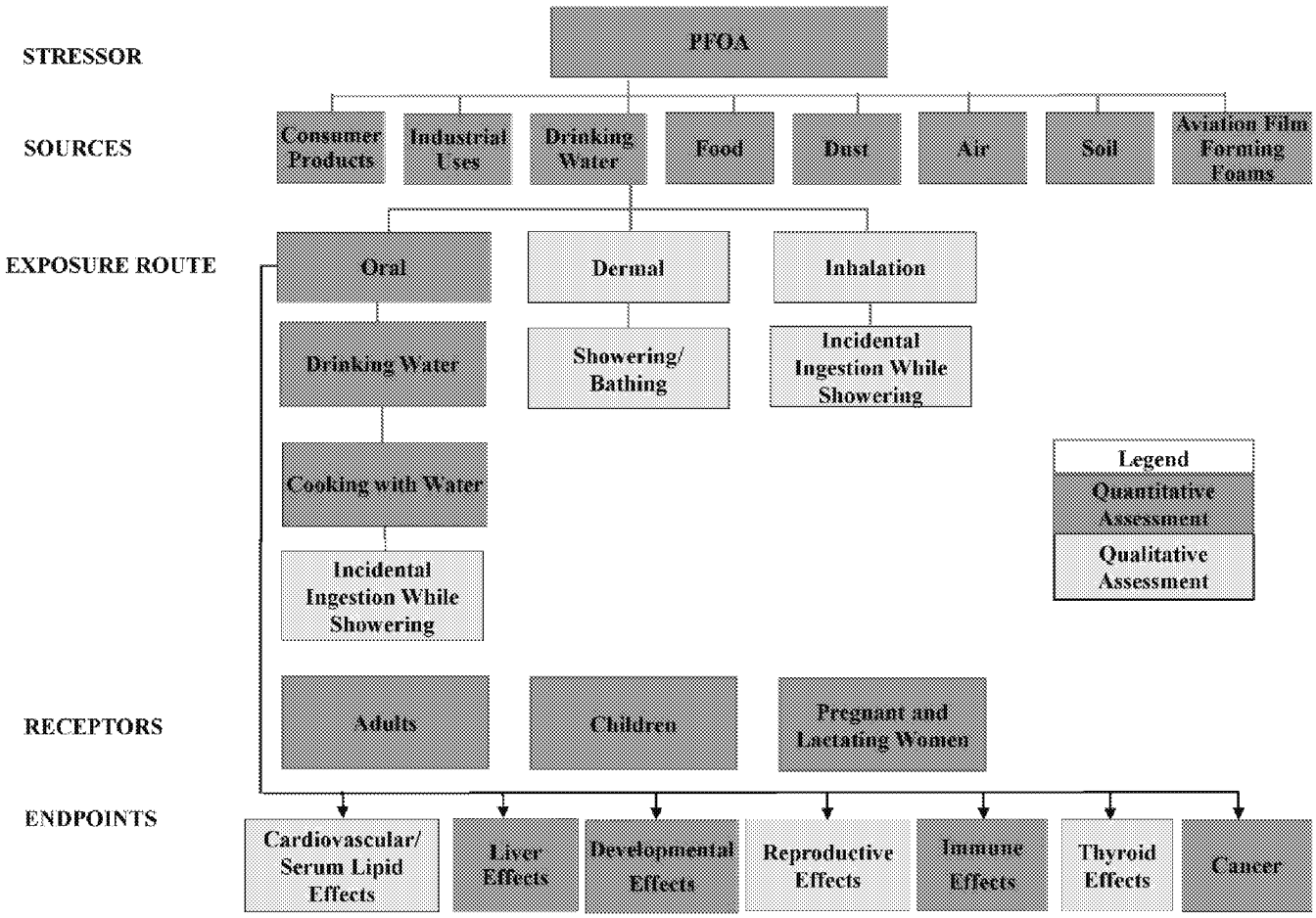


Figure 3-1. Conceptual Model for PFOA in Finished Drinking Water

ED_005024_00000285-00028

3.1.1 Conceptual Model Diagram for Exposure via finished Drinking Water

The conceptual model is intended to explore potential links of exposure to a contaminant or stressor with the adverse effects and toxicological endpoints important for management goals, including the development of drinking water HA values. Boxes that are more darkly shaded indicate pathways that were considered quantitatively in estimating the advisory level, whereas the lightly shaded boxes were only considered from a qualitative perspective.

3.1.2 Factors Considered in the Conceptual Model for PFOA

Stressors: For this HA, the stressor is PFOA in drinking water from public water facilities or private wells.

Sources: Sources of PFOA include both ground and surface waters used for drinking. Multiple potentially important sources of PFOA and precursors exist in addition to drinking water, such as foods, indoor dust in a home or work environment, indoor and outdoor air, soil, consumer products within the homes or place of work including children's schools, and industrial products. The relative contribution of drinking water versus other sources is addressed in the Relative Source Contribution section of the document (section 3.2.5). This HA applies only to drinking water.

Routes of exposure: Exposure to PFOA from contaminated drinking water sources can occur via oral exposure (drinking water, cooking with water, and incidental ingestion from showering); dermal exposure (contact of exposed parts of the body with water containing PFOA during bathing or showering, dishwashing); and inhalation exposure (during bathing or showering or using a humidifier or vaporizer). There is limited information identifying health effects from inhalation or dermal exposures to PFOA in humans and animals. Therefore, these routes of exposure are not quantitatively used in the derivation of the HA. PFOA has a low vapor pressure and is not expected to be present in air except as bound to particulate matter and in aerosols formed from devices such as shower heads and humidifiers that aerosolize tap water. Toxicity data are available for oral exposure from drinking water, but not the other exposure routes (inhalation and dermal exposures). PFOA is not removed by heating water and can increase in concentration when the water is boiled.

Receptors: The receptors are those in the general population (adults, infants and children) who could be exposed to PFOA from tap water through dermal contact and inhalation and/or ingestion at their homes, workplaces, schools, and daycare centers.

Endpoints: Epidemiology data report associations between PFOA exposure and high cholesterol, increased liver enzymes, decreased vaccination response, thyroid disorders, pregnancy-induced hypertension and preeclampsia, and cancer (testicular and kidney) (see section 4.1.2). These studies provide varying levels of support for the effects associated with PFOA exposure in the animals studies used for quantification of the HA. Cholesterol, liver enzymes, and thyroid effects were examined in numerous studies in different populations, but the pregnancy complications of hypertension and preeclampsia in women and testicular cancer in young men were only studied in a high-exposure community (located in the vicinity of a PFOA production plant in West Virginia; i.e., C8 Health Project). The C8 Science Panel assessed the links between PFOA and several diseases and concluded that a probable link existed between

ED_005024_00000285-00029

PFOA and the observed kidney and testicular tumors among the population evaluated (see section 4.1.2).

The associations for most epidemiology endpoints are mixed. Although mean serum values are presented in the human studies, actual estimates of PFOA exposure (i.e., doses/duration) are not currently available. Thus, the serum level at which the effects were first manifest and whether the serum had achieved steady state at the point the effect occurred cannot be determined. It is likely that some of the human exposures that contribute to serum PFOA values come from PFOA derivatives or precursors that break down metabolically to PFOA. These compounds could originate from PFOA in diet and materials used in the home, which creates potential for confounding. In addition, most of the subjects of the epidemiology studies have many PFASs and/or other contaminants in their blood. Although the study designs adjust for other potential toxicants as confounding factors, their presence constitutes a level of uncertainty that is usually absent in the animal studies.

Taken together, the weight of evidence for human studies supports the conclusion that PFOA exposure is a human health hazard. At this time, EPA concludes that the human studies are adequate for use qualitatively in the identification hazard and are supportive of the findings in laboratory animals. EPA plans to begin another effort to determine the range of perfluoroalkyl compounds for which an Integrated Risk Information System (IRIS) assessment is needed, as indicated in the 2015 IRIS Multi Year Agenda.[5]

For PFOA, oral animal studies of short-term, subchronic, and chronic duration are available in multiple species including monkeys, rats, and mice (see section 4.1.1). Adverse effects observed following exposure to PFOA include liver toxicity (hypertrophy, necrosis, and effects on the metabolism and deposition of dietary lipids), kidney toxicity, and developmental effects (survival, body weight changes, reduced ossification, altered puberty, and retarded mammary gland development), immune effects, and cancer. EPA quantitatively evaluated (i.e., modeled serum concentrations) for the liver, developmental, immune, and cancer effects.

In most animal studies, changes in relative and/or absolute liver weight appears to be the most common effect observed with or without other hepatic indicators of adversity identifying increased liver weight as a common indicator of PFOA exposure. The liver also contains the highest levels of PFOA when analyzed after test animal sacrifice. The increases in liver weight and hypertrophy, however, also can be associated with activation of cellular PPARα receptors, making it difficult to determine if this change is a reflection of PPARα activation or an indication of PFOA toxicity. The PPARα response is greater in rodents than it is in humans. EPA evaluated liver disease and liver function resulting from PFOA exposure in studies where liver weight changes and other indicators of adversity such as necrosis, inflammation, fibrosis and/or steatosis (fat accumulation in the liver) or increases in liver or serum enzymes indicative of liver damage were observed. Only the doses associated with the adverse effects were used for the quantification of risk.

[5] For more information on the IRIS agenda see https://www.epa.gov/iris/iris-agenda.

ED_005024_00000285-00030

3.2 Analysis Plan

3.2.1 Health Advisory Guidelines

Assessment endpoints for HAs can be developed for both short-term (1-day and 10-day) and lifetime exposure periods using information on the noncarcinogenic and carcinogenic toxicological endpoints of concern. Where data are available, endpoints reflect susceptible and/or more highly exposed populations.

- A 1-day HA is typically calculated for an infant (0 to 12 months or a 10-kg child), assuming an acute exposure to the chemical; it is generally derived from a study of less than 7 days duration.
- A 10-day HA is typically calculated for an infant (0 to 12 months or a 10-kg child), assuming a limited period of exposure of 1 to 2 weeks; it is generally derived from a study of 7 to 30 days duration.
- A lifetime HA is derived for an adult (> 21 years old or an 80-kg adult), and assumes an exposure period over a lifetime (approximately 70 years). It is usually derived from a chronic study of 2 years duration, but subchronic studies can be used by adjusting the uncertainty factor employed in the calculation. For carcinogens, the HA documents typically provide the concentrations in drinking water associated with a range of risks (from one excess cancer case per 10,000 persons exposed to one excess cancer case per million persons exposed) for Group A and B carcinogens and those classified as known or likely carcinogens (USEPA 1986, 2005). Cancer risks are not provided for Group C carcinogens, or those classified as "suggestive," unless the cancer risk has been quantified.

3.2.2 Establishing the Data Set

The *Health Effects Support Document for Perfluorooctanoic Acid (PFOA)* (USEPA 2016a) provides the health effects basis for development of the HA, including the science-based decisions providing the basis for estimating the point of departure (POD). To develop the HESD for PFOA, EPA assembled available information on toxicokinetics, acute, short-term, subchronic and chronic toxicity along with developmental and reproductive toxicity, neurotoxicity, immunotoxicity, genotoxicity and cancer in humans and animals. For a more detailed description of the literature review search and strategy for inclusion and exclusion of studies, see the Forward and Appendix A of the HESD for PFOA.

Briefly, through a literature search, studies were identified for retrieval, review, and inclusion in the document using the following criteria:

- The data contribute substantially to the weight of evidence for any of the toxicity endpoints.
- Elements of the study design merit its inclusion in the draft assessment based on its contribution to the mode of action (MOA) or the quantification approach.
- The study elucidates the MOA for any toxicity endpoint or toxicokinetic property associated with PFOA exposure.
- The effects observed differ from those in other studies with comparable protocols.

ED_005024_00000285-00031

- The study was relevant to drinking water exposures and to the U.S. population.

In addition, an evaluation of available data was performed by EPA to determine data acceptability. The following study quality considerations from U.S. EPA's (2002) *A Review of the Reference Dose and Reference Concentration Processes* were used in selection of the studies for inclusion in the HESD and development of the HA.

- Clearly defines and states hypothesis.
- Adequately describes the study protocol, methods, and statistical analyses.
- Evaluates appropriate endpoints. Toxicity depends on the amount, duration, timing and pattern of exposure, and could range from frank effects (e.g., mortality) to more subtle biochemical, physiological, pathological or functional changes in multiple organs and tissues.
- Applies appropriate statistical procedures to determine an effect.
- Establishes dose-response relationship (i.e., no observed adverse effect level (NOAEL) and/or lowest observed adverse effect level (LOAEL) or data amenable to modeling of the dose response to identify a POD for a change in the effect considered to be adverse [out of the range of normal biological viability]. The NOAEL is the highest exposure level at which there are no biologically significant increases in the frequency or severity of adverse effects between the exposed population and its appropriate control. The LOAEL is the lowest exposure level at which there are biologically significant increases in frequency or severity of adverse effects between the exposed population and its appropriate control group.

The studies included in the HESD and HA were determined to provide the most current and comprehensive description of the toxicological properties of PFOA and the risk it poses to humans exposed through their drinking water.

After the available, reliable studies were evaluated for inclusion in the HESD and HA, critical studies were selected for consideration based on factors including exposure duration (comparable to the duration of the HAs being derived), route of exposure (e.g., oral exposure via drinking water, gavage, or diet), species sensitivity, comparison of the POD with other available studies demonstrating an effect, and confidence in the study (USEPA 1999). Uncertainty factors appropriate for the studies selected are then applied to the potential PODs to account for variability and uncertainty in the available data.

3.2.3 Approach for HA Calculation

For PFOA, toxicity and exposure data were used to develop a lifetime HA. EPA used measures of effect and estimates of exposure to derive the lifetime HA using the following three-step process:

Step 1: Adopt a Reference Dose (RfD) or calculate an RfD using the appropriate point of departure (POD). The RfD is an estimate (with uncertainty spanning perhaps an order of magnitude) of a daily human exposure to the human population (including sensitive subgroups) that is likely to be without an appreciable risk of deleterious effects during a lifetime. In the case of PFOA, the POD is the human equivalent dose (HED) derived from the modeled serum

ED_005024_00000285-00032

concentration representing either an NOAEL or LOAEL experimental dose after applying uncertainty factors established following EPA guidelines.

$$RfD = \frac{HED_{NOAEL} \text{ or } HED_{LOAEL}}{UF}$$

Where:

HED_{NOAEL} = The HED from the modeled average serum representing the highest of the given doses that lacked adverse effects (mg/kg/day).

HED_{LOAEL} = The HED from the modeled average serum representing the lowest of the given doses that results in adverse effects (mg/kg/day) and of an appropriate duration and endpoint to use for a lifetime HA.

UF = Total Uncertainty Factor established in accordance with EPA guidelines considering variations in sensitivity among humans, differences between animals and humans, the duration of exposure in the key study compared to a lifetime of the species studied, whether the HED is a dose that caused an effect or no effect, and the completeness of the toxicology database.

Step 2: Calculate a Drinking Water Equivalent Level (DWEL) from the RfD. The DWEL assumes that 100% of the exposure comes from drinking water.

$$DWEL = \frac{RfD \times bw}{DWI}$$

Where:

RfD = Reference dose (mg/kg bw/day)
bw = Assumed body weight (kg)
DWI = Assumed human daily drinking water intake (L/day)

Step 3: Calculation of the Lifetime HA. The lifetime HA is calculated by factoring in other sources of exposure (e.g., air, food, soil) in addition to drinking water using the methodology described for calculation of an RSC described in USEPA (2000) and section 6.1.

$$\text{Lifetime HA} = DWEL \times RSC$$

Where:

DWEL = Drinking water equivalent level calculated from step 2 (mg/L)
RSC = Relative source contribution

3.2.4 Measures of Effect

The animal toxicology studies were used in the dose-response assessment for PFOA. These studies demonstrated dose-related effects on systemic and developmental endpoints in multiple species (monkeys, rats, mice) following exposure to PFOA for durations of 11 to 84 days; these are described in detail in the HESD for PFOA. The studies selected for pharmacokinetic analysis were chosen based on their experimental design, data quality, dose-response data identified through the range of experimental NOAELs/LOAELs, and serum measurements of PFOA.

ED_005024_00000285-00033

EPA used a peer-reviewed pharmacokinetic model developed by Wambaugh et al. (2013) to calculate the average serum concentrations associated with the candidate NOAELs and LOAELs from the toxicological database. Average serum levels of PFOA from the model were used to determine the HED associated with the study NOAEL and LOAEL. The Wambaugh et al. (2013) model is based on the Andersen et al. (2006) concept that saturable renal resorption is responsible for the long serum half-lives seen in humans and animals.

A unique feature of the pharmacokinetic approach is the use of a single model for the three species and reliance on the serum PFOA level as the measure of exposure. For each species the model accommodated the appropriate toxicokinetic variables for the species/strain. The pharmacokinetic analysis facilitated examination for consistency in the average serum values associated with effect and no-effect doses from the animal PFOA studies. A nonhierarchical model for parameter values was assumed wherein a single numeric value represented all individuals of the same species, gender, and strain. Body weight, the number of doses, and magnitude of the doses were the only parameters that varied.

3.2.5 Relative Source Contribution

The RSC is applied in the HA calculation to ensure that an individual's total exposure from a contaminant (i.e., PFOA) does not exceed the RfD. The RSC is the portion of the RfD attributed to drinking water (directly or indirectly in beverages like coffee, tea, or soup); the remainder of the RfD is allocated to other potential exposure sources. In the case of PFOA, other potential sources include ambient air, foods, incidental soil/dust ingestion, consumer products, and others (see sections 2.2 and 6.1). The RSC for the HA is based on exposure to the general population.

EPA derived an RSC for PFOA by using the Exposure Decision Tree approach (USEPA 2000) (see section 6.1). To use that approach, EPA compiled information for PFOS on its uses, chemical and physical properties, occurrences in other potential sources (e.g., air, food), and releases to the environment. To determine the RSC to be used in the HA calculation for PFOA, EPA then used the information to address the questions posed in the Exposure Decision Tree. Some of the important items evaluated in the Exposure Decision Tree are:

- Adequacy of data available for each relevant exposure source and pathway.
- Availability of information sufficient to characterize the likelihood of exposure to relevant sources.
- Whether there are significant known or potential uses/sources other than the source of concern (i.e., ambient water and fish/seafood from those waters).
- Whether information on each source is available to characterize exposure.

In cases where environmental or exposure data are lacking, the Exposure Decision Tree approach results in a recommended RSC of 20%. This 20% RSC value may be replaced where sufficient data are available to develop a scientifically defensible alternative value. When appropriate, if scientific data demonstrating that sources and routes of exposure other than drinking water are not anticipated for the pollutant in question, the RSC may be raised to 80% based on the available data (USEPA 2000).

ED_005024_00000285-00034

4.0 EFFECTS ASSESSMENT

The database for PFOA includes a large number of laboratory animal toxicity studies, as well as numerous epidemiology studies. The most extensive epidemiology studies were conducted by the C8 Science Panel for a highly exposed population in West Virginia. These animal and human studies are described below and in greater detail in the HESD for PFOA (USEPA 2016a). Because of uncertainties associated with the human data (described above), EPA is relying on animal data to quantitatively assess effects; however, the epidemiology studies provide important data to establish probable links between PFOA exposure to humans and health effects. In particular, effects on the liver enzymes indicative of liver effects, low birthweight, antibody response, and cancer in laboratory animals are supported by human epidemiology studies.

4.1 Noncancer Health Effects

4.1.1 Animal Toxicity Studies

The database of animal toxicology studies is extensive with short term, subchronic, and chronic toxicity and cancer studies; developmental and reproductive toxicity, neurotoxicity, and immunotoxicity studies; and mechanistic studies.

Developmental Effects

Both rats and mice showed developmental toxicity based on low birth weights, skeletal effects (reduced ossification), altered onset of puberty (Butenhoff et al. 2004a; Lau et al. 2006; Wolf et al. 2007). Doses that elicited a response were higher in rats compared with those in mice. Meta-analyses were conducted to determine whether developmental exposure to PFOA was associated with fetal growth effects in animals (Koustas et al. 2014). Eight animal studies identified in the published literature met the criteria of the Navigation Guide systematic review methodology as developed and published by Woodruff and Sutton (2014) for inclusion in the analyses. The animal data sets included mouse gavage studies with maternal PFOA doses from 0.01 to 20 mg/kg/day. The results from the meta-analysis showed that a 1 mg/kg/day increase in PFOA dose was associated with a -0.023 g (95% CI [-0.029, -0.016]) difference in pup birth weight. The MOA for decreased pup body weight is not known, but receptor-activated changes in metabolism, hormonal perturbations, and impeded intercellular communication might play a role.

One animal neurological study (Johansson et al. 2009) showed effects on habituation and activity patterns in NMRI (Naval Medical Research Institute) mice treated on post-natal day (PND)10 with a single dose of PFOA and evaluated at 2 and 4 months of age (LOAEL = 0.58 mg/kg). The in vivo observations are supported by changes in the expression of a variety of neurologically active brain proteins in the treated pups (Johansson et al. 2009). The offspring of C57BL/6/Bkl dams fed diets that provided a dose of 0.3 mg PFOA/kg/day throughout gestation had detectable levels of PFOA in their brains at birth (Onishchenko et al. 2011). Behavioral assessments of the offspring starting at 5 weeks of age revealed sex-related differences in exploratory behavior patterns. In the social group setting, the PFOA-exposed males were more active and PFOA-exposed females were less active than their respective controls. The PFOA-exposed males also had increased activity counts compared to control males in circadian activity

ED_005024_00000285-00035

experiments. The results of an *in vitro* study of hippocampal synaptic transmission and neurite growth in the presence of long chain perfluorinated compounds showed that 50 or 100 micromolar PFOA increased spontaneous synaptic current and had an equivocal impact on neurite growth (Liao et al. 2009a, 2009b). These data suggest a need for additional studies of the effects of PFASs, including PFOA, on the brain.

The developmental impacts of PFOA exposure ranged from delayed mammary gland development in pups (Albrecht et al. 2013; Macon et al. 2011; Tucker et al. 2015; White et al. 2009, 2011; Wolf et al. 2007) to delays in attaining developmental milestones (Lau et al. 2006; White et al. 2009; Wolf et al. 2007). The LOAEL for the mammary gland developmental effects in female offspring from dams given 0.01 mg/kg/day for 8 days from Macon et al. (2011) is of unknown biological significance. The same study showed no effects on offspring body weight at maternal doses up to 3 mg/kg/day for 17 days (Macon et al. 2011). Data from White et al. (2011) showed no significant effects on body weight gain in pups nursing from dams treated with 1 mg/kg/day, despite these dams having less fully developed mammary glands compared to controls. Similarly, no differences in response to a lactational challenge were seen in PFOA exposed dams with morphologically delayed mammary gland development (White et al. 2011).

Immune Function

Several animal studies demonstrate effects on the spleen and thymus as well as their cellular products (B lymphocytes and T-helper cells) in several strains of mice. Studies by Yang et al. (2000, 2001, 2002b) and DeWitt et al. (2008) were conducted using relatively high PFOA doses (~30 to 40 mg/kg/day). In each study, the PFOA-treated animals exhibited significant decreases in spleen and thymus weights as well as splenocyte and thymocyte populations at various stages of differentiation. Recovery usually occurred within several days of cessation of PFOA dosing. When the response of C57BL/6Tac PPARα mice were compared to wild type of the same strain, the knockout mice showed no response with both spleen and thymus weights at 30 mg/kg/day, whereas there was a response in the wild-type strain (DeWitt et al. 2015), suggesting an impact of PPARα. Both strains showed an increase in immunoglobulin M (IgM) in response to a sheep red blood cell (SRBC) injection. The 30 mg/kg/day dose was the LOAEL for the knockout mice and 7.5 mg/kg/day was the response level for the wild-type strain. Thus, the suppression of the immune system is not totally a PPARα-related response.

DeWitt et al. (2008) used different functionality assays in their study in C57Bl/6 mice. The IgM response to SRBCs was suppressed by 20% when mice were immunized immediately after exposure to the initial dose of 30 mg PFOA/kg/day ceased. However, no significant increase occurred in the response to bovine serum albumin 4 days post-PFOA exposure, or in the immunoglobulin G (IgG) response to SRBC 15 days post-PFOA exposure. These results are indicative of recovery once PFOA exposures have ceased. DeWitt et al. (2008) followed their initial study of PFOA with one designed to examine the dose response for a 15-day drinking water exposure in a slightly different mouse strain, C57Bl/6N. The LOAEL was 3.75 mg/kg/day based on a significant decrease in IgM response, and the NOAEL was 1.88 mg/kg/day indicating the inability to respond to an immunological challenge.

ED_005024_00000285-00036

Liver Disease and Liver Function

Hepatocellular hypertrophy and an increased liver-to-body weight ratio are common findings in rodents, but are considered non-adverse if there is evidence for PPARα activation. These effects are considered adverse if accompanied by necrosis, fibrosis, inflammation, and steatosis (Hall et al. 2012). Low-level necrotic cell damage was observed in the Palazzolo et al. (1993) rat study and in the Loveless et al. (2008) studies in CD rats and CD-1 mice. Palazzo et al. (1993) is an unpublished report that was later published as Perkins et al. (2004). The liver histopathology details of this study were only presented in Palazzolo et al. (1993). This study will be referred to throughout the rest of the document as Palazzolo et al. (1993)/Perkins et al. (2004). In this study there was a slight increase in coagulative necrosis at 1.94 and 6.5 mg/kg/day when compared to the control and lower dose (0.94 mg/kg/day). Some hepatocellular necrosis was also observed in conjunction with hepatocellular hypertrophy and increased liver weight at a dose of 3 mg/kg/day in F1 male rats from the Butenhoff et al. (2004a) two-generation study.

In general, effects on organs other than the liver tend to occur at doses higher than those that affect the liver. Lung effects including pulmonary congestion were observed in male Sprague-Dawley rats (LOAEL = 5 mg/kg/day) by Cui et al. (2009). Increased thickness and prominence of the adrenal zona glomerulosa and vacuolization in the cells of the adrenal cortex were observed in male rats fed 10 mg/kg/day for approximately 56 days (Butenhoff et al. 2004a).

Kidney Function

Some studies have shown effects on the kidney of male rats at doses similar to those resulting in liver effects. Increases in absolute and relative-to-body kidney weights occurred in rats given 5 mg/kg/day (lowest dose tested) via gavage for 28 days (Cui et al. 2009). In a two-generation gavage study, F0 and F1 males had significantly increased absolute kidney weight at 1 and 3 mg/kg/day, but significantly decreased kidney weight at 30 mg/kg/day. Organ weight-to-terminal body weight ratios for the kidney were statistically significantly increased at ≥ 1 mg/kg/day. Kidney weight-to-brain weight ratios were significantly increased at 1, 3, and 10 mg/kg/day, but decreased at 30 mg/kg/day (Butenhoff et al. 2004a). In the high-dose group, absolute and relative kidney weight changes occurred in a pattern typically associated with decrements in body weight and are indicative of systemic toxicity. In the lower-dose groups, the consistently increased absolute and relative to body and brain weights suggest a cellular response, whereby the kidney tubular cells upregulate expression of transporter proteins to facilitate the PFOA excretion. This is adverse because it is a biomarker for systemic PFOA bioaccumulation. The differential expression of transporters in the kidney of male rats is under hormonal control with males having lower levels of export transporters compared to females (Kudo et al. 2002). No dose-related changes in kidney weight or histopathology were found in male rats at the end of 2 years with a dose of 14.2 mg/kg/day (Butenhoff et al. 2012).

Diabetes

Hines et al. (2009) found no differences in glucose tolerance tests at 15–16 weeks and at 17 months of age in PFOA-exposed CD-1 mice, but did observe significantly increased serum leptin and insulin levels at 21 and 31 weeks of age suggesting that the insulin resistance mechanistic pathway could be affected by PFOA and a connection between PFOA and increased body weight. Leptin is a hormone secreted by adipose tissue that is associated with weight gain.

ED_005024_00000285-00037

Conversely, Quist et al. (2015) found no dose-related impact on serum leptin in CD-1 pups on PND 91. Quist et al. (2015) found that when mice were on a high-fat diet and not fasted before serum collection and these were compared to the same mice that were fasted before serum collection, leptin increased, thereby suggesting that the leptin change could be temporary and dependent on the fat content of the diet and the timing of serum collection.

Thyroid

Effects of PFOA on thyroid hormones in animals are not as well characterized as those of PFOS. Butenhoff et al. (2002) evaluated the toxicity of PFOA in a small number of male monkeys during 6 months of oral administration and reported that levels of total T3 and free T3 in circulation were reduced significantly in the 30/20 mg/kg/day treatment group, beginning at 5 weeks after initiation of treatment but accompanied by other signs of systemic toxicity. Recovery of T3 deficits was noted when PFOA returned to baseline 90 days later. Serum total thyroxine (T4), free T4, and thyroid-stimulating hormone (TSH) were not altered throughout the study. The preferential effects of PFOA on serum T3 and a lack of a TSH compensatory response are similar to those observed with PFOS, and are possibly a consequence of PFOA binding to the T3 receptor (Ren et al. 2015). None of the thyroid hormones were affected by PFOA in mature female rats (Butenhoff et al (2002), primarily because these animals were able to clear the chemical effectively (with half-life estimate of 2 to 4 hours, compared to that of 6 to 7 days for male rats). This suggests that the thyroid disrupting effects of PFOA are directly related to endogenous accumulation of the chemical and might be relevant to humans because of the long PFOA human half-life.

Fertility, Pregnancy, and Birth Outcomes

Among animal studies there was no effect of PFOA on reproductive or fertility parameters in rats (Butenhoff et al. 2004a), but effects on male fertility were observed in mice given a dose of 5 mg/kg/day for 28 days prior to mating (Lu et al. 2015). A NOAEL of 2.5 mg/kg/day and a LOAEL of 5 mg/kg/day were reported for reduced sperm counts and changes in testicular morphology after a 14-day exposure by Liu et al. (2015); 2 mg/kg/day led to significantly increased serum estradiol and increased hepatic aromatase activity in the same study. Gender differences in dose response are likely related to half-life differences of hours for the female rat and days-to-weeks for the female mouse.

Serum Lipids

Information on serum lipids from animal studies has received less attention than in the human population because decreases in triglycerides, cholesterol, and lipoprotein complexes are an expected consequence of PPARα activation in rodents. PFOA is an activator of the PPARα nuclear receptor in both humans and animals, but activation in humans does not increase the cellular levels of peroxisomes to the same extent it does in rodents. The PPARα response in animals tends to lower rather than raise serum cholesterol and associated lipid levels. PFOA is known to activate the PPAR pathway by increasing transcription of mitochondrial and peroxisomal lipid metabolism, sterol, and bile acid biosynthesis and retinol metabolism genes. However, based on transcriptional activation of many genes in PPARα null mice, the effects of PFOA involve more than activation of PPAR. Also activated are the constitutive androstane receptor (CAR), farnesoid X receptor (FXR), and pregnane X receptor (PXR).

ED_005024_00000285-00038

Cholesterol and/or triglycerides were monitored in few animal studies. Nakamura et al. (2009) found that mice with a normal PPARα receptor had significantly increased levels of cholesterol and triglycerides in liver, but not plasma, at a LOAEL of 0.3 mg/kg/day. However, no differences were observed in serum or liver cholesterol or triglycerides between PFOA-treated mice with a humanized PPARα receptor or PPARα null mice (NOAEL = 0.3 mg/kg/day) and their respective controls. A study by Minata et al. (2010) used higher doses and found that total cholesterol was significantly decreased and total triglycerides significantly increased in wild-type mice. In the PPARα null mice, total triglycerides were significantly increased at all doses.

In animal studies, serum levels of alanine aminotransferase (ALT) and/or aspartate aminotransferase (AST) were significantly increased indicative of apoptosis or necrosis of liver cells (Butenhoff et al. 2012; Minata et al. 2010; Son et al. 2008). Increased levels of ALT were observed at a LOAEL of 2.65 mg/kg/day in ICR mice by Son et al. (2008). Yahia et al. (2010) reported significantly increased ALT, gamma-glutamyl transferase (GGT), AST, and alkaline phosphatase (ALP) in PFOA-exposed (10 mg/kg) pregnant ICR mice. Total protein, albumin, and globulin were significantly decreased in the same mice.

4.1.2 Human Epidemiology Studies

Numerous epidemiology studies evaluating large cohorts of highly exposed occupational and general populations have examined the association of PFOA exposure to a variety of health endpoints. Health outcomes assessed include blood lipid and clinical chemistry profiles; reproductive parameters; thyroid effects; diabetes; immune function; birth, fetal, and developmental growth measures; and cancer. Members of the general population living in the vicinity of the West Virginia DuPont Washington Works PFOA production plant in Parkersburg, West Virginia, are the focus of an ongoing study titled the C8 Health Project. Releases from the Washington Works plant, where PFOA was used as a processing aid in the manufacture of fluoropolymers, contaminated the ground water from six water districts near the plant, resulting in exposures to the general population. The C8 Health Project is the largest study evaluating human exposure and health endpoints for PFOA; the study included more than 65,000 people in Mid-Ohio Valley communities who were exposed to PFOA for longer than 1 year.

As part of the C8 Health Project, a panel of expert epidemiologists reviewed the epidemiological and other data available in 2011 and 2012 to assess probable links between PFOA exposure and disease.[6] The C8 Science Panel concluded that a probable link existed between PFOA exposure and the following conditions: high cholesterol, thyroid disease, pregnancy-induced hypertension, ulcerative colitis, and kidney and testicular cancer. The C8 Science Panel did not find a probable link between PFOA exposure and multiple other conditions, including other autoimmune diseases (rheumatoid arthritis, lupus, Type I diabetes, Crohn's disease, multiple sclerosis), Type II diabetes, high blood pressure, coronary artery disease, infectious disease, liver disease, Parkinson's disease, osteoarthritis, neurodevelopmental disorders in children (attention deficit hyperactivity disorder, learning disabilities), chronic kidney disease, stroke, asthma or chronic obstructive airways disease (COPD), and birth defects,

[6] For more information see http://www.c8sciencepanel.org/prob_link.html.

ED_005024_00000285-00039

miscarriage or stillbirth, preterm birth or low birth weight, and other types of cancer. The summary below focuses on the endpoints highlighted as "probable links" by the C8 Science panel, and on other epidemiology studies published after the 2011–2012 reports.

Serum Lipids

The association between PFOA and serum lipids has been examined in several studies in different populations. Cross-sectional and longitudinal studies in occupational settings (Costa et al. 2009; Olsen et al. 2000, 2003; Olsen and Zobel, 2007; Sakr et al. 2007a, 2007b; Steenland et al. 2015) and in the high-exposure community (the C8 Health Project study population) (Fitz-Simon et al. 2013; Frisbee et al. 2010; Steenland et al. 2009; Winquist and Steenland 2014a) generally observed positive associations between serum PFOA and total cholesterol in adults and children (ages 1 to < 18 years); most of these effect estimates were statistically significant. Although exceptions to this pattern are present (i.e., some of the analyses examining incidence of self-reported high cholesterol based on medication use in Winquist and Steenland, 2014a, and in Steenland et al. 2015), the results are relatively consistent and robust. Similar associations were seen in analyses of low-density lipoprotein (LDL), but were not seen with high-density lipoprotein (HDL). The range of exposure in occupational studies is large (means varying between 0.4 and > 12 micrograms per milliliter [μg/mL]), and the mean serum levels in the C8 population studies were around 0.08 μg/mL. Positive associations between serum PFOA and total cholesterol (i.e., increasing lipid level with increasing PFOA) were observed in most of the general population studies at mean exposure levels of 0.002 to 0.007 μg/mL (Eriksen et al. 2013; Fisher et al. 2013; Geiger et al. 2014; Nelson et al. 2010; Starling et al. 2014). The interpretation of these general population results is limited, however, by the moderately strong correlations (Spearman r > 0.6) and similarity in results seen for PFOS and PFOA.

Liver Disease and Liver Function

Few studies pertaining to the relation between PFOA and liver disease are available. The C8 Health Project did not observe associations with hepatitis, fatty liver disease, or other types of liver disease. In the studies of PFOA exposure and liver enzymes (measure in serum), positive associations were seen. The results of the occupational studies provide evidence of an association with increases in serum AST, ALT, and GGT, with the most consistent results seen for ALT. The associations were not large, and the associations could depend on the co-variates in the models, such as body mass index, use of lipid lowering medications, and triglycerides (Costa et al. 2009; Olsen et al 2000, 2003; Olsen and Zobel, 2007; Sakr et al. 2007a, 2007b).

Two population-based studies of highly exposed C8 area residents evaluated associations with liver enzymes, and the larger of the two studies reported associations of increasing serum ln ALT and ln GGT levels with increasing serum PFOA concentrations (Emmett et al. 2006; Gallo et al. 2012). A cross-sectional analysis of data from NHANES, representative of the U.S. national population, also found associations with ln PFOA concentration with increasing serum ALT and ln GGT levels. Serum bilirubin was inversely associated with serum PFOA in the occupational studies. A U-shaped exposure-response pattern for serum bilirubin was observed among the participants in the C8 Health Project which might explain the inverse associations reported for occupational cohorts. Overall, an association of serum PFOA concentration with elevations in serum levels of ALT and GGT was consistently observed in occupational, highly

ED_005024_00000285-00040

exposed residential communities, and the U.S. general population. The associations are not large in magnitude, but indicate the potential to affect liver cells.

Immune Function

Three studies examined associations between maternal and/or child serum PFOA levels and vaccine response (measured by antibody levels) in children (Grandjean et al. 2012; Granum et al. 2013) and adults (Looker et al. 2014). The study in adults was part of the high-exposure community C8 Health Project; a reduced antibody response to one of the three influenza strains tested after receiving the flu vaccine was seen with increasing levels of serum PFOA. The studies in children were conducted in general populations in Norway and in the Faroe Islands. As observed in the animal studies, decreased vaccine response in relation to PFOA levels was seen in these studies, but similar results also were seen with other correlated PFASs (e.g., PFOS).

Thyroid

Three studies reported an increased risk of thyroid disease in women or girls, but not men (Lopez-Espinosa et al. 2012; Melzer et al. 2010; Winquist and Steenland, 2014b). A fourth study also reported a trend of elevated TSH and decreased T4 (hypothyroidism) in pregnant women testing positive for hypothyroid autoimmune disease (Webster et al. 2014). Similarly, the C8 Panel concluded there was strong evidence to link PFOA exposure to thyroid disease in its population. Hypothyroxinemia (decreased free thyroxine (FT4) without concomitant elevation of TSH) was measured in one study of pregnant women showing null findings for hypothyroxinemia incidence versus controls; hypothyroxinemia is not typically studied in the clinic as TSH and T4 concomitantly inversely shift with thyroid disease. Looking at thyroid hormone levels, some studies found changes in thyroid hormone levels associated with PFOA (de Cock et al. 2014; Shrestha et al. 2015; Webster et al. 2014; Wen et al. 2013,); others found null effects of PFOA in association with thyroid hormones. Generally null associations were found in other studies on the general population, pregnant women, and patients in association with thyroid hormone levels or one portion of the thyroid panel was outside of control range. Across multiple studies, thyroid hormone concentrations have mixed evidence (associations and null findings) in association with PFOA concentrations. Increased risk for thyroid disease in women appears to be associated with PFOA serum concentration; evidence is weaker or null in men.

Diabetes

No associations were observed between serum PFOA levels and type II diabetes incidence rate in general or worker populations with mean serum PFOA up to 0.0913–0.113 µg/mL (MacNeil et al. 2009; Steenland et al. 2015). PFOA was not associated with measures of metabolic syndrome in adolescents or adults (Lin et al. 2009). However, one study found an increased risk for developing gestational diabetes in women with mean serum PFOA (measured at preconception) of 0.00394 µg/mL (Zhang et al. 2015).

Fertility, Pregnancy, and Birth Outcomes

The association between PFOA and birth weight has been examined in numerous studies (see section 4.1.1.7 in USEPA 2016a). Most studies measured PFOA in the general population using

ED_005024_00000285-00041

maternal blood samples taken in the second or third trimester or in cord blood samples. One study was able to collect samples earlier in the pregnancy (4–14 weeks) (Fei et al. 2007), and another study in the high-exposure community (the C8 Health Project population) modeled exposure based on data on residential history and environmental data (Savitz et al. 2012). Two meta-analyses of these studies have been conducted (Johnson et al. 2014; Verner et al. 2015), with similar results: mean birthweight reduction of 19 g (95% CI [-30, -9]) per each one unit (ng/mL) increase in maternal or cord serum PFOA levels in Johnson et al. (2014), and a mean birthweight reduction of 15 g (95% CI [-22, -8]) based on seven of these nine studies in Verner et al. (2015). It has been suggested that low glomerular filtration rate (GFR) can affect birth weight (Morken et al 2014). Verner et al (2015) conducted a meta-analysis based on physiologically based pharmacokinetic model (PBPK) simulations and found that some of the association reported between PFOA and birth weight is attributable to GFR and that the actual association may be closer to a 7 gram reduction (95% CI [-8, -6]). Verner et al. (2015) showed that in individuals with low GFR there are increased levels of serum PFOA and lower birth weights. Although some uncertainty exists in the interpretation of the observed association between PFOA and birth weight given the potential impact of low GFR, the available information indicate that the association between PFOA exposure and birth weight cannot be ruled out. In humans with low GFR (which includes women with pregnancy-induced hypertension or preeclampsia) the impact on body weight is likely due to a combination of the low GFR and the serum PFOA.

Two studies examined development of puberty in girls in relation to prenatal exposure to PFOA as measured through maternal or cord blood samples in follow-up of pregnancy cohorts conducted in England (Christensen et al. 2011) and in Denmark (Kristensen et al. 2013). The results of these two studies are conflicting, with no association (or a possible indication of an earlier menarche seen with higher PFOA) in Christensen et al. (2011), and a later menarche seen with higher PFOA in Kristensen et al. (2013). Another study examined PFOA exposure measured concurrently with the assessment of pubertal status (Lopez-Espinosa et al. 2011). An association between later age at menarche and higher PFOA levels was observed, but the interpretation of this finding is complicated by the potential effect of puberty on the exposure biomarker levels (i.e., reverse causality). Menstruation is a route of excretion for albumin-bound PFOA; thus, the beginning of menstruation will remove serum PFOA when the menstruation periods begin during puberty and its cessation at menarche will decrease the loss of PFOA in blood and allow serum levels to increase.

Limited data suggest a correlation between higher PFOA levels (>0.02 μg/mL) in women and decreases in fecundity and fertility (Fei et al. 2009; Vélez et al. 2015), but there are no clear effects of PFOA on male fertility endpoints (0.0035–0.005 μg/mL; Joensen et al. 2009, 2013).

4.1.3 Noncancer Mode of Action

No published cohesive MOA exists that accounts for the varied toxicological properties of PFOA. However, a number of the unique properties of the compound contribute to its toxicity:

- Metabolic stability accompanied by persistence in tissues as an apparent consequence of saturable renal resorption.

ED_005024_00000285-00042

- Electrostatic binding to biopolymers with areas of positive charge, especially proteins (MacManus-Spencer et al. 2009; Salvalaglio et al. 2010; Wu et al. 2009b; L. Zhang et al. 2013).
- Displacement of endogenous/exogenous substances normally bound to serum albumin such as fatty acids, bile acids, pharmaceuticals, and T3 (Fasano et al. 2005; Qin et al. 2010; Wu et al. 2009a).
- Renal resorption (Andersen et al. 2006) and biliary excretion that are dependent on transporters genetically encoded for management of natural substances (endogenous and exogenous) that prolong systemic retention of absorbed PFOS and explain its long half-life
- Binding to and activating receptors such as PPAR, thereby initiating activation or suppression of gene transcription related to fatty acid metabolism and lipid transport (Nakamura et al. 2009; Rosen et al. 2007, 2009a, 2009b; Takacs and Abbott 2007).
- Interference with intercellular communication (Upham et al. 1998, 2009).

The renal resorption and biliary competition between natural substrates and PFAS contribute to ambiguity in some of the epidemiology study outcomes where serum levels of endogenous or dietary-transported substrates are altered because of preferential removal or resorption of the PFOA, or the PFOA serum level increases because of the preferred excretion of the natural material. Physiological status also has an impact on the epidemiology results given that blood loss through menstruation is an excretory pathway for serum-albumin-bound PFOA. Thus, serum levels will be lower in girls after puberty than before, and will increase in women after menopause. In pregnant women, increased blood volume as well as cessation of monthly menstrual blood flow route also influences serum levels.

The outcome from studies of antibodies or immunoglobulins can be confounded by PFOA protein binding depending on the impact of morphological changes caused by PFOA binding on the sensitivity of the assay. Interaction of PFOA and other PFASs (Ren et al. 2015) with the T3 receptor has the potential to influence cellular uptake of T3. Binding to thyroid hormone transport protein or transthyretin (TTR) can displace T4 increasing the unbound level in serum (Weiss et al. 2009).

There are no cohesive studies designed to identify modes of action for the liver weight and hypertrophy endpoints represented in the animal studies. Both effects are clearly, in part, an outcome of PPARα activation. They become adverse when accompanied by inflammation, fibrosis, steatosis, or necrosis (Hall et al. 2012) as seen in Palazzolo et al. (1993)/Perkins et al. (2004), Loveless et al. (2008), and Butenhoff et al. (2004a).

The MOA for decreased pup body weight observed in the animal studies is unknown (Butenhoff et al. 2004a; White et al. 2009; Wolf et al. 2007). The observed effects on birth weight in animals are supported by evidence of an association between PFOA and low birth weight in humans (Johnson et al. 2014). Receptor-activated changes in metabolism, hormonal perturbations, and impeded intercellular communication could play a role in this effect. It has been suggested that GFR can affect birth weight (Morken et al 2014). Verner et al (2015) conducted a meta-analysis based on PBPK simulations and found that some of the association reported between PFOA and birth weight could be partially attributable to low GFR and that the actual association might be closer to a 7 gram reduction (95% CI [-8, -6]). However, the study

ED_005024_00000285-00043

authors demonstrated that individuals with low GFR have increased levels of serum PFOA and lower birth weights. Although some uncertainty exists in the interpretation of the observed association between PFOA and birth weight given the potential impact of low GFR, the available information indicate that the association between PFOA exposure and birth weight cannot be ruled out. In humans with low GFR (which includes women with pregnancy-induced hypertension or preeclampsia) the impact on body weight is likely due to a combination of the low GFR and the serum PFOA.

Women with hypertension during pregnancy are a susceptible population that could have an increased risk for having a low birth weight baby.

There also is a lack of data relative to the MOA for immunological effects of PFOA as seen in animal studies. Some of the responses are PPARα linked (increased spleen and thymus weights) but not all as demonstrated by DeWitt et al. (2015). Effects on serum immunoglobulins observed in humans could be a reflection of analytical method interference as a result of PFOA binding to the immunoglobin (Kerstner-Wood et al. 2003).

PFOA is associated with delayed breast tissue development (reduced ductal branching and numbers of terminal endbuds) in CD-1 mice (Albrecht et al. 2013; Macon et al. 2011; Tucker et al. 2015; White et al. 2009); however, no functional impacts on the ability of the dams to provide nourishment were observed based on the weight increases in the pups reared by the impacted dams (Macon et al. 2011; White et al. 2011). CD-1 mice seem to be more sensitive for this effect than other mice strains evaluated (Tucker et al. 2015). No mechanistic studies exist that inform the MOA for the mammary gland development effects.

Quist et al. (2015) found that the level of dietary fat in an animal diet is an important variable that influences liver lipid levels. At PFOA doses < 0.3 mg/kg/day, the LDL and total serum cholesterol levels in the fasted and nonfasted high-fat diet animals were greater than in the untreated Purina controls. Tan et al. (2013) found that the fat content of the diet was an important variable in determining the impact of PFOA (5 mg/kg/day) on liver and serum lipids. Intake of a high-fat diet plus PFOA increased liver triglycerides and serum free fatty acids as compared to a regular fat diet plus PFOA, but it had no impact on liver cholesterol concentrations. Serum cholesterol was not monitored. A high-fat diet predisposes animals and possibly humans to hepatic steatosis.

4.2 Cancer

4.2.1 Animal Cancer Bioassays

The only animal carcinogenicity studies available for PFOA indicate that exposure can lead to liver adenomas (Biegel et al. 2001), Leydig cell adenomas (Biegel et al. 2001; Butenhoff et al. 2012), and pancreatic acinar cell tumors (PACT) (Biegel et al. 2001) in male Sprague-Dawley rats. In the Butenhoff et al. (2012) study there was an increase in liver carcinomas at the high dose (14.2 mg/kg/day) in the males compared to controls (6% versus 10%). For the females receiving 16.1 mg/kg/day (i.e., the high dose) the tumor incidence compared to controls was 0% versus 2%. The increase in liver tumors did not show a direct relationship to dose in the male rats and was not statistically significantly elevated in either males or females at the high dose when

ED_005024_00000285-00044

compared to controls (Butenhoff et al. 2012). Liver adenomas were observed in the Biegel et al. (2001) study at an incidence of 10/76 (13%) at 20 mg/kg/day. The incidence in the control group was 2/80 (3%).

Butenhoff et al. (2012) also observed increased incidence of testicular Leydig cell tumors (LCTs) in rats. At the 1-year sacrifice, testicular masses were found in 7/50 (14%) high-dose and 2/50 (4%) low-dose rats, but not in any of the controls. A significant increase ($p < 0.05$) in the incidence of testicular (Leydig) cell adenomas was also observed in the high-dose male rats at the end of the study. The LCT incidence in the control, low-, and high-dose groups was 0/50 (0%), 2/50 (4%), and 7/50 (14%), respectively. Biegel et al. (2001) observed a significant increase in the incidence of Leydig cell adenomas in the treated rats (11%, 8/76) when compared to the pair-fed control rats (3%, 2/78) supporting the observations from the Butenhoff et al. (2012) study. The LCTs in the Butenhoff et al. (2012) study were accompanied by statistically significant testicular vascular mineralization and by Leydig cell hyperplasia in the Biegel et al. (2001) study.

PACTs were only observed in the Biegel et al. (2001) study, with an incidence of 11% at 20 mg/kg/day compared to controls. Although no PACTs were observed by Butenhoff et al. (2012), pancreatic acinar hyperplasia was observed at 1.3 and 14.2 mg/kg/day at incidences of 6% and 2%, respectively. Re-examination of the pancreatic lesions in Butenhoff et al. (2012) and Biegel et al. (2001) resulted in the conclusion that the high dose increased the incidence of proliferative acinar cell lesions in both studies. Some lesions in the Biegel et al. (2001) study had progressed to adenomas but not those in the Butenhoff et al. (2012) study.

The initial findings from the Butenhoff et al. (2012) study were equivocal for mammary fibroadenomas in female rats. However, a re-examination of the tissues by a pathology working group (PWG) found no statistically significant differences in the incidence of fibroadenomas or other neoplasms of the mammary gland between control and treated animals (Hardisty et al. 2010). The PWG used the diagnostic criteria and nomenclature of the Society of Toxicological Pathologists for the re-examination.

4.2.2 Human Epidemiology Studies

Evidence of carcinogenic effects of PFOA in epidemiology studies is based on studies of kidney and testicular cancer. These cancers have relatively high survival rates (2005–2011 5-year survival rates are 73% and 95%, respectively, for kidney and testicular cancer, based on NCI Surveillance, Epidemiology and End Results data); therefore, studies that examine population cancer incidence are particularly useful for these types of cancers. For testicular cancer, the high-exposure community studies also have the advantage of including the age period of greatest risk, as the median age at diagnosis is 33 years. The two occupational cohorts in Minnesota and West Virginia (most recently updated, respectively, in Raleigh et al. 2014 and Steenland and Woskie, 2012) do not support an increased risk of these cancers, but each of these is limited by a small number of observed cases (six kidney cancer deaths, 16 incident kidney cancer cases, and five incidence testicular cancer cases in Raleigh et al. [2014]; 12 kidney cancer deaths and one testicular cancer death in Steenland and Woskie [2012]). Two studies involving members of the C8 Health Project showed a positive association between PFOA levels (mean at enrolment 0.024 μg/mL) and kidney and testicular cancers (Barry et al. 2013; Vieira et al. 2013);

ED_005024_00000285-00045

some of the cases included in these studies overlap. None of the general population studies examined kidney or testicular cancer, but no associations were found in the general population between mean serum PFOA levels up to 0.0866 μg/mL and colorectal, breast, prostate, bladder, and liver cancer (Bonefeld-Jørgensen et al. 2014; Eriksen et al. 2009; Hardell et al. 2014; Innes et al. 2014).

4.2.3 Cancer Mode of Action

The mode of carcinogenic action of PFOA is not clearly understood. Some researchers have concluded from the available data that the liver tumors observed in the cancer bioassays can be attributed mostly to the impact of PFOA on peroxisome proliferation based on a hypothesized lower sensitivity of humans to this MOA (Klaunig et al. 2003, 2012). Some data support the hypothesis that PPARα agonism MOA could be responsible for observed liver tumors in animals. Rosen et al. (2008a, 2008b) examined transcript profiles in the livers of wild-type and PPARα-null mice dosed with PFOA for up to 7 days. This study showed that animal responses were consistent with PPARα agonism, but evidence also shows PPARγ agonism (down-regulation of cholesterol synthesis) and activation of CAR and PXR-related genes (Martin et al. 2007). There is evidence that PFOA is a potent peroxisome proliferator inducing peroxisome formation in the livers of rats and mice (Elcombe et al. 2010; Minata et al. 2010; Pastoor et al. 1987; Wolf et al. 2008; Yang et al. 2001). Beyond activation of PPARα, few studies have evaluated whether additional steps (i.e., cell proliferation and apoptosis) are in the hypothesized PPARα agonism MOA (Elcombe et al. 2010; Minata et al. 2010; Wolf et al. 2008). For example, no studies were identified that focused specifically on preneoplastic foci and clonal expansion of altered cells after PPAR activation.

The proposed MOA for testicular LCTs is linked to decreased serum testosterone levels and signaling of the hypothalamus to produce gonadotropin releasing hormone (GnRH), a signaling agent for the pituitary to release luteinizing hormone which upregulates testosterone production in Leydig cells. Administering PFOA to adult male rats by gavage for 14 days was shown to decrease testosterone levels and increase serum estradiol levels (Cook et al. 1992). These endocrine changes correlated with its potency to induce LCTs in rats and were hypothesized to play a role in the PFOA-induction of LCTs (Biegel et al. 2001). Support for PPARα-mediated inhibition of testosterone production is found in Li et al. (2011). However, some researchers have proposed that data are not currently sufficient to demonstrate that the other key steps in the postulated MOA are present in PFOA-treated animals following exposures that lead to tumor formation (Klaunig et al. 2012).

Two hypothetical MOAs have been proposed for PACTs (Klaunig et al. 2003, 2012; Obourn et al. 1997). In one case, growth factors such as cholecystokinin (CCK) and/or gastrin activate a feedback loop resulting in proliferation of the secretory pancreatic acinar cells leading to tumors. The other proposed MOA suggests that increased serum testosterone supports the growth of acinar cell preneoplastic foci.

Li et al. (2011) found that serum testosterone levels were decreased, not increased, in wild-type, PPARα- null and mice with humanized PPARα. Biegel et al. (2001) found no change in serum testosterone in their bioassay. Obourn et al. (1997) studied the impact of PFOA on CCK and trypsin using *in vitro* assays and found that PFOA was not an agonist for the cholecystokinin

ED_005024_00000285-00046

agonism receptor receptor that activated CCK release. Plummer et al. (2007) reported on gene expression changes induced in pancreatic acinar cells isolated from Sprague-Dawley rats fed diets containing 300 parts per million (~20 mg/kg/day) PFOA for 28 days. Expression of genes regulated by PPARα, γ, δ in pancreatic acinar cells was directly opposite of the expression of those same genes in liver tissue. At the present time, data are insufficient to demonstrate a MOA that can account for the PACTs identified in the chronic study by Biegel et al. (2001).

The mutagenicity data on PFOA are largely negative, although some evidence shows clastogenicity in the presence of microsomal activation and at cytotoxic concentrations (Murli 1996a, 1996b). PFOA's clastogenic effects are likely the result of an indirect mechanism, given the chemical and physical properties of PFOA (i.e., it is not metabolized, it binds to cellular proteins, and it carries a net negative electrostatic surface charge). PFOA has the potential to interfere with the process of DNA replication because of its protein-binding properties and the fact that histone proteins, spermine, and spermidine carry a net positive surface charge. Involvement of reactive oxygen species (ROS) in the MOA as a result of PFOA alone is unlikely because of its metabolic stability. Conditions leading to ROS would be a function of metabolic responses perturbed by PFOA, rather than PFOA alone.

A compound that is not metabolized will not be able to covalently alter the structure of DNA or intercalate because of electrostatic repulsion between the aromatic base pi bond electrons and the partial negative charges on the PFOA fluoride atoms. Because of its protein-binding properties, PFOA could affect one or more of the proteins involved in the process of DNA replication or cell division (cytoskeletal proteins), however, no mechanistic studies were identified that examined the biochemical effects of PFOA on DNA replication or cell division. No data support this as a MOA for clastogenic effects.

4.2.4 Weight of Evidence Classification

Under EPA's *Guidelines for Carcinogen Risk Assessment* (USEPA 2005) there is Suggestive Evidence of Carcinogenic Potential of PFOA in humans. The bioassay findings for Leydig cell testicular tumors in rats, combined with the C-8 Panel finding of a probable link to testicular and renal tumors among the members of the C8 Health Project, support this conclusion.

In June 2014, 20 experts met at the International Agency for Research on Cancer (IARC) in Lyon, France, to assess the carcinogenicity of perfluorooctanoic acid (PFOA), among other chemicals. Although the assessments have not yet been published (they are expected to be published in volume 110 of the IARC monographs), the expert findings from this meeting are available in a peer-reviewed publication (Benbrahim-Tallaa et al. 2014), and their determination is on the IARC website. The working group classified PFOA as *possibly carcinogenic to humans* (*Group 2B*) and considered the evidence regarding mechanisms of PFOA-associated carcinogenesis to be moderate. This assessment did not lead to a change in the overall classification of PFOA by IARC.

ED_005024_00000285-00047

5.0 DOSE-RESPONSE ASSESSMENT

As an initial step in the dose-response assessment, EPA identified a suite of animal studies with NOAELs and/or LOAELs that identified them as potential candidates for development of the RfD for PFOA. These studies included short-term, subchronic, and chronic exposures, including developmental and reproductive toxicity studies. The available studies evaluated endpoints including liver effects (weight changes with histopathology), body weight changes in adults and offspring, reproductive outcomes such as fertility, developmental effects (altered puberty, survival, and developmental delays such as eye opening), and immune effects. The candidate studies were selected based on their NOAEL and/or LOAEL values, a duration of 11 to 91 days, use of a control, and two or more doses. From these studies, those that presented serum data amenable for modeling (i.e., determination of HEDs) were selected for dose-response analysis. The subset of studies amenable for use in deriving HED based on average serum measurements from the pharmacokinetic model is limited because of the need to have dose and species-specific serum values for model input as well as exposure durations of sufficient length to achieve values near to steady-state projections or applicable to developmental endpoints with lifetime consequences following short-term exposures. The pharmacokinetically modeled average serum values from the animal studies are restricted to the animal species selected for their low dose response to oral PFOA intakes.

As described in section 3.2.4, EPA used the Wambaugh et al. (2013) pharmacokinetic model to derive the average serum concentrations associated with the candidate NOAELs and LOAELs from the toxicological database. Studies with serum information for each of the doses that demonstrated dose response and were amendable for modeling of the area under the curve (AUC) at the time of sacrifice were used. The AUC results were converted to average serum values at the time of sacrifice with consideration of the duration of exposure. The average serum values were converted to the HED, as described further below.

The data were analyzed within a Bayesian framework using a Markov Chain Monte Carlo sampler implemented as an R statistical analysis package developed by EPA to allow predictions across species, strains, and genders, and to identify serum levels associated with the external doses at the NOAEL and LOAEL. The model predictions were evaluated by comparing each predicted final serum concentration to the serum value measured in the supporting animal studies.

The average serum concentrations were converted into an oral equivalent dose by recognizing that clearance from the body equals dose to the body. Clearance can be calculated if the rate of elimination (derived from half-life) and the volume of distribution are both known. EPA used the Bartell et al. (2010) calculated human half-life of 2.3 years (general population) with the Thompson et al. (2010) volume of distribution (V_d) of 0.17 L/kg body weight (bw) to determine a clearance of 1.4×10^{-4} L/kg bw/day by the following equation:

$$CL = V_d \times (\ln 2 \div t_{½}) = 0.17 \text{ L/kg bw} \times (0.693 \div 839.5 \text{ days}) = 0.00014 \text{ L/kg bw/day}$$

Where:

$V_d = 0.17$ L/kg
$\ln 2 = 0.693$
$t_{½} = 839.5$ days (2.3 years x 365 days/year = 839.5 days)

ED_005024_00000285-00048

Multiplying the derived average serum concentrations (in µg/mL) for the NOAELs and LOAELs identified in the key animal studies by the clearance value predicts oral HEDs in mg/kg bw/day for each corresponding serum measurement. The HED values are the predicted human oral exposures necessary to achieve serum concentrations equivalent to the NOAEL or LOAEL in the animal toxicity studies using linear human kinetic information.

Table 5-1 provides the NOAEL, LOAEL, and effect information from those studies, along with the associated average serum values and the percent of steady state represented by the LOAEL.

Table 5-1. Human Equivalent Doses Derived from the Modeled Animal Average Serum Values

Study	Dosing duration days	NOAEL mg/kg/d	NOAEL Av serum mg/L	HED mg/kg/d	LOAEL mg/kg/d	LOAEL (Av serum) mg/L	HED mg/kg/d
DeWitt et al. (2008): mice; ↓ IgM response to SRBC	15	1.88	38.2	0.0053	3.75	61.9	0.0087
Lau et al. (2006): mice decreased ↓ pup ossification (m, f), accelerated male puberty	17	None	-	-	1	38.0	0.0053
Palazzolo et al. (1993); Perkins et al. (2004): rats; ↑liver weight/necrosis	91	0.64	31.6	0.0044	1.94	77.4	0.0108
Wolf et al. (2007): mice; GD 1–17 ↓Pup body weight	17	None	-	-	3	77.9	0.0109
Wolf et al. (2007): mice; GD 7–17 ↓Pup body weight[1]	11	None	-	-	5	87.9	0.0123
Butenhoff et al. (2004a): ↓ relative body weight/↑ relative kidney weight and ↑kidney: brain weight ratio in F0 and F1 at sacrifice	84	None	-	-	1	45.9	0.0064

Notes:
Significance $p < 0.05$ or $p < 0.01$
m = male; f = female; SRBC = sheep red blood cell; IgM = immunoglobulin M; GD = gestation day
[1] serum from pups on PND 22

The external doses in each of the studies varied. The NOAELs ranged from 0.64 to 1.88 mg/kg/day. The corresponding average serum values ranged from 1.6 mg/L (rat) to 38.2 mg/L (mouse). At the LOAEL, the average serum values range from 38 µg/mL (mouse) to 87.6 µg/mL (monkey) at doses estimated to represent about 56% to 96 % of steady state. At the low end of the range the effects of concern are observed in neonates (low birth weight, delays in developmental endpoints, with increased kidney weight at sacrifice later in life).

ED_005024_00000285-00049

Much of the variability in the average serum levels for the LOAELs was due to differences in the doses used in the individual studies. For example, two of the modeled endpoints (Wolf et al. 2007) identified low birth weights in mouse pups as the critical effect, but had a single external dose that was 3 to 5 times higher than the low dose from the Lau et al. (2006) mouse study (1 mg/kg/day).

Among the studies conducted in mice, dose was a more important variable in determining serum level and percent of steady state than duration of exposure. This is a characteristic of the nonlinear toxicokinetics exhibited by PFOA. The half-life for doses that exceed the resorption capacity of the kidney are shorter than lower doses that can be resorbed and thereby persist in serum over a longer exposure duration. For example, in Wolf et al. (2007), an 11-day dose of 5 mg/kg/day resulted in an average serum of 88 mg/L (82% of concentration at steady state or Css) whereas a 1 mg/kg/day dose for 17 days resulted in an average serum of 38 mg/L (56% of Css). In rats, dosed at 1 mg/kg/day, over two generations (84 days), an average serum of 45.9 mg/L at 87% of steady state was determined (Butenhoff et al. 2004a). A 91-day exposure (Palazzolo et al. 1993/Perkins et al. 2004) to 1.94 mg/kg/day resulted in a serum value of 77 mg/kg/day and was 91% of steady state. The endpoints in Butenhoff et al. (2004a) are effects on body weight and relative kidney weight in the adult F0 and F1 rats, while the endpoint for Palazzolo et al. (1993)/Perkins et al. (2004) was systemic increased liver weight with lower-level necrosis.

Assuming that MOA and susceptibility to toxicity do not vary and that pharmacokinetics alone explains variation, it is reasonable to expect similar concentrations to cause similar effects in humans and are more important than both dose and duration once steady state is attained.

5.1 Uncertainty Factors

An uncertainty factor for intraspecies variability (UF_H) of 10 is assigned to account for variability in the responses within the human populations because of both intrinsic (toxicokinetic genetic, life stage, health status) and extrinsic (life style) factors that can influence the response to dose. No information was available relative to variability in the human population that supports a factor other than 10.

An uncertainty factor for interspecies variability (UF_A) of 3 is applied to account for uncertainty in extrapolating from laboratory animals to humans (i.e., interspecies variability). The 3-fold factor is applied to account for toxicodynamic differences between the animals and humans. The HEDs were derived using average serum values from a model to account for toxicokinetic differences between animals and humans.

An uncertainty factor for LOAEL to NOAEL extrapolation (UF_L) of 10 is applied to all PODs other than the Palazzolo et al. (1993)/Perkins et al. (2004) and DeWitt et al. (2008) studies to account for use of a LOAEL for the POD. The POD for the Palazzolo et al. (1993)/Perkins et al. (2004) and DeWitt et al. (2008) studies are NOAELs for the effect identified as critical.

An uncertainty factor for extrapolation from a subchronic to a chronic exposure duration (UF_S) of 1 is applied because the PODs are based on average serum concentrations and determined to represent >80% of steady state for each study (81–91%), except for the Lau et al. (2006) developmental study (56%). The Lau et al. (2006) developmental HED was not adjusted

ED_005024_00000285-00050

for lifetime exposures because the average serum values associated with the developmental studies are more protective than those for the longer-term studies of systemic toxicity. A UF_S of 10 was applied to the DeWitt et al. (2008) study serum derived HED reflecting (74%) of steady state because the data suggest that longer term exposures to the same dose have the potential to increase serum values beyond the levels indicated by the 15-day study. In addition, the NOAEL for immunological effects (0.94 mg/kg/day) was a LOAEL for effects on liver weight in the absence of histological evaluation on both days 16 and 31 following a 15-day exposure (DeWitt et al. 2008). Thus, there is a potential that lifetime exposures at steady state can affect the liver and increase the risk for tissue damage.

A database uncertainty factor (UF_D) of 1 was applied to account for deficiencies in the database for PFOA. There are extensive human data from epidemiological data from the general population as well as worker cohorts. The epidemiology data provide strong support for the identification of hazards observed following exposure to PFOA in the laboratory animal studies and human relevance. However, uncertainties in the use of the available epidemiology data precluded their use at this time in the quantification of the effect level for derivation of the drinking water HA. In animals, acute, short term, subchronic and chronic studies, including a long term cancer study, are available. In addition, several developmental studies and a two-generation reproductive toxicity study evaluating exposure of pregnant dams and offspring to PFOA are available.

5.2 RfD Determination

Table 5-2 provides the calculations for candidate RfDs using the HEDs derived from the NOAEL or LOAEL average serum concentrations using pharmacokinetic modeling based on the serum values measures collected at animal sacrifice. Uncertainty factors (see section 5.1) were applied to each POD, and Table 5-2 illustrates the array of candidate RfD outcomes. Each POD is affected by the doses used in the subject study, the endpoints monitored, and the animal species/gender studied. Thus, the array of outcomes, combined with knowledge of the individual study characteristics helps to inform selection of an RfD that will be protective for humans. Other than DeWitt et al. (2008) and Lau et al. (2006), all of the selected studies had serum levels that had reached > 80% of Css. It is important to note the relatively narrow range of RfDs across the multiple endpoints and study durations evaluated.

Using the pharmacokinetic model of Wambaugh et al. (2013), average serum PFOA concentrations were derived from AUC considering the number of days of exposure before sacrifice. The predicted serum concentrations were converted as described above to oral HEDs mg/kg/day for each corresponding serum measurement. The candidate RfDs in Table 5-2 range from 0.00002 to 0.00015 mg/kg/day across multiple endpoints. The RfD of 0.00002 mg/kg/day calculated from HED average serum values from Lau et al. (2006) was selected. This RfD is derived from reduced ossification of the proximal phalanges (forelimb and hindlimb) and accelerated puberty in male pups (4 days earlier than controls) as the critical effects. The POD for the derivation of the RfD for PFOA is the HED of 0.0053 mg/kg/day that corresponds to a LOAEL that represents approximately 60% of steady-state concentration. An UF of 300 (10 UF_H, 3 UF_A, and 10 UF_L) was applied to the HED LOAEL to derive an RfD of 0.00002 mg/kg/day.

ED_005024_00000285-00051

Table 5-2. Candidate RfDs Derived from the HEDs from the Pharmacokinetic Model Average Serum Values

POD	HED POD mg/kg/day	UF_H	UF_A	UF_L	UF_S	UF_D	UF_{total}	Candidate RfD mg/kg/day
PK-HED_{NOAEL} Palazzolo et al. (1993)/Perkins et al. (2004) rats; ↑liver weight/necrosis	0.0044	10	3	-	-	-	30	0.00015
PK-HED_{LOAEL} Wolf et al. (2007) GD1-17 mice; ↓Pup body weight	0.0109	10	3	10	-	-	300	0.00004
PK-HED_{LOAEL} Wolf et al. (2007) GD 7-17 mice; ↓Pup body weight (serum from pups on PND 22)	0.0123	10	3	10	-	-	300	0.00004
PK-HED_{NOAEL} DeWitt et al. (2008) mice; ↓ IgM response to SRBC	0.0053	10	3	-	10	-	300	0.00002
PK-HED_{LOAEL} Lau et al. (2006) mice decreased ↓ pup ossification (m, f), accelerated male puberty	0.0053	10	3	10	-	-	300	0.00002
PK-HED_{LOAEL} Butenhoff et al. (2004a) ↓ relative body weight/↑ relative kidney weight and ↑kidney: brain weight ratio in F0 and F1 at sacrifice	0.0064	10	3	10	-	-	300	0.00002

Notes:
PK-HED = pharmacokinetic human equivalent dose; NOAEL = no observed adverse effect level; LOAEL = lowest observed adverse effect level; GD = gestation day; IgM = immunoglobulin M; m = male; f = female; SRBC = sheep red blood cell; UF_H = intraindividual uncertainty factor; UF_A = interspecies uncertainty factor; UF_S = subchronic to chronic uncertainty factor; UF_L = LOAEL to NOAEL uncertainty factor; UF_D = incomplete database uncertainty factor; UF_{total} = total (multiplied) uncertainty factor

Decreased pup body weights also were observed in studies conducted by Wolf et al. (2007), White et al. (2009), and Lu et al. (2015) using mice receiving external doses within the same order of magnitude (1, 3, and 5 mg/kg/day respectively) as those chosen for the RfD. The selected RfD from the reproductive and developmental studies is supported by the longer term RfD for effects on the response of the immune system to external challenges as observed following the short-term exposures to mature mice and the effects on kidney weight observed at the time of sacrifice in the F0 and F1 adult males that provided the serum in the Butenhoff et al. (2004a) study (DeWitt et al. 2008).

Support for the selected RfD also is provided by other key studies with NOAELs and LOAELs similar to those used for quantification, but lacking serum data that could be used for modeling. There were effects on liver weight and hepatic hypertrophy in the Perkins et al. (2004) and DeWitt et al. (2008) studies that were modeled but not considered in the derivation of the

ED_005024_00000285-00052

RfD because of a lack of data to demonstrate adversity as determined by the Hall et al. (2012) criteria at the dose causing the liver effects but not the effects identified as critical. The LOAEL for evidence of hepatic necrosis and other signs of tissue damage in the F1 male rat pups from the Butenhoff et al. (2004a) study was 3 mg/kg/day; the NOAEL was 1 mg/kg/day. In the Loveless et al. (2008) study, the LOAEL for increased relative liver weight accompanied by focal liver necrosis in male rats was 10 mg/kg/day and the NOAEL was 1 mg/kg/day, while in male mice, the LOAEL for the same effect was 1 mg/kg/day and the NOAEL was 0.3 mg/kg/day following a 29-day exposure. In the study by Tan et al. (2013), the degree of damage to the liver at 5 mg/kg/day became more severe with increased necrosis, inflammation, and steatosis when animals were given a high-fat diet. The HED modeled from the average serum value in mice for the LOAEL (3 mg/L) from Wolf et al. (2007) and White et al. (2009) was 0.0110 mg/kg/day, about twice that for the rats in the Lau et al. (2006) study (0.0053 mg/kg/day). Both studies lacked a NOAEL. Each of these data sets support LOAELs for the critical study by Lau et al. (2006) selected for RfD derivation and, as a consequence, the HED derived from modeled average serum values.

6.0 HEALTH ADVISORY VALUES

6.1 Relative Source Contribution

As described in section 2.2 and below, humans can be exposed to PFOA and precursor chemicals via multiple sources, including air, food, and consumer and industrial products (including textiles and rugs). The most common route of exposure to PFOA is via the diet, followed by indoor dust, especially for children.

Food is a significant source of exposure to PFOA: It has been detected in a variety of foods including snack foods, vegetables, meat, dairy products, human breast milk, and fish. Occurrence in food products can result from the use of contaminated water in processing and preparation; growth of food in contaminated soils; direct and indirect exposures of domestic animals to PFOA from drinking water, consumption of plants grown in contaminated soil, and through particulate matter in air; fish from contaminated water ways; and packaging materials.

PFOA has been detected in finished drinking water samples collected by EPA and others. PFOA is not regulated under the SDWA and was included in EPA's UCMR 3. PFOA was detected at a small number of PWSs (0.9%) through this monitoring program. Therefore, there is potential exposure to PFOA from drinking water ingestion.

The vapor pressure of PFOA indicates that volatilization is low; however, PFOA can be released into the atmosphere from industrial and municipal waste incinerators and adsorb to airborne particulates. It can be transported long distances through the atmosphere and has been detected globally at low concentrations. Inhalation of PFOA is possible, and it has been measured in indoor air in residential, commercial, and office settings because of its use in carpets, textiles, paint, furniture, and other consumer products. Both air and dust can be a vehicle for volatile telomer alcohols that metabolically degrade to PFOA. Given the widespread commercial and industrial use of PFOA and its physical properties, air is a potential source of exposure to it and the C8:2 telomer alcohol precursors.

ED_005024_00000285-00053

PFOA also has been detected in soils and dust from carpets and upholstered furniture in homes, offices, and vehicles. Incidental exposure from soils and dust is an important exposure route, particularly for small children because of their hand-to-mouth behaviors. Also, the levels in soils and surface waters can affect the concentrations in local produce, meat/poultry, dairy products, fish, and particulates in the air.

In summary, based on the physical properties and available exposure information for PFOA, there are many are potential sources. Following EPA's Exposure Decision Tree in its 2000 methodology (USEPA 2000), significant potential sources other than drinking water ingestion exist; however, information is not available to quantitatively characterize exposure from all of these different sources (Box 8B in the Decision Tree). Therefore, EPA recommends an RSC of 20% (0.20) for PFOA.

6.2 Lifetime Health Advisory

Based on the consistency of the responses across the chronic studies and those for reproductive and developmental endpoints, and with recognition of the use of developmental toxicity as the most sensitive endpoint, 0.00002 mg/kg/day was selected as the RfD for PFOA. This value is based on the HED for developmental effects (reduced ossification in male and female pups and accelerated puberty in male pups) from the Lau et al. (2006) study. The RfD that serves as the POD for the lifetime HA is applicable for effects other than those occurring during development. The candidate RfD values derived from the two-generation study by Butenhoff et al. (2004a) for effects on adult body weight plus relative liver and kidney weights in F0 and F1 male rats is the same as the value based on the developmental effects observed by Lau et al (2006). The candidate RfD from the DeWitt et al. (2008) study for suppression of the immunological response to a challenge is the same as that from Lau et al. (2006).

Due to the potential increased susceptibility during the time period of pregnancy and lactation, EPA used drinking water intake and body weight parameters for lactating women in the calculation of a lifetime HA for this target population during this potential critical time period. EPA used the rate of 54 mL/kg-day representing the consumers only estimate of combined direct and indirect community water ingestion at the 90th percentile for lactating women (see Table 3-81 in USEPA 2011b). Comparing the pregnant woman and the lactating woman, the lactating woman is the more protective scenario given her increased water intake rate for her body weight needed to support milk production. Additionally, human studies demonstrate that PFOA is transferred from mother to infant via cord blood and breast milk. A recent study showed that breast milk contributed > 83% of the PFOA exposure in 6-month-old infants (Haug et al. 2011).

The exposure factors applied to the RfD to derive the lifetime HA are specific to the most sensitive population and will be protective of pregnant women as well as of the general population. Thus, the protection conferred by the lifetime HA is broadly protective of public health.

The lifetime HA for PFOA is calculated as follows:

A DWEL is derived from the RfD and assumes that 100% of the exposure comes from drinking water.

ED_005024_00000285-00054

$$DWEL = \frac{RfD \times bw}{DWI}$$

$$DWEL = \frac{0.00002 \text{ mg/kg/day}}{0.054 \text{ L/kg-day}} = 0.00037 \text{ mg/L}$$

Where:

RfD = 0.00002 mg/kg/day; based on the LOAEL for reduced ossification of the proximal phalanges (forelimb and hindlimb) in male and female pups and accelerated (4 days earlier than controls) puberty in male pups of dams exposed to PFOA by gavage on gestation days 1 to 17 and sacrificed at weaning (Lau et al. 2006).

DWI/bw = 0.054 L/kg-day; 90th percentile consumers only estimate of combined direct and indirect community water ingestion for lactating women (see Table 3-81 in USEPA 2011b).

The lifetime HA is calculated after application of a 20% RSC (see section 6.1) as follows:

Lifetime HA = DWEL x RSC
= 0.00037 mg/L x 0.2
= 0.000074 mg/L (rounded to 0.00007 mg/L)
= 0.07 µg/L

The lifetime HA for PFOA is based on effects (reduced ossification in male and female pups and accelerated puberty in male pups) on the developing fetus resulting from exposures that occur during gestation and lactation. These developmental endpoints are the most protective for the population at large and are effects that can carry lifetime consequences for a less than lifetime exposure. Developmental toxicity endpoints (following less than chronic exposures during a defined period of gestation or lactation) can be analyzed in both acute and chronic exposure scenarios. Because the developing organism is changing rapidly and is vulnerable at various stages in development, a single exposure at a critical time in development can produce an adverse effect (USEPA 1991). Additionally, PFOA is extremely persistent in both the human body and the environment; thus, even a short-term exposure results in a body burden that persists for years and can increase with additional exposures.

Because the critical effect identified for PFOA is a developmental endpoint and can potentially result from a short-term exposure during a critical period of development, EPA concludes that the lifetime HA for PFOA is applicable to both short-term and chronic risk assessment scenarios. Thus, the lifetime HA of 0.07 µg/L also applies to short-term exposure scenarios (weeks to months) to PFOA in drinking water, including during pregnancy and lactation.

Adverse effects observed following exposures to PFOA and PFOS are the same or similar and include effects on serum lipids, birth weight, and serum antibodies in humans. Among the animal studies, there are common effects on the liver, neonate development, and responses to immunological challenges. Both compounds also were associated with tumors in long-term animal studies. The effects that serve as the basis for the RfDs for both PFOA and PFOS are developmental endpoints (reduced ossification and accelerated puberty in males for PFOA and decreased pup birth weight for PFOS (USEPA 2016a, 2016b). Because the RfDs for both PFOA

ED_005024_00000285-00055

and PFOS are based on similar developmental effects and are numerically identical, where these two chemicals co-occur at the same time and location in a drinking water source, a conservative and health protective approach that EPA recommends would be to compare the sum of the concentrations ([PFOA] + [PFOS]) to the HA (0.07 μg/L).

7.0 QUANTIFICATION OF CANCER RISK

The evidence for the carcinogenicity of PFOA is considered suggestive because only one species has been evaluated, and the tumor responses (liver, testicular Leydig cell, and pancreatic acinar cell tumors) occurred primarily in males. Dose-response data are only available for the LCTs in one study. The dose-response data on LCTs from the (Butenhoff et al. (2012) studies were modeled to provide a perspective on the magnitude of the potential cancer risk as it compares with the level of protection provided by the RfD.

Under EPA's *Guidelines for Carcinogen Risk Assessment* (USEPA 2005), when there is Suggestive Evidence for Carcinogenic Potential for a chemical, a dose-response assessment would generally not be attempted. The guidelines state that, when the evidence includes a well-conducted study, quantitative analyses could be useful for some purposes—for example, by providing a sense of the magnitude and uncertainty of potential risks, ranking potential hazards, or setting research priorities. The data from the Butenhoff et al. (2012) study are adequate to support a quantitative cancer dose-response assessment for PFOA's testicular tumors. The epidemiology studies demonstrate an association of serum PFOA with kidney and testicular tumors among highly exposed members of the general population. Thus, EPA concluded that a quantitative analysis could be useful by providing a sense of the magnitude of potential carcinogenic risk.

The dose-response data for LCTs in rats was analyzed using the multistage cancer model for a dichotomous data set to predict the dose at which a 4% increase in tumor incidence would occur (see appendix A). A benchmark response of 4% was chosen as the low end of the observed response range within the study results. The resulting benchmark dose level ($BMDL_{04}$) was 1.99 mg/kg/day, which yields a HED of 0.58 mg/kg/day and a slope factor of 0.07 $(mg/kg/day)^{-1}$. The cancer slope factor was calculated to determine if a lifetime HA derived from the RfD would be protective for the cancer endpoint. As a comparative analysis, the concentration of PFOA in drinking water that would have a one-in-1-million chance of an increased cancer risk was calculated using the oral slope factor for testicular tumors, assuming a default adult body weight of 80 kg (mean weight for adults ages 21 and older) (Table 8.1 in USEPA 2011b) and a default drinking water intake rate of 2.5 L/day (consumers only estimate of combined direct and indirect community water ingestion at the 90th percentile for adults ages 21 and older) (Table 3-33 in USEPA 2011b). The resultant 0.5 μg/L value is greater than the lifetime HA (0.07 μg/L) based on noncancer effects (see section 6.22.2.), indicating that the HA derived based on the developmental endpoint is protective for the cancer endpoint.

$$10^{-6} \text{ cancer risk} = \frac{0.000001 \times 80 \text{ kg}}{(0.07 \text{ mg/kg/day} \times 2.5 \text{L/d})} = 0.00046 \text{ mg/L rounded to } 0.5\ \mu\text{g/L}$$

ED_005024_00000285-00056

8.0 EFFECTS CHARACTERIZATION

8.1 Uncertainty and Variability

The variability and uncertainty in the lifetime HA is a function of both intrinsic and extrinsic factors. EPA's HESD for PFOA identified 20 short- or long-term studies that provided dose-response information and were considered during the quantitative assessment of risk (USEPA 2016a). The range of external dose NOAELs among the 20 studies is 0–30 mg/kg/day for females and 7.5 mg/kg/day for males. The LOAELs range from zero to 30 mg/kg/day for females and zero to 14.2 mg/kg/day for males (USEPA 2016a). Six dose-response data sets included the serum data necessary for modeling to derive HEDs for use as the POD for the RfD. Average serum values from those studies were chosen for use in the derivation of the RfD. The external dose range for the NOAELs in the modeled studies is 0–1.88 mg/kg/day and the LOAEL range is 1–5 mg/kg/day (USEPA 2016a). EPA believes the uncertainty in the chosen POD and the reliance on studies with serum data is minimized because of the large and extensive database examining the PFOA hazard and the selection of reduced ossification, and accelerated male puberty as the critical effects with lifetime implications at a LOAEL dose (1 mg/kg/day) from the low end of the narrow range of values evaluated.

The intrinsic uncertainties in the risk assessment reflect the fact that the NOAELs and LOAELs are derived using central tendency estimates for variables such as body weight, food and drinking water intakes, and dose. The central tendency estimates are derived from small numbers of genetically, relatively similar animals representing one or more strains of rats or mice living in controlled environments. The animals lack the heterogeneous genetic complexity, behavioral diversity, and complex habitats experienced by humans. These differences, to some extent, are minimized through the use of the modeled central tendency outcomes and their standard deviations to help inform the application of the uncertainty factors.

Variability in the study outcomes is extrinsically a function of study design and the endpoints monitored. Studies of systemic toxicity monitor an array of endpoints that are not evaluated in studies of reproductive, developmental, neurological, and immunological toxicity. The reverse is true for the other types of toxicity studies compared to standard short-term to long-term systemic studies. Studies of systemic toxicity do not often examine neurological or immunological endpoints. Increases in liver weight were seen in many of the studies with dose-response information but only a few of the studies carried out a histological evaluation of the liver to support a determination of whether the increase in liver weight could be classified as adverse according to the Hall et al. (2012) criteria.

The RfD is based on the HED derived from serum levels at the LOAEL from developmental study in mice with application of an uncertainty factor of 300 to cover extrapolation from a LOAEL to a NOAEL, variability in the human population, and differences in the ways humans and rodents respond to the PFOA that reaches their tissues (Lau et al. 2006). The selected RfD is based on the developmental effects in neonates to provide protection to both the sensitive life stages and the general population. The RfD is supported by the outcomes from two other studies based on different endpoints, Butenhoff et al. (2004a) and DeWitt et al. (2008), with RfDs for systemic effects on liver, kidney and the immune system. These data increase the confidence in the RfD.

ED_005024_00000285-00057

8.2 Use of Human Epidemiology Data

The human epidemiology studies provide evidence of an association between PFOA exposure and health effects in humans, and is another line of evidence supporting this assessment. The human data demonstrate an association between PFOA exposure and endpoints, including effects on serum lipids, antibody responses, fetal growth and development, and the liver. They provide support for identification of hazards of PFOA exposure. The associations observed for serum lipids, and reproductive parameters and immunotoxicity are the strongest. For many endpoints, however, the results are inconsistent. Although the human studies collectively support the conclusion that PFOA exposure is a hazard, EPA concluded that, based on several uncertainties associated with the database, the human studies are adequate for use qualitatively in the identification hazard at this time. These considerations are discussed below.

Although mean serum values are presented in the human studies, actual estimates of exposure (i.e., doses/duration) are not available. Thus, the serum level at which the effects were first manifest and whether the serum had achieved steady state or was in decline at the point the effect was evaluated cannot be determined. The NHANES and C8 study data indicate that serum levels in the general population are declining. Because epidemiology data are a reflection of the serum concentration at the time the sample was collected, there is no way to determine if levels were previously higher and had decreased. All of the C8 study serum samples were collected after the PFOA peak exposures had presumably passed. The half-life measurement for the general population of the Little Hocking area was derived from declining serum concentrations over time, demonstrating that serum levels among that population were not constant (Bartell et al. 2010).

Some of the human exposure that results in serum PFOA can come from telomer alcohol PFOA derivatives that break down metabolically to PFOA (Gebbink et al. 2015; Jogsten et al. 2012). The derivatives do not originate from PFOA in drinking water; they usually originate from diet and materials used in the home. Thus, there is added uncertainty in the observed epidemiological associations between serum PFOA and health effects.

Although the epidemiology studies provide valuable associations between exposure to PFOA and the effects seen in animal studies, most of the subjects of the epidemiology studies had other perfluorinated carboxylates and sulfonates and/or other biopersistent contaminants in their blood. Although the study designs adjust for other potential toxicants as confounding factors, their presence constitutes a level of uncertainty that is usually absent in the animal studies.

The database for PFOA includes extensive human data from epidemiology studies of the general population as well as worker cohorts. Data from oral short-term, subchronic, chronic (including evaluation of cancer), reproductive, and developmental studies in laboratory animals also are available. Many of the effects observed in the human epidemiology studies are similar to those seen in the animal studies.

8.3 Consideration of Immunotoxicity

Both human and animal studies have demonstrated the potential effect of PFOA on the immune system. However, there are uncertainties related to MOA and the level, duration, and/or

ED_005024_00000285-00058

timing of exposure that are not yet clearly delineated. As a result, EPA used the animal data rather than the human data to quantify the dose response for immunotoxicity for PFOA.

Taken together, available human studies do not provide consistent evidence of a significant association between PFOA exposure and serological vaccine responses in general (Grandjean et al. 2012; Granum et al. 2013; Looker et al. 2014). Within each study, most estimated associations were statistically nonsignificant, and results were inconsistent by vaccine type and by outcome classification. Authors provided no *a priori* biological hypothesis to explain why PFOA exposure would impair the antibody response to one vaccine type but not another. Some authors suggested that their results could be explained by different immunostimulatory effects of different vaccines, but they did not elaborate on this hypothesis or provide supporting mechanistic evidence.

One issue related to use of immune biomarkers and antibody levels in human studies is whether small but statistically significant changes in these endpoints, when analyzed on a continuous scale, are clinically meaningful, particularly when most or all subjects are within the normal range. For PFOA, some epidemiology studies attempted to address this issue by analyzing outcomes dichotomized relative to standard reference values, with the implication that values outside the reference range indicate immune abnormalities (Emmett et al. 2006; Grandjean et al. 2012; Looker et al. 2014). A limitation of this approach is that a reference range is typically determined based on the mean plus or minus two standard deviations calculated from a group of healthy adults or children. By definition, 5% of the normal population falls outside of such a reference range (AACC 2015, cited in Chang et al. 2016). The only way to determine whether a given value outside a reference range is truly abnormal is to associate it with a clinical abnormality; this has not been done in most epidemiology studies of immune biomarkers.

Although Grandjean et al. (2012) found fairly consistent, albeit mostly statistically nonsignificant, intrastudy associations between childhood serum PFOA levels and poorer antibody responses against tetanus and diphtheria toxoids, associations with maternal prenatal serum PFOA and PFOS levels were inconsistent between vaccine types. Two studies were strengthened by their measurement of PFOA levels prior to ascertaining vaccine response (Grandjean et al. 2012; Granum et al. 2013), and one had the additional advantage of collecting exposure and outcome information at two time points each (Grandjean et al. 2012). However, the variability in findings by timing of exposure and outcome measurement in the latter study (e.g., mostly nonsignificant associations with prenatal PFOA concentrations, but several significant associations between higher PFOA concentrations at age 5 years and poorer vaccine response at age 7 years) makes the results difficult to interpret. This pattern of results could reflect a window of susceptibility in early childhood, but such an explanation remains conjectural.

None of the studies demonstrated a clinically recognizable increased risk of infectious diseases as a consequence of a diminished vaccine response. Overall, although these results are not sufficient to establish a causal effect of PFOA exposure on an impaired serological vaccine response, some of the positive associations are striking in magnitude and require replication in independent studies.

Chang et al. (2016) recently completed and published a systematic review of 24 epidemiology studies that reviewed a variety of endpoints among the general population,

ED_005024_00000285-00059

occupationally exposed workers, children, and adults and concluded that the available epidemiologic evidence is insufficient to reach a conclusion about a causal relationship between exposure to PFOA and PFOS and any immune-related, health condition in humans. The majority of the studies reviewed by the authors are included in EPA's HESDs for PFOA and PFOS (USEPA 2016a, 2016b). The authors identified numerous weaknesses in study designs, including lack of validation of self-reported medical conditions, basing conclusions on significant associations without considering statistical significance, inadequate consideration of confounding factors, bias, and the role of chance being responsible for outcomes. After application of the Hill et al. (1965) criteria, they faulted the studies for "generally weak associations, no specific endpoints with consistent findings across all relevant studies, uncertainty about any critical duration of exposure and window(s) of susceptibility, mixed exposure-response trends, and a dearth of supportive animal and mechanistic data."

There remains a need for additional research on MOA, key biomarkers that are reliable indicators for the upstream effects elicited by the PFASs, the temporal relationship between exposure and outcome, plus the analytical and functional impact of PFASs binding to serum immunoglobins and/or related proteins.

8.4 Effects on Mammary Gland Development

Several studies in mice have examined postnatal mammary gland development in female mice. A qualitative/qualitative assessment found delayed mammary gland development of female CD-1 mouse pups following maternal doses ≥ 0.01 mg PFOA/kg in Macon et al. (2011) and Tucker et al. (2015). Macon et al. (2011) also found significant differences from controls in quantitative measures of longitudinal and lateral growth and numbers of terminal end buds at 1 mg/kg/day. However, Albrecht et al. (2013) found no significant differences in the average length of mammary gland ducts and the average number of terminal end buds per mammary gland per litter in female pups of PPARα wild type, PPARα-null, or hPPARα sv/129 following a maternal dose of 3 mg/kg using an approach to scoring that lacked a qualitative component adjustment such as that used by Macon et al. (2011) in identifying the 0.01 mg/kg/day dose as a LOAEL.

The approach to scoring mammary gland development was not consistent across studies and little information was provided on the qualitative components of the scores. This makes comparisons across studies difficult. Statistical significance was attained at higher dose levels for the quantitative portion of the Macon et al. (2011) scoring protocol than for the qualitative component of the score. Tucker et al. (2015) found that CD-1 mice were considerably more sensitive to effects on mammary gland development (LOAEL 0.01 mg/kg/day) than C57BL/6 mice (NOAEL 0.1 mg/kg/day). Scoring was conducted using the Macon et al. (2011) approach.

White et al. (2011) used doses of 0 or 1 mg PFOA/kg/day for F0 dams throughout gestation with and without the addition of drinking water containing 5-ppb PFOA beginning on gestation day 7 and continuing the contaminated drinking water during the production of two more generations; no persistent significant differences were found in the body weights of the pups in the F1 and F2 generations for the pups receiving 1 mg/kg/day, indicating a poor correlation between mammary duct branching patterns and the ability to support pup growth during lactation. The 5-mg/kg/day dose did affect body weight. Albrecht et al. (2013) also found no

ED_005024_00000285-00060

significant impacts on pup body weight in their one-generation assay at a dose of 3 mg/kg/day. Despite the diminished ductal network assessed in the qualitative mammary gland developmental score of the dams in White et al. (2011), milk production was sufficient to nourish growth in the exposed pups as reflected in the body weight measurements compared to controls at the 1-mg/kg/day dose. The MOA for PFOA-induced delayed mammary gland development is unknown and requires further investigation.

8.5 Alternative Exposure Scenarios

EPA is issuing a lifetime HA for PFOA of 0.07 µg/L to prevent a variety of adverse developmental effects to fetuses during pregnancy and to infants during breast feeding. Due to the potential increased susceptibility during this critical time period, EPA used drinking water intake and body weight parameters for lactating women to calculate the lifetime HA (see section 6.2). Specifically, EPA used the rate of 54 mL/kg-day representing the consumers only estimate of combined direct and indirect community water ingestion at the 90th percentile for lactating women (see Table 3-81 in USEPA [2011b]).

As a comparative analysis, EPA calculated a lifetime HA value for alternative exposure scenarios for the general population. Calculation of a lifetime HA value for the general population (adults ages 21 and older) is 0.1 µg/L, assuming a drinking water rate of 2.5 L/day and a mean body weight of 80 kg (see Tables 3-33 and 8-1 in USEPA[2011b]).

PFOA is extremely persistent in both the human body and the environment; thus, even a short-term exposure results in a body burden that persists for years and can increase if additional exposure occurs later. Human studies have shown that PFOA is transferred from mother to infant via cord blood and breast milk. The exposure scenario for the lactating woman is the most protective given her increased water intake rate to support milk production and thus is the basis for EPA's recommended lifetime HA for PFOA of 0.07 µg/L. The lifetime HA for PFOA is also protective of adverse health effects in the adult general population (e.g., testicular and kidney cancer, liver damage, immune effects).

8.6 Relative Source Contribution Considerations

EPA used the Exposure Decision Tree methodology to derive the RSC for this HA (USEPA 2000). Findings from studies on populations in the United States, Canada, and Western Europe support the conclusion that diet is the major contributor to total PFOA exposure, typically with drinking water and/or dust as important additional exposure routes, especially for sensitive subpopulations. Estimates of relative exposure from different sources vary widely, as described below.

- Tittlemier et al. (2007) conducted a total diet study, focused on collection and analysis of different food items. They concluded that diet represented approximately 60% of exposure to total PFASs, with a negligible contribution from drinking water, based on samples collected from two cities in Canada.
- Lorber and Egeghy (2011) used models to estimate exposures for adults and 2-year-olds. The data and analysis identify dietary ingestion as the major contributor to adult intake of PFOA, and dust and diet for young children in different media. The authors estimated

ED_005024_00000285-00061

PFOA exposure from drinking water at 17 ng/day or approximately 24% of total intake for both adults and children. As background concentrations of PFOA in water increase, drinking water represents a greater source of total dietary intake.

- Gebbink et al. (2015) estimated the relative contributions of the major exposure media to total direct and indirect PFOA exposures under assumptions of low (5th percentile), intermediate (median values), and high (95th percentile) exposures. The authors used a scenario-based risk assessment modeling approach with data collected in 2007 to estimate the relative contributions of diet, dust, water, and air to total exposures. Only data for samples collected in North America, Europe, Korea, and Japan were included in the evaluation. The authors point out that both the blood serum concentrations and the temporal trends of PFASs in the United States, Europe, and Japan are similar. The data for direct and indirect contributors to serum PFOA are presented graphically in the published paper. They are consistent with the following exposure patterns for the combination of direct and indirect (precursor) exposures in adults:
  - Low-exposure scenario: diet (~50%) > air (~25%) > dust (~15%) > water (~10%);
  - Intermediate-exposure scenario: diet (~45%) > dust (~35%) > water (~10%) ≈ air (~10%); and
  - High-exposure scenario: dust (~65%) > diet (~20%) > water (10%) > air (~5%).

As the environmental level increases, so does the contribution of precursors to total exposure, increasing from about 15% to 30% to 60% as the exposure increases from low to high.

The approaches and assumptions used in these studies vary widely; some uncertainties associated with these data include:

- Many of the data are obtained from review papers or individual studies conducted at single locations often in Europe and are not nationally representative.
- Concentrations range widely in exposure estimates.
- The ambient air and dust exposure estimates are limited, regional, and variable.
- Drinking water exposure varies among age groups and individuals.
- Because of recent reductions in use of PFOA and its precursors, it is difficult to assess current relative exposures to the general population.

Additionally, there is a lack of data on other routes of exposure:

- Estimates of dermal exposure to treated fabrics and inhalation exposure associated with contaminated water are not available.
- Drinking water exposure estimates apply only to direct ingestion of tap water and beverages or soups prepared locally. They do not generally include PFOA in water that becomes incorporated in solid foods during home preparation and cooking or that is present in commercial beverages.
- Transformation of PFOA precursors that decay or are metabolized to PFOA is a route that is rarely evaluated in dietary studies yet can contribute to total exposure. Air and dust can be vehicles for derivatives that metabolically degrade to PFOA.

Given these uncertainties, EPA used the Exposure Decision Tree methodology, described in section 6.1, to estimate an RSC of 20% for drinking water for the general population.

ED_005024_00000285-00062

8.7 Sensitive Populations: Gender Differences

Some animal species have gender differences that affect toxicity of PFOA. Sexually mature female rats excreted almost all of a 10-mg/kg dose of PFOA within 48 hours compared to only 19% excreted by male rats. Male hamsters excrete PFOA faster than female hamsters, and female rabbits excrete PFOA slightly faster than male rabbits. Male and female mice excrete PFOA at approximately the same rate (Hundley et al. 2006). Studies of the transporters involved in the toxicokinetics of PFOA demonstrate that they are differentially affected by the presence of male and female sex hormones (Cheng et al. 2006; Kudo et al. 2002). As studied in rats (Kudo et al. 2002), the male sex hormones increased half-life (decreased excretion) of PFOA while the female hormones were associated with shorter half-lives (increased excretion). The gender differences in toxicokinetics in mice are not as pronounced as those in rats. Work by Cheng et al. (2006) and Cheng and Klaassen (2009) demonstrated that the hormones affected transporters in the liver and kidney, protecting the females and increasing the sensitivity for males. Results of the NHANES data on PFOA suggest that in humans, serum levels are lower in females (Calafat et al. 2007a, 2007b; Jain 2014); both menstruation and lactation are excretory routes in females and shorten the half-life of PFOA during associated life phases.

In studies where both male and female rats were used, the males were more sensitive to toxicity than the females (Butenhoff et al. 2004a). Mice displayed similar sensitivities following PFOA exposure (Kennedy 1987). In the monkey studies, the number of animals per gender per dose group was too small to reveal a difference related to gender.

Unfortunately, much work remains to be done to determine whether the gender difference seen in rats is relevant to humans. Similarities are possible because the long half-life in humans suggests that they might be more like the male rat than the female rat. The broad range of half-lives in human epidemiology studies suggests a variability in human transport capabilities resulting from the isomeric composition of the PFOA and genetic variations in transporter structures and consequently in function (Y. Zhang et al. 2013, 2014). Genetic variation in human transporters are identified in a review by Zaïr et al. (2008).

8.8 Sensitive Populations: Developmental Effects

PFOA-exposure during development in rats and mice resulted in increased resorptions (mouse), increased fetal skeletal variation (rats, mouse), decreased neonatal survival (rat, mouse), decreased postnatal body weight (mouse), delayed eye opening and body hair growth (rat, mouse), delayed vaginal opening (mouse), accelerated preputial separation (mouse), and delayed mammary gland development (mouse) (Butenhoff et al. 2004a; Lau et al. 2006; Macon et al. 2011; Tucker et al. 2015; White et al. 2007, 2009, 2011; Wolf et al. 2007). Some effects were seen as low-dose exposures such as the ossification delays and accelerated puberty in male mice exposed via their dams to a dose of 1 mg/kg/day during gestation (Lau et al. 2006), the mammary gland effects (0.01 mg/kg/day) (Macon et al. 2011), and the postnatal effects on body weight in pups exposed to PFOA during gestation and lactation to doses of 3 or 5 mg/kg/day (White et al. 2009; Wolf et al. 2007). Only the low birth weight receives support from the epidemiology studies. The other effects generally lack correlates among the effects evaluated by the studies.

ED_005024_00000285-00063

In the Wolf et al. (2007) study, pup postnatal body weights were lower than controls for all exposure durations during the last 10 days of gestation evaluated. The authors found that the magnitude of the body weight effect was directly related to the days of exposure (i.e., 3, 5, 7, or 10); the longer the exposure, the greater the body weight deficit in the male and female pups during the PND 2–22 time period. In male but not female pups, the exposure duration deficits in body weight persisted up to PND 92. The difference in the male rat response over the PND 29–92 period likely reflects their longer half-life than females.

Both gestational and lactational exposures contribute to the impact of PFOA on body weight during early life as illustrated by cross-fostering control unexposed female pups with those dosed with PFOA. Three cross-fostering combinations were evaluated by White et al (2009): control pups nursed by exposed dams, exposed pups nursed by control dams, and exposed pups nursed by exposed dams. Two doses were evaluated: 3 and 5 mg/kg/day. The PND 1–10 body weight data were only provided for the 5-mg/kg/day dose. PFOA exposures significantly reduced pup body weights and increased liver weights. The body weight deficits compared to control were greatest for the gestation and lactation exposure combination and lowest for the lactation-only group.

Diet can influence the risk associated with PFOA exposures. Animal studies demonstrate an increased risk for liver steatosis in animals on a high-fat diet and possibly for insulin resistance (Hines et al. 2009; Quist et al. 2015; Tan et al. 2013). The epidemiology data are not supportive of a correlation with insulin resistance, but the observations of elevated serum triglycerides, especially among a highly exposed population, could be viewed as a risk factor for steatosis. Most of the epidemiology studies did not evaluate dietary factors as part of the study design for either birth weight or serum lipids (e.g., cholesterol, triglycerides, LDL).

9.0 ANALYTICAL METHODS

EPA developed a liquid chromatography/tandem mass spectrometry (LC/MS/MS) analytical method— Method 537—to monitor drinking water for 14 select perfluorinated alkyl acids that include PFOA (USEPA 2009b). Accuracy and precision data were generated for PFOA, PFOS, and the other 12 PFASs in reagent water, finished ground water, and finished surface water. This method identifies a single laboratory lowest concentration minimum reporting level or quantitation limit for PFOA at 5.1 ng/L (0.0051 μg/L) and for PFOS at 6.5 ng/L (0.0065 μg/L). The method-published detection limit for PFOA is 1.7 ng/L (0.0017 μg/L).

In this method, PFAS standards, extracts, and samples should not come into contact with any glass containers or pipettes because PFASs can potentially adsorb to the surface of the glassware. Polypropylene containers should be used instead. Also, these compounds can be found in commonly used laboratory supplies and equipment, such as PTFE products, liquid chromatograph solvent lines, methanol, aluminum foil, and solid phase extraction (SPE) sample transfer lines. These materials need to be routinely demonstrated to be free of interferences per the guidelines for laboratory reagent blanks described in the method. In summary, the method procedure involves passing a preserved 250-mL water sample (fortified with an extraction surrogate) through a SPE cartridge containing polystyrenedivinylbenzene (SDVB) to extract the method analytes and surrogates.

ED_005024_00000285-00064

The compounds are eluted from the SPE with a small amount of methanol. The extract is concentrated to dryness with nitrogen in a heated water bath, and then adjusted to a 1-mL volume with 96%:4% (vol/vol) methanol:water after adding the internal standards. The extract is injected into a liquid chromatograph that is interfaced to an MS/MS. The analytes are separated and identified by comparing the acquired mass spectra and retention times to reference spectra and retention times for calibration standards acquired under identical LC/MS/MS conditions. The concentration of each analyte is determined by using the internal standard technique. Surrogate analytes are added to all field and quality control samples to monitor the extraction efficiency of the method analytes. *Method 537: Determination of Selected Perfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)* (USEPA 2009b) is available for download at http://www.epa.gov/nerlcwww/ordmeth.htm.

10.0 TREATMENT TECHNOLOGIES

As mentioned above, PFOA is an organic compound in which the carbon-hydrogen bonds are replaced by carbon-fluorine bonds. This influences the chemical characteristics of both molecules and, therefore, will impact the effectiveness of any given drinking water treatment process. The characteristics of organic contaminants that treatment processes take advantage of include molecular size, solubility, ionic form, volatility, oxidizability, hydrolysis, photolysis, and biodegradability. Because fluorine is the most electronegative element, the carbon-fluorine bond will be one of the strongest bonds in nature, making it exceedingly resistant to biodegradation, hydrolysis, oxidation, and photolysis. Also, because PFOA is a dissolved contaminant that is resistant to oxidation to an insoluble form, treatment processes that are designed for particulate control such as conventional treatment will not be effective. This leaves adsorption, ion exchange resins, and high-pressure membranes as the technologies that can be effective. The following subsections discuss the effectiveness of commonly used drinking water technologies in rough order of applicability for PFOA and PFOS removal. Additional information can be found on EPA's Drinking Water Treatability Database (USEPA 2015b) at https://iaspub.epa.gov/tdb/pages/general/home.do.

To varying degrees of applicability, the technologies discussed below can be employed in centralized drinking water facilities or in a distributed fashion such as point-of-entry (POE) or point-of-use (POU) applications in buildings and homes. As they imply, POE systems treat the water as it enters the building or house, and POU systems treat the water where used, such as a kitchen or bathroom sink. Although the cost of treatment varies with scale, the following general discussion on the relative effectiveness of each technology applies regardless of scale. One reference below specifically addresses POU systems (MDH 2008).

Activated Carbon Adsorption

Activated carbon is applied in either powdered or granular form. Either can be effective; however, because PFOA and PFOS have moderate adsorbability, the specifics of the design are very important for achieving successful treatment.

ED_005024_00000285-00065

Powdered Activated Carbon

Powdered activated carbon (PAC) is often applied prior to, or within a, conventional treatment train. The contaminant-loaded PAC is then removed along with the other particulates. Although some studies have shown limited PFOA and PFOS removal in plants using PAC (Quiñones and Snyder 2009), in general, PAC can be an effective treatment strategy for the removal of PFOA and PFOS given the correct choice of carbon type, high enough carbon doses, and adequate contact time (Dudley et al. 2015; Hansen et al. 2010).

Granular Activated Carbon

Granular activated carbon is applied as a filtration step either as a filter adsorber where a relatively short carbon cap is added to an existing sand filter, or as a post-filter adsorber where a deeper bed is employed as a stand-alone unit following a typical sand filter. Because PFOA and PFOS have moderate adsorbability, a post-filter adsorber with a deeper bed is considered a safer approach. In general, granular activated carbon treatment was found to be effective given the correct choice of carbon, adequate bed depth, moderate or low hydraulic loading rate, and frequent replacement or regeneration of the carbon (Appleman et al. 2013, 2014; MDH 2008; Shivakoti et al. 2010; Takagi et al. 2008).

Membrane Technologies

There are many types of membrane technologies. They can be broadly classified as either low-pressure or high-pressure systems. This distinction also corresponds to the general effectiveness of removing PFOA and PFOS with low-pressure membranes being ineffective, while high-pressure membranes are effective.

Low-pressure Membranes

Low-pressure systems incorporating cartridge, microfiltration, or ultrafiltration membranes are designed for particulate control. They have relatively large pore structures through which water and dissolved contaminants can easily flow, leaving behind larger particulate matter that includes turbidity and microbiological agents. Low-pressure membranes have been found to be ineffective for PFOA and PFOS control (McLaughlin et al. 2011; Thompson et al. 2011). This is consistent with other treatment processes (e.g., conventional treatment) that target particulate contaminants but not dissolved contaminants. However, as with conventional treatment, low-pressure membranes can be effective if they are used in conjunction with PAC. The PAC will adsorb the PFOA and PFOS, and the low-pressure membrane will remove the spent PAC. Care should be taken in the design of the system, including the choice of the PAC as mentioned above (Dudley et al. 2015).

High-pressure Membranes

High-pressure systems have a much tighter pore structure, relying on water diffusing through the membrane material. High-pressure systems such as nanofiltration and reverse osmosis can reject not only particulates, but also dissolved constituents such as organic contaminants and salts. Reverse-osmosis membranes are the tightest of the high-pressure systems, having the ability to reject monovalent salts such as sodium chloride (e.g., sea water desalination). High-pressure membrane systems have been shown to be very effective for PFOA and PFOS

ED_005024_00000285-00066

(Appleman et al. 2013, 2014; MDH 2008; Quiñones and Snyder 2009; Tang et al. 2006, 2007; Thompson et al. 2011).

Ion Exchange Resin Treatment

There are two broad categories of ion exchange resins: cationic and anionic. Cationic exchange resins are effective for removing positively charged contaminants. Anion exchange resins are effective for negatively charged contaminants. Because PFOA and PFOS are negatively charged in drinking waters, cation-exchange resins will not be effective, and therefore, have not been studied. There have been studies that have evaluated different anion exchange resins (macroporous styrenedivinylbenzene, gel-type polystyrene divinylbenzene, and polyacrylic quaternary amine resins). Generally, anion exchange resins have been found to be effective for PFOA and PFOS removal (Appleman et al. 2014; Carter and Farrell 2010; Chularueangaksorn et al. 2013; Dudley et al. 2015), although the design of the system including regeneration effectiveness is important. Special consideration should be given to dealing with the regenerate brine waste, and if frequent regenerations are needed, to the amount of operator effort and expertise required.

Oxidation / Disinfection

Oxidation/disinfection processes can transform certain contaminants into different molecules, which ideally have less toxicity. It also can transform certain dissolved constituents into a higher oxidation state that might be less soluble (e.g., iron, manganese). The less soluble form can then be precipitated and removed in the floc or on a media filter of a conventional treatment system. Due to the strength of the carbon-fluorine bond, all drinking water oxidants or disinfectants have been shown to be ineffective in reacting PFOA or PFOS. This has been shown numerous times for common oxidative/disinfection agents such as packed tower aeration, chloramination, chlorination, ozonation, potassium permanganate, and ultraviolet (UV) treatment (Appleman et al. 2014; Hori et al. 2004; Liu et al. 2012; McLaughlin et al. 2011; Quiñones and Snyder 2009; Schröder and Meesters 2005; Shivakoti et al. 2010; Thompson et al. 2011). It also is true for advanced oxidation processes (AOPs) that use the nonselective hydroxyl radical as an oxidative agent. There are many ways of producing hydroxyl radicals, usually combining technologies such as hydrogen peroxide plus iron (Fenton's reagent), ozone plus peroxide, UV plus titanium dioxide, UV plus ozone, and UV plus peroxide. All of these combinations have been shown to be ineffective for PFOA and PFOS control at reasonable contact times (Benotti et al. 2009; Hori et al. 2004; Schröder and Meesters 2005; Tellez 2014).

Biological Treatment

Similar to the discussion on oxidation processes, because of the strength of the carbon-fluorine bond, it is expected that both aerobic and anaerobic biological treatment processes (e.g., biofiltration, bioreactors) are expected to be ineffective for PFOA and PFOS removal. A number of researchers have found this to be the case (Kwon et al. 2014; Saez et al. 2008; Thompson et al. 2011). Some results have shown that specific microbes could have the ability to break the carbon-to-carbon bonds in PFOS, albeit slowly; however, this cannot be engineered into a consistent and robust treatment process (Kwon et al. 2014).

ED_005024_00000285-00067

Conventional Treatment

Conventional treatment is commonly defined as a series of successive steps: rapid mix, coagulation, flocculation, sedimentation, and filtration. Certain variations exist, such as direct filtration, which does not employ a sedimentation step. Regardless of the configuration, conventional treatment is designed to remove particulates (e.g., turbidity and microbiological agents). Dissolved contaminants, however, will not be removed by conventional treatment. The exception is when the contaminants are first oxidized to an insoluble form (e.g., iron, manganese), or if they are exceedingly hydrophobic as evidenced by an extremely low solubility. Therefore, because of the resistance of PFOA and PFOS to oxidation to an insoluble form and their moderately high solubility, conventional treatment is not expected to be effective in their removal, even in enhanced coagulation conditions. Numerous studies have confirmed this statement (Appleman et al. 2014; Loos et al. 2007; Quiñones and Snyder 2009; Shivakoti et al. 2010; Skutlarek et al. 2006; Tabe et al. 2010; Takagi et al. 2008; Thompson et al. 2011; Xiao et al. 2013).

Similar to low-pressure membranes, conventional treatment can be effective if it is used in conjunction with PAC (see above). The PAC will adsorb the PFOA and PFOS, and the conventional treatment system will remove the spent PAC in the sedimentation and filtration steps. Care should be taken in the design of the system, including the choice of the PAC (Dudley et al. 2015).

ED_005024_00000285-00068

11.0 REFERENCES

AACC (American Association for Clinical Chemistry). 2015. *Reference Ranges and What They Mean. Lab Tests Online*. Accessed May 2016. https://labtestsonline.org/understanding/features/ref-ranges/start/6.

Abbott, B.D., C.J. Wolf, J.E. Schmid, K.P. Das, R.D. Zehr, L.Helfant, S. Nakayama, A.B. Lindstrom, M.J. Styrnar, and C. Lau. 2007. Perfluorooctanoic acid-induced developmental toxicity in the mouse is dependent on expression of peroxisome proliferator-activated receptor-alpha. *Toxicological Science* 98:571–581.

Ahrens, L., M. Shoeib, T. Harner, S.C. Lee, R. Guo, and E.J. Reiner. 2011. Wastewater treatment plant and landfills as sources of polyfluoroalkyl compounds to the atmosphere. *Environmental Science & Technology* 45:8098–8105.

Albrecht, P.P., N.E. Torsell, P. Krishnan, D.J. Ehresman, S.R. Frame, S.-C. Chang, J.L. Butenhoff, G.L. Kennedy, F.J. Gonzalex, and J.M. Peters. 2013. A species difference in the peroxisome proliferator-activated receptor α-dependent response to the developmental effects of perfluorooctanoic acid. *Toxicological Science* 131: 568–582.

Andersen, M.E., H.J. Clewell, III, Y-M. Tan, J.L. Butenhoff, and G.W. Olsen. 2006. Pharmacokinetic modeling of saturable, renal resorptions of perfluoroalkylacids in monkeys-probing the determinants of long plasma half-lives. *Toxicology* 227:156–164.

Anzai, N., Y. Kanai, and H. Endou. 2006. Organic anion transporter family: Current knowledge. *Journal of Pharmacological Science* 100:411–426.

Apelberg, B.J., L.R. Goldman, A.M. Calafat, J.B. Herbstman, Z. Kuklenyik, J. Heidler, L.L. Needham, R.U. Halden, and F.R. Witter. 2007. Determinants of fetal exposure to polyfluoroaklyl compounds in Baltimore, Maryland. *Environmental Science & Technology* 41:3891–3897.

Appleman, T.D., E.R.V. Dickenson, C. Bellona, and C.P. Higgins. 2013. Nanofiltration and Granular Activated Carbon Treatment of Perfluoroalkyl Acids. *Journal of Hazardous Materials* 260:740–746.

Appleman, T.D., C.P. Higgins, O. Quiñones, B.J. Vanderford, C. Kolstad, J.C. Zeigler-Holady, and E.R.V. Dickenson. 2014. Treatment of Poly- and Perfluoroalkyl Substances in U.S. Full-Scale Water Treatment Systems. *Water Research* 51: 246–55.

Armstrong, D.L., N. Lozano, C.P. Rice, M. Ramirez, and A. Torrents. 2016. Temporal trends of perfluoroalkyl substances in limed biosolids from a large municipal water resource recovery facility. *Journal of Environmental Management.* 165:88–95.

ATSDR (Agency for Toxic Substances and Disease Registry). 2005. *Health Consultation, 3M Chemolite, Perfluorochemical Releases at the 3M – Cottage Grove Facility*. City of Cottage Grove, Washington County, Minnesota. EPA Facility ID: MND006172969, February 18, 2005. Accessed May 2016. http://www.atsdr.cdc.gov/HAC/pha/3M-CGF021805-MN/3M-CGF021805-MN_pt1.pdf.

ATSDR (Agency for Toxic Substances and Disease Registry). 2015. *Toxicological Profile for Perfluoroalkyls*. Draft for Public Comment. Agency for Toxic Substances and Disease Registry, Public Health Service, United States Department of Health and Human Services, Atlanta, GA. Accessed May 2016. http://www.atsdr.cdc.gov/ToxProfiles/tp200.pdf.

Barry, V., A. Winquist, and K. Steenland. 2013. Perfluorooctanoic acid (PFOA) exposures and incident cancers among adults living near a chemical plant. *Environmental Health Perspectives* 121:1313–1318.

Bartell, S., A. Calafat, C. Lyu, K. Kato, P.B. Ryan, and K. Steenland. 2010. Rate of decline in serum PFOA concentrations after granular activated carbon filtration at two public water systems in Ohio and West Virginia. *Environmental Health Perspectives* 118:222–228.

Beesoon, S., and J.W. Martin. 2015. Isomer-specific binding affinity of perfluorooctanesulfonate (PFOS) and perfluorooctanoate (PFOA) to serum proteins. *Environmental Science & Technology* 49(9):5722–5731.

Begley, T.H., K. White, P. Honigfort, M. L. Twaroski, R. Neches, and R. A. Walker. 2005. Perfluorochemicals: Potential sources of and migration from food packaging. *Food Additives and Contaminants* 22(10):1023–1031.

Benbrahim-Tallaa, L., B. Laubry-Secretan, D. Loomis, K.Z. Guyton, Y. Grosse, F. El Ghissassi, V. Bouvard, N. Guha, H. Mattock, and K. Straif, on behalf of the International Agency for Research on Cancer Monograph Working Group. 2014. Carcinogenicity of perfluorooctanoic acid, tetrafluoroethylene, dichloromethane, 1,2-dichloropropane, and 1,3-propane sultone. *Lancet Oncology* 15(9):924–925.

Benotti, M.J., B.D. Stanford, E.C. Wert, and S.A. Snyder. 2009. Evaluation of a photocatalytic reactor membrane pilot system for the removal of pharmaceuticals and endocrine disrupting compounds from water. *Water Research* 43:1513–1522.

Beser, M.I., O. Pardo, J. Beltran, and V. Yusa. 2011. Determination of per- and polyfluorinated substances in airborne particulate matter by microwave-assisted extraction and liquid chromatography–tandem mass spectrometry. *Journal of Chromatography A* 1218:4847–4855.

Bhavsar, S.P., X. Zhang, R. Guo, E. Braekevelt, S. Petro, N. Gandhi, E.J. Reiner, H. Lee, R. Bronson, and S. A. Tittlemier. 2014. Cooking fish is not effective in reducing exposure to perfluoroalkyl and polyfluoroalkyl substances. *Environmental International* 66:107–114.

Biegel, L.B., M.E. Hurtt, S. R. Frame, J.C. O'Connor, and J.C. Cook. 2001. Mechanisms of extrahepatic tumor induction by peroxisome proliferators in male CD rats. *Toxicological Science* 60:44–55.

Blaine, A.C., C.D. Rich, E.M. Sedlacko, L.S. Hundal, K. Kumar, C. Lau, M.A. Mills, K.M. Harris, and C.P. Higgins. 2014. Perfluoroalkyl acid distribution in various plant compartments of edible crops grown in biosolids-amended soils. *Environmental Science & Technology* 48:7858–7865.

ED_005024_00000285-00070

Bonefeld-Jørgensen, E.C., M. Long, S.O. Fredslund, R. Bossi, and J. Olsen. 2014. 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* 25(11):1439–1448.

Boulanger, B., J. Vargo, J.L. Schnoor, and K.C. Hornbuckle. 2004. Detection of perfluorooctane surfactants in Great Lakes water. *Environmental Science & Technology* 38(15):4064–4070.

Buck, R.C., J. Franklin, U. Berger, J.M. Conder, I.T. Cousins, P. de Voogt, A.A. Jensen, K. Kannan, S.A. Mabury, and S.P.J. van Leeuwen. 2011. Perfluoroalkyl and polyfluoroalkyl substances in the environment: Terminology, classification, and origins. *Integrated Environmental Management and Assessment* 7(4):513–541.

Butenhoff, J., G. Costa, C. Elcombe, D. Farrar, K. Hansen, H. Iwai, R. Jung, G. Kennedy Jr., P. Lieder, G. Olsen, and P. Thomford. 2002. Toxicity of ammonium perfluorooctanoate in male cynomolgus monkeys after oral dosing for 6 months. *Toxicological Science* 69:244–257.

Butenhoff, J.L., G.L. Kennedy, Jr., S.R. Frame, J.C. O'Connor, and R.G. York. 2004a. The reproductive toxicology of ammonium perfluorooctanoate (APFO) in the rat. *Toxicology* 196:95–116.

Butenhoff, J.L., G.L. Kennedy, P.M. Hinderliter, P.H.Lieder, R. Jung, K.J. Hansen, G.S. Gorman, P.E. Noker, and P.J. Thomford. 2004b. Pharmacokinetics of perfluorooctanoate in Cynomolgus monkeys. *Toxicological Science* 82:394–406.

Butenhoff, J.L., G.L. Kennedy, Jr., S.C. Chang, and G.W. Olsen. 2012. Chronic dietary toxicity and carcinogenicity study with ammonium perfluorooctanoate in Sprague-Dawley rats. *Toxicology* 298:1–13.

Calafat, A.M., L.Y. Wong, Z. Kuklenyik, J.A. Reidy, and L.L. Needham. 2007a. 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. *Environmental Health Perspectives* (115):1596–1602.

Calafat, A.M., Z. Kuklenyik, J.A. Reidy, S.P. Caudill, J.S. Tully, and L.L. Needham. 2007b. Serum concentrations of 11 polyfluoroalkyl compounds in the US population: data from the National Health and Nutrition Examination Survey (NHANES) 1999-2000. *Environmental Science & Technology* 41(7):2237–2242.

Cariou, R., B. Veyrand, A. Yamada, A, D. Zalko, S. Durand, C. Pollono, P. Marchand, J.C. Leblanc, J.P. Antignac, and B. Le Bizec. 2015. Perfluoroalkyl acid (PFAA) levels and profiles in breast milk, maternal and cord serum of French women and their newborns. *Environment International* 84:71–81.

Carter, K.E., and J. Farrell. 2010. Removal of perfluorooctane and perfluorobutane sulfonate from water via carbon adsorption and ion exchange. *Separation Science and Technology* 45(6):762–767.

ED_005024_00000285-00071

CDC (Centers for Disease Control and Prevention). 2009. *Fourth National Report on Human Exposure to Environmental Chemicals*. Department of Health and Human Services, Centers for Disease Control and Prevention. Accessed May 2016. http://www.cdc.gov/exposurereport/.

CDC (Centers for Disease Control and Prevention). 2015. *Fourth National Report on Human Exposure to Environmental Chemicals, Updated Tables, February*. Department of Health and Human Services, Centers for Disease Control and Prevention. Accessed May 2016. http://www.cdc.gov/exposurereport/.

Chang, E.T., H.O. Adami, P. Boffetta, H.J. Wedner, and J.S. Mandel. 2016. A critical review of perfluorooctanoate and perfluorooctanesulfonate exposure and immunological health conditions in humans. *Critical Reviews in Toxicology* 46(4):279–331.

Cheng, X., J. Maher, H. Lu, and C.D. Klaassen. 2006. Endrocrine regulation of gender-divergent mouse organic anion-transporting polypeptide (Oatp) expression. *Molecular Pharmacology* 70:1291–1297.

Cheng, X., and C.D. Klaassen. 2008. Critical role of PPAR-α in perfluorooctanoic acid–and perfluorodecanoic acid–induced downregulation of Oatp uptake transporters in mouse livers. *Toxicological Sciences* 106(1):37–45.

Cheng, X., and C.D. Klaassen. 2009. Tissue distribution, ontogeny, and hormonal regulation of xenobiotic transporters in mouse kidneys. *Drug Metabolism and Disposition* 37:2178–2185.

Christensen, K.Y., M. Maisonet, C. Rubin, A. Holmes, A.M. Calafat, K. Kato, W.D. Flanders, J. Heron, M.A. McGeehin, and M. Marcus. 2011. Exposure to polyfluoroalkyl chemicals during pregnancy is not associated with offspring age at menarche in a contemporary British cohort. *Environment International* 37(1):129–135.

Chularueangaksorn, P., S. Tanaka, S. Fujii, and C. Kunacheva. 2013. Regeneration and reusability of anion exchange resin used in perfluorooctane sulfonate removal by batch experiments. *Journal of Applied Polymer Science* 130(2):884–890.

Clegg, L.X., E.J. Feuer, D.N. Midthune, M.P. Fay, and B.F. Hankey. 2002. Impact of reporting delay and reporting error on cancer incidence rates and trends. *Journal of the National Cancer Institute* 94(20):1537–1545.

Cook, J.C., S.M. Murray, S.R. Frame, and M.E. Hurtt. 1992. Induction of Leydig cell adenomas by ammonium perfluorooctanoate: A possible endocrine-related mechanism. *Toxicology and Applied Pharmacology* 113: 209–217.

Costa G., S. Sartori, and D. Consonni. 2009. Thirty years of medical surveillance in perfluorooctanoic acid production workers. *Journal of Occupational and Environmental Medicine* 51:364–372.

ED_005024_00000285-00072

Cui, L., Q. Zhou, C. Liao, J. Fu, and G. Jiang. 2009. Studies on the toxicological effects of PFOA and PFOS on rats using histological observation and chemical analysis. *Archives of Environmental Contamination and Toxicology* 56:338–349.

D'eon, J.C., and S. A. Mabury. 2011. Is indirect exposure a significant contributor to the burden of perfluorinated acids observed in humans? *Environmental Science & Technology* 45(19):7974–7984.

D'Hollander, W., L. Roosens, A. Covaci, C. Cornelis, H. Reynders, K. Van Campenhout, P. de Voogt, and L. Vervoets. 2010. Brominated flame retardants and perfluorinated compounds in indoor dust from homes and offices in Flanders, Belgium. *Chemosphere* 81:478–487.

Danish Ministry of the Environment. 2015. *Perfluoroalkylated substances: PFOA, PFOS and PFOSA: Evaluation of Health Hazards and Proposal of a Health Based Quality Criterion for Drinking Water, Soil and Ground Water*. Environmental project No. 1665, authors: P.B. Larsen and E. Giovalle. Copenhagen, Denmark: The Danish Environmental Protection Agency. Accessed May 2016. http://www2.mst.dk/Udgiv/publications/2015/04/978-87-93283-01-5.pdf.

Darrow, L.A., C.R. Stein, and K. Steenland. 2013. Serum perfluorooctanoic acid and perfluorooctane sulfonate concentrations in relation to birth outcomes in the Mid-Ohio Valley, 2005-2010. *Environmental Health Perspectives* 121(10):1207–1213.

de Cock, M., M.R. de Boer, M. Lamoree, J. Legler, and M. van de Bor. 2014. Prenatal exposure to endocrine disrupting chemicals in relation to thyroid hormone levels in infants – a Dutch prospective cohort study. *Environmental Health* 13(1):1–10.

Del Vento, S., C. Halsall, R. Gioia, K. Jones, and J. Dachs. 2012. Volatile per- and polyfluoroalkyl compounds in the remote atmosphere of the western Antarctic Peninsula: an indirect source of perfluoroalkyl acids to Antarctic waters? *Atmospheric Pollution Research* 3(4):450–455.

Denys, S., S. Fraize-Frontier, O. Moussa, B. le Bizec, B. Veyrand, and J.-L. Volatier. 2014. Is the fresh water fish consumption a significant determinant of the internal exposure to perfluoroalkylated substances (PFAS)? *Toxicology Letters* 231:233–238.

DeWitt, J.C., C.B. Copeland, M.J. Strynar, and R.W. Luebke. 2008. Perfluorooctanoic acid-induced immunomodulation in adult C57BL/6J or C57BL/6N female mice. *Environmental Health Perspectives* 116:644–650.

DeWitt, J.C., W.C. Williams, J. Creech, and R.W. Luebke. 2015. Suppression of antigen-specific antibody responses in mice exposed to perfluorooctanoic acid: Role of PPARα and T- and B-cell targeting. *Journal of Immunotoxicology* 13(1):38–45.

DNREC (Delaware Department of Resources and Environmental Control). 2016. *Reporting Level Table*. Accessed May 2016. http://www.dnrec.delaware.gov/dwhs/sirb/Documents/Notification%20Guidance.pdf.

ED_005024_00000285-00073

Dudley, L., E.C. Arevalo, and D.R.U. Knappe. 2015. *Removal of Perfluoroalkyl Substances by PAC Adsorption and Anion Exchange*. Web Report #4344, Water Research Foundation.

EFSA (European Food Safety Authority). 2008. Opinion of the scientific panel on contaminants in the food chain on perfluorooctane sulfonate (PFOS) and perfluorooctanoic acid (PFOA) and their salts. *EFSA Journal* 653:1–131.

Egeghy, P., and M. Lorber. 2011. An assessment of the exposure of Americans to perfluorooctane sulfonate: A comparison of estimated intake with values inferred from NHANES data. *Journal of Exposure Science and Environmental Epidemiology* 21:150–168.

Elcombe, C.R., B.M. Elcombe, J.R. Foster, D.G. Farrar, R. Jung, S.C. Chang, G.L. Kennedy, and J.L. Butenhoff. 2010. Hepatocellular hypertrophy and cell proliferation in Sprague-Dawley rats following dietary exposure to ammonium perfluorooctanoate occurs through increased activation of the xenosensor nuclear receptors PPARα and CAR/PXR. *Archives of Toxicology* 84(10):787–798.

Emmett, E.A., H. Zhang, F.S. Shofer, D. Freeman, N.V. Rodway, C. Desai, and L.M. Shaw. 2006. Community Exposure to Perfluorooctanoate: Relationships between Serum Concentrations and Certain Health Parameters. *Journal of Occupational Medicine* 48:771–779.

Environment Canada and Health Canada. 2012. *Screening Assessment Report, Perfluorooctanoic Acid, its Salts, and its Precursors*. Accessed May 2016. https://www.ec.gc.ca/ese-ees/370AB133-3972-454F-A03A-F18890B58277/PFOA_EN.pdf.

Eriksen, K.T., M. Sørensen, J.K. McLaughlin, L. Lipworth, A. Tjønneland, K. Overvad, and O. Raaschou-Nielsen. 2009. Perfluorooctanoate and perfluorooctanesulfonate plasma levels and risk of cancer in the general Danish population. *Journal of the National Cancer Institute* 101:605–609.

Eriksen, K.T., O. Raaschou-Nielsen, J.K. McLaughlin, L. Lipworth, A. Tjønneland, K. Overvad, and M. Sørensen. 2013. Association between plasma PFOA and PFOS levels and total cholesterol in a middle-aged Danish population. *PLoS ONE* 8:e56969.

EWG (Environmental Working Group). 2015. *National Drinking Water Database*. Accessed May 2016. http://www.ewg.org/tap-water/chemical-contI6:J7aminants/Perfluorooctanoic-Acid-PFOA/E207/.

Fasano, W.J., G.L. Kennedy, B. Szostek, D.G. Farrar, R.J. Ward, L. Haroun, and P.M. Hinderliter. 2005. Penetration of ammonium perfluorooctanoate through rat and human skin in vitro. *Drug and Chemical Toxicology* 28(1):79–90.

Fei, C., J.K. McLaughlin, R.E. Tarone, and J. Olsen. 2007. Perfluorinated chemicals and fetal growth: a study within the Danish National Birth Cohort. *Environmental Health Perspectives* 115:1677–1682.

ED_005024_00000285-00074

Fei, C., J.K. McLaughlin, L. Lipworth, and J. Olsen. 2009. Maternal levels of perfluorinated chemicals and subfecundity. *Human Reproduction* 24:1200–1205.

Fenton, S.E., J.L. Reiner, S.F. Nakayama, A.D. Delinsky, J.P. Stanko, E.P. Hines, S.S. White, A.B. Lindstrom, M.J. Strynar, and S.-S.E. Petropoulou. 2009. Analysis of PFOA in dosed CD-1 mice. Part 2: Disposition of PFOA in tissues and fluids from pregnant and lactating mice and their pups. *Reproductive Toxicology* 27(3):365–372.

Filipovic, M., A. Woldegiorgis, K. Norstrom, M. Bibi, M. Lindberg, and A.H. Osteras. 2015. Historical usage of aqueous film forming foam: A case study of the widespread distribution of perfluoroalkyl acids from a military airport to groundwater, lakes, soils and fish. *Chemosphere* 129:39–45.

Fisher, M., T.E. Arbuckle, M. Wade, and D.A. Haines. 2013. Do perfluoroalkyl substances affect metabolic function and plasma lipids?—analysis of the 2007–2009, Canadian Health Measures Survey (CHMS) Cycle 1. *Environmental Research* 121:95–103.

Fitz-Simon, N., T. Fletcher, M.I. Luster, K. Steenland, A.M. Calafat, K. Kato, and B. Armstrong. 2013. Reductions in serum lipids with a 4-year decline in serum perfluorooctanoic acid and perfluorooctanesulfonic acid. *Epidemiology* 24(4):569–576.

Fraser, A.J., T.F. Webster, D.J. Watkins, M.J. Strynar, K. Katod, A.M. Calafat, V.M. Vieira, and M.D. McClean. 2013. Polyfluorinated compounds in dust from homes, offices, and vehicles as predictors of concentrations in office workers' serum. *Environment International* 60:128–136.

Frisbee, S.J., A. Shankar, S.S. Knox, K. Steenland, D.A. Savitz, T. Fletcher, and A. Ducatman. 2010. Perfluorooctanoic acid, perfluorooctanesulfonate, and serum lipids in children and adolescents: results from the C8 health project. *Archives of Pediatrics and Adolescent Medicine* 164:860–869.

Fromme, H., O. Midasch, D. Twardella, J. Angerer, S. Boehmer, and B. Liebl. 2007. Occurrence of perfluorinated substances in an adult German population in southern Bavaria. *International Archives of Occupational and Environmental Health* 80(4):313–319.

Fromme, H., S.A. Tittlemier, W. Völkel, M. Wilhelm, and D. Twardella. 2009. Perfluorinated compounds–exposure assessment for the general population in Western countries. *International Journal of Hygiene and Environmental Health* 212(3):239–270.

Gallo, V., G. Leonardi, B. Genser, M.J. Lopez-Espinosa, S.J. Frisbee. L. Karlsson, A.M. Ducatman, and T. Fletcher. 2012. Serum perfluorooctanoate (PFOA) and perfluorooctane sulfonate (PFOS) concentrations and liver function biomarkers in a population with elevated PFOA exposure. *Environmental Health Perspectives* 120:655–660.

Gebbink, W.A., U. Berger, and I.T. Cousins. 2015. Estimating human exposure to PFOS isomers and PFCA homologues: The relative importance of direct and indirect (precursor) exposure. *Environment International* 74:160–169.

ED_005024_00000285-00075

Geiger, S.D., J. Xiao, A. Ducatman, S. Frisbee, K. Innes, and A. Shankar. 2014. The association between PFOA, PFOS and serum lipid levels in adolescents. *Chemosphere* 98:78–83.

Genuis, S.J., D. Birkholz, M. Ralitsch, and N. Thibault. 2010. Human detoxification of perfluorinated compounds. *Public Health* 124:367–375.

German Ministry of Health. 2006. *Assessment of PFOA in the Drinking Water of the German Hochsauerlandkreis. Provisional Evaluation of PFT in Drinking Water with the Guide Substances Perfluorooctanoic acid (PFOA) and Perfluorooctane Sulfonate (PFOS) as Examples*. Accessed May 2016. http://www.umweltbundesamt.de/sites/default/files/medien/pdfs/pft-in-drinking-water.pdf.

Gobas, F.A.P.C., W. de Wolf, L.P. Burkhard, E. Verbruggen, and K. Plotzke. 2009. Revisiting bioaccumulation criteria for POPs and PBT assessments. *Integrated Environmental Assessment and Management* 5(4):624–637.

Goeden, H., and J. Kelly. 2006. Targeted Sampling 2004-2005. Perfluorochemicals in Minnesota, MN, Department of Health.

Goosey, E., and S. Harrad. 2012. Perfluoroalkyl substances in UK indoor and outdoor air: Spatial and seasonal variation, and implications for human exposure. *Environment International* 45:86–90.

Grandjean, P., E.W. Andersen, E. Budtz-Jørgensen, F. Nielsen, K. Mølbak, P. Weihe, and C. Heilmann. 2012. Serum vaccine antibody concentrations in children exposed to perfluorinated compounds. *Journal of the American Medical Association* 307:391–397.

Granum, B., L.S. Haug, E. Namork, S.B. Stølevik, C. Thomsen, I.S. Aaberge, H. van Loveren, M. Løvik, and U.C. Nygaard. 2013. Pre-natal exposure to perfluoroalkyl substances may be associated with altered vaccine antibody levels and immune-related health outcomes in early childhood. *Journal of Immunotoxicology* 10(4):373–379.

Hall, A.P., C.R. Elcombe, J.R. Foster, T. Harada, W. Kaufmann, A. Knippel, K. Küttler, D.E. Malarkey, R.R. Maronpot, A. Nishikawa, T. Nolte, A. Schulte, V. Strauss, and M.J. York. 2012. Liver hypertrophy: a review of adaptive (adverse and non-adverse) changes – conclusions from the 3rd International ESTP Expert Workshop. *Toxicologic Pathology* 40:971–994.

Hansen, K.J., H.O. Johnson, J.S. Elridge, J.L. Butenhoff, and L.A. Dick. 2002. Quantitative characterization of trace levels of PFOS and PFOA in the Tennessee River. *Environmental Science & Technology* 36(8):1681–1685.

Hansen, M., M. Borreson, M. Schlabach, and G.Cornelissen. 2010. Sorption of perfluorinated compounds from contaminated water to activated carbon. *Journal of Soils and Sediments* 10:179–185.

ED_005024_00000285-00076

Hardell, E., A. Kärrman, B. van Bavel, J. Bao, M. Carlberg, and L. Hardell. 2014. Case–control study on perfluorinated alkyl acids (PFAAs) and the risk of prostate cancer. *Environment International* 63:35–39.

Hardisty, J.F., G.A. Willson, W.R. Brown, E.E. McConnell, S.R. Frame, D.W. Gaylor, G.L. Kennedy, and J.L. Butenhoff. 2010. Pathology Working Group review and evaluation of proliferative lesions of mammary gland tissues in female rats fed ammonium perfluorooctanoate (APFO) in the diet for 2 years. *Drug and Chemical Toxicology* 33(2):131–137.

Haug, L.S., S. Salihovic, I.E. Jogsten, C. Thomsen, B. van Bavel, G. Lindstrom, and G. Becher. 2010. Levels in food and beverages and daily intake of perfluorinated compounds in Norway. *Chemosphere* 80:1137–1143.

Haug, L.S., S. Huber, G. Becher, and C. Thomsen. 2011. Characterization of human exposure pathways to perfluorinated compounds—comparing exposure estimates with biomarkers of exposure. *Environment International* 37:687–693.

Hekster, F.M., R.W. Laane, and P. de Voogt. 2003. Environmental and toxicity effects of perfluoroalkylated substances. *Reviews of Environmental Contamination and Toxicology* 179:99–121.

Higgins C., and R. Luthy. 2006. Sorption of Perfluorinated Surfactants on Sediments. *Environmental Science & Technology* 40(23):7251–7256.

Hill, A.B. 1965. The environment and disease: Association or causation? In *Proceedings of the Royal Society of Medicine* 58(5):295–300.

Hinderliter, P.M. 2004. Ammonium perfluorooctanoate: Age effect on the PFOA plasma concentration in post-weaning rats following oral gavage. E.I. du Pont de Nemours and Company. Laboratory Project ID: Dupont-15302. December 2, 2004.

Hinderliter, P.M., E. Mylchreest, S.A. Gannon, J.L. Butenhoff, and G.L. Kennedy, Jr. 2005. Perfluroroocctanoate: Placental and lactational transport pharmacokinetics in rats. *Toxicology* 211:139–148.

Hinderliter, P.M., X. Han, G.L. Kennedy, Jr., and J.L. Butenhoff. 2006. Age effect on perfluorooctanoate (PFOA) plasma concentration in post-weaning rats following oral gavage with ammonium perfluorooctanoate (APFO). *Toxicology* 225:195–203.

Hines, E.P., S.S. White, J.P. Stanko, E.A. Gibbs-Flournoy, C. Lau, and S.E. Fenton. 2009. Phenotypic dichotomy following developmental exposure to perfluorooctanoic acid (PFOA) in female CD-1 mice: Low doses induce elevated serum leptin and insulin, and overweight in mid-life. *Molecular and Cellular Endocrinology* 304:97–105.

Hlouskova, V., P. Hradkova, J. Poustka, B. Gianfranco, S.P. De Filipps, W. D'Hollander, L. Bervoets, D. Herzke, S. Huber, P. de Voogt, and J. Pulkrabova. 2013. Occurrence of perfluoroalkyl substances (PFAS) in various food items of animal origin collected in four European countries. *Food Additives and Contaminants: Part A* 30(11):1918–1932.

ED_005024_00000285-00077

Hori, H., E. Hayakawa, H. Einaga, S. Kutsuna, K. Koike, T. Ibusuki, H. Kiatagawa, and R. Arakawa. 2004. Decomposition of environmentally persistent perfluorooctanoic acid in water by photochemical approaches. *Environmental Science & Technology* 38:22:6118.

Houde, M., T.A. Bujas, J. Small, R.S. Wells, P.A. Fair, G.D. Bossart, K.R. Solomon, and D.C. Muir. 2006. Biomagnification of perfluoroalkyl compounds in the bottlenose dolphin (*Tursiops truncatus*) food web. *Environmental Science & Technology* 40(13):4138–4144.

HSDB (Hazardous Substances Data Bank). 2012. *Perfluorooctanoic acid*. Accessed May 2016. http://toxnet.nlm.nih.gov/cgi-bin/sis/search2/r?dbs+hsdb:@term+@DOCNO+7137.

Hundley, S.G., A.M. Sarrif, and G.L. Kennedy, Jr. 2006. Absorption, distribution, and excretion of ammonium perfluorooctanoate (APFO) after oral administration to various species. *Drug and Chemical Toxicology* 29(2):137–145.

Innes, K.E., J.H. Wimsatt, S. Frisbee, and A.M. Ducatman. 2014. Inverse association of colorectal cancer prevalence to serum levels of perfluorooctane sulfonate (PFOS) and perfluorooctanoate (PFOA) in a large Appalachian population. *BMC Cancer* 14:45.

Jain, R.B. 2014. Contribution of diet and other factors to the levels of selected polyfluorinated compounds: Data from NHANES 2003-2008. *International Journal of Hygiene and Environmental Health* 217:52–61.

Joensen, U.N., R. Bossi, H. Leffers, A.A. Jensen, N.E. Skakkbæk, and N. Jørgensen. 2009. Do perfluoroalkyl compounds impair human semen quality? *Environmental Health Perspectives* 117: 923–927.

Joensen, U.N., B. Veyrand, J.-P. Antignac, M.B. Jensen, J.H. Petersen, P. Marchand, N.E. Skakkebæk, A.-M. Andersson, B. Le Bizec, and N. Jørgensen. 2013. PFOS (perfluorooctanesulfonate) in serum is negatively associated with testosterone levels, but not with semen quality, in healthy men. *Human Reproduction* 28:599–608.

Jogsten, I.E., M. Nadal, B. van Bavel, G. Lindström, and J.L. Domingo. 2012. Per-and polyfluorinated compounds (PFCs) in house dust and indoor air in Catalonia, Spain: implications for human exposure. *Environment International* 39(1):172–180.

Johansson, N., P. Eriksson, and H. Viberg. 2009. Neonatal exposure to PFOS and PFOA in mice results in changes in proteins which are important for neuronal growth and synaptogenesis in the developing brain. *Toxicological Science* 108: 412–418.

Johansson, J.H., U. Berger, R. Vestergren, I.T. Cousins, A. Bignert, A. Glynn, and P.O. Darnerud. 2014. Temporal trends (1999–2010) of perfluoroalkyl acids in commonly consumed food items. *Environmental Pollution* 188:102–108.

Johnson, P.I., P. Sutton, D.S. Atchley, E. Koustas, J. Lam, S. Sen, K.A. Robinson, D.A. Axelrad, and T.J. Woodruff. 2014. The Navigation Guide – evidence-based medicine meets environmental health: systematic review of human evidence for PFOA effects on fetal growth. *Environmental Health Perspectives* 122:1028–1039.

ED_005024_00000285-00078

Kaiser, M.A., B.S. Larsen, C-P.C. Kao, and R.C. Buck. 2005. Vapor pressures of perfluoro-octanoic, -nonanoic, -decanoic, undecanoic, and dodecanoic acids. *Journal of Chemical and Engineering Data* 50(6):1841–1843.

Kauck, E.A., and A.R. Diesslin. 1951. Some Properties of Perfluorocarboxylic Acids. *Industrial and Engineering Chemical Research* 43(10):2332–2334.

Kelly, B.C., M.G. Ikonomou, J.D. Blair, B. Surridge, D. Hoover, R. Grace, and F.A.P.C. Gobas. 2009. Perfluoroalkyl contaminants in an Arctic Marine Food Web: Trophic Magnification and Wildlife Exposure. *Environmental Science & Technology* 43:4037–4043.

Kemper, R.A. 2003. Perfluorooctanoic acid: Toxicokinetics in the rat. Laboratory Project ID: Dupont-7473. Haskell Laboratory for Health and Environmental Sciences, E.I. du Pont de Nemours and Company. April 2, 2003. U.S. Environmental Protection Agency Administrative Record 226-1499.

Kennedy, G.L. 1987. Increase in mouse liver weight following feeding of ammonium perfluorooctanoate and related fluorochemicals. *Toxicology Letters* 39(2):295–300.

Kerstner-Wood, C., L. Coward, and G. Gorman. 2003. Protein binding of perfluorohexane sulfonate, perfluorooctane sulfonate and perfluorooctanoate to plasma (human, rat, and monkey), and various human-derived plasma protein fractions. Southern Research Institute. Study ID 9921.7. U.S. Environmental Protection Agency Administrative Record 226-1354.

Kirk-Othmer. 1994. Encyclopedia of Chemical Technology. 4th ed. Volumes 1: New York, NY. John Wiley and Sons, 1991-Present. V11:551.

Klaassen, C.D., and L.M. Aleksunes. 2010. Xenobiotic, bile acid, and cholesterol transporters: Function and regulation. *Pharmacology Review* 62:1–96.

Klaunig, J.E., M.A. Babich, L.P. Baetcke, J.C. Cook, J.C. Corton, R.M. David, J.G. DeLuca, D.Y. Lai, R.H. McKee, J.M. Peters, R.A. Roberts, and P.A. Fenner-Crisp. 2003. PPARα agonist-induced rodent tumors: modes of action and human relevance. *Critical Reviews in Toxicology* 33: 655–780.

Klaunig, J.E., B.A. Hocevar, and L.M. Kamendulis. 2012. Mode of action analysis of perfluorooctanoic acid (PFOA) tumorigenicity and human relevance. *Reproductive Toxicology* 33:410–418.

Knobeloch, L., P. Imm, and H. Anderson. 2012. Perfluoroalkyl chemicals in vacuum cleaner dust from 39 Wisconsin homes. *Chemosphere* 88:779–783.

Konwick, B.J., G.T. Tomy, N. Ismail, J.T. Peterson, R.J. Fauver, D. Higginbotham, and A.T. Fisk. 2008. Concentrations and patterns of perfluoroalkyl acids in Georgia, USA source waters near and distant to a major use source. *Environmental Toxicology & Chemistry* 27(10):2011–2018.

ED_005024_00000285-00079

Koustas, E., J. Lam, P. Sutton, P.I. Johnson, D.S. Atchley, S. Sen, K.A. Robinson, D.A. Axelrad, and T.J. Woodruff. 2014. The Navigation Guide – evidence-based medicine meets environmental health: systematic review of nonhuman evidence for PFOA effects on fetal growth. *Environmental Health Perspectives* 122:1015–1027.

Krippner, J., H. Brunn, S. Falk, S. Georgii, S. Schubert, and T. Stahl. 2014. Effects of chain length and pH on the uptake and distribution of perfluoroalkyl substances in maize (*Zea mays*). *Chemosphere* 94:85–90.

Kristensen, S.L., C.H. Ramlau-Hansen, E. Ernst, S.F. Olsen, J.P. Bonde, A. Vested, T.I. Halldorsson, G. Becher, L.S. Haug, and G. Toft. 2013. Long-term effects of prenatal exposure to perfluoroalkyl substances on female reproduction. *Human Reproduction* 0:1–12.

Kudo, N., M. Katakura, Y. Sato, and Y. Kawashima. 2002. Sex hormone-regulated renal transport of perfluorooctanoic acid. *Chemico-Biological Interactions* 139:301–316.

Kwon, B.G., H.J. Lim, S.H. Na, B.I. Choi, D.S. Shin, and S.Y. Chung. 2014. Biodegradation of perfluorooctanesulfonate (PFOS) as an emerging contaminant. *Chemosphere* 109: 221–225.

Lange F.T., C. Schmidt, and H.J. Brauch. 2006. Perfluoroalkyl Carboxylates and Sulfonates, Rhine Water Works, The Netherlands, Association of River Waterworks – RIWA. Accessed May 2016. http://www.riwa-rijn.org/wp-content/uploads/2015/05/137_ptfe_report.pdf.

Langer, V., A. Dreyer, and R. Ebinghaus. 2010. Polyfluorinated compounds in residential and nonresidential indoor air. *Environmental Science & Technology* 44(21):8075–8081.

Lau, C., J.R. Thibodeaux, R.G. Hanson, J.M. Rogers, B.E. Grey, M.E. Stanton, J.L. Butenhoff, and L.A. Stevenson. 2003. Exposure to perfluorooctane sulfonate during pregnancy in rat and mouse. II: postnatal evaluation. *Toxicological Science* 74:382–392.

Lau, C., J.R. Thibodeaux, R.G. Hanson, M.G. Narotsky, J.M. Rogers, A.B. Lindstrom, and M.J. Strynar. 2006. Effects of perfluorooctanoic acid exposure during pregnancy in the mouse. *Toxicological Science* 90:510–518.

Lewis, R.J., Sr., ed. 2004. *Sax's Dangerous Properties of Industrial Materials*. 11th ed. Wiley-Interscience, Wiley & Sons, Inc., Hoboken, N.J V3:2860.

Li, X., S. Chen, X. Quan, and Y. Zhang. 2011. Enhanced adsorption of PFOA and PFOS on multiwalled carbon nanotubes under electrochemical assistance. *Environmental Science & Technology* 45(19):8498–8505.

Liao, C., T. Wang, L. Cui, Q. Zhou, S. Duan, and G. Jiang. 2009a. Changes in synaptic transmission, calcium current, and neurite growth by perfluorinated compounds are dependent on the chain length and functional group. *Environmental Science & Technology* 43:2099–2104.

ED_005024_00000285-00080

Liao, C., T. Wang, L. Cui, Q. Zhou, S. Duan, and G. Jiang. 2009b. Supporting Information: Changes in synaptic transmission, calcium current, and neurite growth by perfluorinated compounds are dependent on the chain length and functional group. *Environmental Science & Technology.*

Lide, D.R. 2007. *CRC Handbook of Chemistry and Physics.* 88th ed. CRC Press, Taylor & Francis, Boca Raton, FL. 3–412.

Lin, C-Y., Y-C Lin, P-C Chen, and L-Y Lin. 2009. Association among serum perfluoroalkyl chemicals, glucose homeostasis, and metabolic syndrome in adolescents and adults. *Diabetes Care* 32:702–707.

Lindstrom, A.B., M.J. Strynar, and E.L. Libelo. 2011a. Polyfluorinated compounds: past, present, and future. *Environmental Science & Technology* 45:7954–7961.

Lindstrom, A.B., M.J. Strynar, A.D. Delinsky, S.F. Nakayama, L. McMillan, E.L. Libelo, M. Neill, and L. Thomas. 2011b. Application of WWTP Biosolids and Resulting Perfluorinated Compound Contamination of Surface and Well Water in Decatur, Alabama, USA. *Environmental Science & Technology* 45:8015–8021.

Liu, C.S., K. Shih, and F. Wang. 2012. Oxidative decomposition of perfluorooctane sulfonate in water by permanganate. *Separation and Purification Technology* 87:95–100.

Liu, B., H. Zhang, D.Yao, J. Li, L. Xie, X. Wang, Y. Wang, G. Liu, and B. Yang. 2015. Perfluorinated compounds (PFCs) in the atmosphere of Shenzhen, China: Spatial distribution, sources and health risk assessment. *Chemosphere* 138:511–518.

Livsmidelsverket. 2014. Perfluorerade alkylsyror i drickvatten. 2014-02-21. Komplettering, 2014-01-08; Riskhanteringsrapport, 24-03-12, cited in Danish Ministry of the Environment. 2015. *Perfluoroalkylated substances: PFOA, PFOS and PFOSA: Evaluation of health hazards and proposal of a health based quality criterion for drinking water, soil and ground water*. Environmental project No. 1665, authors: P.B. Larsen and E. Giovalle. Copenhagen, Denmark: The Danish Environmental Protection Agency. Accessed May 2016. http://www2.mst.dk/Udgiv/publications/2015/04/978-87-93283-01-5.pdf.

Loi, E.I., L.W. Yeung, S. Taniyasu, P.K.S. Lam, K. Kannan, and N. Yamashita. 2011. Trophic magnification of poly- and perfluorinated compounds in a subtropical food web. *Environmental Science & Technology* 45(13):5506–5513.

Looker, C., M.I. Luster, A.M. Calafat, V.J. Johnson, G.R. Burleson, F.G. Burleson, and T. Fletcher. 2014. Influenza vaccine response in adults exposed to perfluorooctanoate and perfluorooctanesulfonate. *Toxicological Science* 138:76–88.

Loos, R., J. Woollgast, T. Huber, and G. Hanke. 2007. Polar herbicides, pharmaceutical products, perfluorooctanesulfonate (PFOS), perfluorooctanoate (PFOA), and nonylphenol and its carboxylates and ethoxylates in surface and tap waters around Lake Maggiore in Northern Italy. *Analytical and Bioanalytical Chemistry* 387:1469.

ED_005024_00000285-00081

Lopez-Espinosa, M.-J., T. Fletcher, B. Armstron, B. Genser, K. Dhatariya, D. Mondal, A. Ducatman, and G. Leonardi. 2011. Association of perfluorooctanoic acid (PFOA) and perfluorooctane sulfonate (PFOS) with age of puberty among children living near a chemical plant. *Environmental Science & Technology.*

Lopez-Espinosa, M.-J., D. Mondal, B. Armstrong, M.S. Bloom, and T. Fletcher. 2012. Thyroid function and perfluoroalkyl acids in children living near a chemical plant. *Environmental Health Perspectives* 120:1036–1041.

Lorber, M., and P.P. Egeghy. 2011. Simple intake and pharmacokinetic modeling to characterize exposure of Americans to perfluoroctanoic acid, PFOA. *Environmental Science & Technology* 45:8006–8014.

Loveless, S.E., D. Hoban, G. Sykes, S.R. Frame, and N.E. Everds. 2008. Evaluation of the immune system in rats and mice administered linear ammonium perfluorooctanoate. *Toxicological Science* 105:86–96.

Lu, Y., B. Luo, J. Li, and J. Dai. 2015. Perfluorooctanoic acid disrupts the blood-testes barrier and activates TNFα/p38 MAPK signaling pathway in vivo and in vitro. *Archives of Toxicology* 90(4):971–983.

Luebker, D.J., K.J. Hansen, N.M. Bass, J.L. Butenhoff, and A.M. Seacat. 2002. Interactions of fluorochemicals with rat liver fatty acid-binding protein. *Toxicology* 176: 175–185.

MacNeil, J., N.K. Steenland, A. Shankar, and A. Ducatman. 2009. A cross-sectional analysis of type II diabetes in a community with exposure to perfluorooctanoic acid (PFOA). *Environmental Research* 109: 997–1003.

Macon, M.B., L.R. Villanueva, K. Tatum-gibbs, R.D. Zehr, M.J. Strynar, J.P. Stanko, S.S. White, L. Helfant, and S.E. Fenton. 2011. Prenatal perfluorooctanoic acid exposure in CD-1 mice: low dose developmental effects and internal dosimetry. *Toxicological Science* 122(1):135–145.

MacManus-Spencer, L.A., M.L. Tse, P.C. Hebert, H.N. Bischel, and R.G. Luthy. 2010. Binding of perfluorocarboxylates to serum albumin: a comparison of analytical methods. *Analytical Chemistry* 82(3):974–981.

Maine DHHS (Department of Health and Human Services). 2014. *Maximum Exposure Guideline for Perfluorooctanoic Acid in Drinking Water*. CAS Registry Number (Free Acid): 335-67-1. Augusta, ME: Environmental and Occupational Health Program. Accessed May 2016. https://www1.maine.gov/dhhs/mecdc/environmental-health/eohp/wells/documents/pfoameg.pdf.

Martin, J.W., S.A. Mabury, K.R. Solomon, and D.C. Muir. 2003a. Bioconcentration and tissue distribution of perfluorinated acids in rainbow trout (*Oncorhynchus mykiss).* *Environmental Toxicology and Chemistry* 22(1):196–204.

ED_005024_00000285-00082

Martin, J.W., S.A. Mabury, K.R. Solomon, and D.C. Muir. 2003b. Dietary accumulation of perfluorinated acids in juvenile rainbow trout (*Oncorhynchus mykiss*). *Environmental Toxicology and Chemistry* 22(1):189–195.

Martin, J.W., S.A. Mabury, K.R. Solomon, and D.C. Muir. 2003c. Progress toward understanding the bioaccumulation of perfluorinated alkyl acids. *Environmental Toxciology and Chemistry* 32(11):2421–2423.

Martin, J.W., M.M. Smithwick, B.M. Braune, P.F. Hoekstra, D.C.G. Muir, and S.A.Mabury. 2004. Identification of long-chain perfluorinated acids in biota from the Canadian Arctic. *Environmental Science & Technology* 38(2):373–380.

Martin, M.T., R.J. Brennan, W. Hu, E. Ayanoglu, C. Lau, H. Ren, C.R. Wood, J.C. Corton, R.J. Kavlock, and D.J. Dix. 2007. Toxicogenomic study of triazole fungicides and perfluoroalkyl acids in rat livers predicts toxicity and categorizes chemicals based on mechanisms of toxicity. *Toxicological Science* 97:595–613.

McLaughlin, C., S. Blake, T. Hall, M. Harman, R. Kanda, J. Foster, and P. Rumsby. 2011. Perfluorooctane sulphonate in raw and drinking water sources in the United Kingdom. *Water and Environment Journal* 25:1:13.

McMurdo, C.J., D.A. Ellis, E. Webster, J. Butler, R.D. Christensen, and L.K. Reid. 2008. Aerosol enrichment of the surfactant PFO and mediation of the water– air transport of gaseous PFOA. *Environmental Science & Technology* 42(11):3969–3974.

MDH (Minnesota Department of Health). 2008. *Removal of Perfluorochemicals (PFC's) with Point-of-Use (POU) Water Treatment Devices.* Accessed May 2016. http://www.health.state.mn.us/divs/eh/wells/waterquality/poudevicefinal.pdf.

MDH (Minnesota Department of Health). 2009. *Health Risk Limits for Groundwater 2008 Rule Revision*. Accessed May 2016. http://www.health.state.mn.us/divs/eh/risk/guidance/gw/pfoa.pdf.

Melzer, D., N. Rice, M.H. Depledge, W.E. Henley, and T.S. Galloway. 2010. Association between serum perfluorooctanoic acid (PFOA) and thyroid disease in the NHANES study. *Environmental Health Perspectives* 118: 686–692.

Michigan DEQ (Department of Environmental Quality). 2013. *Rule 57 Water Quality Values, Surface Water Assessment Section.* Accessed May 2016. http://www.michigan.gov/documents/deq/wrd-swas-rule57_372470_7.pdf.

Minata, M., K.H. Harada, A. Kärrman, T. Hitomi, M. Hirosawa, F.J. Gonzales, and A. Koizumi. 2010. Role of peroxisome proliferator-activated receptor-α in hepatobiliary injury induced by ammonium perfluorooctanoate in mouse liver. *Industrial Health* 48:96–107.

ED_005024_00000285-00083

Mondal, D., R.H. Weldon, B.G. Armstrong, L.J. Gibson, M-J. Lopez-Espinosa, H-M. Shin, and T. Fletcher. 2014. Breastfeeding: a potential excretion route for mothers and implications for infant exposure to perfluoroalkyl acids. *Environmental Health Perspectives* 122:187–192.

Monroy, R., K. Morrison, K. Two, S. Atkinson, C. Kubwabo, B. Steward, and W.G, Foster. 2008. Serum levels of perfluoroalkyl compounds in human maternal and umbilical cord blood samples. *Environmental Research* 108:56–62.

Moody, C.A., J.W. Martin, W.C. Kwan, D.C.G. Muir, and S.C. Mabury. 2002. Monitoring perfluorinated surfactants in biota and surface water samples following an accidental release of fire-fighting foam into Etobicoke Creek. *Environmental Science & Technology* 36(4):545–551.

Moody, C.A., G.N. Hebert, S.H. Strauss, and J.A. Field. 2003. Occurrence and persistence of perfluorooctanesulfonate and other perflourinated surfactants in groundwater at a fire-training area at Wurtsmith Air Force Base, Michigan, USA. *Journal of Environmental Monitoring* 5:341-345.

Morikawa, A., N. Kamei, K. Harada, K. Inoue, T. Yoshinaga, N. Saito, and A. Koizumi. 2005. The bioconcentration factor of perfluorooctane sulfonate is significantly larger than that of perfluorooctanoate in wild turtles (*Trachemys scripta elegans* and *Chinemys reevesii*): an Ai river ecological study in Japan. *Ecotoxicology and Environmental Safety* 65(1):14–21.

Morken, N.-H., G.S. Travlos, R.E. Wilson, M. Eggesbø, and M.P. Longnecker. 2014. Maternal glomerular filtration rate in pregnancy and fetal size. *PLOS One* 9:e101897.

Murli, H. 1996a. *Mutagenicity test on T-6342 measuring chromosomal aberrations in human whole blood lymphocytes with a confirmatory assay with multiple harvests*. Corning-Hazelton, Inc., Vienna, VA. Study No. 17073-0-449CO, November 1, 1996. U.S. Environmental Protection Agency Administrative Record 226-0433.

Murli, H. 1996b. *Mutagenicity test on T-6564 measuring chromosomal aberrations in Chinese Hamster Ovary (CHO) cells with a confirmatory assay with multiple harvests.* Corning Hazleton Inc., Vienna, VA. Study No. 17750-0-437CO, September 16, 1996. U.S. Environmental Protection Agency Administrative Record 226-0431.

Nakagawa, H., T. Hirata, T. Terada, P. Jutabha, D. Miura, K.H. Harada, K. Inoue, N. Anzai, H. Endou, K. Inui, Y. Kanai, and A. Koizumi. 2007. Roles of organic anion transporters in the renal excretion of perfluorooctanoic acid. *Basic and Clinical Pharmacology and Toxicology* 103:1–8.

Nakagawa, H., T. Terada, K.H. Harada, T. Hitomi, K. Inoue, K. Inui, and A. Koizumi. 2009. Human organic anion transporter hOAT4 is a transporter of perfluorooctanoic acid. *Basic and Clinical Pharmacology and Toxicology* 105:136–138.

ED_005024_00000285-00084

Nakamura, F., Y. Ito, Y. Yanagiba, D.H. Ramdhan, Y. Kono, H. Naito, Y. Hayashi, Y. Li, T. Aoyam, F.J. Gonzalez, and T. Nakajima. 2009. Microgram-order ammonium perfluorooctanoate may activate mouse peroxisome proliferator-activated receptor α, but not human PPARα. *Toxicology* 9: 27–33.

Nakayama, S.F., M.J. Strynar, J.L. Reiner, A.D. Delinsky, and A.B. Lindstrom. 2010. Determination of perfluorinated compounds in the Upper Mississippi River Basin. *Environmental Science & Technology* 44(11):4103–4109.

NCDEQ (North Carolina Department of Environmental Quality). 2013. *Interim Maximum Allowable Concentration for Perfluorooctanoic Acid (PFOA) in Groundwater*. April 2013. Accessed May 2016. https://ncdenr.s3.amazonaws.com/s3fs-public/documents/files/IMAC%20table_5-22-13.pdf.

Nelson, J.W., E.E. Hatch, and T.F. Webster. 2010. Exposure to polyfluoroalkyl chemicals and cholesterol, body weight, and insulin resistance in the general U.S. population. *Environmental Health Perspectives* 118:197–202.

NJDEP (New Jersey Department of Environmental Protection). 2007. *Determination of Perfluorooctanoic Acid (PFOA) in Aqueous Samples: Final Report*. January 2007. NJDEP, Division of Water Supply.

NJDEP (New Jersey Department of Environmental Protection). 2014. *Occurrence of Perfluorinated Chemicals in Untreated New Jersey Drinking Water Sources: Final Report*. April 2014, NJDEP Division of Water Supply & Geoscience. Accessed May 2016. http://www.nj.gov/dep/watersupply/pdf/pfc-study.pdf.

Noorlander, C.W., S.P. van Leeuwen, J.D. te Biesebeek, M.J. Mengelers, and M.J. Zeilmaker. 2011. Levels of perfluorinated compounds in food and dietary intake of PFOS and PFOA in the Netherlands. *Journal Of Agricultural And Food Chemistry* 59(13):7496–7505.

Obourn, J.D., S.R. Frame, R.H Bell, Jr,, D.S. Longnecker, G.S. Elliott, and J.C. Cook. 1997. Mechanisms for the pancreatic oncogenic effects of the peroxisome proliferator Wyeth-14,643. *Toxicology and Applied Pharmacology* 145: 425–436.

Okada, E., S. Sasaki, Y. Saijo, N. Washino, C. Miyashita, S. Kobayashi, K. Konishi, Y.M. Ito, R. Ito, A. Nakata, Y. Iwasaki, K. Saito, H. Nakazawa, and R. Kishi. 2012. Prenatal exposure to perfluorinated chemicals and relationship with allergies and infectious diseases in infants. *Environmental Research* 112:118–125.

Olsen, G.W., J.M. Burris, M.M. Burlew, and J.H. Mandel. 2000. Plasma cholecystokinin and hepatic enzymes, cholesterol and lipoproteins in ammonium perfluorooctanoate production workers. *Drug and Chemical Toxicology* 23:603–620.

ED_005024_00000285-00085

Olsen, G.W., M.M. Burlew, J.M. Burris, and J.H. Mandel. 2001. *A Cross-Sectional Analysis Of Serum Perfluorooctanesulfonate (PFOS) And Perfluorooctanoate (PFOA) In Relation To Clinical Chemistry, Thyroid Hormone, Hematology And Urinalysis Results From Male And Female Employee Participants Of The 2000 Antwerp And Decatur Fluorochemical Medical Surveillance Program*. 3M Company. Final Report. October 11, 2001. U.S. Environmental Protection Agency Administrative Record 226-1087.

Olsen, G.W., J.M. Burris, M.M. Burlew, and J.H. Mandel. 2003. Epidemiologic assessment of worker serum perfluorooctanesulfonate (PFOS) and perfluorooctanoate (PFOA) concentrations and medical surveillance examinations. *Journal of Occupational and Environmental Medicine* 45:260–270.

Olsen, G.W., and L.R. Zobel. 2007. Assessment of lipid, hepatic, and thyroid parameters with serum perfluorooctanoate (PFOA) concentrations in fluorochemical workers. *International Archives of Occupational and Environmental Health* 81:231–246.

Olsen, G.W., J.M. Burris, D.J. Ehresman, J.W. Froehlich, A.M. Seacat, J.L. Butenhoff, and L.R. Zobel. 2007. Half-life of serum elimination of perfluorooctanesulfonate, perfluorohexanesulfonate and perfluorooctanoate in retired fluorochemical production workers. *Environmental Health Perspective* 115:1298–1305.

Onishchenko, N., C. Fischer, W.N.W. Ibrahim, S. Negri, S. Spulbur, S. Cottica, and S. Ceccatelli. 2011. Prenatal exposure to PFOS or PFOA alters motor function in mice in a sex-related manner. *Neurotoxicity Research* 19:452–461.

Palazzolo, M.J. 1993. *Thirteen-Week Dietary Toxicity Study With T-5180, Ammonium Perfluorooctanoate (CAS No. 3825-26-1) In Male Rats*. Final Report. Laboratory Project Identification HWI 6329-100. Hazleton Wisconsin, Inc. U.S. Environmental Protection Agency Administrative Record 226-0449.

Pastoor, T.P., K.P. Lee, M.A. Perri, and P.J. Gillies. 1987. Biochemical and morphological studies of ammonium perfluorooctanoate-induced hepatomegaly and peroxisome proliferation. *Experimental and Molecular Pathology* 47(1):98–109.

Perkins, R., J. Butenhoff, G. Kennedy, and M. Palazzolo. 2004. 13-Week dietary toxicity study of ammonium perfluorooctanoate (APFO) in male rats. *Drug and Chemical Toxicology* 27: 361–378.

Plummer, S.M., D.G. Farrar, and C.R. Elcombe. 2007. Comparison of gene expression changes in whole pancreas with isolated pancreatic acinar cells of rats fed diets containing Wyeth-14,643 or ammonium perfluorooctanoate. *Toxicology* 240: 171–172.

Post, G.B., J.B. Louis, K.R. Cooper, B.J. Boros-Russo, and R.L. Lippincott. 2009. Occurrence and potential significance of perfluorooctanoic acid (pfoa) detected in New Jersey public drinking water systems. *Environmental Science & Technology* 43(12):4547–4554.

Qin, P., R. Liu, X. Pan, X. Fang, and Y. Mou. 2010. Impact of carbon chain length on binding of perfluoroalkyl acids to bovine serum albumin determined by spectroscopic methods. *Journal of Agricultural and Food Chemistry* 58(9):5561–5567.

ED_005024_00000285-00086

Quinete, N., Q. Wu, T. Zhang, S.H. Yun, I. Moreira, and K. Kannan. 2009. Specific profiles of perfluorinated compounds in surface and drinking waters and accumulation in mussels, fish, and dolphins from southeastern Brazil. *Chemosphere* 77(6):863–869.

Quiñones, O., and S.A. Snyder. 2009. Occurrence of perfluoroalkyl carboxylates and sulfonates in drinking water utilities and related waters from the United States. *Environmental Science & Technology* 43 (24):9089–9095.

Quist, E.M., A.J. Filgo, C.A. Cummings, G.E. Kissling, and M.J. Hoenerhoff. 2015. Hepatic mitochondrial alteration in CD-1 mice associated with prenatal exposures to low doses of perfluorooctanoic acid (PFOA). *Toxicologic Pathways* 41:546–557.

Raleigh, K.K., B.H. Alexander, G.W. Olsen, G. Ramachandran, S.Z. Morey, T.R. Church, P.W. Logan, L.L.F. Scott, and E.M. Allen. 2014. Mortality and cancer incidence in ammonium perfluorooctanoate production workers. *Occupational Environmental Medicine* 0:1–7.

Ren, X.-M., Y.-F. Zhang, L.-H. Guo, Z.-F. Qin, Q.-Y. Lv, and L.-Y. Zhang. 2015. Structure-activity relations in binding of perfluoroalkyl compounds to human thyroid hormone T3 receptor. *Archives of Toxicology* 89:233–242.

Renner, R. 2009. EPA finds record PFOS, PFOA levels in Alabama grazing fields. *Environmental Science & Technology* 43(3):1245–1246.

Renzi, M., C. Guerranti, A. Giovani, G. Perra, and S.E. Focardi. 2013. Perfluorinated compounds: Levels, trophic web enrichments and human dietary intakes in transitional water ecosystems. *Marine Pollution Bulletin* 76:146–157.

Rosen, M.B., J.R. Thibodeaux, C.R. Wood, R.D. Zehr, J.E. Schmid, and C. Lau. 2007. Gene expression profiling in the lung and liver of PFOA-exposed mouse fetuses. *Toxicology* 239:15–33.

Rosen, M.B., B.A. Abbott,D.C. Wolf, J.C. Corton, C.R. Wood, J.E. Schmid, K.P. Das, R.D. Zehr, E.T. Blair, and C. Lau. 2008a. Gene profiling in the livers of wild-type and PPARα-null mice exposed to perfluorooctanoic acid. *Toxicological Pathology* 36:592–607.

Rosen, M.B., J.S. Lee, H. Ren, B. Vallanat, J. Liu, M.P. Waalkes, B.D. Abbott, C. Lau, and J.C. Corton. 2008b. Toxicogenomic dissection of the perfluooctanoic acid transcript profile in mouse liver: evidence for the involvement of nuclear receptors PPARα and CAR. *Toxicological Science* 103: 46–56.

Rosen, M.B., C. Lau, and J.C. Corton. 2009a. Does exposure to perfluoroalkyl acids present a risk to human health? *Toxicological Sciences* 111(1):1–3.

Rosen, M.B., J.E. Schmid, K.P. Das, C.R. Wood, R.D. Zehr, and C. Lau. 2009b. Gene expression profiling in the liver and lung of perfluorooctane sulfonate-exposed mouse fetuses: comparison to changes induced by exposure to perfluorooctanoic acid. *Reproductive Toxicology* 27(3):278–288.

ED_005024_00000285-00087

Saez, M., P. de Voogt, and J.R. Parsons. 2008. Persistence of perfluoroalkylated substances in closed bottle tests with municipal sewage sludge. *Environmental Science and Pollution Research* 15:472–477.

Sakr, C.J., R.C. Leonard, K.H. Kreckmann, M.D. Slade, and M.R. Cullen. 2007a. Longitudinal study of serum lipids and liver enzymes in workers with occupational exposure to ammonium perfluorooctanoate. *Journal of Occupational and Environmental Medicine* 49: 872–879.

Sakr, C.J., K.H. Kreckmann, J.W. Green, P.J. Gillies, J.L. Reynolds, and R.C. Leonard. 2007b. Cross-sectional study of lipids related to a serum biomarker of exposure (ammonium perfluorooctanoate or APFO) as part of a general health survey in a cohort of occupationally exposed workers. *Journal of Occupational and Environmental Medicine* 49:1086–1096.

Salvalaglio, M., I. Muscionico, and C. Cavallotti. 2010. Determination of energies and sites of binding of PFOA and PFOS to human serum albumin. *Journal of Physical Chemistry B* 114:14860–14874.

Savitz, D.A., C.R. Stein, S.M. Bartell, B. Elston, J. Gong, H.M. Shin, and G.A. Wellenius. 2012. Perfluorooctanoic acid exposure and pregnancy outcome in a highly exposed community. *Epidemiology* 23:386–92.

Schecter, A., J. Colacino, D. Haffner, K. Patel, M. Opel, O. Päpke, and L. Birnbaum. 2010. Perfluorinated compounds, polychlorinated biphenyls, and organochlorine pesticide contamination in composite food samples from Dallas, Texas, USA. *Environmental Health Perspectives* 118(6):796.

Schlummer, M., C. Sölch, T Meisel, M. Still, L. Gruber, and G. Wolz. 2015. Emission of perfluoroalkyl carboxylic acids (PFCA) from heated surfaces made of polytetrafluoroethylene (PTFE) applied in food contact materials and consumer products. *Chemosphere* 129:46–53.

Schröder, H. F., and R.J. Meesters. 2005. Stability of fluorinated surfactants in advanced oxidation processes—a follow up of degradation products using flow injection–mass spectrometry, liquid chromatography–mass spectrometry and liquid chromatography–multiple stage mass spectrometry. *Journal of Chromatography A* 1082(1):110–119.

Seow, J. 2013. *Fire-Fighting Foams with Perfluorochemicals – Environmental Review*. Department of Environment and Conservation Western Australia. Accessed May 2016. http://www.hemmingfire.com/news/fullstory.php/aid/1748/The_final_definitive_version_of__91Fire_Fighting_Foams_with_Perfluorochemicals__96_Environmental_Review_92,_by_Dr_Jimmy_Seow,_Manager,_Pollution_Response_Unit,_Department_of_Environment_and_Conservation_Western_Australia.html.

Shivakoti, B.R., S. Fujii, M. Nozoe, S. Tanaka, and C. Kunacheva. 2010. Perfluorinated chemicals (PFCs) in water purification plants (WPPs) with advanced treatment processes. *Water Science and Technology: Water Supply* 10(1):87–95.

ED_005024_00000285-00088

Shoeib, M., T. Harner, and P. Vlahos. 2006. Perfluorinated chemicals in the Arctic atmosphere. *Environmental Science & Technology* 40:7577–7583.

Shrestha, S., M.S. Bloom, R. Yucel, R.F. Seegal, Q. Wu, K. Kannan, R. Rej, and E.F. Fitzgerald. 2015. Perfluoroalkyl substances and thyroid function in older adults. *Environmental International* 75:206–214

SIAR (SIDS Initial Assessment Profile). 2008. *Final SIDS Assessment Report: PFOA. Organization for Economic Cooperation and Development*. Paris, France. April 16-18. Accessed May 2016. http://webnet.oecd.org/HPV/UI/handler.axd?id=1f391916-96ba-46f6-a7ce-c96712da3b7e.

Skutlarek, D., M. Exner, and H. Farber. 2006. Perfluorinated surfactants in surface and drinking waters. *Environmental Science and Pollution Research International* 13(5):299.

Smithwick M., R.J. Norstrom, S.A. Mabury, K. Solomon, T.J. Evans, I. Stirling, M.K. Taylor, and D.C.G. Muir. 2006. Temporal trends of perfluoroalkyl contaminants in polar bears (*Ursus maritimus*) from two locations in the North American Arctic, 1972-2002. *Environmental Science & Technology* 40(4):1139–1143.

Son, H-Y., A-H Kim, H-I. Shin, H-I. Bae, and J-H. Yang. 2008. Perfluorooctanoic acid-induced hepatic toxicity following 21-day oral exposure in mice. *Archives of Toxicology* 82:239–246.

SRC (Syracuse Research Corporation). 2016. PHYSPROP Database. Accessed May 2016. http://www.srcinc.com/what-we-do/environmental/scientific-databases.html.

Stahl, L.L., B.D. Snyder, A.R. Olsen, T.M. Kincaid, J.B. Wathen, and H.B. McCarty. 2014. Perfluorinated compounds in fish from U.S. urban rivers and the Great Lakes. *The Science of the Total Environment* 499:185–195.

Starling, A.P., S.M. Engel, K.W. Whitworth, D.B. Richardson, A.M. Stuebe, J.L. Daniels, L.S. Haug, M. Eggesbø, G. Becher, A. Sabaredzovic, C. Thomsen, R.E. Wilson, G.S. Travlos, J.A. Hoppin, D.D. Baird, and M.P. Longnecker. 2014. Perfluoroalkyl substances and lipid concentrations in plasma during pregnancy among women in the Norwegian Mother and Child Cohort Study. *Environment International* 62:104–112.

Steenland, K., S. Tinker, S. Frisbee, A. Ducatman, and V. Vaccarino. 2009. Association of perfluorooctanoic acid and perfluorooctane sulfonate with serum lipds among adults living near a chemical plant. *American Journal of Epidemiology* 170:1269–1278.

Steenland, K., and S. Woskie. 2012. Cohort ortality study of workers exposed to perfluorooctanoic acid. *American Journal of Epidemiology* 176:909–917.

Steenland, K., L. Zhao, and A. Winquist. 2015. A cohort incidence study of workers exposed to perfluoroctanoic acid (PFOA). *Occupational and Environmental Medicine* 0:1–8.

Takacs, M.L., and B.D. Abbott. 2007. Activation of mouse and human peroxisome proliferator-activated receptors (α,β/δ, γ) by perfluorooctanoic acid and perfluorooctane sulfonate. *Toxicological Science* 95:108–117.

ED_005024_00000285-00089

Takagi, S., F. Adachi, K. Miyano, Y. Koizumi, H. Tanaka, M. Mimura, I. Watanabe, S. Tanabe, and K. Kannan. 2008. Perfluorooctane sulfonate and perfluorooctanoate in raw and treated tap water from Osaka, Japan. *Chemosphere* 72:1409.

Tan, X., G. Xie, X. Sun, Q. Li, W. Zhong, P. Oiao, X. Sun, W. Jai, and Z. Zhou. 2013. High fat diet feeding exaggerates perfluorooctanoic acid-induced liver injury in mice via modulating multiple metabolic pathways. *PLOS One* 8(4):e61409.

Tang, C.Y., Q.S. Fu, A.P. Robertson, C.S. Criddle, and J.O. Leckie. 2006. Use of reverse osmosis membranes to remove perfluorooctane sulfonate (PFOS) from semiconductor wastewater. *Environmental Science & Technology* 40:23:7343–7349.

Tang, C.Y., Q.S. Fu, C.S. Criddle, and J.O. Leckie. 2007. Effect of flux (transmembrane pressure) and membrane properties on fouling and rejection of reverse osmosis and nanofiltration membranes treating perfluorooctane sulfonate containing wastewater. *Environmental Science & Technology* 41:6:2008–2014.

Tao, L., J. Ma, T. Kunisue, E.L. Libelo, S. Tanabe, and K. Kannan. 2008. Perfluorinated compounds in human breast milk from several Asian countries, and in infant formula and dairy milk from the United States. *Environmental Science & Technology* 42(22):8597–8602.

Tellez, M.H. 2014. Treatment of perfluorinated compounds and nitroaromatics by photocatalysis in the presence of ultraviolet and solar light. Thesis. Air Force Institute of Technology, Wright-Patterson Air Force Base, Ohio.

Thompson, J., M. Lorber, L.-M.L. Toms, K. Kato, A.M. Calafat, and J.F. Mueller. 2010. Use of simple pharmacokinetic modeling to characterize exposure of Australians to perfluorooctanoic acid and perfluorooctane sulfonic acid. *Environment International* 36:390–397.

Thompson, J., G. Eaglesham, J. Reungoat, Y. Poussade, M. Bartkowf, M. Lawrence, and J.F. Mueller. 2011. Removal of PFOS, PFOA and other perfluoroalkyl acids at water reclamation plants in South East Queensland Australia. *Chemosphere* 82:9–17.

Tittlemier, S.A., K. Pepper, C. Seymour, J. Moisey, R. Bronson, X.L. Cao, and R.W. Dabeka. 2007. Dietary exposure of Canadians to perfluorinated carboxylates and perfluorooctane sulfonate via consumption of meat, fish, fast foods, and food items prepared in their packaging. *Journal of Agricultural and Food Chemistry* 55(8):3203–3210.

Tabe, S., P. Yang, X. Zhao, C. Hao, R. Seth, L. Schweizer, and T. Jamal. 2010. Occurrence and Removal of PPCPs and EDCs in the Detroit River Watershed. *Water Practice & Technology* 5(1):1–8.

Trudel, D., L.Horowitz, M. Wormuth, M. Scheringer, I.T. Cousins, and K. Hungerbuhler. 2008. Estimating Consumer Exposure to PFOS and PFOA. *Risk Analysis* 28:251–269.

ED_005024_00000285-00090

Tucker, D.E., M.B. Macon, M.J. Strynar, S. Dragnino, E. Andersen, and S.E. Fenton. 2015. The mammary gland is a pensitive pubertal target in CD-1 and C57BL/6 mice following perinatal perfluorooctamoic acid (PFOA) exposure. *Reproductive Toxicology* 54:26–36.

UK (United Kingdom) Drinking Water Inspectorate. 2009. *Guidance on the Water Supply (Water Quality) Regulations 20001 specific to PFOS (perfluorooctane sulphonate) and PFOA (perfluorooctanoic acid) concentrations in drinking water.* Accessed May 2016. http://www.dwi.gov.uk/stakeholders/information-letters/2009/10_2009annex.pdf.

UNEP (United Nations Environmental Program). 2015. *Proposal to list pentadecafluorooctanoic acid (CAS No: 335-67-1, PFOA, perfluorooctanoic acid), its salts and PFOA-related compounds in Annexes A, B and/or C to the Stockholm Convention on Persistent Organic Pollutants.*

USEPA (U.S. Environmental Protection Agency). 1986. Guidelines for Carcinogen Risk Assessment. EPA/630/R-00/004. *Federal Register* 51(185):33992-34003

USEPA (U.S. Environmental Protection Agency). 1991. Guidelines for Developmental Toxicity Risk Assessment. *Federal Register* 56(234):63798-63826.

USEPA (U.S. Environmental Protection Agency). 1999. *Drinking Water Health Advisory: Pesticides*. Lewis Publishers, Chelsea, MI. ISBN: 978-0-87371-235-4.

USEPA (U.S. Environmental Protection Agency). 2000. *Methodology for Deriving Ambient Water Quality Criteria for the Protection of Human Health*. EPA-822-B-00-004. U.S. Environmental Protection Agency, Office of Water, Office of Science and Technology, Washington, DC. Accessed May 2016. http://www.nj.gov/drbc/library/documents/EPA_human-health-criteria2000.pdf.

USEPA (U.S. Environmental Protection Agency). 2002. *A Review of the Reference Dose and Reference Concentration Processes*. EPA/630/P-02/0002F. U.S. Environmental Protection Agency, Washington, DC. Accessed May 2016. https://www.epa.gov/sites/production/files/2014-12/documents/rfd-final.pdf.

USEPA (U.S. Environmental Protection Agency). 2005. Guidelines for Carcinogen Risk Assessment. U.S. Environmental Protection Agency. *Federal Register* 70(66):17765–18717.

USEPA (U.S. Environmental Protection Agency). 2006. Letter to Charles O. Holliday, Jr., Chairman and Chief Executive Officer of Dupont, inviting participation in the PFOA Stewardship Program. Accessed May 2016. https://www.epa.gov/sites/production/files/2015-10/documents/dupont.pdf.

USEPA (U.S. Environmental Protection Agency). 2009a. *Final Contaminant Candidate List 3 Chemicals: Screening to a PCCL*. EPA 815-R-09-007. U.S. Environmental Protection Agency, Office of Water. Accessed May 2016. https://www.epa.gov/sites/production/files/2014-05/documents/ccl3chem_screening_to_pccl_08-31-09_508v2.pdf.

ED_005024_00000285-00091

USEPA (U.S. Environmental Protection Agency). 2009b. *Method 537. Determination of Selected Perfluorinated Alkyl Acids in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS)*. EPA/600/R-08/092. U.S. Environmental Protection Agency, Office of Research and Development, National Exposure Research Laboratory, Cincinnati, OH. Accessed May 2016. https://cfpub.epa.gov/si/si_public_file_download.cfm?p_download_id=525468.

USEPA (United States Environmental Protection Agency). 2009c. *Provisional Health Advisories for Perfluorooctanoic Acid (PFOA) and Perfluorooctane Sulfonate (PFOS)*. Office of Water. US Environmental Protection Agency. Washington, D.C. Accessed May 2016. http://www.epa.gov/waterscience/criteria/drinking/pha-PFOA_PFOS.pdf.

USEPA (U.S. Environmental Protection Agency). 2011a. *Perfluorochemical (PFC) Contamination of Biosolids Near Decatur, Alabama (Fact Sheet)*. U.S. Environmental Protection Agency, Washington, DC. Accessed May 2016. https://archive.epa.gov/pesticides/region4/water/documents/web/pdf/epa_decatur_fact_sheet_final.pdf.

USEPA (U.S. Environmental Protection Agency). 2011b. *Exposure Factors Handbook: 2011 Edition (Final)*. EPA/600/R-09/052F. U.S. Environmental Protection Agency, Office of Research and Development, National Center for Environmental Assessment. Washington, DC. Accessed May 2016. https://cfpub.epa.gov/ncea/risk/recordisplay.cfm?deid=236252.

USEPA (U.S. Environmental Protection Agency). 2014a. *Framework for Human Health Risk Assessment to Inform Decision Making*. EPA/100/R-14/001. U.S. Environmental Protection Agency, Office of the Science Advisor, Washington, DC. Accessed May 2016. https://www.epa.gov/sites/production/files/2014-12/documents/hhra-framework-final-2014.pdf.

USEPA (U.S. Environmental Protection Agency). 2014b. *Emerging Contaminants – Perfluorooctane Sulfonate (PFOS) and Perfluorooctanoic Acid (PFOA)* (Fact Sheet). EPA 505-F-14-001. U.S. Environmental Protection Agency, Solid Waste and Emergency Response. Accessed May 2016. http://nepis.epa.gov/Exe/ZyPDF.cgi/P100LTG6.PDF?Dockey=P100LTG6.PDF.

USEPA (U.S. Environmental Protection Agency). 2015a. *Draft Contaminant Candidate List 4 (CCL4)*. EPA-505-F-14-001. U.S. Environmental Protection Agency. Washington, DC. Accessed May 2016. https://www.gpo.gov/fdsys/pkg/FR-2015-02-04/pdf/2015-02210.pdf.

USEPA (U.S. Environmental Protection Agency). 2015b. Drinking Water Treatability Database. U.S. Environmental Protection Agency, Washington, DC. Accessed May 2016. https://iaspub.epa.gov/tdb/pages/general/home.do.

USEPA (U.S. Environmental Protection Agency). 2015c. *Perfluorooctanoic Acid (PFOA) and Fluorinated Telomers, Basic Information*. U.S. Environmental Protection Agency, Washington, DC. Accessed May 2016. http://www.epa.gov/oppt/pfoa/pubs/pfoainfo.html.

ED_005024_00000285-00092

USEPA (U.S. Environmental Protection Agency). 2016a. *Health Effects Support Document for Perfluorooctanoic Acid (PFOA)*. EPA 822R16003. U.S. Environmental Protection Agency, Washington, DC. Accessed May 2016. https://www.epa.gov/safewater.

USEPA (U.S. Environmental Protection Agency). 2016b. *Health Effects Support Document for Perfluorooctane Sulfonate (PFOS)*. EPA 822R16002. U.S. Environmental Protection Agency, Washington, DC. Accessed May 2016. https://www.epa.gov/safewater.

USEPA (U.S. Environmental Protection Agency). 2016c. *Drinking Water Health Advisory for Perfluorooctanoic Acid (PFOA)*. EPA 822R16005. U.S. Environmental Protection Agency, Washington, DC. Accessed May 2016. https://www.epa.gov/safewater.

USEPA (U.S. Environmental Protection Agency). 2016d. *Drinking Water Health Advisory for Perfluorooctane Sulfonate (PFOS)*. EPA 822R16004. U.S. Environmental Protection Agency, Washington, DC. Accessed May 2016. https://www.epa.gov/safewater.

USGS (U.S. Geological Survey). 2011. *Report as of FY2011 for 2010MD207B: "Source Characterization of Contamination by Poly- and Per-Fluorinated Chemicals (PFCs) in Maryland Waterways."* Accessed May 2016. http://water.usgs.gov/wrri/10grants/progress/2010MD207B.pdf.

Upham, B.L., N.D. Deocampo, B. Wurl, and J.E. Trosko. 1998. Inhibition of gap junctional intercellular communication by perfluorinated fatty acids is dependent on the chain length of the fluorinated tail. *International Journal of Cancer* 78(4):491–495.

Upham, B.L., J.S. Park, P. Babica, I. Sovadinova, A.M. Rummel, J.E. Trosko, A. Hirose, R. Hasegawa, J. Kanno, and K. Sai. 2009. Structure-activity-dependent regulation of cell communication by perfluorinated fatty acids using in vivo and in vitro model systems. *Environmental Health Perspectives* 117(4):545.

Vélez, M.P., T.E. Arbuckle, and W.D. Fraser. 2015. Maternal exposure to perfluorinated chemicals and reduced fecundity: The MIREC study. *Human Reproduction* 30:701–709.

Venkatesan, A.K., and R.U. Halden. 2013. National inventory of perfluoroalkyl substances in archived U.S. biosolids from the 2001 EPA National Sewage Sludge Survey. *Journal of Hazardous Materials* 252–253:413–418.

Vermont ANR (Agency of Natural Resources). 2016. Summary of perfluorooctanoic acid (PFOA) drinking water contamination. March 10, 2016. Vermont Agency of Natural Resources, Department of Health. Accessed May 2016. http://healthvermont.gov/enviro/pfoa.aspx.

Verner, M.A., A.E. Loccisano, N.H. Morken, M. Yoon, H. Wu, R. McDougall, M. Maisonet, M. Marcus, R. Kishi, C. Miyashita, M.H. Chen, W.S. Hsieh, M.E. Andersen, H.J. Clewell, III, and M.P. Longnecker. 2015. Associations of perfluoroalkyl substances (PFAS) with lower birth weight: An evaluation of potential confounding by glomerular filtration rate using a physiologically based pharmacokinetic model (PBPK). *Environmental Health Perspectives* 123:1317–1324.

ED_005024_00000285-00093

Vestergren, R., F. Orata, U. Berger, and I.T. Cousins. 2013. Bioaccumulation of perfluoroalkyl acids in dairy cows in a naturally contaminated environment. *Environmental Science and Pollution Research* 20:7959–7969.

Vieira, V.M., K. Hoffman, H.M. Shin, J.M. Weinberg, T.F. Webster, and T. Fletcher. 2013. Perfluorooctanoic acid exposure and cancer outcomes in a contaminated community: A geographic analysis. *Environmental Health Perspectives* 121(3).

Vierke, C., C. Staude, A. Biegel-Engler, W. Drost, and C. Schulte. 2012. Perfluorooctanoic acid (PFOA) — Main concerns and regulatory developments in Europe from an environmental point of view. *Environmental Sciences Europe* 24:16.

Völkel, W., O. Genzel-Boroviczeny, H. Demmelmair, C. Gebauer, B. Koletzko, D. Twardella, U. Raab, and H. Fromme. 2008. Perfluorooctane sulphonate (PFOS) and perfluorooctanoic acid (PFOA) in human breast milk: Results of a pilot study. *International Journal of Hygiene and Environmental Health* 211(3):440–446.

Wallington, T.J., M.D. Hurley, J. Xia, D.J. Wuebbles, S. Sillman, A. Ito, J.E. Penner, D.A. Ellis, J. Martin, S.A. Mabury, O.J.Nielsen, and M.P. Sulbaek Andersen. 2006. Formation of C7F15COOH (PFOA) and Other Perfluorocarboxylic Acids during the Atmospheric Oxidation of 8:2 Fluorotelomer Alcohol. *Environmental Science & Technology* 40:924–930.

Walters, A., and D. Santillo. 2006. *Uses of Perfluorinated Substances*. GRL-TN-06-2006. Greenpeace Research Laboratories Technical Note 06/2006. Accessed May 2016. http://www.greenpeace.to/publications/uses-of-perfluorinated-chemicals.pdf.

Wambaugh, J.F., R.W. Setzer, A.M. Pitruzzello, J. Liu, D.M. Reif, N.C. Kleinstreuer, N. Ching, Y. Wang, N. Sipes, M. Martin, K. Das, J.C. DeWitt, M. Strynar, R. Judson, K.A. Houck, and C. Lau. 2013. Dosimetric anchoring of *in vivo* and *in vitro* studies for perfluorooctanoate and perfluorooctanesulfonate. *Toxicological Science* 136:308–327

Washington, J.W., J.J. Ellington, T.M. Jenkins, J.J. Evans, H. Yoo, and S.C. Hafner. 2009. Degradability of an acrylate-linked, fluorotelomer polymer in soil. *Environmental Science & Technology* 43:6617–6623.

Washington, J.W., J.J. Ellington, T.M. Jenkins, and M.P. Neill. 2010a. Concentrations, distribution and persistence of fluorotelomer alcohols in sludge-applied soils near Decatur, Alabama, USA. *Environmental Science & Technology* 44:8397–8402.

Washington, J.W., H. Yoo. J.J. Ellington, T.M. Jenkins, and E.L. Libelo. 2010b. Concentrations, distribution and persistence of perfluoroalkylates in sludge-applied soils near Decatur, Alabama, USA. *Environmental Science & Technology* 44:8390–8396.

Washington, J.W., and T.M. Jenkins. 2015a. Abiotic hydrolysis of fluorotelomer polymers as a source of perfluorocarboxylates at the global scale. *Environmental Science & Technology* 49:14129–14135.

ED_005024_00000285-00094

Washington, J.W., T.M. Jenkins, K. Rankin, and J.E. Naile. 2015b. Decades-Scale Degradation of Commercial, Side-Chain, Fluorotelomer-based Polymers in Soils & Water. *Environmental Science & Technology* 49:915–923.

Weaver, Y.M., D.J. Ehresman, J.L. Butanhoff, and B. Hagenbuch. 2009 (epub). Roles of renal organic anion transporters in transporting perfluorinated carboxylates with different chain lengths. *Toxicological Science* 113:305–314.

Weaver, J.D., B.J. Ka, D.K. Morris, W. Thompson, and J.A. Tunge. 2010. Stereospecific decarboxylative allylation of sulfones. *Journal of the American Chemical Society* 132(35):12179–12181.

Webster, G.M., S.A. Venners, A. Mattman, and J.W. Martin. 2014. Associations between perfluoroalkyl acids (PFASs) and maternal thyroid hormones in early pregnancy: A population-based cohort study. *Environmental Research* 133:338–347.

Weiss, J.M., P.L. Andersson, M.H. Lamoree, P.E.G. Leonards, S.P.J. van Leeuwen, and T. Hamers. 2009. Competitive binding of poly- and perfluorinated compounds to the thyroid hormone transport protein transthyretin. *Toxicological Science* 109: 206–216.

Wen, L.-L., L.-Y. Lin, T.-C. Su, P.-C. Chen, and C.-Y. Lin. 2013. Association between serum perfluorinated chemicals and thyroid function in U.S. adults: The National Health and Nutrition Examination survey 2007-2010. *The Journal of Clinical Endocrinology and Metabolism* 98(9):E1456–E1464.

White, S.S., A.M. Calafat, A. Kuklenyik, L. Villanueva, R.D. Zehr, L. Helfant, M.J. Strynar, A.B. Lindstrom, J.R. Thibodeaux, C. Wood, and S.E. Fenton. 2007. Gestational PFOA exposure of mice is associated with altered mammary gland development in dams and female offspring. *Toxicological Science* 96:133–144.

White, S.S., K. Kato, L.T. Jia, B.J. Basden, A.M. Calafat, E.P. Hines, J.P. Stanko, C.J. Wolf, B.D. Abbott, and S.E. Fenton. 2009. Effects of perfluorooctanoic acid on mouse mammary gland development and differentiation resulting from cross-foster and restricted gestational exposures. *Reproductive Toxicology* 27:289–298.

White, S.S., J.P. Stanko, K. Kato, A.M. Calafat, E.P. Hines, and S.E. Fenton. 2011. Gestional and chronic low-dose PFOA exposures and mammary gland growth and differentiation in three generations of CD-1 mice. *Environmental Health Perspectives* 119(8):1070–1076.

Winquist, A., and K. Steenland. 2014a. Modeled PFOA exposure and coronary artery disease, hypertension, and high cholesterol in community and worker cohorts. *Environmental Health Perspectives* 122:1299–1305.

Winquist, A., and K. Steenland. 2014b. Perfluorooctanoic acid exposure and thyroid disease in community and worker cohorts. *Epidemiology* 25:255–264.

ED_005024_00000285-00095

Wolf, C.J., S.E. Fenton, J.E. Schmid, A.M. Calafat, Z. Kuklenyik, X.A. Bryant, J. Thibodeaux, K.P. Das, S.S. White, C.S. Lau, and B.D. Abbott. 2007. Developmental toxicity of perfluorooctanoic acid in the CD-1 mouse after cross-foster and restricted gestational exposure. *Toxicological Science* 95:462–473.

Wolf, C.J., M.L. Takacs, J.E. Schmid, C. Lau, and B.D. Abbott. 2008. Activation of mouse and human peroxisome proliferator- activated receptor alpha by perfluoroalkyl acids of different functional groups and chain lengths. *Toxicological Science* 106:162–171.

Woodruff, T.J., and P. Sutton. 2014. The Navigation Guide Systematic Review Methodology: a rigorous and transparent method for translating environmental health science into better health outcomes. *Environmental Health Perspectives* 122:1007–1014.

Wu, L., H. Gao, N. Gao, F. Chen, and L. Chen. 2009a. Interaction of perfluorooctanoic acid with human serum albumin. *BMC Structural Biology* 9:31.

Wu, J., H.M. Zhou, H.Z. Li, P.C. Zhang, and J. Jiang. 2009b. Impacts of hydrodynamic shear force on nucleation of flocculent sludge in anaerobic reactor. *Water Research* 43(12):3029–3036.

Xiao, F., M.F. Simcik, and J.S. Gulliver. 2013. Mechanisms for removal of perfluorooctane sulfonate (PFOS) and perfluorooctanoate (PFOA) from drinking water by conventional and enhanced coagulation. *Water Research* 47:49–56.

Xiao, F., M.F. Simcik, T.R. Halbach, and J.S. Gulliver. 2015. Perfluorooctane sulfonate (PFOS) and perfluorooctanoate (PFOA) in soils and groundwater of a U.S. metropolitan area: Migration and implications for human exposure. *Water Research* 72:64–74.

Xu, Z., S. Fiedler, G. Pfister, B. Henkelmann, C. Mosch, W. Volkel, H. Fromme, and K.W. Schramm. 2013. Human exposure to fluorotelomer alcohols, perfluorooctane sulfonate and perfluorooctanoate via house dust in Bavaria, Germany. *The Science of the Total Environment* 443:485–490.

Yahia, D., M.A. El-Nasser, M. Abedel-Latif, C. Tsukuba, M. Yoshida, I. Sato, and S. Tsuda. 2010. Effects of perfluorooctanoic acid (PFOA) exposure to pregnant mice on reproduction. *Journal of Toxicological Science* 35: 527–533.

Yamada, A., N. Bemrah, B. Veyrand, C. Pollono, M. Merlo, V. Desvignes, and J.P. Antignac. 2014. Dietary exposure to perfluoroalkyl acids of specific French adult sub-populations: high seafood consumers, high freshwater fish consumers and pregnant women. *Science of the Total Environment* 491:170–175.

Yamada, T., P.H. Taylor, R.C. Buck, M.A. Kaiser, and R.J. Giraud. 2005. Thermal degradation of fluorotelomer treated articles and related materials. *Chemosphere* 61(7):974–84.

Yamashita, N., K. Kannan, S. Taniyasu, Y. Horii, G. Petrick, and T. Gamo. 2005. A global survey of perfluorinated acids in oceans. *Marine Pollution Bulletin* 51(8):658–668.

Yang, Q., Y. Xie, and W. Depierre. 2000. Effects of peroxisome proliferators in the thymus and spleen of mice. *Clinical and Experimental Immunology* 122:219–226.

ED_005024_00000285-00096

Yang, Q., Y. Xie, A.M. Ericksson, B.D. Nelson, and J.W. DePierre. 2001. Further evidence for the involvement of inhibition of cell proliferation and development in thymic and splenic atrophy induced by the peroxisome proliferator perfluoroctanoic acid in mice. *Biochemical Pharmacology* 62:1133–1140.

Yang, Q., M. Abedi-Valugerdi, Y. Xie, X. Zhao, G. Molle, B.D. Nelson, and J.W. DePierre. 2002a. Potent suppression of the adaptive immune response in mice upon dietary exposure to the potent peroxisome proliferator, perfluorooctanoic acid. *International Immunopharmacology* 2:389–397.

Yang, Q., Y. Xie, S.H.E. Alexson, B.D. Nelson, and J.W. DePierre. 2002b. Involvement of the peroxisome proliferator-activated receptor alpha in the immunomodilation caused by peroxisome proliferators in mice. *Biochemical Pharmacology* 63:1893–1900.

Yang, C., Y.S. Tan, J.R. Harkema, and S.Z. Haslam. 2009. Differential effects of peripubertal exposure to perfluorooctanoic acid on mammary gland development in C57Bl/6 and Balb/c mouse strains. *Reproductive Toxicology* 27:299–306.

Yang, C.-H., K.P. Glover, and X. Han. 2010. Characterization of cellular uptake of perfluorooctanoate via organic-anion transporting polypeptide 1A2, organic anion transporter 4, and urate transporter 1 for their potential roles in mediating human renal reabsorbtion of perfluorocarboxylates. *Toxicological Science* 117:294–302.

Yoo, H., J.W. Washington, T.M. Jenkins, and J.J. Ellington. 2011. Quantitative determination of perfluorochemicals and fluorotelomer alcohols in plants from biosolid-amended fields using LC/MS/MS and GC/MS. *Environmental Science & Technology* 45(19):7985–7990.

Young, C.J., V.I. Furdui, J. Franklin, R.M. Koerner, D.C.G. Muir, and S.A. Mabury. 2007. Perfluorinated acids in arctic snow: new evidence for atmospheric formation. *Environmental Science & Technology* 41(10):3455–3461.

Zaïr, Z.M., J.J. Eloranta, B. Stieger, and G.A. Kullak-Ublick. 2008. Pharmacogenetics of OATP (SLC21/SLCO), OAT and OCT (SLC22) and PRPT (SLC15) transporters in the intestine, liver, and kidney. *Pharmacogenomics* 9:597–624.

Zareitalabad P., J. Siemens, M. Hamer, and W. Amelung. 2013. Perfluorooctanoic acid (PFOA) and perfluorooctanesulfonic acid (PFOS) in surface waters, sediments, soils and wastewater – A review on concentrations and distribution coefficients. *Chemosphere* 91:725–732.

Zhang, L., X.-M. Ren, and L.-H. Guo. 2013. Structure-based investigation on the interaction of perfluorinated compounds with human liver fatty acid binding protein. *Environmental Science & Technology* 47:11293–11301.

Zhang, T., H. Sun, Y. Lin, Y. Qin, X. Geng, and L. Kannan. 2013. Distribution of poly- and perfluoroalkyl substances in matched samples from pregnant women and carbon chain length related maternal transfer. *Environmental Science & Technology* 47:7974–7981.

ED_005024_00000285-00097

Zhang, Y., S. Beesoon, L. Zhu, and J.W. Martin. 2013. Biomonitoring of perfluoroalkyl acids in human urine and estimates of biological half-life. *Environmental Science & Technology* 47(18):10619–10627.

Zhang, T., H. Sun, X. Qin, Z. Gan, and K. Kannan. 2014. PFOS and PFOA in paired urine and blood from general adults and pregnant women. *Environmental Science and Pollution Research* 22(7):5572–5579.

Zhang, C., R. Sundaram, J. Maisog, A.M. Calafat, D. Boyd Barr, and G.M. Buck Louis. 2015. A prospective study of prepregnancy serum concentrations of perfluorochemicals and the risk of gestational diabetes. *Fertility Sterility* 103:184–189.

ED_005024_00000285-00098

12.0 APPENDIX A-QUANTITATIVE CANCER ASSESSMENT MODELING

Multistage Model for Leydig Cell Tumors

```
 ======================================================================
   Multistage Cancer Model. (Version: 1.9;  Date: 05/26/2010)
   Input Data File: C:/1Data/MyFiles/PFOA-PFOS/PFOA Docs/msc_Leydig_Opt.(d)
   Gnuplot Plotting File:  C:/1Data/MyFiles/PFOA-PFOS/PFOA Docs/msc_Leydig_Opt.plt
                                            Thu May 09 11:59:27 2013
 ======================================================================

                                   BMDS_Model_Run
~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~

   The form of the probability function is:

   P[response] = background + (1-background)*[1-EXP(-beta1*dose^1-beta2*dose^2)]

   The parameter betas are restricted to be positive


   Dependent variable = Col2
   Independent variable = Col1

 Total number of observations = 3
 Total number of records with missing values = 0
 Total number of parameters in model = 3
 Total number of specified parameters = 0
 Degree of polynomial = 2


 Maximum number of iterations = 250
 Relative Function Convergence has been set to: 1e-008
 Parameter Convergence has been set to: 1e-008



                              Default Initial Parameter Values

                                     Background =      0.0132945
                                        Beta(1) =      0.0097738
                                        Beta(2) =              0


           Asymptotic Correlation Matrix of Parameter Estimates

           ( *** The model parameter(s)  -Beta(2)
                 have been estimated at a boundary point, or have been specified by
the user,
                 and do not appear in the correlation matrix )


                    Background         Beta(1)

 Background                  1           -0.64

     Beta(1)             -0.64               1
```


ED_005024_00000285-00099

Parameter Estimates

Variable	Estimate	Std. Err.	95.0% Wald Confidence Interval Lower Conf. Limit	Upper Conf. Limit
Background	0.00409839	*	*	*
Beta(1)	0.0116288	*	*	*
Beta(2)	0	*	*	*

* - Indicates that this value is not calculated.

Analysis of Deviance Table

Model	Log(likelihood)	# Param's	Deviance	Test d.f.	P-value
Full model	-28.6454	3			
Fitted model	-29.3468	2	1.40286	1	0.2362
Reduced model	-34.0451	1	10.7995	2	0.004518

AIC: 62.6936

Goodness of Fit

Dose	Est._Prob.	Expected	Observed	Size	Scaled Residual
0.0000	0.0041	0.205	0.000	50	-0.454
1.3000	0.0190	0.952	2.000	50	1.084
14.2000	0.1557	7.784	7.000	50	-0.306

Chi^2 = 1.48 d.f. = 1 P-value = 0.2245

Benchmark Dose Computation

Specified effect = 0.04

Risk Type = Extra risk

Confidence level = 0.95

BMD = 3.51044

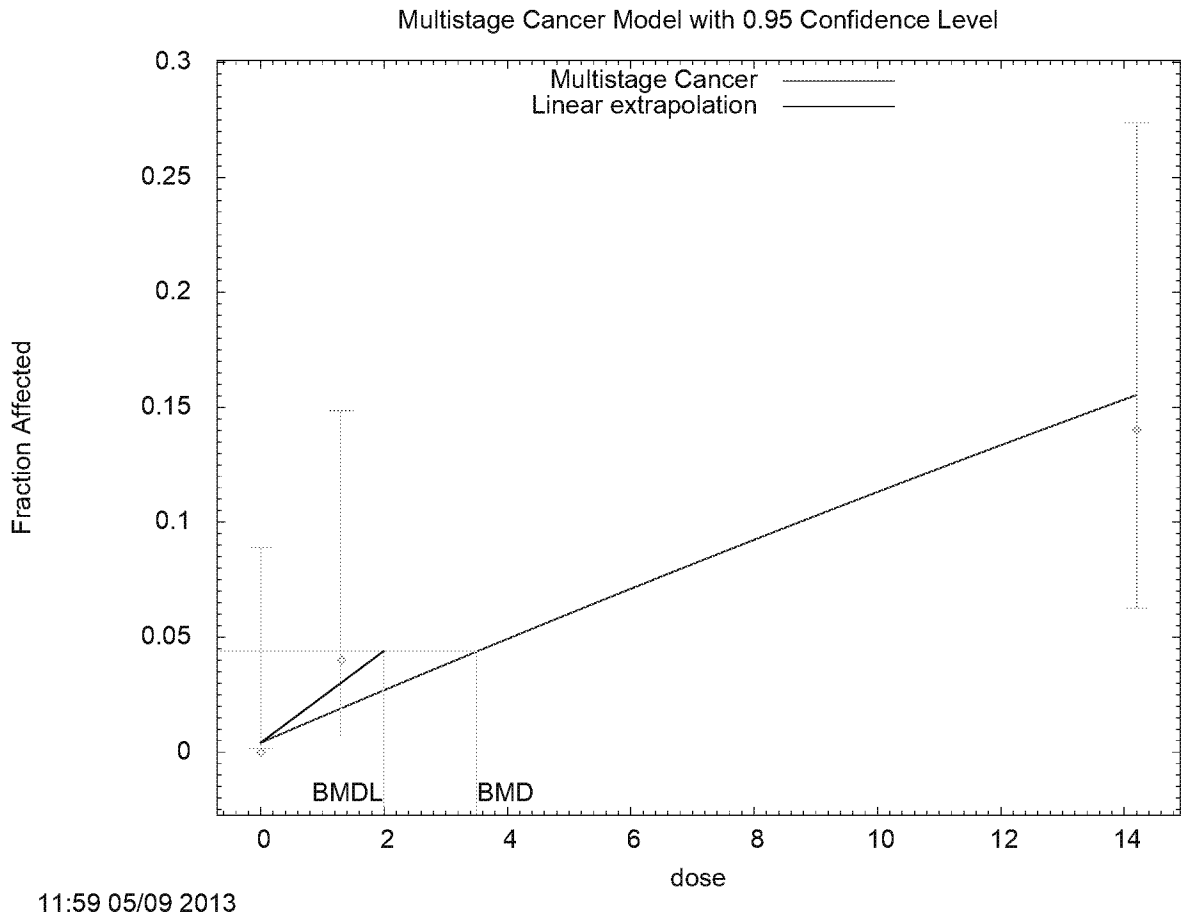
BMDL = 1.99346

BMDU = 10.7788

Taken together, (1.99346, 10.7788) is a 90% two-sided confidence interval for the BMD

Multistage Cancer Slope Factor = 0.0200656

ED_005024_00000285-00100




```
=====================================================================
  Multistage Cancer Model. (Version: 1.9;  Date: 05/26/2010)
  Input Data File: C:/1Data/MyFiles/PFOA-PFOS/PFOA Docs/msc_Leydig_Opt.(d)
  Gnuplot Plotting File:  C:/1Data/MyFiles/PFOA-PFOS/PFOA Docs/msc_Leydig_Opt.plt
                                          Thu May 09 12:05:42 2013
=====================================================================

BMDS_Model_Run
~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~

  The form of the probability function is:

  P[response] = background + (1-background)*[1-EXP(-beta1*dose^1)]

  The parameter betas are restricted to be positive


  Dependent variable = Col2
  Independent variable = Col1

Total number of observations = 3
Total number of records with missing values = 0
Total number of parameters in model = 2
Total number of specified parameters = 0
Degree of polynomial = 1
```


ED_005024_00000285-00101

Maximum number of iterations = 250
Relative Function Convergence has been set to: 1e-008
Parameter Convergence has been set to: 1e-008

Default Initial Parameter Values

Background =	0.0132945
Beta(1) =	0.0097738

Asymptotic Correlation Matrix of Parameter Estimates

	Background	Beta(1)
Background	1	-0.64
Beta(1)	-0.64	1

Parameter Estimates

Variable	Estimate	Std. Err.	95.0% Wald Confidence Interval Lower Conf. Limit	Upper Conf. Limit
Background	0.00409839	*	*	*
Beta(1)	0.0116288	*	*	*

* - Indicates that this value is not calculated.

Analysis of Deviance Table

Model	Log(likelihood)	# Param's	Deviance	Test d.f.	P-value
Full model	-28.6454	3			
Fitted model	-29.3468	2	1.40286	1	0.2362
Reduced model	-34.0451	1	10.7995	2	0.004518

AIC: 62.6936

Goodness of Fit

Dose	Est._Prob.	Expected	Observed	Size	Scaled Residual
0.0000	0.0041	0.205	0.000	50	-0.454
1.3000	0.0190	0.952	2.000	50	1.084
14.2000	0.1557	7.784	7.000	50	-0.306

Chi^2 = 1.48 d.f. = 1 P-value = 0.2245

ED_005024_00000285-00102

```
Benchmark Dose Computation

         Specified effect =           0.04
         Risk Type        =     Extra risk
         Confidence level =           0.95
                      BMD =        3.51044
                     BMDL =        1.99346
                     BMDU =         8.7003

Taken together, (1.99346, 8.7003) is a 90% two-sided confidence interval for the BMD

Multistage Cancer Slope Factor =     0.0200657
```


ED_005024_00000285-00103