GUIDANCE DOCUMENT

Guidance for Assessing the Ecological Risks of PFASs to Threatened and Endangered Species at Aqueous Film Forming Foam-Impacted Sites

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14. ABSTRACT
This guidance document, developed under SERDP to aid the DoD, presents a current state-of-the-practice overview of available methods, best practices, and key data gaps in assessing the potential for risks from exposure to perfluoroalkyl and polyfluoroalkyl substances (PFAS) for threatened and endangered (T&E) species at aqueous film forming foam (AFFF) impacted sites. It is intended to provide clear guidance to quantitatively evaluate ecological risks to PFAS, and enable site managers to make defensible, risk-based management decisions using the best available information and approaches. The guidance presents clear and specific quantitative recommendations for ecological risk assessments. This information is based on a mid-2018 to early 2019 review of publicly available information, and the values and recommendations herein should be viewed in context of future additional technical and regulatory information. Accordingly, the approaches and recommendations in this guidance are not intended to be absolute and are subject to change based on new information, site-specific regulatory and scientific considerations, and common sense.

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LIST OF ACRONYMS AND ABBREVIATIONS

AFFF	Aqueous film forming foam
ATSDR	Agency for Toxic Substances and Disease Registry
AUF	Area use factor
BAF	Bioaccumulation factor
BCF	Bioconcentration factor
BERA	Baseline Ecological Risk Assessment
BMF	Biomagnification factor
BSAF	Biota-sediment accumulation factor
$BSAF_{AP}$	Biota-sediment accumulation factor aquatic plants
$BSAF_{BI}$	Biota-sediment accumulation factor benthic invertebrates
$BSAF_{TI}$	Biota-sediment accumulation factor terrestrial invertebrates
$BSAF_{TP}$	Biota-sediment accumulation factor terrestrial plants
BW	Body weight
CERCLA	Comprehensive Environmental Response, Compensation, and Liability Act
COPEC	Chemical of Potential Ecological Concern
CSM	Conceptual site model
EcoRAGs	Ecological Risk Assessment Guidance
EcoSSL	Ecological Soil Screening Levels
EIA	Environmental impact assessments
EPC	Exposure point concentrations
ERA	Ecological risk assessments
ESA	Endangered Species Act of 1973
HC1	1% hazardous concentration
HC5	5% hazardous concentration
HQ	Hazard Quotient
HI	Hazard Indices
HPFO-DA	Hexafluoropropylene oxide dimer acid
HQ	Hazard Quotient
HHRA	Human health risk assessment
INRMP	Integrated Natural Resource Management Plans
ITRC	Interstate Technology & Regulatory Council
kg	Kilogram
K_{OC}	Organic carbon-water partitioning coefficients

LIST OF ACRONYMS AND ABBREVIATIONS (continued)

K_{ow}	Octanol- water partitioning coefficients
LOAEL	Lowest observed adverse effect level
LOEC	Lowest observed effect concentration
LOEL	Lowest observed effect level
MDEP	Massachusetts Department of Environmental Protection
mg/kg	Milligram per kilogram
mg/kg-day	Milligrams per kilogram body weight per day
MIL	Military
N-EtFOSAA	2-(N-Ethyl perfluorooctane sulfonamido) acetic acid
N-MeFOSAA	2-(N-Methyl perfluorooctane sulfonamido) acetic acid
NAVFAC	Naval Facilities Engineering Command
NMFS	National Marine Fisheries Service
NOAEL	No observed adverse effect level
NOEC	No observed effect concentration
NOEL	No observed effect level
OC	Organic carbon
PFAA	Perfluoroalkyl acids
PFAS	Perfluoroalkyl and polyfluoroalkyl substances
PFCA	Perfluoroalkyl carboxylic acids
PFBA	Perfluorobutanoic acid
PFBS	Perfluorobutanesulfonic acid
PFDA	Perfluorodecanoic acid
PFDoDA	Perfluorododecanoic acid
PFDS	Perfluorodecane sulfonic acid
PFHpA	Perfluoroheptanoic acid
PFHxA	Perfluorohexanoic acid
PFHxS	Perfluorohexanesulfonic acid
PFNA	Perfluorononanoic acid
PFOA	Perfluorooctanoic acid
PFOS	Perfluorooctanesulfonic acid
PFOSA	Perfluorooctane sulfonamide
PFPeA	Perfluoropentanoic acid
PFTeDA	Perfluorotetradecanoic acid
PFTrDA	Perfluorotridecanoic acid
PFUnDA	Perfluoroundecanoic acid

LIST OF ACRONYMS AND ABBREVIATIONS (continued)

PFSA	Perfluoroalkyl sulfonic acids
PTFE	Polytetrafluoroethylene
QA	Quality assurance
QC	Quality control
QSAR	Quantitative-structure activity relationships
SDSs	Product Data Safety Sheets
SERDP	Strategic Environmental Research and Development Program
SLERA	Screening level ERAs
SSD	Species sensitivity distribution
TDD	Total daily dose
TDI	Total daily intake
T&E	Threatened and endangered
TOC	Total organic carbon
TOPA	Total Oxidable Precursor Assay
TRV	Toxicity reference values
TSERAWG	Tri-Service Environmental Risk Assessment Working Group
UCL	Upper confidence limit
UCMR	Unregulated Contaminant Monitoring Rule
UF	Uncertainty factor
µg/L	Micrograms per liter
USEPA	U.S. Environmental Protection Agency
USFWS	U.S. Fish and Wildlife Service
ww	Wet weight

EXECUTIVE SUMMARY

Guidance for Assessing the Ecological Risks of PFAS to Threatened and Endangered Species at Aqueous Film Forming Foam Impacted Sites

Goal of the Guidance

- This focused guidance provides key recommendations and information to support quantitative ecological risk assessment (ERA) for threatened and endangered (T&E) species of 18 commonly occurring PFAS at aqueous film forming foam (AFFF)-impacted sites.

PFAS Ecological Conceptual Site Model

- Off-site habitats and aquatic food webs downgradient of AFFF release areas are particularly susceptible to potential AFFF-derived PFAS due to the soluble nature of PFAS and their ability to travel to these habitats via water transport.

PFAS Exposure Assessment

- Smaller mammals and birds with small home ranges are key T&E species exposed to PFAS at AFFF sites, and their exposures to PFAS in diet items and incidental soil/sediment ingestion can be evaluated using traditional ERA wildlife exposure modeling.
- Wildlife exposure modeling can use empirical bioaccumulation modeling of terrestrial and aquatic food webs based on measured concentrations of PFAS in soil, sediment, water, and organic carbon content of soil/sediment; a recommended model approach and model parameters are provided.

PFAS Effects Assessment

- Effects assessment for ecological risk assessments of T&E species generally involve selection of no-effect toxicity benchmarks to which site-specific exposures are compared.
- Effects to mammalian and avian wildlife, aquatic life (e.g., invertebrates and fish), terrestrial invertebrates, and terrestrial plants can be evaluated using recommended benchmarks provided in this guidance, although information is largely limited to PFOA and PFOS for many receptors.

Risk Evaluation and Interpretation

- The comparison of site-specific exposures to effects benchmarks for T&E species risk assessments at AFFF sites follows general ERA procedures.
- Site-specific exceedances of no-effects benchmarks do not imply the presence of adverse effects and may indicate the need for further evaluation of the ERA model approaches and assumptions, collection of additional data to refine the assessment, and/or site-specific ecological evaluations.

Uncertainties and Data Gaps

- There is a robust body of literature regarding fate and toxicity of PFOS and PFOA, but far less information on other PFAS.
- In terrestrial ecosystems, data gaps for PFHxA, PFDA and PFHxS have been identified as most critical based on the occurrence and behavior of these PFAS in terrestrial systems. In aquatic ecosystems, data gaps for PFHxA, PFDA, and PFDoDA have been identified as most critical based on the occurrence and behavior of these PFAS in aquatic systems.

1. INTRODUCTION

This guidance document[1], developed under the Strategic Environmental Research and Development Program (SERDP) to aid the United States Department of Defense (DoD), presents a current state-of-the-practice overview of available methods, best practices, and key data gaps in assessing the potential for risks from exposure to perfluoroalkyl and polyfluoroalkyl substances (PFAS) for threatened and endangered (T&E) species at aqueous film forming foam (AFFF) impacted sites. It is intended to provide clear guidance to quantitatively evaluate ecological risks to PFAS, and enable site managers to make defensible, risk-based management decisions using the best available information and approaches. This guidance represents recommendations and suggestions for best practices based on the current state-of-the-science; is not intended as regulation or a binding set of procedures. The guidance presents clear and specific quantitative recommendations for ecological risk assessments. This information is based on a mid-2018 to early 2019 review of publicly available information, and the values and recommendations herein should be viewed in context of future additional technical and regulatory information. Accordingly, the approaches and recommendations in this guidance are not intended to be absolute and are subject to change based on new information, site-specific regulatory and scientific considerations, and common sense.

The key target audience for this document is ecological risk assessors tasked with conducting ecological risk assessments, although the guidance will also be useful to overall site managers, as well as the broader groups within DoD that are involved in strategic management and research of PFAS. Site managers and technical specialists at AFFF-impacted sites can use this guidance to evaluate risks to T&E species exposed to PFAS, reduce uncertainty, and improve the evaluation of impacts to T&E species with the overall goals of reducing overly protective assumptions that lead to inefficient and/or potentially unnecessary remediation efforts.

Much of this guidance can be applicable for ecological risk assessments for common species; however, some of the quantitative ecological risk modeling tools, parameters, and receptors are specifically selected for assessing Federally listed T&E species present at AFFF release sites, particularly with regard to the characterization of effects, which are much more conservative for T&E species assessments. Receptor selection is based on T&E species that generally drive risks at AFFF sites, and chemical parameters (bioaccumulation factors and toxicity factors) are limited to PFAS typically found at AFFF-impacted sites. Where appropriate, this guidance highlights methodological differences specific to T&E species and provides additional information that can be applied to non-T&E species at AFFF-impacted sites.

In addition to guidance that can be applied on a site-specific basis, we highlight and prioritize key data gaps that should be communicated to the overall PFAS management and research community.

[1] The term guidance is used within this document in a general manner to represent the authors recommendations on best practices; it is not mandatory or officially binding rules to be applied by DoD services.

Addressing these data gaps will improve ecological risk assessment practice for PFAS at AFFF sites.

The key objectives of this guidance are:

- To provide a framework for the evaluation of T&E species found to be present during ecological risk assessments (ERAs) at AFFF-impacted DoD sites that reflects the level of protection and conservatism needed for T&E species;
- To provide the reader with an understanding of the specific T&E species or general feeding guilds typically expected to be considered most exposed at AFFF-impacted DoD sites;
- To provide the reader with recommended parameters (exposure factors, toxicity reference values [TRVs], uptake factors) to perform a food web model-based ERA for wildlife T&E species and aquatic life evaluations at AFFF-impacted DoD sites; and
- To provide the reader with an understanding of key data gaps and uncertainties when evaluating Federally listed T&E species found to be present at AFFF-impacted DoD sites.

The remainder of the document is organized as follows:

- **Section 1.1 Perfluoroalkyl and Polyfluoroalkyl Substances in Aqueous Film Forming Foam:** A background on PFAS in AFFF, as well as the specific PFAS addressed in this document.
- **Section 1.2 Ecological Risk Assessment Background:** A brief overview of ecological risk assessment, especially as it pertains to T&E species.
- **Section 2.1 T&E Ecological Risk Assessments at DoD Facilities**: Context for T&E species ecological risk assessments at DoD facilities.
- **Section 3.1 Generalized AFFF Conceptual Site Model:** Discussion of key ecological exposure pathways for PFAS at AFFF sites.
- **Section 3.2 Overview of T&E Species Risk Assessment:** Introduction to the overall approach for ecological risk assessments for T&E species.
- **Section 3.3 T&E Species Exposure Assessment:** Approaches for selecting representative species, collecting site-specific data, and food web modeling for predicting exposures to vertebrate wildlife.
- **Section 3.4 T&E Species Effects Assessment:** Guidance on selecting assessment endpoints and effects benchmarks with which to characterize predicted site-specific exposures.
- **Section 3.5 T&E Risk Evaluation and Interpretation:** Direction on comparing predicted site-specific exposures to effects benchmarks, including next steps for refining estimates or potential management of risks.
- **Section 4.1 Key Uncertainties:** Discussion of key uncertainties for the current exposure and effects guidance.

- **Section 4.2 Research Needs and Critical Data Gaps for Ecological Risk Assessment of PFAS:** Identification of critical information needs that would improve ecological risk assessment of PFAS at all AFFF sites.

1.1 Perfluoroalkyl and Polyfluoroalkyl Substances in Aqueous Film Forming Foam

AFFF is a synthetic Class B firefighting foam developed in the 1950s to quickly suppress hydrocarbon fires such as those that occur at airports, military sites, or refineries. AFFFs are the most effective fire suppression tool available for hydrocarbon fires and are critical components of site safety systems at many industrial and military facilities. AFFF used at military installations must meet the criteria for efficacy (extinguishment time, corrosion rate) and environmental safety outlined in military specification (Mil-Spec) MIL-F-24385. Although it is used in some building-mounted fire systems, AFFF is also used in mobile firefighting settings where fires occur in large open spaces (e.g., fuel spill or aircraft fires on a runway). DoD was a frequent user of AFFF due to the need for hydrocarbon fuels in many activities, and due to the pattern of AFFF uses, AFFF-impacted areas are found at many DoD facilities (Anderson et al., 2016; Field et al., 2017).

Due to their unique water repellency and surfactant properties, PFAS were and continue to be a key component of AFFF (Field et al., 2017; ITRC, 2018). PFAS are a family of several hundred different organic substances whose molecular structures contain one or more carbon (C) atoms with fluorine (F) atoms in the place of hydrogen (H) atoms (Buck et al., 2011). Many environmental professionals and stakeholders are familiar with perfluoroalkyl acids (PFAAs) particularly two key classes of PFAAs: 1) perfluoroalkyl carboxylic acids (PFCAs), which includes perfluorooctanoic acid (PFOA); and 2) perfluoroalkyl sulfonates (perfluoroalkyl sulfonic acids [PFSAs]), a class that includes perfluorooctanesulfonic acid (PFOS). However, the larger class of PFAS includes up to 3,000 chemicals, including many more complex and intermediate polyfluoroalkyl substances that may degrade to the persistent perfluoroalkyl acids (Wang et al., 2013; 2017).

Many AFFF formulations contain a broad spectrum of both long carbon-fluorine chain and short carbon-fluorine chain PFAS, including dozens to hundreds of PFAS that are of potential environmental concern (Buck et al., 2011; D'Agostino and Mabury, 2014). These include (but are not limited to) PFCAs, PFSAs, fluorotelomer sulfonic acids/sulfonates, fluorotelomer carboxylic acids, fluorotelomermer-captoalkylamido sulfonate, and many intermediate and precursor compounds that may transform to persistent PFCAs and PFSAs (Buck et al., 2011; Young and Mabury, 2010; Weiner et al., 2013; D'Agostino and Mabury, 2014). In general, AFFF formulations contain several percent (by weight) of PFAS (Weiner et al., 2013; Backe et al., 2013; Barzen-Hanson et al., 2015; 2017; Place and Field, 2012).

PFAS Analyte List for T&E Assessment Guidance

It is not possible to provide guidance on all potential PFAS that may be present in environmental media at AFFF sites due to the present lack of data. Information on some groups of PFAS currently inform risk assessment practices. For example, efforts to control exposure to long-chain PFAS

have resulted in PFOS and its precursors being included under the Stockholm Convention on Persistent Organic Chemicals, as well as other national and regional regulatory and voluntary initiatives. PFOA and its precursors are being considered for inclusion under the Stockholm Convention and have been subject to voluntary phase out initiatives in the United States. This has resulted in a relatively robust level of information on PFOS and PFOA. In response to regulatory actions, there has been a shift towards PFOA and PFOS production in less regulated countries in Asia as well as towards production of short-chain PFAS and other fluorinated alternatives (Ritter, 2010; Wang et al., 2013). Estimates suggest that at least 3,000 PFAS are currently on the global market (Swedish Chemical Agency (KEMI), 2015); although not all of these are associated with AFFF. A 2012 study by Place and Field (2012) identified 10 subclasses of PFAS in multiple AFFF formulations, and D'Agostino and Mabury (2014) reported 22 classes of PFAS in AFFF and commercial products. In a follow-up to these efforts, Barzen-Hanson et al. (2017) documented an additional 40 classes of PFAS in AFFFs used by the U.S. military and in AFFF-impacted groundwater. It should be noted that there are many non-AFFF sources of PFAS and it may not be appropriate to assume all PFAS detected in environmental media at a Site are related to AFFF use or release.

Despite these production shifts and analytical discoveries, toxicity information for wildlife and aquatic life is primarily limited to only a few PFAS (particularly PFOA and PFOS); therefore, specific guidance regarding the assessment and uncertainty of other commonly encountered PFAS at AFFF sites, as well as PFAS precursors, may be provided based on the availability of technical information needed for ecological risk assessments. For example, AFFF based on gaseous fluorinated ketone, PFBS derivatives, or pure 6:2 fluorotelomers are being developed to replace the early generation of PFAS used in AFFFs (Wang et al., 2013), but ecological data on these newer compounds are largely nonexistent. Currently, there does not appear to be evidence to suggest that the PFAS alternatives produced as part of the GenX processing technology (perfluoro-2-propoxypropanoic acid (hexafluoropropylene oxide dimer acid, HPFO-DA) and the ammonium salt of HPFO-DA), which have gained relevance in the recent scientific literature and media, are present in AFFF formulations.

This guidance covers a limited analyte list of 18 PFAS (Table 1; Figure 1). In developing this list, the 14 PFAS quantifiable by the United States Environmental Protection Agency (USEPA) Method 537 (Revision 1.1; 2009) were initially considered, with a particular focus on the six PFAS included in the Third Unregulated Contaminant Monitoring Rule (UCMR3) and PFAS included in the recent (June 2018) Agency for Toxic Substances and Disease Registry (ATSDR) draft toxicological profile for perfluoroalkyl substances (Table 1). Additional PFAS with state guidance or standards are also discussed in the document, as data are available (Table 1).

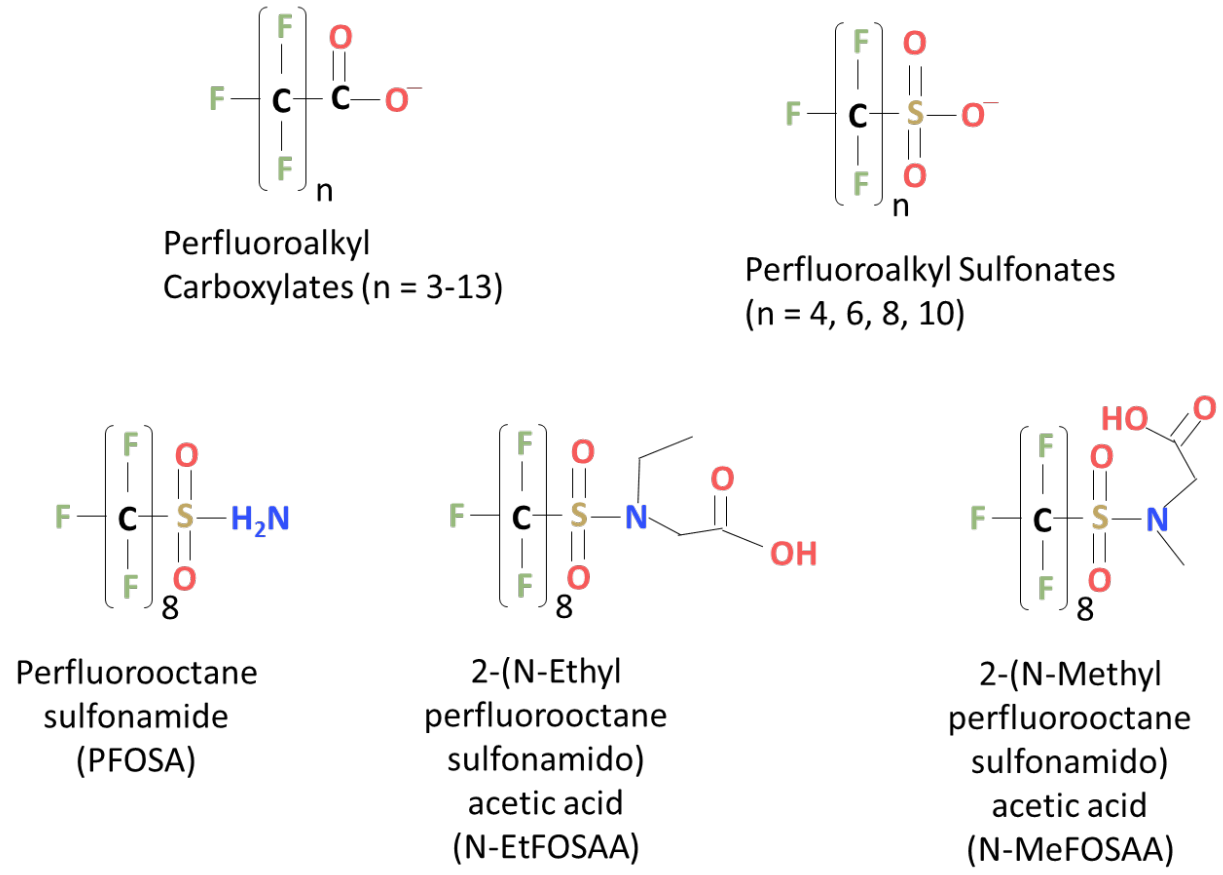

Figure 1: General Classes of PFAS Evaluated

While it is generally recognized that the environmental fate, transport, and risks from PFOA and PFOS are well characterized relative to other PFAS, the additional PFAS identified in Table 1 were considered to have the highest likelihood of empirical data useful for this guidance document, given their attention in the USEPA drinking water method, the UCMR program, and the ATSDR toxicological data profile, as well as individual state efforts to manage PFAS. Additionally, many of the compounds listed in Table 1 also have screening levels for groundwater protection and/or human health screening levels established by one or more US states or USEPA (ITRC, 2018). While most of the research on these compounds and the established guidelines noted above are focused on potential risks to humans, these compounds are likely to be included in the evaluation of ecological risks and are, therefore, included in this guidance document.

1.2 Ecological Risk Assessment Background

Ecological risk assessments (ERAs) are key components of the evaluation of environmental risks and the need for remediation or restoration at DoD Sites. Along with human health risk assessments (HHRAs), ERAs identify the chemicals of concern (COCs) that are posing potentially

unacceptable risks to plants, invertebrates, wildlife, and other ecological receptors and functions. ERAs and HHRAs form the foundation of most remedial planning at impacted sites. The overarching goal of ERAs is to protect ecological resources, including the ecological functions of populations of communities.

Significant guidance on performing ERAs has been developed by the USEPA and the DoD under the Tri-Service Environmental Risk Assessment Working Group (TSERAWG; 1996, 2008). A strong understanding of the conceptual site model (CSM) is the foundation for all risk assessments and outlines the sources, fate and transport pathways, exposure media, and potentially exposed receptors at a site. ERAs are performed initially as screening level ERAs (SLERAs) where conservative assumptions are used to eliminate chemicals and media that can be shown with a high degree of confidence to pose no unacceptable risk (USEPA, 1997). Chemicals and media that cannot be excluded during the SLERA process are carried forward into a baseline ERA (BERA), which refines assumptions and performs additional sampling or modeling evaluations to refine estimates of potential risk. ERAs conducted for the Navy follow a similar three-tiered process: Tier 1 Screening Risk Assessment; Tier 2 Baseline Ecological Risk Assessment; and Tier 3 Evaluation of Remedial Alternatives (NAVFAC, 2018).

Detailed guidance on the performance of ERAs is not within the scope of this document; however, readers are referred to the following for more detail:

- USEPA (1997) Ecological Risk Assessment Guidance (EcoRAGs)
- TSERAWG (1996) Tri-Services Procedural Guidelines for Ecological Risk Assessment
- TSERAWG (2008) A Guide to Screening Level Ecological Risk Assessment

When T&E species are found to be present at a site, additional considerations must be made during the ERA process. Federally listed T&E species are protected under the Endangered Species Act of 1973 (ESA), which provides a program for the conservation of T&E plants and animals and the habitats in which they are found. The ESA requires federal agencies such as DoD to ensure that their actions are not likely to jeopardize the continued existence of any listed species or result in the destruction or adverse modification of designated critical habitat of such species. The U.S. Fish and Wildlife Service (USFWS) and the National Marine Fisheries Service (NMFS) are responsible for administering the ESA, including the listing of species (i.e., the labeling of a species as either threatened or endangered) and designations of critical habitat. Responsibilities are split by habitat type, with USFWS being responsible for the management of terrestrial and freshwater species, and the NMFS responsible for managing marine species and anadromous fish species (species that migrate from saltwater into freshwater to spawn).

The key difference during the ERA process when T&E species are involved are the additional requirements to prevent habitat destruction, the prohibition of any “take” of T&E species (which includes sampling for scientific purposes), and the additional level of protection expected during the ERA process. As noted above, the goals of typical ERAs involving non-T&E species are to protect ecosystem services and functions and to protect the structure and function of populations of ecological communities – this goal can allow for some low-level impacts to individuals so long

as these are not likely to result in impacts to the population or community as a whole. However, when T&E species are considered at a site, the goal of an ERA usually becomes more protective of all individuals of the T&E species and all designated critical habitat for the T&E species. As a result, this guidance has been developed to include a high level of conservatism and protection for T&E species that may not necessarily be appropriate for application at sites where risks to commonly occurring species are being assessed. Where relevant, this guidance notes these key differences and includes how this guidance may be adjusted for application to non-T&E species-related ERAs.

Introduction: Key Points

- AFFF formulations can contain hundreds of PFAS; 18 PFAS are evaluated in this focused T&E guidance.
- ERAs provide a process to evaluate environmental risks of PFAS to plants, invertebrates, aquatic life, and wildlife at sites impacted by AFFF.
- ERAs for T&E species found to be present at Comprehensive Environmental Response, Compensation, and Liability Act (CERCLA) sites are generally more conservative than ERAs for common species and often require additional levels of protection to prevent habitat destruction and disturbance of any T&E species.

2. T&E SPECIES RISK ASSESSMENTS AT DoD FACILITIES

The DoD manages approximately 25 million acres of land in the United States across 420 large military installations (each greater than 500 acres), with 344 of those installations having natural resources significant enough to require active management plans (DoD, 2017). The DoD actually has a higher density of T&E species on their lands than any other federal agency; therefore, the management of T&E species is a considerable task. The DoD considers T&E species management under multiple contexts, including during the development of Integrated Natural Resource Management Plans (INRMP), the development of project-specific environmental impact assessments (EIA), and in the context of potential risks to T&E species from exposure to chemicals as a result of DoD activities under ERAs. INRMPs are planning documents that allow DoD installations to implement landscape-level management of their natural resources and can include captive breeding programs, habitat enhancement, prescribed burning, invasive species management, noise effect studies, monitoring, and inventory (DoD, 2017).

The majority of ERAs on DoD sites identified have been performed under the Comprehensive Environmental Response, Compensation, and Liability Act (CERCLA) program. ERAs under CERCLA follow the USEPA (1997) framework for ERA, as outlined in EcoRAGs (USEPA, 1997). In general, all programs and guidance on ERAs indicate that T&E species should be identified and considered in an ERA. Under the CERCLA program, when T&E species are included as part of an ERA, the following two key issues are relevant:

1. The T&E species is specifically included in the risk modeling and evaluation. If information on the T&E species is unavailable, a surrogate species with a similar life history and exposure parameters is evaluated (USEPA, 1997). If biota sampling is selected as part of an ERA, a surrogate species is used. The surrogate should represent the same feeding guild, be of similar size, and have the same foraging behavior.

2. The assessment endpoints for the ERA often focus on effects to individual organisms, rather than population-level impacts. It is noted in USEPA (1999) that CERCLA remedial actions generally should not be designed to protect organisms on an individual basis (the exception being designated protected status resources, such as listed or candidate Federally listed T&E species or treaty-protected species that could be exposed to site releases), but to protect local populations and communities of biota.

In practice, the use of assessment endpoints of population and community level effects is typically implemented in the toxicity characterization of the ERA via the selection of TRVs or other effect benchmarks developed from controlled laboratory studies with standard test animals (e.g., chicken, rat, mice, etc.). TRVs are identified from exposure doses associated with an absence of statistically detectable differences in effects from controls (termed no observed effect level (NOEL) values) or doses associated with a lack of adverse effects (termed no observed adverse effect level (NOAEL) values). NOAEL and NOEL values are generally more conservative than lowest observed effect level (LOEL) or lowest observed adverse effect level (LOAEL) values associated with a potential low level adverse or statistically detectable differences in effects from controls, respectively. For

vertebrate wildlife species, NOEL, NOAEL, LOEL, and LOEALs are typically determined based on daily oral exposures (dietary intakes) and are expressed on dosage units (e.g., mg chemicals per kilogram body weight per day, mg/kg-day).

A number of examples supporting the use of NOEL or NOAEL values for the assessment of T&E species are available. For example, the Ecological Soil Screening Levels (Eco-SSLs; USEPA, 2005) are based on NOAEL values and note that Eco-SSLs should be protective of rare, endangered, and threatened species. However, the final decision should be made on a site-specific basis in consultation with the USFWS and other natural resource trustees. The use of NOEL values is noted in several state and federal guidance as well. For example, Oregon state law requires that TRVs for the protection of bird populations be identified based on LOAEL exposures; whereas, TRVs for the protection of individual birds (i.e., for threatened and endangered species) must be identified based on NOAEL exposures (Fuchsman et al., 2017). For some chemicals, only a LOEL or LOAEL value may be available. In these cases, some guidance and literature sources recommend consideration of the application of modifying or uncertainty factors to "convert" a LOAEL or LOEL TRVs to lower, more protective value (Giesy and Jones, 2004). The general use of uncertainty factors is controversial (Allard et al., 2009), and risk assessors should consult state and EPA-region guidance and coordinate with regulatory stakeholders on their application for particular sites and exposure scenarios.

The terms no observed effect concentration (NOEC) and lowest observed effect concentration (LOEC) can also apply when considering media-specific concentrations (e.g., concentrations in surface water for characterizing effects of aquatic life, concentrations in soil for characterizing effects for invertebrates and plants in soil, etc.). For the application of these effect benchmarks to T&E species, a NOEC value developed from a controlled laboratory study with a similar organism as the T&E species of interest is generally used. If sufficient information is available, aquatic life benchmarks protective of a hypothetical proportion (i.e., 90%, 95%, or 99%) of all species are developed from Species Sensitivity Distributions that incorporate measured toxicological responses of multiple species. Concentrations in tissue associated with effects (or lack of effects) in controlled laboratory studies can also be used for evaluating toxicity of aquatic life (or soil invertebrates), however, the availability of these values for PFAS are limited.

2.1 T&E Ecological Risk Assessments at DoD Facilities

To provide context for this guidance, available studies and reports related to ERAs with a focus on T&E species found at AFFF-impacted sites were targeted for review, with the aim of providing an overview of current methods and approaches. However, ERAs at DoD facilities impacted by AFFF that have included T&E species could not be identified and obtained for this review. In fact, only one completed ERA specific to PFAS was identified (Salice et al., 2018), and this ERA did not include T&E species. Following use of AFFF at Barksdale Airforce Base, PFAS were identified in the downstream wetland of Cooper Bayou, Louisiana, where exposure to aquatic life and wildlife were evaluated. Due to the lack of TRVs and other effect benchmarks for many PFAS, PFOS was the focus of the risk assessment (Salice et al., 2018). The ERA performed followed

general ERA methodologies, with site and exposure characterization, followed by comparisons of exposure estimates to toxicity benchmarks using multiple benchmarks and lines of evidence to provide context for exposures. Following site characterization and sampling, concentrations of PFOS in surface water, sediment, and tissue were evaluated against media-specific benchmarks for PFOS. To evaluate the potential for adverse effects to aquatic life, NOEC and LOEC values from toxicity studies in the literature were compiled to develop a species sensitivity distribution (SSD) and calculate a 5% hazardous concentration (HC5) which represented a concentration in surface water expected to be protective of 95% of all species. The distribution of concentrations of PFOS in surface water were compared to the HC5 to estimate the probability of potential effects to aquatic life. Additionally, tissue samples from fish collected from the site were evaluated against literature-reported NOEC and LOEC toxicity values for fish. Some potential for adverse effects to aquatic life were noted for the most highly contaminated areas of the site. While this study did not include a specific quantitative evaluation of potential effects to upper trophic level vertebrate wildlife, the authors noted that modeling work has indicated that wildlife exposures are an important consideration for PFAS at AFFF sites (Salice, et al., 2018; Larson et al., 2018). Considerable uncertainties and data gaps are discussed in Salice et al. (2018), including the lack of chronic or multigenerational toxicity studies for PFOS and a lack of toxicity information for most other PFAS.

Examples of ecological risk assessments for T&E species at DoD sites can be obtained for other chemicals and confirm the general guidance for more protective nature of T&E assessments. For example, the Naval Station Treasure Island in San Francisco, California was identified as a site for which a T&E species-specific ERA was conducted. A screening-level ERA was performed under the standard USEPA framework that identified the American peregrine falcon (*Falco peregrinus*) as a potential T&E species receptor (Tri-Eco TT, 2015). The American peregrine falcon roosts on the Bay Bridge and was assumed to use the island and surrounding waters for foraging. While the health of peregrine falcon individuals was not specifically identified as an assessment endpoint, the protection of carnivorous birds was identified as an assessment endpoint, and the Great blue heron was selected as a representative receptor for this class of organisms. Estimated doses of Chemicals of Potential Ecological Concern (COPECs), as modeled for Great blue heron, were compared to a range of no- and low-effect levels from the literature under a risk refinement step (Step 3a in USEPA [1997]). However, the key consideration for this risk assessment focused on predicted no effect level exposures that would be protective of single organisms, rather than using low-effect TRVs as the basis for risk management.

T&E Species Risk Assessment at DoD Facilities: Key Points

- Very few PFAS-specific ERAs have been performed on DoD installations to date due to the emerging nature of the contaminants, and none of the ERAs specifically evaluated a T&E species.
- T&E ERAs generally follow standard ERA evaluations, although no-effect toxicological benchmarks (rather than low-effect benchmarks) are usually used to characterize site-specific exposures.

3. FRAMEWORK FOR EVALUATION OF RISKS TO T&E SPECIES AT AFFF-IMPACTED SITES

3.1 Generalized AFFF Conceptual Site Model for Ecological Risk Assessment

Given the general chemical properties of PFAS, composition of PFAS in AFFF products, and release and/or disposal practices for AFFF (see Buck et al., 2011; D'Agostino and Mabury, 2014), several generic CSM components can be formulated as "default" options with respect to the occurrence of PFAS at sites where AFFF have been used. This generic CSM can provide a basis for prioritizing analytical approaches, determining which environmental media (e.g., surface water, soil, groundwater, fish) to sample, identifying specific sampling locations, and understanding potential exposure routes for ecological receptors.

As part of initial site characterization activities, the components of any AFFF-related equipment, systems, and AFFF training and release practices should be identified. In general, at most AFFF-impacted sites, the primary release mechanisms are:

- Direct discharge of AFFF during fire training activities;
- Direct discharge of AFFF during emergency response activities; and
- Releases/leaks of AFFF from fixed or mobile AFFF systems and storage areas.

Following AFFF releases, PFAAs, particularly PFOS, PFOA, and PFHxS tend to be the most commonly detected PFAS in environmental samples from AFFF sites. Among the PFCAs and PFSAs, PFOS regularly exhibits the highest concentrations (reports of up to several thousand ng/L in water and several thousand mg/kg in soil and sediment; Backe et al., 2013; Ahrens et al., 2015; Anderson et al., 2016). Given the wide range of solubility, sorption, and bioaccumulation properties, PFCAs and PFSAs can be prevalent in a wide variety of environmental media, including groundwater, surface water, soil, sediment, biosolids, landfill leachate, plants, fish, invertebrates, and wildlife (Lau, 2012). PFCAs and PFSAs are considered to be extremely persistent, as they not expected to degrade or transform under in environmental media under typical environmental conditions (Buck et al., 2011).

Most areas at the point of AFFF releases (and many industrial areas where PFAS products were used) do not generally feature favorable ecological habitats that make these areas relevant for ecological risk assessment. For example, fire-fighting training area locations at military and civilian airports are usually located in a manicured area or an impermeable area (paved or cement) adjacent to an airfield. In most cases, these areas are highly disturbed, are not managed or meant to provide habitat for common or T&E species and should not be included in ecological risk assessments. However, these areas may require investigation and management when they serve as sources to downgradient areas that host ecological habitats.

Off-site exposures adjacent to or downgradient of initial AFFF release areas are expected to pose the highest risks to ecological resources (Figure 2). The relatively high water solubility of PFAS (compared to other persistent organic chemicals) results in a high potential for off-site transport

via groundwater, surface water, and stormwater, as well as erosion of impacted soils and sediment. Off-site transport is likely to result in a wide variety of exposure scenarios for ecological receptors.

Aquatic environments located downgradient of AFFF site groundwater or surface water pathways could be habitat to aquatic or aquatic-dependent wildlife T&E species that may be particularly at risk of the following PFAS impacts (Ahrens and Bundschuh, 2014; Larson et al., 2018):

- Aquatic organisms such as invertebrates and fish may be at risk of the direct toxic effects of PFAS in water.
- The accumulation of PFAS in the aquatic food web may result in exposures of higher trophic level mammals and birds, and these animals may also be exposed to PFAS in sediment and surface water when the animals forage for plants or invertebrates.

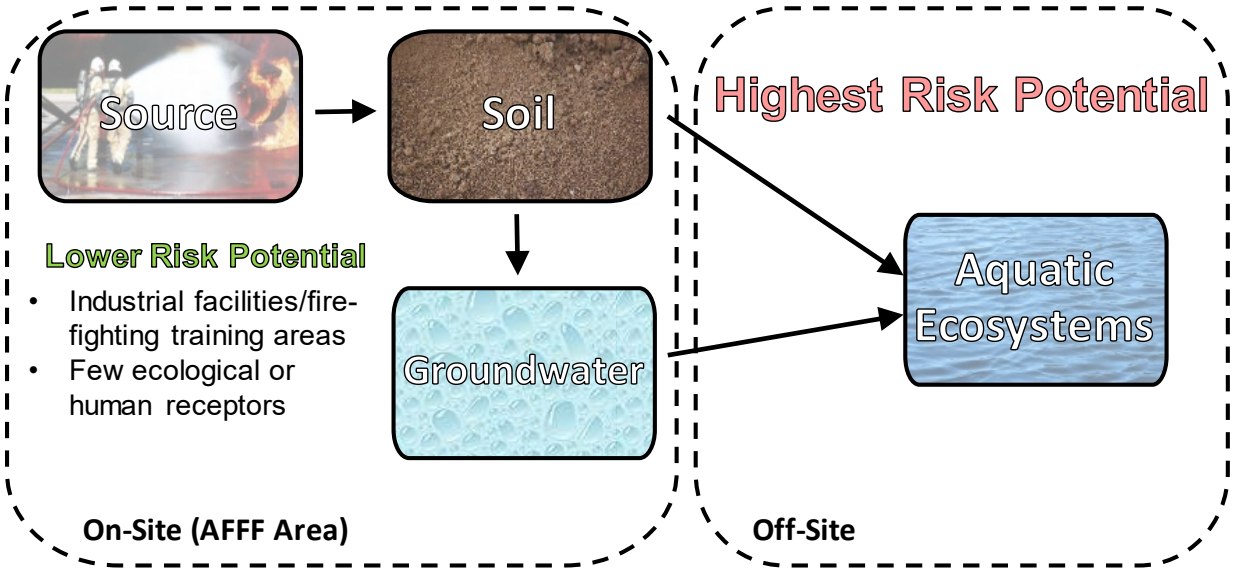

Figure 2: Simplified Conceptual Site Model for Sites Impacted by AFFF or other PFAS Sources.

Contamination of terrestrial ecosystems (or a need to evaluate terrestrial systems) is also possible via a number of hypothetical scenarios, including:

- Flooding of AFFF-impacted waterways that result in deposition of PFAS to adjacent soils.
- Disposal of PFAS-impacted soils or wastes from AFFF-training areas in natural areas that host terrestrial habitat.
- Emergency fire-response activities using AFFF in natural areas that host terrestrial habitat.

- The conversion of airfields or other areas that may have been directly impacted with AFFF to natural areas that will host habitat for ecological receptors in the future (following restoration or natural recovery).

Thus, in most cases, off-site exposures and risks are likely to be more sensitive to lower levels of PFAS relative to on-site exposures and risks and are likely to drive investigations at many AFFF-impacted sites.

Based on the exposure pathways present at AFFF-impacted sites (including off-site areas) and the features that would result in high exposures to ecological receptors, the receptors that would typically require consideration at either terrestrial or aquatic habitats that have been impacted by AFFF include:

1. Terrestrial receptors to be evaluated when AFFF-impacted soils are present:
    - Plants and soil invertebrates exposed directly to soil;
    - Small terrestrial avian and mammalian insectivores or omnivores exposed directly to soil (incidental ingestion), via diet items that have accumulated PFAS from soil, and via ingestion of surface water; and
    - Large carnivorous birds and mammals that consume surface water, and prey on smaller terrestrial birds and mammals.
2. Aquatic receptors to be evaluated when AFFF-impacted surface water bodies are present:
    - Pelagic invertebrates, amphibians, and fish exposed directly to water;
    - Benthic invertebrates exposed directly to sediment; and
    - Aquatic-dependent mammals and avian wildlife exposed to sediment (incidental ingestion), via diet items that have accumulated PFAS from sediment and/or water, and via ingestion of surface water.

Overview of Ecological AFFF PFAS Conceptual Site Model: Key Points

- Many areas in which AFFF is released are not generally targeted for ecological risk assessment because they do not usually provide habitat.
- Exposures to adjacent and proximal habitats, or downgradient areas of initial AFFF release locations are expected to pose the highest risks to ecological resources.
- Aquatic food webs downgradient of AFFF release areas are particularly susceptible to potential AFFF-derived PFAS due to the soluble nature of the contaminants and their ability to travel to these habitats via surface water run-off or groundwater to surface water transport.

3.2 Overview of T&E Species Risk Assessment

The overall framework for evaluating potential risks to T&E species is presented below for terrestrial wildlife, aquatic life, and aquatic-life dependent wildlife. For typical ecological risk assessments, these steps are usually preceded by a comparison of site-specific chemistry data to screening levels indicative of the need for ecological risk assessment. Currently, there are no nationally promulgated screening levels for ecological risk assessment of PFAS. Assuming the presence of PFAS-impacted habitat of sufficient quality and size to support a population of common or T&E species, the presence of widespread detectable concentrations of PFAS in environmental media at the site is usually sufficient to warrant an ecological risk assessment. Guidance on determining if T&E species are present at a DoD facility is not included here; it is assumed that users of this guidance document have resources and guidance for identifying specific T&E species and/or critical habitat at a facility, and that an appropriate Integrated Natural Resource Management Plans or another site-specific management plan has been developed.

Consistent with standard ecological risk assessment practice and USEPA guidance [USEPA, 1997], the following three steps are included in this guidance:

- *Exposure Assessment* – the exposure assessment step of an ERA includes the selection of representative species as assessment endpoints; estimating or measuring concentrations of COCs in diet items of selected representative species; and the estimation of daily intake of COCs from diet items, soil/sediment ingestion, and surface water.
- *Effects Assessment* – the effects or toxicity assessment step of an ERA includes selection of the TRV (based on daily intake of a compound to wildlife) or other media-based toxicity benchmarks (i.e., concentrations in surface water protective of aquatic T&E species).

- *Risk Characterization* – in the risk characterization step, the information provided by the Exposure Assessment and Effects Assessment is combined to yield quantitative risk estimates that characterize the relationship between site-specific exposures and potential toxicity or adverse effects. Typically, the exposure (either as wildlife daily intake or concentrations in exposure media for directly exposed receptors) is divided by the TRV or benchmark to calculate a Hazard Quotient (HQ). If exposure is higher than the TRV or benchmark, the HQ is greater than 1, and the following conclusions are reached:
    - If a NOEL or NOAEL TRV (or other no-effect benchmark) was used to calculate an HQ > 1, the general conclusion is that *the absence of potentially adverse effects at the site cannot be confidently concluded*. HQs > 1, when based on no-effect benchmarks, are not evidence that an adverse effect is predicted.
    - If a LOEL or LOAEL TRV (or other lowest-effect benchmark) was used to calculate an HQ > 1, the general conclusion is that *an effect at the site may be present*. However, HQs > 1, when based on low-effect benchmarks, are not necessarily evidence that an adverse effect is evident. The magnitude and type of the effect predicted based on the measured endpoints in the toxicity study used as the basis for the TRV should be clearly communicated within the risk assessment if a low-effect HQ exceeds 1. The magnitude of the HQ value (when in exceedance of 1) does not necessarily relate to a higher likelihood or severity of hazard.

In general, the above three-step general framework is customized for evaluating potential risks to T&E species at AFFF-impacted sites. For a T&E wildlife receptor (exposed primarily via dietary items), the following steps are used:

- Representative T&E species are identified and selected as assessment endpoints in the ERA. Selection of risk-driving or highly exposed representative organisms allows for the conservative evaluation of other T&E species.
- Abiotic data are collected and used with empirical measures of bioaccumulation in prey items (via chemical analysis of biota or food web modeling) to estimate dietary exposures of PFAS.
- Critical life history parameters and exposure factors to estimate exposure to PFAS are combined with data on abiotic media to estimate total daily exposures to receptors from various exposure media using standard ecological exposure modeling approaches.
- Comparisons are made between estimated daily exposures and TRVs to estimate the potential for effects.
- If unacceptable potential for risks are identified, a number of potential activities can be considered, such as: exposure model estimates can be refined by the collection of additional data, site-specific ecological evaluations of T&E species (non-invasive) or non-T&E surrogate species, and consideration of site-specific management activities to address exposures. It should be noted that the definition of "unacceptable potential risks" is a Site-

specific determination depending on the regulatory/policy framework and stakeholder concerns at a Site.

The framework for evaluating potential risks to T&E species at AFFF-impacted sites is similar for aquatic receptors that are directly exposed to PFAS in surface water or sediment and terrestrial plant and invertebrate communities that are exposed directly to PFAS in soil. In these cases, modeling to evaluate dietary contributions are not needed, and the focus is on evaluating concentrations in abiotic media against robust toxicity benchmarks:

- Representative T&E species are identified and selected as assessment endpoints in the ERA.
- Abiotic media are collected and evaluated for PFAS.
- Concentrations in media are compared to either specific toxicity information for the specific T&E species or a close surrogate. Additionally, species sensitivities distributions (SSD) for aquatic or terrestrial invertebrate or plant communities can be used to estimate a concentration in the exposure media that is protective of all receptors, including T&E species, where species or general specific toxicity information is unavailable.
- If the potential for unacceptable risks are identified, a number of potential activities can be considered, such as: exposure model estimates can be refined by the collection of additional data, predicted effects can be evaluated through toxicity testing with non-T&E surrogate species, site-specific ecological evaluations of T&E species (non-invasive) or non-T&E surrogate species, and site-specific management activities to address exposures.

Where potential risks to a T&E species are identified under this framework, risk management actions are recommended. While risk management likely involves the removal of PFAS exposures, consideration of the potential for harm to T&E species must be considered and weighed carefully.

Overview of T&E Species Risk Assessment: Key Points

- The presence of detectable concentrations of PFAS in environmental media at a site with habitat for T&E species can warrant an ecological risk assessment.
- Risk assessments for T&E species at AFFF sites follow the general approach for risk assessment of non-T&E species but with a higher level of conservatism.

3.3 T&E Species Exposure Assessment

Exposure assessment is the process of measuring or estimating the intensity, frequency, and duration of ecological exposure to a chemical in the environment. This section describes the recommended approach for the selection of representative species, the estimation of concentrations of PFAS in diet items for vertebrate wildlife (herein referred to as "wildlife"), and the estimation of wildlife intake. For receptors directly exposed to PFAS in media only (i.e., aquatic invertebrates, plants, and soil invertebrates), the estimation of dietary exposure is not generally required, as the evaluation of potential risks relies on the comparison of concentrations in external exposure media to protective, media-specific benchmarks.

3.3.1 Selection of Representative T&E Species

The first step to evaluating potential risks to T&E species at AFFF-impacted sites is to develop a site-specific CSM and select an appropriate assessment endpoint from one or more of the significant receptors that may be significantly exposed to PFAS. Generally, for ERAs concerning T&E species, the protection of a T&E species of concern is selected. However, sufficient information on the life history parameters of the species may not be available, and a representative species will need to be selected as a surrogate. It is best to select a representative common species that is a similar feeding guild and trophic level, similar body weight, and with similar exposure routes, exposure frequencies, and exposure durations. The selection of a representative, surrogate species that is considered a highly exposed receptor or "risk driving" species facilitates a conservative evaluation for T&E species.

Considering the generic CSM described above and the physicochemical properties of PFAS, the expectation at most AFFF-impacted sites is that the highest exposed wildlife receptors will exhibit the following characteristics:

- Receptors with a small home range, as they spend a higher proportion of foraging and feeding within impacted areas. Compared to smaller, lower-trophic level organisms, larger mammalian and avian carnivores are expected to have lower exposures from site-specific AFFF PFAS sources, as they forage over larger areas that are relatively unimpacted, as compared to small organisms with small home ranges (Larson et al., 2018). However, for landscape-level PFAS exposures, such as via aerial deposition or other non-point PFAS release scenarios, some PFAS have been observed to biomagnify in higher trophic level organisms (Kelly et al., 2007). The size of the AFFF-impacted habitat relative to the degree of biomagnification risk should be considered, although at most sites, as with other chemicals, the highest risks for PFAS are generally expected for small wildlife (e.g., shrews and other small rodents, small non-migratory birds) with home ranges similar to or smaller in size than the impacted area[2].

- [2] Wide-ranging receptors with large home ranges that are larger than an AFFF-impacted area being evaluated in a typical ERA are difficult to evaluate, as they may be exposed to PFAS from non-Site related (ambient) sources. Site-specific ERAs are not typically required to assess or manage ambient chemical exposures that

- Insectivore or omnivore receptors are exposed via direct soil ingestion during foraging activities and consumption of diet items that have accumulated PFAS from soil, including invertebrates and plants. These receptors are generally lower in trophic level, smaller, and generally spend a higher proportion of their foraging in small areas, though due to the consumption of predatory insects, some small birds and mammals may reach trophic positions similar to apex predators. For some species and sites, ingestion of surface water may also be relevant to evaluate, although for many chemicals, exposure via food and incidental soil/sediment ingestion are such that exposure via surface water ingestion is comparatively insignificant.

To facilitate exposure modeling for highly exposed T&E terrestrial and aquatic birds and mammals, specific T&E species that are commonly encountered within the United States were identified to provide an example within this guidance. For each receptor group (terrestrial birds, terrestrial mammals, aquatic-dependent birds, and aquatic mammals), the relevant exposure factors required for the estimation of total daily dose (TDD) or total daily intake (TDI) were compiled and are presented in Appendix A. For each receptor group, small body-weight receptors with varying feeding preferences (i.e., herbivores, invertivores, omnivores) and life history aspects that correspond to the above characteristics for high exposure potential at AFFF sites were prioritized for review. The selected representative species (selected from within the United States) include the following T&E species:

- Terrestrial mammals: Buena Vista Lake Ornate Shrew (*Sorex ornatus relictus*); Western Pocket Gopher (*Thomomys mazama*); Anastasia Beach Deermouse (*Peromyscus polionotus phasma*).
- Terrestrial birds: Coastal California Gnatcatcher (*Polioptila californica*); Masked Bobwhite Quail (*Colinus virginianus ridgwayi*); Florida Scrub-jay (*Aphelocoma coerulescens*).
- Aquatic mammals: West Indian manatee *(Trichechus manatus*); Southern Sea Otter (*Enhydra lutris nereis*); Stellar sea lion (*Eumetopias jubatus*).
- Aquatic-life dependent birds: Mississippi Sandhill Crane (*Antigone canadensis pulla*); California clapper rail (*Rallus obsoletus*); Hawaiian stilt (*Himantopus mexicanus knudseni*).

These representative species and exposure factors are provided as examples. For T&E species not reflected in Appendix A, this information provides an example for site-specific risk assessors as to the resources available and the types of exposure factors needed to address the potential for risks

originate from other areas; however, it may be useful in some cases to compare Site-specific and ambient exposures (to the extent ambient data are available).

to wildlife. Risk assessors are encouraged to select values for species as closely related to the T&E species at the site as possible.

Reptiles and amphibians are not included in Appendix A, despite being common T&E species at in the United States. Reptiles and amphibians can be exposed at aquatic AFFF impacted sites and potential risks to these species may need to be addressed. For example, Salice et al. (2018) found measurable concentrations of PFAS in fish in a waterway that likely received AFFF discharges indicating potential PFAS exposures to resident aquatic wildlife, such as amphibians and reptiles. Furthermore, research on amphibians show negative effects on growth and development with rapid uptake and depuration (Hoover et al., 2017). Amphibians and reptiles are typically not used as representative ecological receptors in risk modeling, as there is often a lack of reptile bioaccumulation and toxicity studies essential for providing parameters for ecological risk models. Reptiles could be exposed to PFAS via consumption of prey items and sediments, however the life history parameters required for modeling exposure via these routes are often lacking, as are robust toxicity data specific to PFAS. Despite the apparent potential effects on amphibians and reptiles, the current understanding of PFAS toxicity and the availability of modeling parameters for these organisms does not support a modeling approach at this time. However, larval amphibians are exposed to PFAS directly in aquatic systems and can therefore be included in assessments when using an SSD. Toxicological studies based on amphibians were included in the SSD developed by Salice et al. (2018), and in the aquatic life SSD developed in this guidance, and while the number of toxicological studies on amphibians was considerably lower than fish or aquatic invertebrates, these values are anticipated to be protective of amphibians.

As a result of the increasing presence of PFAS and accumulation in receptors at AFFF impacted aquatic sites, a critical data gap for toxicity to higher level aquatic life emerged. Currently, ESTCP is funding research to provide PFAS toxicological data on commonly exposed wildlife under ER-2627 (https://www.serdp-estcp.org/Program-Areas/Environmental-Restoration/Risk-Assessment/ER-2627/ER-2627). The research effort includes both acute and chronic toxicity studies utilizing benchmark dose methods on various aquatic species including reptiles. Until the state of the science includes a more robust evaluation of uptake and toxicity to aquatic reptiles and amphibians, site-specific biota sampling or toxicity testing using non-T&E surrogate species may be required.

3.3.2 Site-specific Exposure Assessment Data Needs and Data Collection Approach

3.3.2.1 Site-specific Data Needs

Following the identification of representative T&E species, the next step to evaluate potential risks to T&E species requires the collection and analysis of abiotic media for concentrations of PFAS.

The framework for assessing potential risks provided herein is a phased approach for data collection and analysis, with a focus on collecting abiotic samples first and evaluating potential risks using modeling approaches. The approach assumes that collection of tissue samples for analysis is only needed in cases in which modeling results are considered to be highly uncertain.

Collection and analysis of tissue samples at sites (or laboratory bioaccumulation tests with site samples) is often difficult in terms of logistics and in terms of experimental design and interpretation. By applying a conservative bioaccumulation and exposure model using measured concentrations in abiotic media, tissue sampling needs can be minimized when model-predicted risks are low. Tissue samples from non-T&E species can be collected and used to verify modeling results where the model-predicted potential for risks is high or there is high uncertainty in model outcomes.

Abiotic sampling should reflect local habitats and the site-specific CSM. The historic uses of AFFF and locations of potential releases should guide preliminary sampling; however, transport of these compounds to off-site habitats should also be evaluated.

- For terrestrial habitats, surface soils are the primary exposure media, and sufficient samples should be collected to represent spatial and temporal variability and provide sufficient statistical power to calculate appropriate exposure point concentrations (EPCs) for modeling use (e.g., 95% Upper Confidence Limits on the mean [95UCLs]). Where impacted groundwater is shallow and interacts with plants, or where T&E species of concern are herbivores, collection of plant tissue samples may be collected and used directly or used to refine risk results from preliminary exposure modeling. Terrestrial invertebrate (e.g., earthworm, arthropod) samples may be collected for evaluating the performance of food web models used to evaluate risks to wildlife. Bioaccumulation tests in the laboratory using standard invertebrate and plant test species (e.g., earthworms, lettuce, etc.) may also be used with site-collected soil.

- For aquatic habitats, surface water and sediment are the primary exposure media. Sufficient samples of both should be collected to represent spatial and temporal variability and provide sufficient statistical power to calculate appropriate EPCs for modeling use. For sediment samples, analyzing samples for total organic carbon (TOC) as additional supporting information is strongly recommended. PFAS, in particular the long chain PFSAs, sorb to organic carbon fractions in sediment, and TOC data support the modeling for uptake from sediments into benthic invertebrates (Larson et al., 2018). Benthic invertebrate, fish, or aquatic plant tissue samples from non-T&E species may be collected for evaluating the performance of food web models used to evaluate risks to wildlife. Bioaccumulation tests in the laboratory using invertebrates and plants may also be used with site-collected sediment.

3.3.2.2 PFAS Sampling and Analysis

Collecting samples for PFAS analysis can pose challenges not common with other analytes, as PFAS may be present in many regularly used sampling materials (e.g., Teflon® tubing, Teflon®-lined lids on sample collection jars), resulting in potential low and high bias issues. Several regulatory agencies provide guidance on the preferred methodology and materials for sampling and analysis of PFAS (Massachusetts Department of Environmental Protection [MDEP], 2017; Transport Canada, 2013; United States Navy, 2012). However, guidance for the sampling of

PFAS, particularly in tissue, is in its infancy, is inconsistent between regulatory agencies, and is often based on anecdotal sampling experiences. Currently, a need for robust, high quality guidance on sampling methods exists.

Site managers are encouraged to discuss sampling and analysis plans with the certified laboratory selected for the PFAS analysis. In general, the materials and activities that may introduce PFAS to sample matrices are discussed below as general guidance for PFAS sampling (summarized from MDEP [2017], ITRC [2018], and Transport Canada [2013]).

- Avoid using polytetrafluoroethylene (PTFE) (tradename Teflon®)-containing sampling equipment and sampling containers. Coordinate with laboratories and field sampling crews to ensure that all materials for collecting, processing, shipping, or storing samples do not contain Teflon®. Although PTFE/Teflon® is not a PFCA or PFSA, which are often the focus on many investigations, trace impurities/residual amounts of PFCAs may be associated with some PTFE (Buck et al., 2011).
- Do not use waterproof or plastic field notebooks, Sharpies or other markers, Post-it Notes, or blue ice packs during sampling, as these materials may contain surface coatings or materials that contain PFAS.
- Do not wear water-resistant, waterproof, or stain-treated clothing such as GOR-TEX or coated Tyvex, or new, never-washed clothing, as these may contain trace amounts of PFAS used as surface treatments.
- Eating food while sampling is never recommended; do not eat food stored in plastic containers, bags, or other polymer products while sampling. Food packaging may be a source of PFAS.
- When decontaminating reusable field equipment, some decontaminating solutions should be avoided; for example, Decon90 contains PFAS, but Alconox or Liquinox do not. Product Safety Data Sheets (SDSs) should be checked to confirm that solutions are PFAS free.
- Avoid using anything with “fluoro” in the name and avoid using materials containing fluorinated ethylene propylene (FEP), ethylene tetrafluoroethylene (ETFE), low-density polyethylene (LDPE) or polyvinylidene fluoride (PVDF).
- As contamination can occur from a variety of common consumer and sampling items, a robust program that includes several field or trip blanks and several equipment blanks is recommended during sampling. Laboratory-certified PFAS-free water should be used for field blanks and equipment decontamination.

As with any environmental investigation, early planning and coordination for sampling, identification of data quality and data use objectives, and appropriate quality assurance/quality control (QA/QC) procedures are needed to generate high-quality usable data.

3.3.3 Food Wed Modeling of PFAS for T&E Wildlife Risk Assessment

As discussed above, for wildlife species where PFAS exposure occurs primarily via dietary uptake, bioaccumulation or food web modeling is the recommended approach for a preliminary evaluation of potential risks. There are currently no mechanistic models for PFAS bioaccumulation (i.e., analogous to models that rely on the octanol-water partition coefficient, K_{OW}, to estimate the bioaccumulation of hydrophobic organic compounds). The best available approach is a stepwise estimation of concentrations of PFAS in each trophic level by applying bioaccumulation metrics.

The site-specific data needs for this type of model are discussed above and consist of abiotic media (concentrations of PFAS soil, surface water, sediment and TOC in sediment) and may include collection of biotic media tissue samples (e.g., plants, invertebrates, fish) to confirm or improve model performance, as needed.

3.3.3.1 Overview of Bioaccumulation Metrics for PFAS

Bioaccumulation occurs when uptake of chemicals exceeds excretion and/or metabolism, resulting in an increase in internal tissue concentrations relative to the environment occurs (Gobas et al., 2009). Bioaccumulation generally refers to two specific processes – bioconcentration and biomagnification. Bioconcentration refers to the uptake of a chemical from the respiratory media of an organism (water or air). Biomagnification refers to accumulation that occurs in the gastrointestinal tract when food is being digested and absorbed, and can result in higher concentrations in tissues of predators than those of its diet/prey (Connolly and Pedersen, 1988; Gobas and Wilcockson, 1999; Gobas et al., 1993).

There are currently multiple measurements of bioaccumulation that can be applied to estimate concentrations in tissues of a receptor from concentrations in exposure media. The application of these bioaccumulation metrics in multiple steps of a food web allows for the estimation of exposures for upper trophic level organisms. Currently, these metrics focus on concentrations in organisms on a wet weight basis, which can be converted to a dry weight basis, rather than evaluation of concentrations on a lipid basis. For PFAS, lipid normalizing of the tissue concentrations is not recommended, as PFAS do not partition preferentially to lipid, as with hydrophobic organics (Conder et al., 2008). The following are important bioaccumulation metrics for use in this modeling framework (summarized from Conder et al., 2012 and Gobas et al., 2009):

- Bioconcentration Factors (BCF), L/kg wet weight [ww]): BCFs are calculated in a laboratory setting under controlled exposures only. Aquatic organisms (typically fish and pelagic invertebrates) are exposed to known concentrations of a chemical, and tissues are sampled at multiple intervals over the exposure and/or at steady state. The BCF is calculated as the concentrations in tissues (wet weight basis) divided by the concentrations in the respired exposure media at steady state. BCFs control for exposure only via respiration (i.e., organisms are not fed a diet containing contaminated food).

- Biomagnification Factors (BMF), kg, ww/kg, ww: BMFs represent the biomagnification portion of bioaccumulation – uptake from diet items into tissues of predators/consumers. BMFs are usually derived from laboratory-controlled exposures where food (containing the chemical(s) of interest) is supplied to an organism (typically fish) in an otherwise clean environment (i.e., no uptake from water). BMFs are calculated as the concentrations in predator divided by the concentrations in their prey/diet. The specific concentration of chemical in the diet must be known. Field studies that measure multiple trophic levels can also be used to estimate BMFs if uptake from the water is assumed to be negligible, as in the case of very hydrophobic organic chemicals that are extremely insoluble in water. In these cases, if the concentration in a prey item is available, a BMF can be calculated using field data. However, given that most PFAS of interest at AFFF sites are relatively more water soluble and a considerable proportion of an aquatic invertebrate's or fish's uptake may occur via absorption through the skin or gills, this assumption may not be appropriate. Due to variability in concentrations between organisms of the same trophic level and lack of control of diet contents, BMFs calculated from field data can be highly uncertain relative to controlled feeding studies.
- Bioaccumulation factors (BAF), L/kg ww: BAFs are very similar to BCFs and both are calculated as the concentration of a chemical in tissues divided by concentrations in the respired exposure media. The key distinction is that BAFs are derived from field-based or mesocosm studies where the intake of chemicals can occur both via the respiratory pathway (bioconcentration) and via the dietary pathway (biomagnification). Therefore, while the ratio is based on concentrations in tissue and water, uptake may have occurred via both water and diet, adding uncertainty to the BAF. Therefore, field studies that report concentrations in tissue and the respiratory medium can only be used to calculate BAFs. It should be noted that BAFs are often misstated as BCFs in the literature.
- Biota-sediment accumulation factors for benthic invertebrates or aquatic plants (e.g., BSAF-BI or BSAF-AP), kg ww/kg OC. BSAFs are calculated from the concentration of a chemical in tissues of an aquatic organism divided by the concentration in sediment to which that organism has been exposed. Ideally, BSAFs are calculated from laboratory-controlled exposures of organisms and chemically spiked sediments. Determining BSAF values from a laboratory setting allows for more control over the sediment concentrations to which organisms are exposed. BSAFs are often calculated from field studies as well, by collecting co-located sediment and tissue samples; however, the spatial variability of sediment concentrations and organism movement adds uncertainty to this method. BSAFs can be calculated for fish, but due to spatial variability of fish foraging activities relative to PFAS sediment contamination, these values can be highly uncertain. It should be noted that chemicals in spiked sediment are typically more bioavailable than in the field, and laboratory BSAFs are often higher than field calculated BSAFs. BSAFs can be calculated on a dry weight sediment basis, but organic carbon-normalized concentrations are preferred for PFAS, as uptake may be more reliably predicted by accounting for the sorption of these compounds to organic carbon.

- Biota-soil accumulation factors for terrestrial invertebrates or terrestrial plants (BSAF-TI, BSAF-TP), kg ww/kg OC: BSAFs are calculated as the concentrations of a chemical in terrestrial invertebrates or plants divided by the concentrations of a chemical in soil. Similar to BSAF values for sediment, laboratory-controlled exposures and organic carbon-normalized concentrations in soil are preferred for calculation of BSAFs.
- Organic carbon-water partitioning coefficients (K_{OC}), L/kg OC: As PFAS sorb to organic carbon of soils and sediments (Higgins & Luthy, 2006), the partitioning coefficients between these media can be used to estimate concentrations in sediment porewater from concentrations in sediment, or vice versa. As with other parameters, laboratory-controlled measurements are preferred.

For all bioaccumulation metrics discussed above, the measurement of concentrations of chemicals in the whole body of animals[3] (rather than organ-specific measurements) is preferred, as wildlife consumption of most animal prey usually occurs on a whole-body basis. This consideration is especially relevant for fish, as they can be an important prey item when evaluating the exposures of PFAS to many aquatic-dependent wildlife. Preliminary research indicates that, although organ tissues such as liver exhibit higher concentrations of PFAS than other fish tissues (e.g., muscle), their contribution in terms of mass relative to the mass of other tissues is small such that concentrations in the whole body roughly approximate those in fillet/muscle tissue (Labadie and Chevreuil, 2011; Martin et al., 2003a; Martin et al., 2003b; Larson et al., 2018). However, a quantitative evaluation of this assumption should be evaluated experimentally or via a thorough multi-study review. For example, bioconcentration studies by Chen et al. (2016) suggested that BCF values for PFAAs with greater than four perfluorinated carbons in whole body fish samples were approximately 20% higher on average compared to BCF values based on muscle-only samples (PFBS and PFBA were a factor of two higher for whole body BCFs). At any rate, bioaccumulation values based on dividing the concentration in an organ (i.e., liver) by the concentration in water or diet should not be used for ecological risk modeling purposes, as this may greatly overestimate the dietary exposure to predators (unless it is assumed that the predator only consumes organs, which is not a viable assumption for most predators).

3.3.3.2 Bioaccumulation Modeling Framework

To facilitate exposure modeling and the assessment of risks for T&E species, bioaccumulation metrics can be applied to measurements of PFAS in abiotic media to estimate concentrations in plants, benthic or pelagic invertebrates, and fish (from respiratory uptake and/or dietary uptake). By applying these metrics in a stepwise manner, concentrations of PFAS can be estimated in benthic and pelagic invertebrates, aquatic plants, forage fish, and larger predatory fish in aquatic systems; and terrestrial plants and terrestrial invertebrates in terrestrial systems.

[3] In many cases, bioaccumulation metrics to predict concentrations in the diets of herbivorous animals focus on the portion of the plants consumed (i.e., leaves, roots, fruits) rather than the entire plant. See the discussion of plant bioaccumulation metrics in Section 3.3.3.4 below.

A CSM for the bioaccumulation modeling is provided in Figure 3. Following the estimation of concentrations of PFAS in aquatic and terrestrial plants and invertebrates and fish, standard dietary exposure modeling can be used to calculate the TDD or TDI intake of PFAS by wildlife consumers of these lower trophic level organisms, including T&E species selected as assessment endpoints. The following equations for estimating the concentrations in food web diet items using bioaccumulation metric values are assumed:

- Concentration in benthic invertebrate = measured concentration in sediment (organic carbon normalized) × BSAF-BI
- Concentration in pelagic invertebrate = measured concentration in water × BCF-PI
- Concentration in fish = sum of:
    - Concentration in benthic invertebrate × Proportion of benthic invertebrate in diet (e.g., 0.5 for forage fish) × BMF-Fish
    - Concentration in pelagic invertebrate × Proportion of pelagic invertebrate in diet (e.g., 0.5 for forage fish) × BMF-Fish
    - Measured concentration in water × BCF-Fish
- Concentration in aquatic plant = measured concentration in water × BCF-AP
- Concentration in terrestrial invertebrate = measured concentration in soil (organic carbon normalized) × BSAF-TI
- Concentration in terrestrial plant = Measured concentration in soil (organic carbon normalized) × BAF-TP

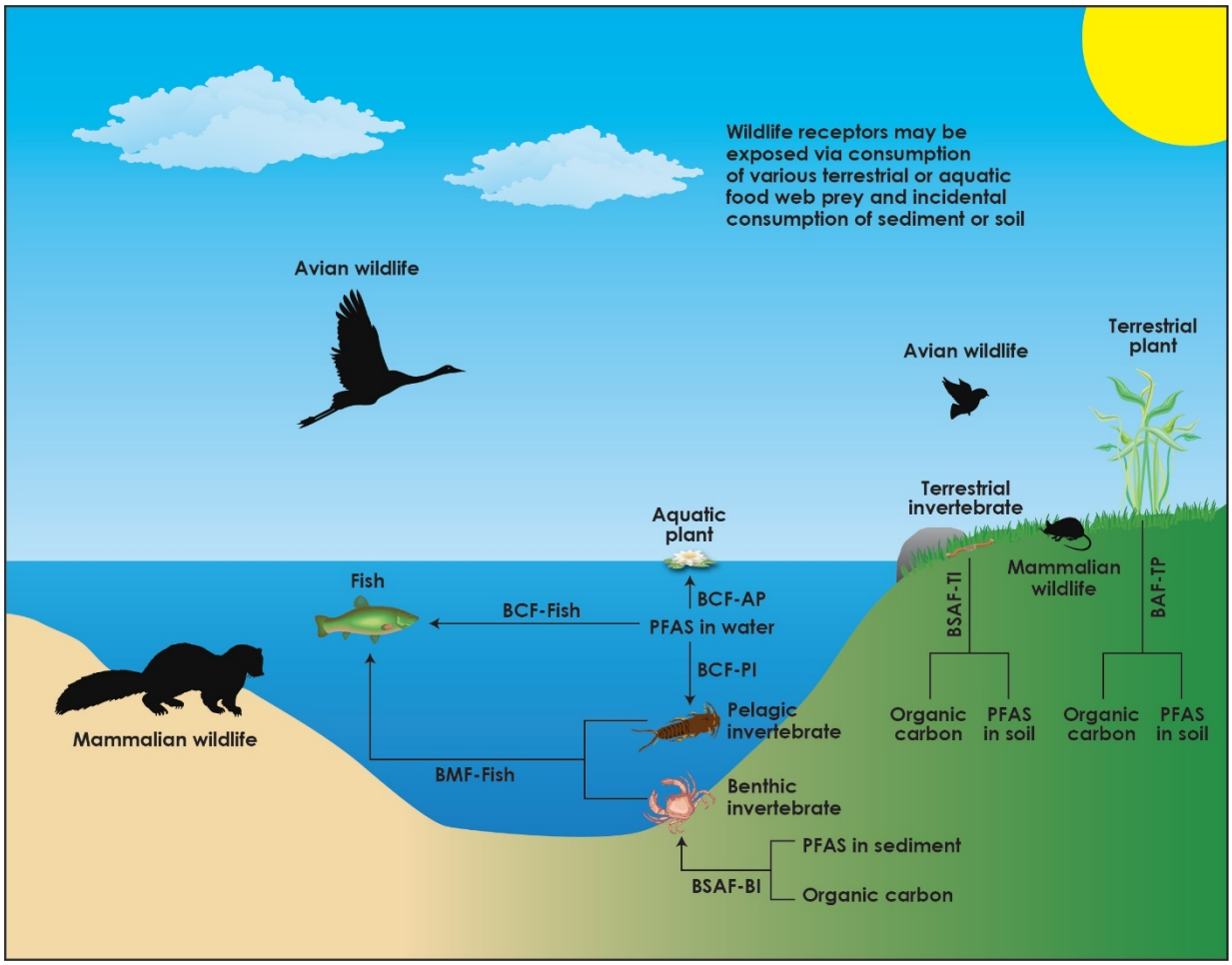

Figure 3: Conceptual Site Model for Empirical Bioaccumulation Modeling in an Aquatic or Terrestrial System

The daily ingested doses from the intake of each media can be calculated for representative wildlife receptors in daily dose rates per unit of body weight (milligram per kilogram per day [mg/kg-day]). For each receptor, the intake of PFAS can be calculated based on ingestion of soil or sediment, ingestion of surface water, and ingestion of diet items, including plants, invertebrates, and/or fish (as estimated using the above equations).

Daily intake can be calculated using standard formulas from the USEPA Wildlife Exposure Factors Handbook [USEPA, 1993]:

$$Intake = \frac{\lfloor(\sum[DFI\ x\ EPC_{diet}\ x\ P] + [DWI\ x\ EPC_{water}] + [DFI\ x\ EPC_{soil}\ x\ P_{soil}])\ x\ AUF\rfloor}{BW}$$

Intake = Daily dietary intake
EPC_{soil} = Exposure Point Concentration in soil or sediment
EPC_{diet} = Exposure Point Concentration in food– estimated using bioaccumulation metrics
EPC_{water} = Exposure Point Concentration in water
DFI = Daily food ingestion rate
DWI = Daily water ingestion rate
P = Proportion of diet composed of the individual food source
P_{soil} = Proportion of diet composed of incidentally consumed soil or sediment
AUF = Area use factor (fraction of time spent foraging at the Site); assumed to equal 1(100%)
BW = Body weight

Additional details on wildlife exposure modeling can be found in USEPA (1997) and USEPA (1993). As discussed in Section 2.3.1, exposure factors (body weight, ingestion rates) can be found for many representative species in the literature and are provided for select T&E species in Appendix A.

3.3.3.3 Selection Process for Recommended Bioaccumulation Metrics for PFAS

The regulatory and peer-reviewed literature was reviewed to identify studies that can provide the best available bioaccumulation metrics for use in ERAs. Appendix B provides a summary of the available literature from which the bioaccumulation metrics can be derived for each PFAS. These studies were reviewed to provide recommended bioaccumulation metrics. K_{OC} values were also reviewed, as these values may be of use in bioaccumulation and fate modeling.

The following guidelines were applied to select the recommended values:

- Laboratory studies using PFAS-spiked media were given the highest priority for selection. Laboratory studies provide a higher level of certainty in bioaccumulation metrics because the exposure time is quantified, the media to which organisms are exposed is homogenous, the organisms are exposed to a known concentration and media (unlike field exposures, where organisms can move and be exposed to varying conditions), organism health and condition is standardized and evaluated, and in most cases, the concentrations in organisms and the media to which they are exposed are measured at steady state, assuring metrics are not misrepresented due to insufficient exposures or spatial uncertainties regarding the movement of organisms. Additionally, laboratory bioaccumulation studies with PFAS-spiked exposure media are expected to yield conservative estimates of bioaccumulation, as PFAS may be more available in freshly spiked environmental media compared to aged PFAS in field samples (research is needed to confirm this hypothesis). The use of

controlled studies avoids uncertainties regarding exposure concentration and the mixtures of linear and branched PFAS isomers. For example, with PFAAs, it is possible that bioaccumulation rates may differ between linear and branched isomer forms, and it is hypothesized that linear PFAS are more bioaccumulative than branched PFAS (Houde et al., 2008; Houde et al., 2011). Thus, spiked, single-compound exposures using linear PFAS isomers would likely result in higher estimates of bioaccumulation compared to field conditions, which may include less bioaccumulative branched PFAS. The use of laboratory studies with spiked compounds also avoids complications with the presence of PFAS precursors which may transform into stable PFAS (such as PFAAs) in the exposure media or within the organism, leading to inaccuracies in estimating bioaccumulation metrics.

- Laboratory studies using field-collected media were generally selected as second priority. These studies may result in less conservative (though potentially more realistic) metrics but include a similar level of control as laboratory studies with PFAS-spiked media.
- Values based on quantitative-structure activity relationships (QSARs) were generally selected next. Specifically, for PFAS, some bioaccumulation studies have interpolated bioaccumulation metric values for additional PFAS based on relationships between specific metrics and fluorinated carbon chain length. For example, Martin et al. (2003a, 2003b) measured BCF and BMF values for a number of PFAAs in fish and developed a regression model (QSAR) that can predict the BCF or BMF value based on the fluorinated carbon chain length of the PFAA. Thus, in this case, a BCF and BMF value for PFNA (and several other PFAAs that were not directly measured in the study) could be predicted.
- Preference was given to studies (or multiple studies by the same author) in which many of the target PFAS were analyzed using consistent species, exposure conditions, and measurement methods, such that most of the recommended values for a metric originated from the fewest numbers of studies. This minimized the variability that could arise from differences in experimental conditions, species, measurement approaches, or other artifacts.
- Best professional judgement and a generally conservative approach was used to select between studies of similar quality. Where multiple bioaccumulation metrics were available for different species within the same group and from studies of similar quality, the more conservative species (higher bioaccumulation metrics) was generally selected. Where multiple exposure groups were used within the same study, the geometric mean of the bioaccumulation metric was calculated and selected.

Where no laboratory-measured or QSAR-derived metrics (e.g. Martin et al. 2003a, 2003b) were available, field studies may be relevant for the selection of BAFs and other parameters for particular ERAs. For this guidance, no recommended BAF values were derived from field studies. If risk assessors find that field studies are the only available source for a parameter for a specific PFAS, it is recommended that the field studies provided in Appendix B be reviewed individually, and a study should be selected that best represents the site and exposure scenario under consideration. For example, if a marine site requires a BSAF-BI for a PFAS where a laboratory

value is not available, then selection of a marine study using an appropriate organism from Appendix B would be most appropriate.

Lastly, it is possible to derive some bioaccumulation metrics based on understanding the partitioning of PFAS between organic carbon and water. For example, a BSAF-BI (kg ww/kg OC) can be used to extrapolate to a BAF (L/kg ww), and vice versa, based on applying the K_{OC} (L/kg OC). This method has a higher level of uncertainty and was not conducted for development of recommended metrics in this guidance review; however, it may be relevant in cases where no other parameters are available. Larson et al. (2018) provides an example of this approach.

It should be noted that the recommended values are subjective to the above general guidelines, and users of these values should exercise their best judgement in application of the values, especially given additional studies which will continue to emerge and refine existing knowledge. Where users of this guidance have detailed information on the dietary components of T&E species, they are encouraged to consider alternate metrics from other studies shown in Appendix B. Appendix B may also be reviewed to obtain secondary bioaccumulation metrics that can be used to perform alternate calculations (sensitivity analyses) with site-specific ecological risk models.

3.3.3.4 Recommended Bioaccumulation Metrics for PFAS

The recommended bioaccumulation metrics are shown in Tables 2a to 2f and discussed briefly below.

Organic Carbon-Water Partition Coefficient (K_{OC}): Recommended values for K_{OC} are provided in Table 2a. K_{OC} values from Guelfo and Higgins (2013), Higgins & Luthy (2006), and Zhao et al. (2012) are recommended. These studies are laboratory-based studies, with K_{oc} measured from PFAS-spiked sediments or soils. Values for two PFAS (PFTrDA, PFTeDA) were not available. Data indicate that K_{oc} values increase with increasing perfluorinated carbon chain length among the PFCAs and PFSAs (with PFBA being a possible exception), and that PFSAs are more sorptive to carbon than PFCAs. This suggests that longer-chain PFSAs may tend to partition to soils and sediments more readily than shorter-chain PFCAs.

Pelagic Invertebrate Bioconcentration Factor (BCF-PI): Recommended values to predict bioaccumulation of PFAS from water to pelagic invertebrates (Water to Pelagic Invertebrate BCF-PI values) are provided in Table 2b and shown in Figure 4. Most of the values were derived from a laboratory study with *Daphnia magna* (water flea) exposed to 25 days in PFAS-spiked laboratory water (Dai et al., 2013). The BCF for PFBS was derived from a laboratory study with *Caenorhabditis elegans* (round worms) exposed to 2 days in PFBS-spiked laboratory water (Chen et al., 2018) – while this was the only study for PFBS, there is some uncertainty regarding the applicability of *C. elegans* as a diet item in food web modeling since they are not a common diet component of higher-level species. Values for 11 of the 18 PFAS measured for in EPA Method 537.1 (PFBA, PFPeA, PFHxA, PFHpA, PFTrDA, PFTeDA, PFHxS, PFDS, PFOSA, N-EtFOSAA, N-MeFOSAA) were unavailable. Field-derived BAF-PI values are not provided as recommended values, as these are derived from field studies and due to variability in field exposures, risk assessors are encouraged to review potentially applicable studies in Appendix B

and select a value that best represents exposure conditions and the organism most relevant to their specific site. Overall, bioaccumulation in pelagic invertebrates tends to increase with increasing perfluorinated carbon chain length among the PFCAs, and there is limited information with regards to PFSAs. This suggests that longer-chain PFCAs may tend to bioaccumulate in pelagic invertebrates more readily than shorter-chain PFCAs.

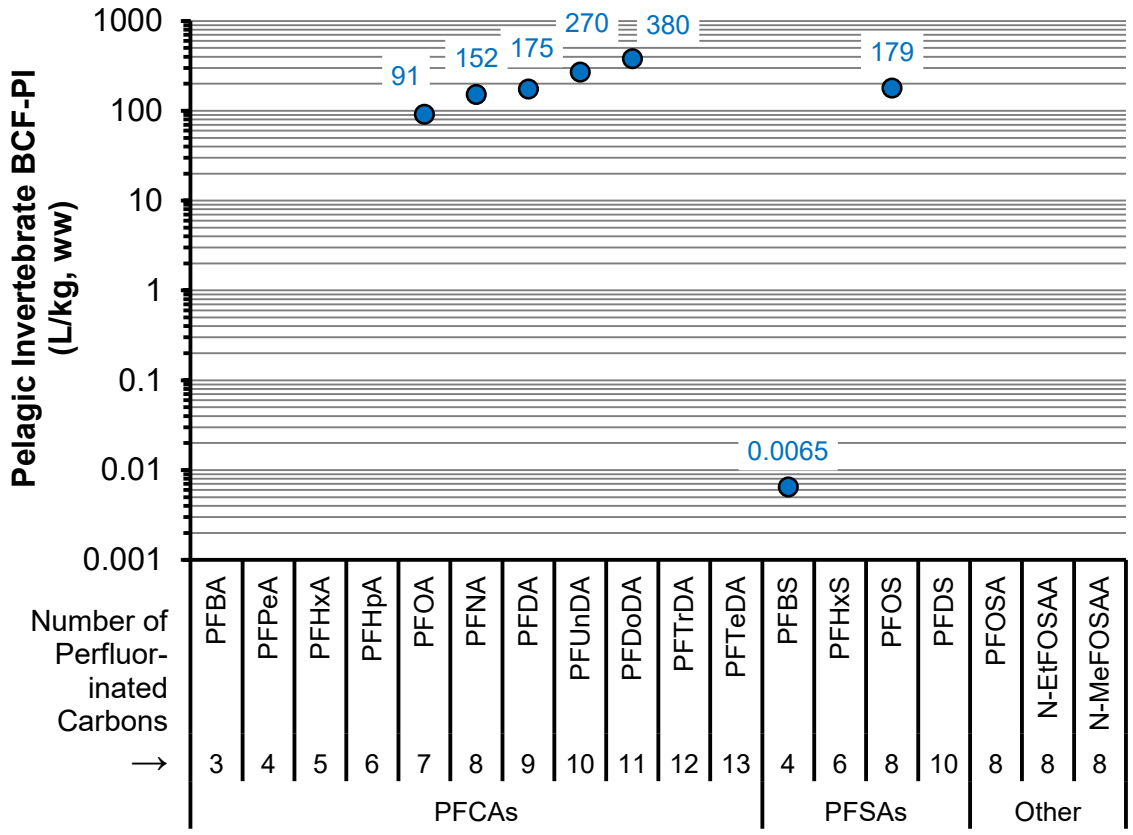

Figure 4: Recommended Water to Pelagic Invertebrate BCF-PI Values

Benthic Invertebrate Biota-Sediment Accumulation Factor (BSAF-BI): Recommended values to predict bioaccumulation of PFAS from sediment to benthic invertebrates (Sediment to Benthic Invertebrate BSAF-BI values) are provided in Table 2b and Figure 5. Most of the values were derived from a laboratory study with *Lumbriculus variegatus* (blackworm) exposed to 56 days in a PFAS-spiked field sediment (Higgins et al., 2007). BSAF values for PFHxA, PFHpA, PFTrDA, PFTeDA, PFBS, and PFHxS were obtained from Lasier et al. (2011), who exposed *L. variegatus* in the laboratory to field sediment impacted with PFAS sources associated with carpet/textile PFAS sources for 28 days. The value for PFOSA was derived from a laboratory study with *Chironomus riparius* (harlequin fly) exposed in the laboratory for 4 days to a field-collected sediment impacted from "industrial PFAS sources" (Bertin et al., 2014). Values for three of the 18

PFAS (PFBA, PFPeA, and N-MeFOSAA) were unavailable. Overall, bioaccumulation in benthic invertebrates tends to increase with increasing perfluorinated carbon chain length among the PFCAs and PFSAs for chain lengths of approximately 4 to 8. For perfluorinated carbon chain lengths greater than 8, the relationship appears to decrease slightly. For PFSAs and PFCAs of the same perfluorinated carbon chain length, BSAF-BI values appear to be similar.

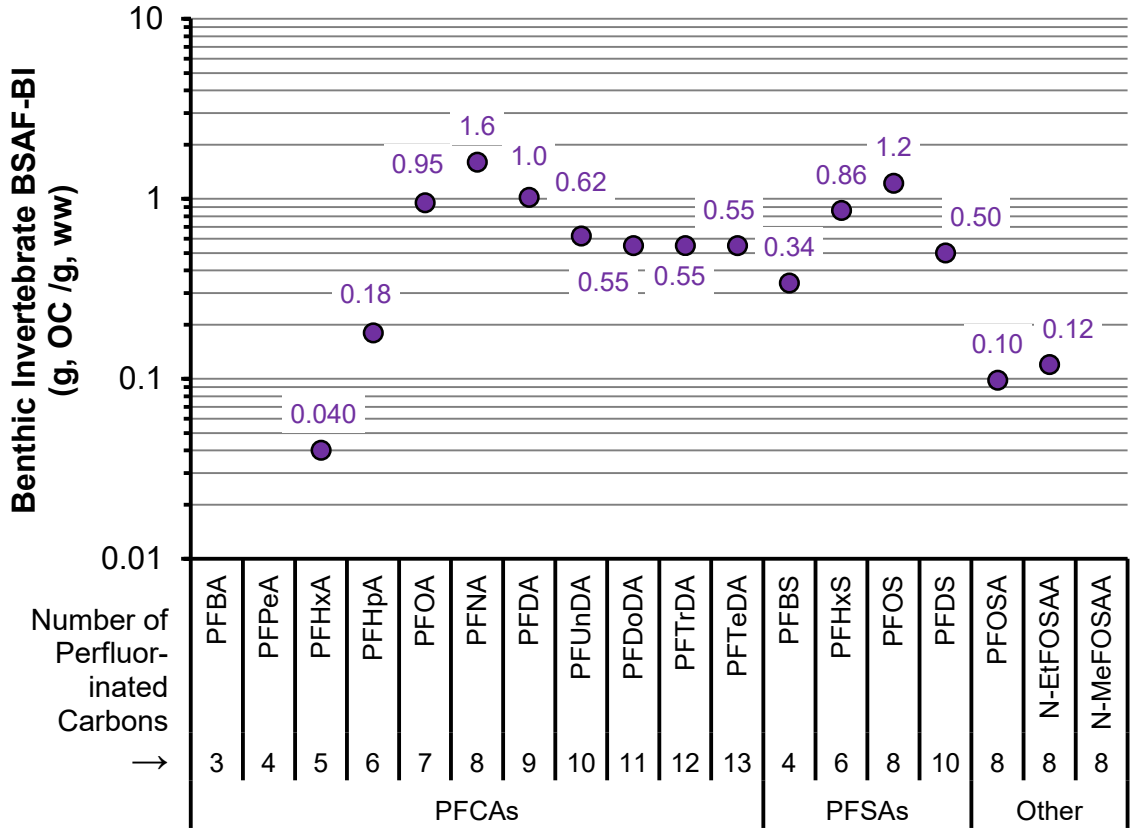

Figure 5: Recommended Sediment to Benthic Invertebrate BSAF-BI Values

Terrestrial Invertebrate Biota-Soil Accumulation Factor (BSAF-TI): Recommended values to predict bioaccumulation of PFAS from soil to terrestrial invertebrates (Soil to Terrestrial Invertebrate BSAF-TI values) are provided in Table 2c and Figure 6. Most of the values were derived from a laboratory study with *Eisenia fetida* (earthworm) exposed to 30 days in PFAS-spiked field soil (Zhao et al., 2014). The PFDS value was determined from a laboratory study with *E. fetida* (earthworm) exposed to 28 days in a field soil contaminated with biosolids impacted by PFAS (Rich et al., 2015). The N-EtFOSAA value was determined from Zhao et al. (2016), who exposed *E. fetida* for 30 days to a field soil spiked with N-ethyl perfluorooctane sulfonamide ethanol (N-EtFOSE). EtFOSAA was the primary degradation product of N-EtFOSE, and the value selected from the study could be considered a BSAF-TI value that could be used for either

compound (development of a value using N-EtFOSAA in soil would be more robust). Uncertainty with precursor chemicals is discussed in more detail in the uncertainty section below in Section 4.1. Values for five of the 18 PFAS (PFBA, PFTrDA, PFTeDA, PFOSA, and N-MeFOSAA) were unavailable.

Overall, bioaccumulation in terrestrial invertebrates tends to increase with increasing perfluorinated carbon chain length among the PFCAs for chain lengths of approximately 4 to 11, and for the PFSAs, from 4 to 8. The BSAF-TI for PFDS appears to be anomalous; however, as noted above, it is from a different study than the other values. Both studies used the same species, but the PFDS value was developed from a study with field soils contaminated by PFAS-impacted biosolids (Rich et al., 2015), whereas the other values were developed from a study with spiked soils (Zhao et al., 2014). In cases where the field and lab studies measured the same PFAA (PFOA, PFNA, PFDA, PFUnDA, PFDoDA, PFHxS, and PFOS), BSAF-TI values from the study with the spiked soils were an average of 30 times higher (range of 10 times to 70 times) than values from the field soils, potentially reflecting the higher bioavailability of PFAS in the freshly spiked soils. Use of the laboratory study BSAFs is therefore likely to yield conservative (higher) estimates of bioaccumulation of terrestrial invertebrates such that bioaccumulation under field conditions may be overestimated. Users of these values may wish to consider evaluating the field-soil derived BSAF-TI values in a sensitivity analysis or measuring site-specific bioaccumulation in site invertebrates or laboratory invertebrates exposed to site soils. For PFAAs of the same perfluorinated carbon chain length, BSAF-TI values appear to be higher for the PFSAs.

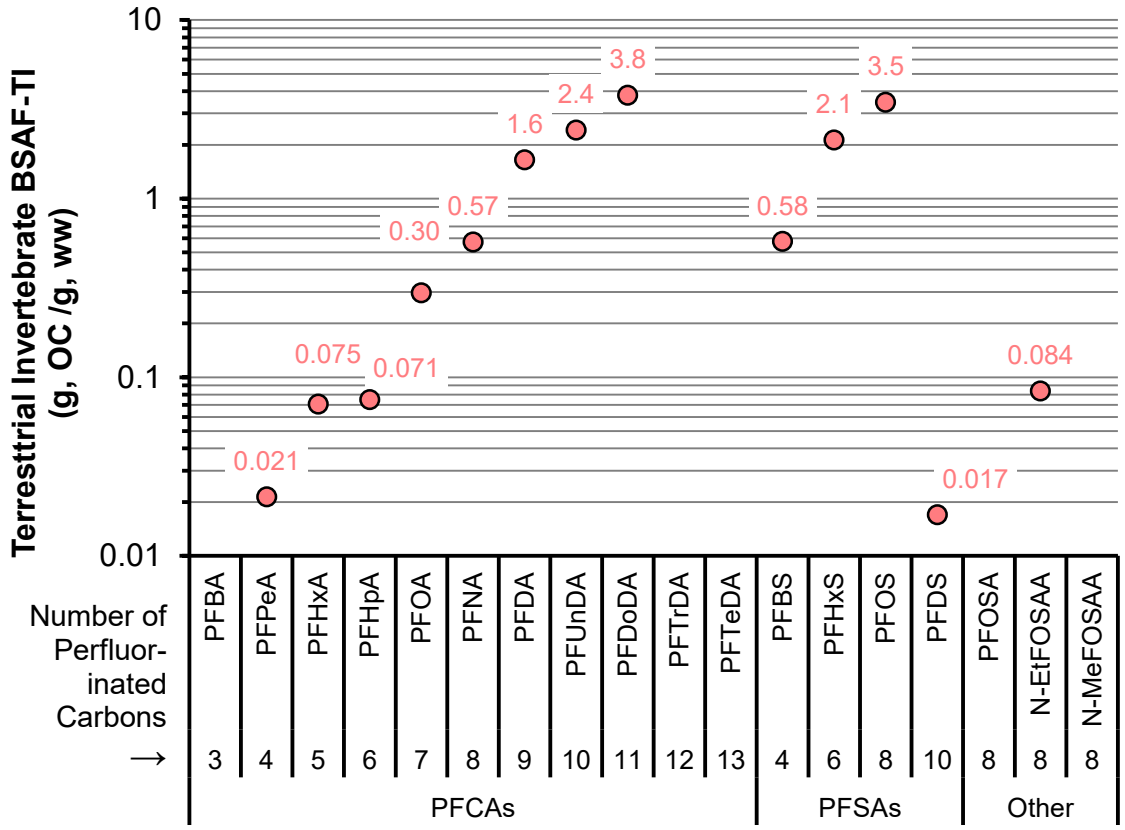

Figure 6: Recommended Soil to Terrestrial Invertebrate BSAF-TI Values

Fish Bioconcentration Factor (Fish BCF) and Fish Biomagnification Factor (Fish BMF): Recommended values to predict bioaccumulation of PFAS from water and diet to fish invertebrates (Water to Fish Tissue BCF and Diet to Fish Tissue BMF values) are provided in Table 2d and Figures 7 and 8. Most of the BCF and BMF were derived from Martin et al. (2003a and 2003b). These paired BCF and BMF studies evaluated these metrics for the same species (juvenile rainbow trout), under similar exposure conditions, and in the same laboratory. These values (as applied in food web models) have also been shown to predict concentrations in fish that correspond to measured concentrations of fish in AFFF site case studies (Larson et al., 2018). Martin et al. (2003a,b) also presented perfluorinated chain length based QSAR equations that were used to estimate BCFs or BMFs from within the range of chain lengths tested (e.g., PFNA). For PFAS with chain lengths outside the range of the Martin et al. (2003a, 2003b) QSARs, BCF values from Wen et al. (2017) and Chen et al. (2016) were selected. These values are from laboratory studies where zebrafish (*Danio rerio*) were exposed to PFAS-spiked water. Measured or QSAR-based values are included for all PFAS. It should be noted that the values for PFOSA, N-EtFOSAA, and N-MeFOSAA are highly uncertain, as these are based on QSARs developed for PFAAs, and it is unclear how the structural changes in these compounds could affect bioconcentration. Additional BMFs for PFNA and PFBS were selected from Goeritz et al. (2013) and are based on laboratory testing with rainbow trout.

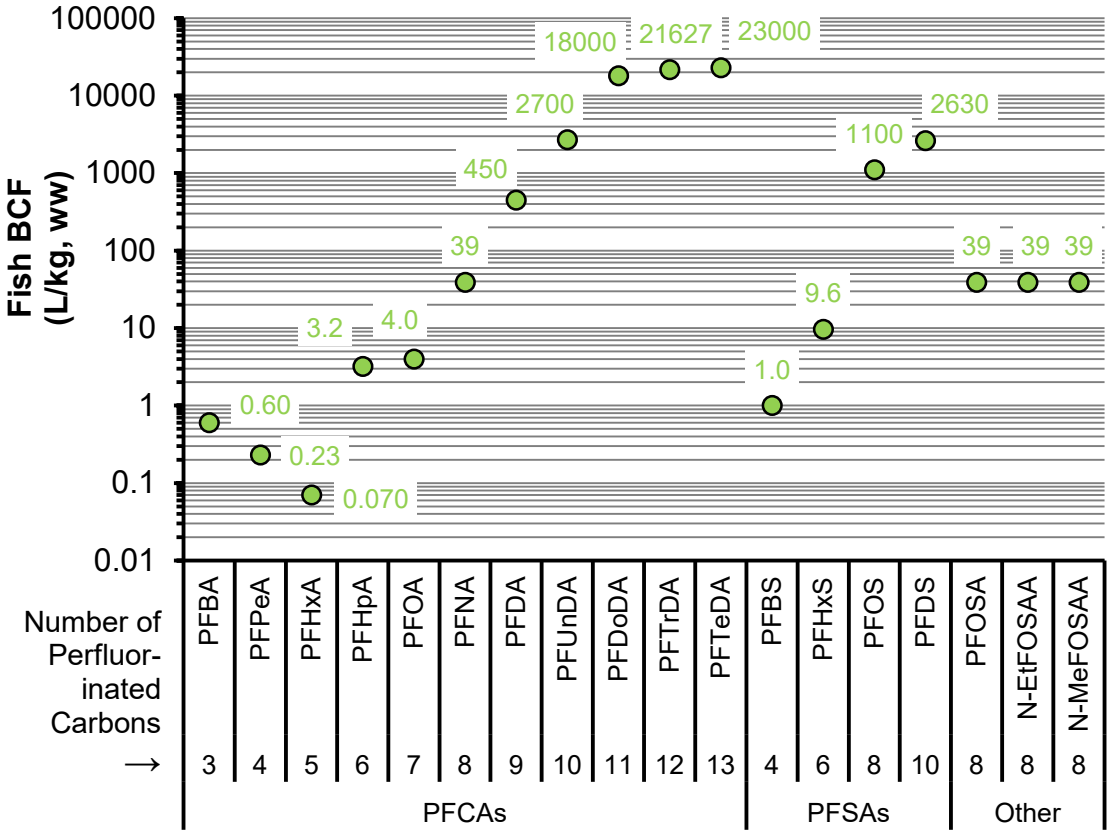

Figure 7: Recommended Water to Fish Tissues BCF Values

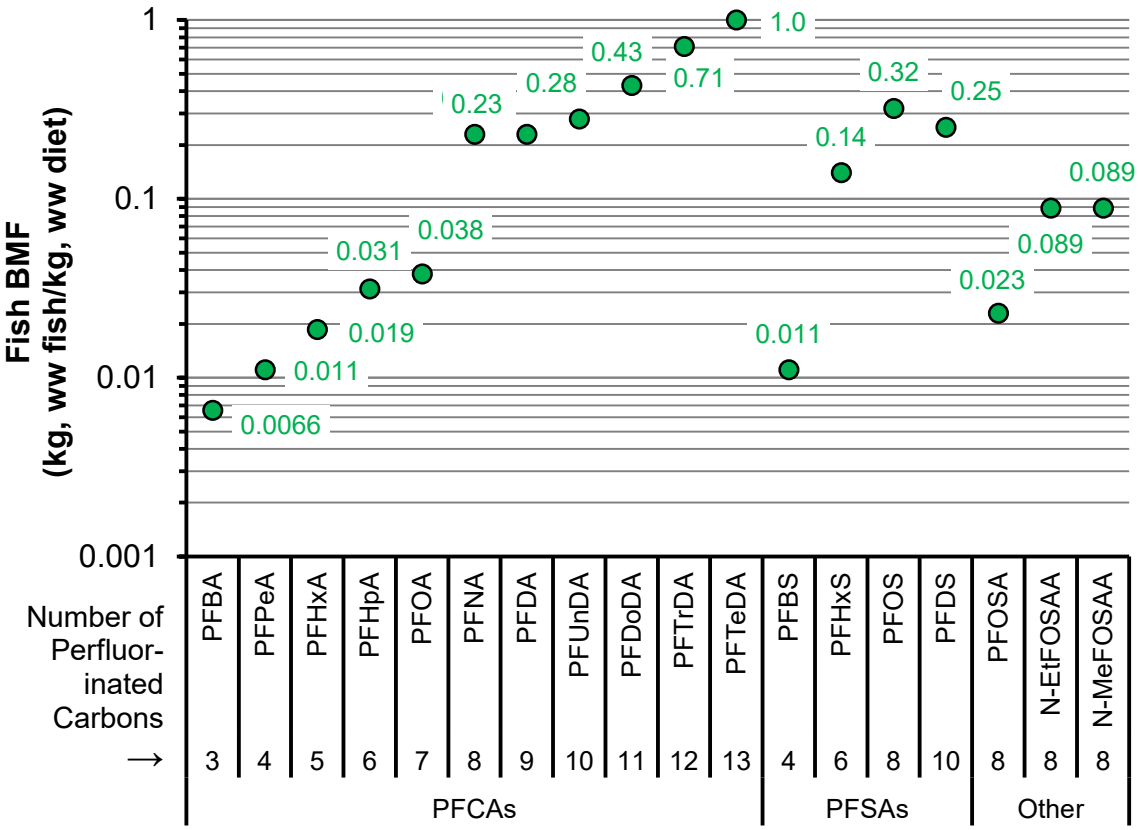

Figure 8: Recommended Diet to Fish Tissue BMF Values

Overall, bioaccumulation in fish tends to increase with increasing perfluorinated carbon chain length among the PFCAs and PFSAs. For PFCAs, BCF values for bioconcentration in fish appear to plateau at a chain length of approximately 11 (Figure 7), although this plateau was not observed for biomagnification from food (BMF values, Figure 8). In contrast, bioconcentration of PFSAs increased from perfluorinated carbon chain lengths of 4 to 10, although biomagnification for the longest PFSA (chain length of 10, PFDS) was lower than that of the PFSA with a chain length of 8 (PFOS). For PFAAs of the same perfluorinated carbon chain length, bioaccumulation values for PFSAs appear to be higher than for PFCAs, especially for bioconcentration.

Terrestrial Plant Bioaccumulation Factor (Terrestrial Plant BAF): Recommended values to predict bioaccumulation of PFAS from soil to terrestrial plants (Soil to Terrestrial Plant BAF values) are provided in Table 2e and Figure 9. There were several available studies that evaluated several different types of plant tissues (e.g., leaves, fruits, roots, shoots). For the purposes of ecological risk assessment, above-ground plant tissues such as leaves and shoots were considered the most applicable plant tissue with which to derive a bioaccumulation metric, as these tissues are likely to comprise a high proportion of most herbivorous wildlife species' diets. Additionally, the

selection of leaves is likely to result in conservative estimates of exposure, as the shorter-chain PFCAs that are predominantly accumulated by plants generally accumulate in the leaves, resulting in the highest levels of PFAAs compared to other tissues. Longer chain PFCAs are accumulated by plants, albeit at lower levels, and these tend to accumulate in root tissues (Blaine et al., 2014). In cases where an ecological risk assessment is considering a herbivorous species that consumes primarily fruit or roots, it is possible to calculate BAFs using fruit or root tissues using information available in several of the references included in Appendix B.

Most of the selected BAF values in Table 2e are from Zhao et al. (2014), who evaluated the uptake of PFAS from spiked soils into wheat. These BAF values are based on the above-ground wheat tissues (leaf plus shoot tissues). Additional soil to terrestrial plant BAFs were selected from Blaine et al. (2013), a laboratory study with lettuce grown in soils amended with PFAS-impacted biosolids. For PFAS measured by both studies, the Blaine et al. (2013) BAF values were lower/less conservative than those measured by Zhao et al. (2014), potentially reflecting higher availability of the spiked PFAS (relative to the biosolids-sourced PFAS) and/or species differences. Values for four PFAS (PFTrDA, PFTeDA, N-EtFOSAA, and N-MeFOSAA) were not identified.

Overall, bioaccumulation in aboveground tissues of terrestrial plants tends to decrease with increasing perfluorinated carbon chain length among the PFCAs and PFSAs. For PFAAs of the same perfluorinated carbon chain length, bioaccumulation values for PFCAs appear to be slightly higher than or similar to those of PFSAs.

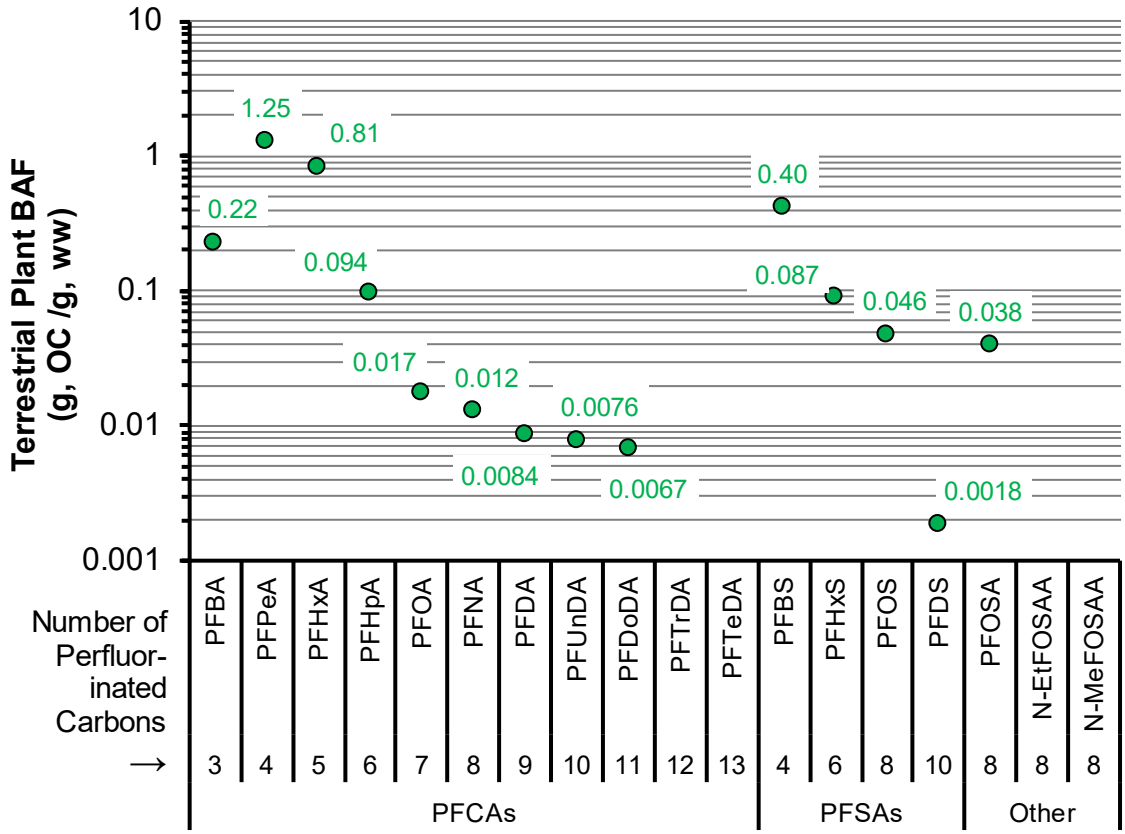

Figure 9: Recommended Soil to Terrestrial Plant BAF Values

Aquatic Plant Bioconcentration Factor (BCF-AP): Recommended values to predict bioaccumulation of PFAS from water to aquatic plants (Water to Aquatic Plant BCF values) are provided in Table 2f and Figure 10. All of the values are recommended from Pi et al. (2017). In this study, *Eichhornia crassipes* (a free-floating macrophyte) were exposed to PFAS in water in a mesocosm study. Unlike bioaccumulation in the aboveground tissues of terrestrial plants, bioaccumulation in aquatic plants tends to increase with increasing perfluorinated carbon chain length among the PFCAs and PFSAs. For PFAAs of the same perfluorinated carbon chain length, bioaccumulation values for PFSAs appears to be slightly higher than or similar to those of PFCAs.

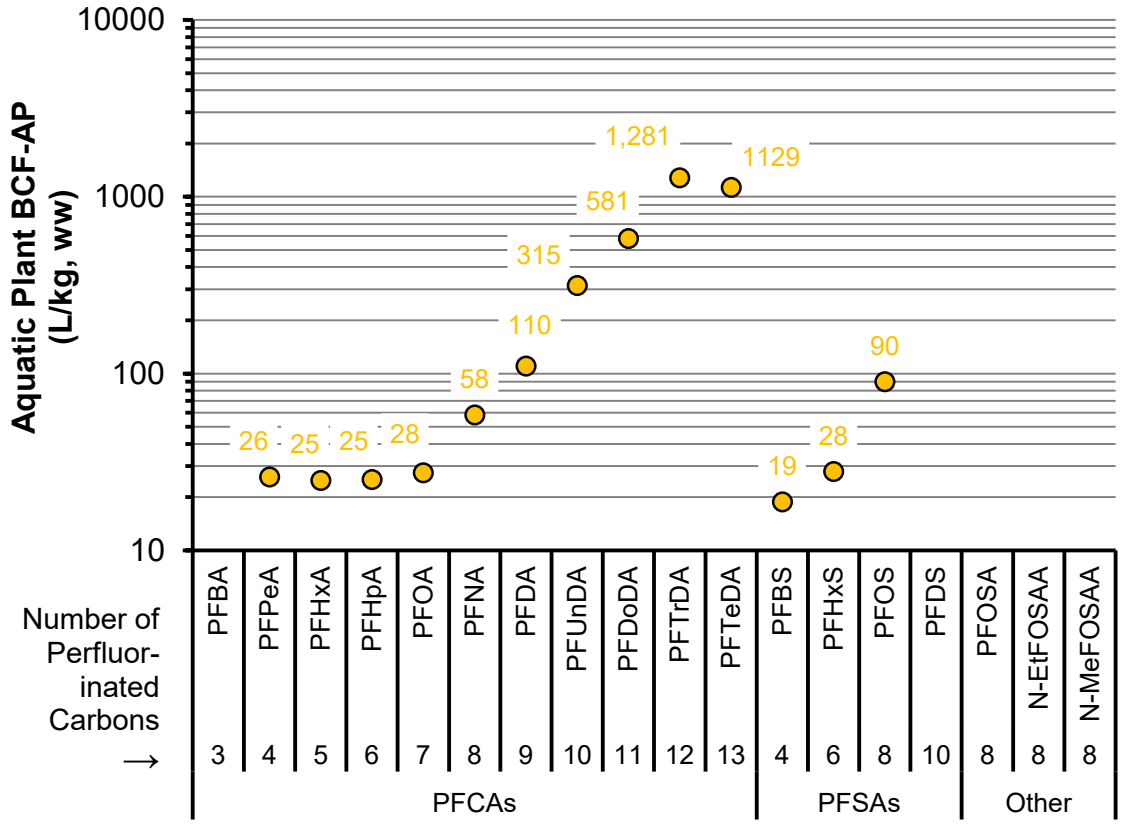

Figure 10: Recommended Water to Aquatic Plant BCF Values

Overview of T&E Species Exposure Assessment: Key Points

- Smaller mammals and birds, with small home ranges, are key wildlife species exposed to PFAS at AFFF sites, and their exposures to PFAS in diet items and incidental soil/sediment ingestion can be evaluated using traditional ecological food web modeling.
- Analysis of PFAS in soil, sediment, water, as well as organic carbon content in soil is recommended to evaluate site-specific wildlife, aquatic life, and terrestrial life exposures.
- As a first step in exposure assessment, PFAS in terrestrial and aquatic food webs can be predicted using empirical bioaccumulation modeling using the recommended approach and values provided.
- As a second step (or concurrent with collection of abiotic samples), tissue samples (wildlife diet items) can also be analyzed for PFAS and used in exposure assessment.

3.4 T&E Species Effects Assessment

The effects or toxicity assessment step in an ERA provides a description of the relationship between a dose of a chemical and the potential likelihood of an adverse health effect. This section provides guidance on the selection of assessment endpoints (i.e., acceptable level of effects) for T&E species; a summary of the toxicity data available in the literature for mammals, birds, aquatic life, and terrestrial plants and invertebrates; the selection and recommendation of wildlife TRVs; and the calculation of T&E specific benchmarks for aquatic life.

3.4.1 Selection of Assessment Endpoints

The selection of an appropriate TRV or benchmark requires the determination of the appropriate assessment endpoint – explicit expressions of the actual environmental values (e.g., ecological resources) that are to be protected. Assessment endpoints focus the risk assessment design and analysis; therefore, appropriate selection and definition of these endpoints are critical to the utility of a risk assessment (USEPA 1997). Generally, for ERAs focused on non-T&E species, assessment endpoints reflect ecosystem function and the sustain structure and function of specific ecological communities. As a result, specific measurement endpoints focus on evaluating key endpoints that relate to overall community function such as growth, reproduction, and development.

The key aspects of selecting a benchmark to meet the assessment endpoint include understanding the magnitude and proportion of an effect from a toxicity study (Suter, 2018). There is often an acceptable level of minor impact without resulting in an ecologically significant population-level or community-level impact. For example, up to a 20% decrease in growth or reproductive output can be considered potentially acceptable, based on the understanding that natural variability and resilience in populations can tolerate a low level of adverse impact to some individuals or at a low magnitude, such that the population or community function will not be affected (Suter et al., 2000). For example, a reported lowest observed effect concentration (LOEC) could relate to a 5% decrease in growth to 15% of a population (not an ecologically significant effect and would meet the assessment endpoint of community protection) or a 50% decrease in growth to 80% of a population (an ecologically significant effect that would not meet the assessment endpoint of community protection).

As it relates to T&E species, the assessment endpoints outlined in the U.S. ESA and are far more stringent than typical assessment endpoints in ERAs. For T&E species, the U.S. ESA assessment endpoint is protection of the individual T&E organism and its critical habitat. These assessment endpoints can be challenging to quantify within ecotoxicology and ecological risk assessment paradigms. This guidance focuses on the application of no observed effect levels (NOEL) or no observed adverse effect levels (NOAEL) for this purpose. These values are derived from laboratory toxicity studies in which standard (non-T&E) test organisms are exposed to a range of chemical dose levels, including a control (zero dose) level. Typically, the highest dose level in which organisms exhibit a lack of a statistically significant difference in effect from controls can be considered NOEL. The next highest dose level (which does exhibit a statistically significant difference in effect from controls) is the LOEL. This approach is conservative and does not take into account the magnitude of the effect. For example, it is possible for a study to identify a LOEL dose that elicits a 5% adverse effect, which would not necessarily be considered ecologically meaningful to populations and communities given the 20% effect threshold discussed above. In this case, the LOEL could be considered a NOAEL if the next highest dose level resulted in greater than a 20% effect (i.e., the LOAEL). However, due to the uncertainty of a 20% effect on the individual level focus for T&E species, it was considered that dose levels that exhibit any statistically significant difference from controls could be associated with a potentially meaningful dose level for a T&E species. Therefore, NOELs/NOECs were used in this guidance to identify doses for T&E species. As discussed in Section 2.1, this approach is consistent with ERAs for T&E species and associated guidance documents (USEPA, 1997, 1999). It should be noted that NOEL- and LOEL-based approaches are encumbered with uncertainty and many functional issues (Landis and Chapman, 2011). Benchmark-dose modeling and the identification of a benchmark-dose level is a robust method to quantify specific magnitudes of effects during toxicity studies, and Site managers are encouraged to consider benchmark-dose levels where available and appropriate. However, at this time, the majority of toxicity tests report NOAEL and LOAEL values and the number of doses in many studies may not be optimal for modeling, limiting the availability of the benchmark-dose level approach in ERAs at this time. Examination of dose response curves when presented in literature and communication of the magnitude of expected effects at the

predicted exposure doses is recommended for communication of ecological risk assessments, as discussed below.

The general framework outlined in this guidance is applicable to population and community level assessments for non-T&E species at sites that would select NOAEL- or LOAEL-based TRVs. And in some cases, risk managers may even wish to consider alternate risk assessment approaches for T&E species that use NOAEL- and LOAEL-based TRVs (or other dose benchmarks) in consideration of balancing the management of predicted chemical risks to T&E species with the risk of remedy. For example, protection of critical habitat is an important regulatory requirement to balance in the risk assessment and management process. Risk management of chemical exposure can often contemplate the active management of soils or aquatic sediments, and actions often result in destruction of habitat that, in some cases, is permanent despite attempts at post-remedy restoration. At a minimum, active management can result in habitat alteration and short-term impacts to species. It is uncertain how long-term chemical risk reduction outweighs these considerations at many contaminated sites. Risk management decisions are highly site-specific, and recommendations are out of scope for this guidance, but users are encouraged to consider the costs and benefits of remediation alternatives and balance the potential for adverse risks for chemical exposure and direct impacts from habitat destruction that is associated with many current active remedial technologies.

3.4.2 Selection Process for Recommended Wildlife (Avian and Mammalian) Effects Values for PFAS

The regulatory and peer-reviewed literature was reviewed for both mammalian and avian toxicity studies for the various PFAS included in Table 1. All reviewed studies are provided in Appendix C. For each study, the key parameters (species, test duration, measurement endpoint, and ecological endpoint) are presented, and the PFAS evaluated are provided. Each wildlife toxicity study was reviewed and scored on a 10-points scoring system (Table 3), which was modified from approaches used in USEPA (2005) to help identify TRVs for use in USEPA's Ecological Soil Screening Levels (EcoSSLs). EcoSSLs are currently applied in risk assessments for both common and T&E species in the US.

From these studies, recommended TRVs (mg/kg-day) were selected for use in T&E ERAs, considering the following study aspects:

- Overall score (based on the scoring system detailed in Table 3);
- General level of regulatory acceptance (i.e., studies that have been supported by USEPA for use in other guidance documents such as the Lifetime Health Advisory Levels for PFOS and PFOA);
- Duration of exposure (generally prioritizing chronic studies over subchronic and acute studies to best represent long-term exposures of resident species[4]);

[4] Sub-chronic or acute studies may be preferable for characterizing exposures, such as in the case of short-term exposures, such as sporadic Site inhabitation by migratory wildlife.

- Endpoint (prioritizing ecologically significant endpoints consistent with USEPA guidance [USEPA 2005] such as growth[5], reproduction and lethality); and
- Magnitude of the TRV (lower, more protective values from robust studies were prioritized for selection).

3.4.3 Recommended Wildlife (Avian and Mammalian) Effects Values for PFAS

Recommended TRVs for mammals and avian are presented in Tables 4 and 5, respectively. The NOEL and LOEL values for mammals are presented on Figure 11 (avian TRVs were limited to PFOS and PFBS and a figure is not presented). Detailed discussion of each selected TRV is provided below.

Recommended Mammalian TRVs:

- The mammalian TRV for PFBA was based on a study by van Otterdiijk (2007b), who reported an unbounded NOEL value of 30 mg/kg-day, associated with a lack of statistically significant differences in growth (body weight) over a 90-day exposure period with Sprague Dawley rats. This study was one of the two highest scoring studies. The other study was for a shorter duration and indicated similar results for the NOEL, which was among the lowest values for the studies reviewed.

- The mammalian TRV for PFHxA was based on a study by Klaunig et al. (2015), who reported a NOEL value of 30 mg/kg-day, associated with a lack of statistically significant differences in survival over a 728-day exposure period with Sprague Dawley rats (no significant effects were detected for growth (body weight) at the highest dosing level of 200 mg/kg-day). This study was the highest scoring study, was conducted for the longest duration, and indicated the lowest NOEL. The next highest exposure level in the Klaunig et al. (2015) study, 200 mg/kg-day, was associated with a statistically significant difference in survival compared to controls in latter stages of the study (when animals were succumbing to mortalities associated with old age). The difference between survival of the controls and animals at the 200 mg/kg-day dose level was slight (36% of control animals surviving to this life stage, versus 22% of 200 mg/kg-day dosed animals). It is uncertain if the additional 14% difference in survivorship (a relative difference of approximately 40% between controls and the LOEL) at later stages in the lifespan would result in an ecologically significant effect on populations. The 200 mg/kg-day effect level may be considered as a potentially relevant TRV for some risk assessments.

- The mammalian TRV for PFOA was based on a study by Butenhoff et al. (2012), who reported a NOEL value of 1.3 mg/kg-day, associated with a lack of statistically significant

[5] Growth has typically been considered an ecologically relevant endpoint on the presumptions that limited growth may impair overall reproductive fitness, result in less favorable interactions with predators or other abiotic stressors, and other aspects that may manifest in significant effects to local populations or communities. However, there are instances a smaller size can be advantageous to fitness.

differences in growth (body weight) over a 730-day exposure period with Sprague Dawley rats. This study scored highly with respect to the evaluation and is consistent with other NOEL values for PFOS (ranging from approximately 1 to 3 mg/kg-day) in Appendix C. The Butenhoff et al. (2012) value is similar to the 1 mg/kg-day LOAEL value for developmental effects in rats (reduced ossification of the proximal phalanges (forelimb and hindlimb)), as described in Lau et al. (2006), which was selected by USEPA to develop the human health reference dose (USEPA, 2016). The Butenhoff et al. (2012) was selected over the Lau et al. (2006) value because of the higher certainty in ecological relevance of the growth endpoint. The next highest exposure level in the Butenhoff et al. (2012) study, 14 mg/kg-day, resulted in an approximate 10% decrease in growth compared to controls in latter stages of the study (when animals were at their heaviest). This effect level is below the commonly accepted 20% level of effects for populations (Suter et al., 2000) and may be considered as a potentially relevant TRV for some risk assessments.

- The mammalian TRV for PFNA was based on a study by Wolf et al. (2010), who reported a NOEL value of 0.83 mg/kg-day, associated with a lack of statistically significant differences in reproduction (number of live pups at birth) for a 18-day exposure period with pregnant mice. This study scored the highest with respect to the TRV evaluation, and the NOEL value is lower than other NOEL values from other studies (Appendix C). The next highest exposure level in the Wolf et al. (2010) study, 1.1 mg/kg-day, resulted in an approximate 46% reduction in the number of live pups at birth, compared to controls.

- The mammalian TRV for PFDA was based on a study by Harris and Birnbaum (1989). A NOEL value of 0.3 mg/kg-day, associated with a lack of statistically significant differences in growth (fetal body weight per litter) for a 10-day exposure period (18-day study) with pregnant mice. A lower dose (0.1 mg/kg-day) resulted in a higher 0.9% reduction in fetal body weight per litter (compared to the 0.3 mg/kg-day dose) that was reported to differ statistically from the controls by the study authors. Given the lack of dose response and very slight level of the effect, the 0.1 mg/kg-day was considered a no-effect result; this interpretation was also made by ATSDR (2018). The Harris and Birnbaum (1989) study scored equivalent to another available study on PFDA with respect to the TRV evaluation, and the NOEL value from this study is lower than other NOEL value (Appendix C). The next highest exposure level in the Harris and Birnbaum (1989), 1 mg/kg-day, was statistically significantly different from controls, and resulted in an approximate 4% reduction in fetal body weight per litter. This effect level is very low and is below the commonly accepted 20% level of effects for populations (Suter et al., 2000). Higher dosing levels in the study, 3, 6.4, and 12.8 mg/kg-day, resulted in effects (compared to controls) of 6%, 23%, and 50%, respectively. The 3 and 6.4 mg/kg-day values may be appropriate as NOAEL and LOAEL TRVs for some risk assessments.

- The mammalian TRV for PFUnDA was based on a study by Takahashi et al. (2014), who reported a NOEL value of 0.3 mg/kg-day, associated with a lack of statistically significant differences in growth (body weight in adults and pups) for a 42-day exposure period with

rats. This was the only candidate study evaluated. The next highest exposure level in the Takahashi et al. (2014) study, 1 mg/kg-day, resulted in a statistically significantly difference in reductions in the body weights of pups of 13% to 19% relative to controls.

- The mammalian TRV for PFDoA was based on a study by Kato et al. (2015). A NOEL value of 0.5 mg/kg-day was derived from this study, associated with a lack of statistically significant differences in growth of adult rats (body weight) and pups (body weight) for a 42-day exposure period with rats. This study was the highest scoring study and produced the lowest NOEL. The next highest exposure level in the Kato et al. (2015) study, 2.5 mg/kg-day, was statistically significantly different from controls, and resulted in reductions in the body weights of adults and pups of approximately 20 to 40% relative to controls.

- The mammalian TRV for PFTeDA was based on a study by Hirata-Koizumi et al. (2015), who reported a NOEL value of 3 mg/kg-day, associated with a lack of statistically significant differences in growth of adult rats (body weight) and pups (body weight) for a 42-day exposure period with rats. This was the only candidate study evaluated. The next highest exposure level in the Hirata-Koizumi et al. (2015) study, 10 mg/kg-day, was statistically significantly different from controls, and resulted in reductions in the body weights of adults of approximately 5% relative to controls, and a reduction in body weights of pups of 8 to 18% relative to controls.

- The mammalian TRV for PFBS was based on a study by Lieder et al. (2009b). A NOEL value of 300 mg/kg-day was derived from this study, associated with a lack of statistically significant differences in growth of adult rats (body weight) for a 120-day exposure period with Sprague Dawley rats. This study was the highest scoring study, produced the lowest NOEL, and was the longest duration (multi-generational study) of the studies evaluated. The next highest exposure level in the Lieder et al. (2009b) study, 1000 mg/kg-day, was statistically significantly different from controls, and resulted in reductions in the body weights of males of approximately 8% relative to controls. This effect level is very low and is below the commonly accepted 20% level of effects for populations (Suter et al., 2000).

- The mammalian TRV for PFHxS was based on a study by Chang et al. (2018). A NOEL value of 0.3 mg/kg-day was derived from this study, associated with a lack of statistically significant differences in reproduction (mean live litter size) for a 77-day exposure period with mice. This study was one of two highest scoring studies. The other study (Butenhoff et al., 2009) was of shorter duration and indicated a higher (unbounded) NOEL value (10 mg/kg-day) for growth (body weight) and reproductive (number of pups per litter) endpoints in Sprague Dawley rats. The next highest exposure level in the Chang et al. (2018) study, 1 mg/kg-day, was statistically significantly different from controls, and resulted in reductions in the litter size of approximately 14% relative to controls. This effect level is below the commonly accepted 20% level of effects for populations (Suter et al., 2000).

- The mammalian TRV for PFOS was based on a study by Luebker et al. (2005). A NOEL value of 0.1 mg/kg-day was derived from this study, associated with a lack of statistically significant differences in growth (body mass gains over the study) for an 84-day exposure period with rats. This study was one of many studies with PFOS. Although it scored slightly lower than other studies, the Luebker et al. (2005) NOEL value is the next-to-lowest NOEL value among those reviewed. Dong et al. (2009) indicated a slightly lower NOEL of 0.08 mg/kg-day for growth in mice in a shorter-duration study. The Luebker et al. (2005) study is also the basis for the USEPA reference dose for PFOS (USEPA, 2016). The next highest exposure level in the Luebker et al. (2005) study, 0.4 mg/kg-day, was statistically significantly different from controls, and resulted in reductions of 14% in body mass gains of relative to controls. This effect level is below the commonly accepted 20% level of effects for populations (Suter et al., 2000). The 1.6 mg/kg-day exposure level in the Luebker et al. (2005) study was statistically significantly different from controls and resulted in reductions in body mass gains of approximately 21% relative to controls. An approximate 20% level of effect for growth in mice was evident at the 0.83 mg/kg-day dose (Dong et al., 2009). These additional TRVs (0.4, 0.83, and 1.6 mg/kg-day) may be useful in risk assessments for PFOS.

- For PFPeA, PFHpA, PFTrDA, PFDS, PFSOA, N-EtFOSAA and N-MeFOSAA, no toxicity information was available to characterize effects to mammals. In the absence of information, it may be possible to use the TRV for PFOS (the lowest, most conservative TRV identified) or TRVs with similar perfluorocarbon chain lengths as potential surrogate TRVs. These approaches are highly uncertain but may be an option for T&E species assessments at some sites with significant exposures of these uncharacterized PFAS. Additionally, exposure to other polyfluoroalkyl substances that may be PFAA precursors can occur. Little information is available on the toxicity of these compounds or on the rate at which precursors degrade to PFAAs for which toxicity data are available. As a significant data uncertainty, the incorporation of PFAA precursors into ecological risk modeling is discussed further in the uncertainty section below (Section 4.1).

Overall, the mammalian TRVs suggest that toxicity is higher for PFAAs with longer perfluorinated carbon chain lengths. For PFAAs of the same perfluorinated carbon chain length, it is difficult to discern whether PFSAs and PFCAs differ in toxicity. PFOS exhibits the highest toxicity.

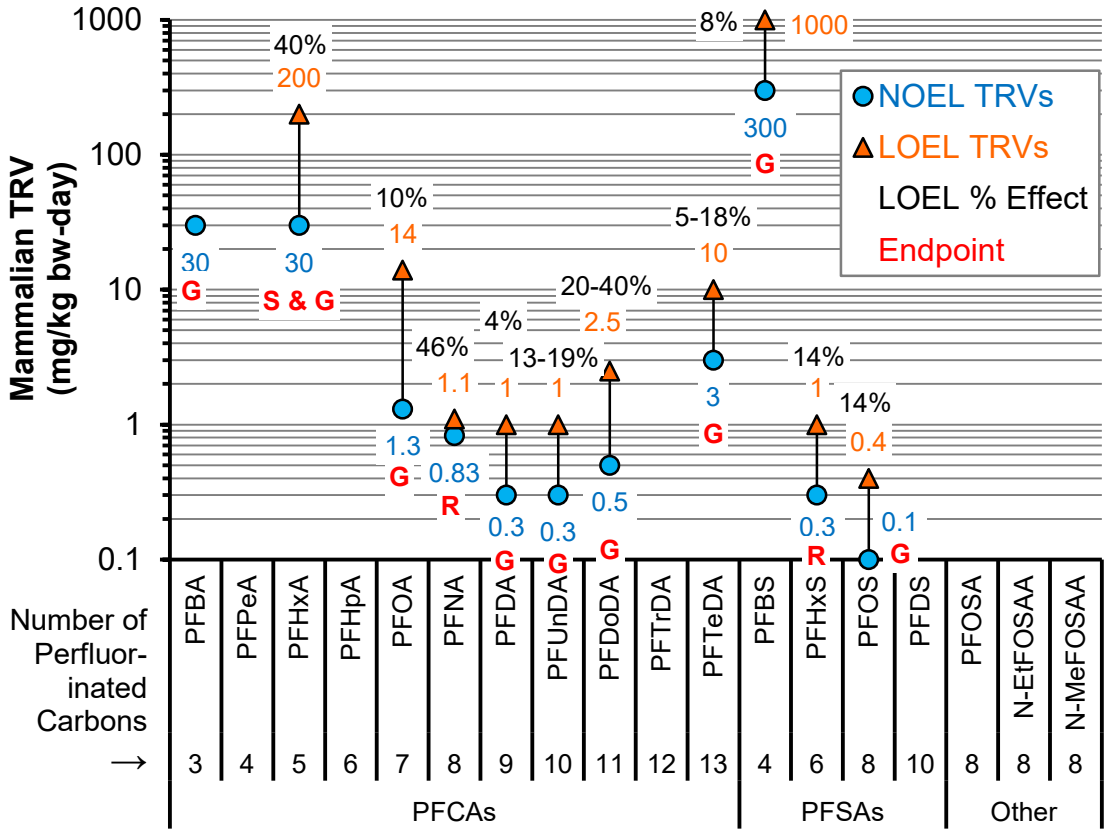

Figure 11: Recommended NOEL and LOEL Values for Mammals for various PFAS. Endpoints are based on growth (G), survival (S) or reproduction (R).

Recommended Avian TRVs:

- The avian TRV for PFBS was based on a study by Gallagher et al. (2005), as detailed in Newsted et al. (2008), who reported an unbounded NOEL value of 88 mg/kg-day, associated with a lack of statistically significant differences in growth (body weight) over a 147-day exposure period with northern bobwhite quail (Colinus virginianus). This study scored highly with respect to the evaluation and is the only source of sublethal toxicity information identified in Appendix C-2.

- The avian TRV for PFOS was based on a study detailed by Newsted et al. (2005, 2007), who reported an unbounded LOEL value of 0.77 mg/kg-day, associated with "less than 20% for the affected reproductive endpoints," with effects including testis size (length) and survivorship of hatchlings relative to number of eggs set over a 147-day exposure period with northern bobwhite quail (Colinus virginianus). The next highest exposure level in the Newsted et al. (2007) study, 2.64 mg/kg-day, resulted in lethality in adult birds (approximately 16% of the birds in the study), and this was considered sufficiently significant to discontinue exposures at this dose level. This study scored highly with

respect to the evaluation and is the longest ecologically relevant sublethal toxicity information identified in Appendix C-2. The effect associated with the 0.77 mg/kg-day LOEL (14-day old survivors/eggs set) was 17% lower than the controls, below the commonly accepted 20% level of effects for populations (Suter et al., 2000). Interpretations may vary in application of the TRVs when true NOELs are unavailable, however. For example, Giesy et al (2010) applied a lowest-effect-to-no-effect Uncertainty Factor (UF) of 2 to the 0.77 mg/kg-day TRV, resulting in a TRV of 0.39 mg/kg-day (additional uncertainty factors were also applied by Giesy et al (2010) to account for inter-taxon extrapolation and exposure durations). The application of UFs remains controversial (Allard et al., 2009) and varies among regulatory jurisdictions. The recommended (LOEL) avian TRV for PFOS is 0.77 mg/kg-day based on a statistically significant, but less than 20% effect level observed in northern bobwhite quail. Users may consider the application of an UF for T&E avian species, though the use of this LOEL is considered appropriate for non-T&E species based on the magnitude of effect.

- It is possible to derive a semi-quantitative TRV for PFOA and PFDA from Yeung et al. (2009). This study exposed one-day old male chicks (Gallus gallus) to two doses plus a control for three weeks. The two doses were comprised of a mixture of PFOA, PFDA and PFOS: a high dose (1 mg/kg bw-day of each of the three PFAS, equivalent to 3 mg total PFAS mg/kg-day) and a low dose (0.1 mg/kg-day of each of the three PFAS, equivalent to 0.3 mg total PFAS mg/kg-day). No significant effects on growth, organ weight or measured histological and plasma biochemical parameters was observed. Typically, TRVs are derived from single-chemical exposures to enable single-chemical exposure and effects modeling in ERAs. Given the absence of PFOA and PFDA avian toxicity test information from the literature, a semi-quantitative, high-uncertainty NOEL TRV based on the lack of effects in the 1 mg/kg-day dose group could be considered for PFOA and PFDA (this TRV is not recommended for PFOS, as a more robust TRV is available). Assuming mixture antagonism did not reduce the potency of the three-PFAS mixture in this study, it is likely that single-PFOA or single-PFDA exposures more than 1 mg/kg-day would result in a lack of adverse effects as the 1 mg/kg-day NOEL reflects combined exposure of the three PFAS. Until single-chemical TRVs are available, the TRVs of 1 mg/kg bw-day for PFOA and 1 mg/kg bw-day for PFDA could be considered for use in ERAs, but should be used with extreme caution, and risk assessors should clearly communicate the high uncertainty associated with conclusions based on their application.

- Avian TRVs for other PFAS were not identified. In the absence of information, it may be possible to use the TRV for PFOS (the lowest, most conservative TRV identified) a potential surrogate TRV. This approach is highly uncertain. TRVs based on concentrations of PFAS in eggs are available for PFHxA, PFOA, and PFHxS (Table 5), but are only useful if egg samples from the site of interest are available. Additionally, similar to mammals, exposure to PFAA precursors for which little toxicity information is available is likely to occur at AFFF impacted sites. The lack of avian toxicity information for PFAA precursors is addressed below in the uncertainty section (Section 4.1).

It should be acknowledged that the TRVs selected here are based on ecologically significant effects of growth, reproduction, and lethality, following the approach used for derivation of TRVs by USEPA in EcoSSLs (USEPA, 2005). EcoSSLs and the TRVs upon which they are derived are applied in risk-based decision making for ecological risks of common and T&E species. Growth, reproduction, and lethality have typically been considered primary ecologically relevant endpoints. Additionally, there may be other effect endpoints that may be considered ecologically relevant for some species, such as lethargy or significant behavioral changes. TRV development for particular sites should specify assessment and management endpoints and these other potentially adverse endpoints (aside from growth, reproduction, and lethality) may need to be considered (Allard et al., 2009). For example, McCarthy et al. (2017) reviewed a variety of additional endpoints for development, liver function, sexual maturation and other endpoints (in addition to growth, reproduction, and lethality) for PFOA and PFOS and noted no-effect and lowest effect TRV ranging from 1 to 30 mg/kg-day and 0.1 to 25 mg/kg-day, respectively. This range is consistent with our NOEL to LOEL range of 1.3 to 14 mg/kg-day for PFOA and 0.1 to 0.4 mg/kg-day for PFOS (reproduction and growth endpoints, Table 4). In contrast, McCarthy et al. (2017) noted that effects on the liver were found for PFNA (low-effect TRV of 0.1 mg/kg-day) and PFBS (100-300 mg/kg-day) at levels that were lower than the TRV ranges we noted for reproductive and growth effects (0.8-1.1 mg/kg-day for PFNA and 300-1000 mg/kg-day for PFBS). With regards to PFAS, the liver is considered a target organ for PFAS accumulation and effects, and field studies at PFAS-impacted sites are needed to confirm the potential linkages between "first-order" organ-level effects and "second order" adverse effects to individual and population health.

For ecological risk assessments that must proceed in the absence of such research, it should be carefully considered whether "first-order" organ-level effects should apply to truly adverse effects on the scale of individual organisms or populations (Tannenbaum, 2004). TRVs associated with these "first order" toxicological effects (changes in organ weights, biochemical levels, physiology, etc.) can be lower and more attractive for site stakeholders because they result in more conservative risk assessments. However, risk managers should balance this conservatism (and potential toxicological disconnect) of these assessments with the potential adverse impacts associated with active remediation, especially in the case of intact and functioning habitats that may be difficult to restore following actions to address the potential impacts of PFAS.

3.4.4 Aquatic Life Risk Assessment Approaches

3.4.4.1 Selection Process for Recommended Pelagic Aquatic Life Effects Values for PFAS

Considerable research has been performed and reviewed to understand the aquatic toxicity of PFOS and other PFAS to pelagic/water column species (Beach et al., 2006; Ahrens, 2011; McCarthy et al., 2017). The larval stages of midges (small flies) appear to be the most sensitive aquatic species tested for exposure to PFOS, with decreased development rates observed below 2.3 µg/L (MacDonald et al., 2004). Recently, the use of SSDs[6] have indicated that adverse effects

[6] A species sensitivity distribution is the cumulative distribution function of multiple toxicological data points for various species. The most sensitive species are in the lowest percentiles of the distribution, and guidelines can be

to the majority (95%) of aquatic species are not expected in freshwater systems below approximately 5 µg/L for PFOS (Arblaster et al., 2017; Environmental Canada, 2017; Giesy et al., 2010), or below 220 µg/L for PFOA and 2,400 µg/L for PFBS (CRC CARE, 2017; Giesy et al., 2010). A review of aquatic toxicity test results on select marine species has indicated a lack of adverse effects below 15 µg/L for PFOS, and 1,500 µg/L for PFOA (Mhadhbi et al., 2012). Similarly, SSDs on marine species indicate that adverse effects to the majority (95%) of species are not expected below 8 µg/L for PFOS or below 9 µg/L for PFOA (CRC CARE, 2017).

The regulatory and peer-reviewed literature was extensively reviewed for information on effects to aquatic life following exposure to PFAS. The majority of peer-reviewed literature and regulatory environmental quality benchmarks have been developed for PFOS and PFOA; however, other select PFAAs have been included in aquatic life evaluations thus far. A summary of papers evaluating adverse effects to aquatic life from PFAS are provided in Appendix D-1. For wildlife TRVs, endpoints included for consideration were limited to growth, reproduction and survival consistent with USEPA guidance on wildlife TRVs (USEPA, 2005). However, aquatic organisms can be exposed to chemicals in surface water during developmental stages, and developmental endpoints are often used in aquatic toxicity testing. Therefore, developmental endpoints with clear links to adult survival (i.e., shell development for crustaceans, normal larval development) were included in this evaluation but preference was given to survival and growth endpoints where studies presented both types of information.

Site risk assessors are encouraged to review Appendix D-1 for surrogate species for specific aquatic T&E species, as toxicity benchmarks for aquatic life are highly variable. However, for cases where toxicological info for PFOS or PFOA is not available for a specific T&E species or surrogate species, a NOEC-based SSD has been developed for PFOS and PFOA to calculate T&E species protective values.

SSDs provide an approach for determining concentrations of a chemical that are protective of multiple species of varying sensitivities and are a commonly used approached for deriving aquatic life benchmarks (USEPA, 2010; Posthuma et al., 2002). Raimondo et al. (2008) showed that the 1% and 5% effect concentrations derived by SSDs using acute lethal toxicity data were below 99.5% and 97% of all endangered species effect levels, respectively, indicating that the use of SSDs as distribution-based risk assessment and criteria development approaches can generally be protective of T&E species. To calculate 1% and 5% effect concentrations that are protective of T&E species, acute and chronic NOEC values for PFOS and PFOA for USA resident species from the literature were compiled for freshwater and marine aquatic organism (Appendix D-2).

It should be noted that while non-resident species are included in Appendix D for informational purposes, they were not included in the development of SSDs per USEPA guidance (USEPA, 2010). Non-resident species in some studies have indicated a higher level of sensitivity to PFOS

developed for the protection of most species (i.e., 95%) by selecting a concentration equal to a conservative percentile (i.e., 5%) generally referred to as Hazardous Concentration 5% or HC5. Details on supporting theory and rationale are provided elsewhere: USEPA, 2010, Posthuma et al., 2002.

or PFOA than the most sensitive resident species. Studies with zebrafish (*Dario rerio*; Keiter et al., 2012) and Mediterranean mussel (*Mytilus galloprovincialis;* Fabbri et al., 2014) have indicated lower NOEC values than those previously observed for PFOS and PFOA. These organisms are not resident to waters of the United States and, therefore, would not be considered T&E species. Consideration of these studies for future research or site-specific considerations may be warranted.

Calculation of the 1% and 5% percentiles from the SSDs as 1% hazardous concentration (HC1) and HC5 values followed USEPA (2010) guidance. Acute and chronic NOECs or a concentration resulting in a 10% effect (EC10) were selected for the SSDs and compiled for freshwater and marine species separately. Only NOEC and EC10 values were considered to reflect the level of protection required for T&E species; however, these values can be applied more widely (i.e., at a site with non-T&E aquatic life exposures). Site risk assessors dealing with non-T&E species may prefer to select a NOEC/LOEC value more applicable to their site based on species and assessment endpoint or a less conservative benchmark from the NOEC-only based SSD (for example, 20% threshold based on NOEC values may be appropriate for non-T&E species).

Acute values were converted to chronic values using mean acute-to-chronic ratios derived from Giesy et al. (2010). Studies of 48 to 96 hours in length were typically considered acute, except for tests covering critical life stages and tests on single-celled organisms which were considered chronic (USEPA, 2010). Chronic studies covered most of the organism life cycle or critical life-stages (Suter and Tsao, 1996). As noted by Salice et al. (2018), there are limited chronic studies for PFAS currently, which adds considerable uncertainty due to the persistence of these compounds and the expectation that multi-generation exposures are likely to occur at AFFF-impacted sites. While Salice et al. (2018) opted to include only chronic studies, it was felt that the application of the acute-to-chronic ratio would be an appropriate mechanism to include acute data, which allowed for a wider inclusion of species.

Consistent with USEPA (2010) guidance, for each species with multiple endpoints, the geometric mean of NOEC values was calculated for the species' mean value. The geometric mean of multiple species within the same genus was then calculated for the genus mean values. Genus mean values were then ordered from lowest to highest, assigned ranks, and the cumulative probability was calculated for each genus mean value, and lastly the HC1 and HC5 were calculated using the equations described in USEPA (2010).

For other PFAS, too few studies are available to calculate an SSD. Therefore, is it recommended that site risk assessors review Appendix D-1 for freshwater and marine aquatic toxicity studies and select a study that best reflects the PFAS and species of concern at a site. Recommended values are not provided here, as the test species are more variable than those used in wildlife toxicity testing, and a site-specific selection is recommended. Where no studies are available for a particular PFAS of concern, it is generally recommended that the toxicity benchmark value for PFOS be considered as a highly conservative surrogate, or the toxicity benchmark value for another PFAS with a similar perfluorinated carbon chain length be considered.

3.4.4.2 Recommended Pelagic Aquatic Life Effects Values for PFAS

The distributions of NOEC genus mean values for freshwater and marine organisms following exposure to PFOS and PFOA are provided in Appendix D-3. From these distributions, the HC1 and HC5 values for T&E species were calculated generally following USEPA (2010) methods as previously described in Section 3.4.4.1 and are recommended for evaluating the potential for risks to T&E aquatic life species from PFOS and PFOA (Table 6). SSDs were calculated for freshwater and marine organisms separately and for PFOS and PFOA; however, too few marine studies were available for PFOA to develop an SSD. These values are intended as conservative screening-level values and should be applied to indicate the need for additional site-specific or receptor-specific evaluation in cases where the levels are exceeded. Exceedance of these values do not necessarily imply adverse aquatic life effects are evident at a site. At sites that exhibit concentrations below these values, it is likely that no adverse effects on aquatic life are expected.

The calculated HC5 for PFOS is approximately 6 µg/L for freshwater species and 8 µg/L for marine species; with HC1 values of 0.5 µg/L and 2.6 µg/L, respectively. The freshwater HC5 for PFOS shows good agreement with other SSD-calculated HC5 benchmarks for PFOS, including those developed by Environment Canada (HC5 = 6.8 µg/L; Environment Canada, 2017), Giesy et al. (HC5 = 5.1 µg/L; Giesy et al., 2010), and Qi et al. (HC5 = 6.66 µg/L; Qi et al., 2011). The PFOS HC5 values for marine species show good agreement with values calculated by CRC CARE (2017) of 7.8 µg/L. The HC5 values calculated from an SSD are unique to that specific selection of data, therefore, variability is expected. Salice et al. (2018) prioritized chronic NOECs and included some non-resident species data that were more sensitive, resulting in a calculated HC5 of 1.12 µg/L. However, generally these values are converging on an HC5 in the 5 to 6 µg/L range, indicating good agreement between different studies and robust datasets.

The calculated HC5 and HC1 values for PFOA are approximately 1100 µg/L and 540 µg/L for freshwater species (marine data were insufficient to derive HC5 and HC1 values) and indicate PFOA is several orders of magnitude less toxic than PFOS. The freshwater HC5 value shows good agreement with an SSD-calculated aquatic life benchmark for PFOA developed by Giesy et al. (HC5 = 2900 µg/L; Giesy et al., 2010).

3.4.4.3 Recommended Benthic Effects Values for PFAS

The SSD and supporting tables developed for PFOS and PFOA exposures to aquatic life are representative of aquatic life exposed directly to PFAS in the water column. Benthic invertebrate organisms are also exposed via direct contact with sediment and sediment porewater. Potential adverse effects to sediment-dwelling (benthic) invertebrates from direct exposure to sediment has been minimally studied (McCarthy et al., 2017) with no published benchmarks. Potential adverse effects to terrestrial invertebrates from direct exposure to soils has been studied more frequently because of implications for land application of biosolids (McCarthy et al., 2017). The direct measurement of effects to benthic invertebrates via spiked-sediment assays was a distinct data gap during this evaluation, and no direct toxicity studies on benthic invertebrates were identified.

To evaluate the potential for adverse effects to benthic invertebrates, two approaches are recommended here.

1. PFAS, in particular PFOS and other long-chain sulfonates, partition into the organic carbon phase of sediments, which reduces bioavailability. Equilibrium-partitioning benchmarks have been calculated for sediments based on partitioning between water or sediment porewater and sediment. Using K_{OC} values (Table 2a) which describe the partitioning between sediment and water, and a concentration in surface water that has been determined protective of aquatic life (such as HC1 or HC5 values for surface water from Table 6), a corresponding protective concentration in sediments can be calculated, based on the assumption that benthic invertebrate species are of similar sensitivity to PFAS as pelagic species.

2. Site risk managers can apply an upper-bound estimate of no-effects following exposure to spiked sediments in a controlled laboratory setting that was not specifically designed to reflect a toxicity testing approach so long as the appropriate endpoints were assessed. For example, Higgins et al. (2007) exposed *Lumbriculus variegatus* to PFAS-spiked sediment to measure bioaccumulation between sediment and tissue. While not designed to be a toxicity study, and thus potentially lacking specific QA/QC steps, this study did also measure weight loss during exposure and, therefore, the highest concentrations where no adverse impact to growth could be selected as a benchmark, though with high uncertainty.

Both of these methods include a higher level of uncertainty due to the inherent assumptions needed for these calculations. In general, no studies were identified that indicated benthic invertebrates would be more sensitive than pelagic invertebrates, although research is needed.

3.4.5 T&E Terrestrial Plants and Invertebrates Risk Assessment Approaches

The regulatory and peer-reviewed literature was reviewed for information on effects to terrestrial plants and invertebrates following direct exposure to PFAS in soil. The majority of peer-reviewed literature and regulatory environmental quality benchmarks have been developed for PFOS and PFOA; however, many other PFAAs have been included in at least one toxicity assessment thus far. A summary of studies evaluating the adverse effects to terrestrial plants and invertebrates from PFAS is provided in Appendix E.

For terrestrial invertebrates, the majority of studies have been performed on earthworms (*Eisenia fetida*). Some studies have focused on non-earthworm species, such as bees, that may be important for T&E species consideration, but the exposure routes applied (oral and contact paper) make including this toxicity testing in ERAs challenging due to uncertainties relating contact- or oral-based toxicity information to soil-based values. Applying toxicity values for earthworms to specific terrestrial invertebrate T&E species has uncertainties due to the potential for interspecies differences; however, data for other species are sparse. Chronic studies that evaluated growth,

development, or reproductive endpoints were preferred for the recommended toxicity values (Table 7).

- The recommended toxicity value for PFOS, a NOEC of 80 mg/kg, is from Xu et al. (2013), where earthworms were exposed to PFOS-spiked artificial soils for 42 days, with growth evaluated as an endpoint.
- The recommended toxicity value for PFOA, a NOEC of 10 mg/kg, is from He et al. (2013), where earthworms were exposed to PFOA-spiked soils for 28 days, with growth evaluated as an endpoint.
- No toxicity specific studies were identified that evaluated additional PFAS, other than PFOS and PFOA. One study from Zhao et al. (2014) evaluated uptake of multiple PFAS into plants and terrestrial invertebrates and noted changes to growth during exposure. Similar to the approach for sediment noted above, this study could be used as a NOEC, as the highest test concentration did not result in statistically significant changes to growth compared to controls. However, the values from this study reflect unbounded NOECs (i.e., no effect at the highest test concentration). Generally, unbounded NOEC are not preferred for use in ERAs, as they fail to provide a range of the threshold of potential effects in the same manner as a bounded NOEC-LOEC pair. The unbounded NOEC is considerably lower than the NOEC for PFOS or PFOA and likely does not represent an accurate NOEC value.

The potential for adverse effects to terrestrial plants has only been evaluated in a small number of species as well, primarily wheat and other produce items, with a focus on PFOS and PFOA. The recommended values for terrestrial plants are provided in Table 7.

- The recommended values for PFOS are from Brignole et al. (2003). This study evaluated the widest range of species with 21-day NOEC values for PFOS based on emergence ranging from 62.5 mg/kg to 1,000 mg/kg for all seven species of plants. The NOEC based on height measurements and shoot weight ranged from less than 3.91 to 62.5 mg/kg among all the tested plant species. In general, based on height and shoot weight, lettuce was the most sensitive plant species tested. Effects were observed at the lowest concentration tested (3.91 mg/kg), with an effective concentration 25% (EC25) of 6.79 mg/kg.
- Few chronic studies have evaluated exposure to PFOA (Yang et al., 2015; González-Naranjo et al., 2015; Zhou et al., 2016). The recommended value is from Zhou et al. (2016), in which wheat was exposed to PFOA spiked soil for 28 days, evaluating multiple growth metrics (root length, shoot length, etc.), and reported an EC10 value of 84 mg/kg. As this was the only chronic plant toxicity study for PFOA that evaluated growth on a common United States plant species, this EC10 was selected over the NOECs for Bok choy, Thale cress, or Sorghum.

Similar to terrestrial invertebrates, only one study (Zhao et al., 2014) evaluated additional PFAS, other than PFOS and PFOA. As noted above regarding earthworms, the NOEC values from this study represent unbounded NOECs, and so are considered very conservative and with high uncertainty but could be applied as a NOEC for the additional PFAS evaluated. The potential for adverse effects to non-T&E species could be evaluated by selecting a NOEC or LOEC value from a study provided in Appendix E.

Overview of T&E Species Effects Assessment: Key Points

- Effects assessment for ecological risk assessments of T&E species generally involve selection of no-effect toxicity benchmarks to which site-specific exposures are compared.
- No-effect toxicity benchmarks for T&E species are more conservative than those for ecological risk assessments that do not consider T&E species.
- Potential effects to mammalian and avian wildlife can be assessed using recommended Toxicity Reference Values provided.
- Potential effects to aquatic life, terrestrial invertebrates, and terrestrial plants can be assessed using recommended benchmarks provided, although information is largely limited to PFOA and PFOS.

3.5 T&E Risk Evaluation and Interpretation

Following exposure characterization and toxicity characterization, the next step in most ERAs is to evaluate risk by comparing the exposure concentrations to the TRV or other benchmark identified as being protective of T&E species.

In general, the potential for adverse effects is quantified as a Hazard Quotient (HQ) and is calculated as the ratio of exposure concentrations (either as concentrations in directly exposed media (soil or water) or as an internal dose for wildlife) to the "safe" concentration established by the TRV or benchmark. Generally, if the HQ is below 1, then exposures are below conservative safe toxicity thresholds and no further consideration is needed. When HQs are greater than 1, this indicates that exposure is above the benchmark value. In these cases, additional evaluation is recommended to refine the HQ. SSDs can be interpreted two ways: 1) the HC1 or HC5 threshold can be applied as a benchmark for the calculation of HQs; and 2) the exposure concentration can be used to identify the number and specific species where exposure may be causing potential adverse effects based on the SSD. In the cases for wildlife ecological risk assessment, if an HQ is above 1, it is recommended that the site- and receptor-specific predicted dose be compared to the

dose levels in the study (or studies) from which the TRV was derived so that the expected magnitude of potential adverse effects can be clearly communicated to stakeholders.

Two examples of risk evaluation – one for wildlife and one for a directly exposed organism – are provided below.

- Example 1: Evaluation of potential risks to wildlife: In this example, the bioaccumulation parameters discussed in Section 2.4 are used with measured concentrations in soil, sediment, and/or surface water to estimate concentrations in diet items for a T&E bird species. The concentrations in diet items, along with ingestion rates and body weights for a similar T&E species presented in Section 2.4, are used to calculate a daily intake for the T&E avian receptor. This daily intake is divided by the TRV identified in Section 2.5 for birds, and the HQ is calculated. If the HQ is greater than 1, additional evaluations are needed, as discussed below.
- Example 2: Evaluation of potential risks to soil invertebrates: In this example, no food web or bioaccumulation modeling is needed. Concentrations of PFAS in soil are directly compared to the benchmarks in Section 2.5 for soil invertebrates, and HQs are calculated. If all HQs are below 1, no further evaluation is needed.

Under Example 1, where HQs are greater than 1, further evaluations may be prudent to refine the model-estimated results. The use of bioaccumulation factors from spiked laboratory studies often results in higher estimates of bioaccumulation than observed in field studies. Thus, the model is conservative and likely results in slight overpredictions of potential risks. Any remediation of soil or sediment as a result of model predicted estimates of risk, needs to be carefully weighed against the potential for habitat and species loss during remediation.

There are a few options to further refine model-estimated risks should model-estimated HQs indicate a potential for adverse effects.

- To confirm model estimates of concentration in diet items, non-T&E species from the site that are known diet items for wildlife species of interest can be sampled and analyzed for concentrations of PFAS in tissue. Measured concentrations of PFAS in diet items can be used to both evaluate the model performance and to directly evaluate exposure to wildlife to refine the HQs.
- Ecological surveys may be performed to evaluate site-specific presence, number of organisms, and potentially changes to T&E species communities. Where model-estimated risks are low and the potential habitat destruction for remediation is high (i.e., for PFAS in sediments where dredging would result in significant species and habitat loss), monitoring for changes to the T&E species population using site-specific ecological surveys may be a preferable option to active remediation.

- Laboratory toxicity testing with non-T&E surrogate species (for aquatic life, benthic life, soil invertebrates, and/or plants) using sediment, water, or soil collected from the site can provide an additional line of evidence to understand the potential for adverse effects. If toxicity testing indicates no effects to surrogate species are occurring, there is more confidence in a lack of effects to T&E species than modeling results alone can provide.

Overview of T&E Risk Evaluation and Interpretation: Key Points

- The comparison of site-specific exposures to effects benchmarks for T&E species risk assessments at AFFF sites follow general ecological risk assessment procedures.
- Site-specific exceedances of effects benchmarks do not necessarily imply the presence of adverse effects and may indicate the need for further evaluation of the risk assessment procedures and assumptions, collection of additional data to refine the risk assessment, or other ecological evaluation.

3.6 Key Summary Points for the Evaluation of Risks to T&E Species at AFFF-impacted Sites

As noted in the following section, there are many uncertainties and data gaps to address with regards to the ecological risk assessment of PFAS at AFFF sites. However, risk-based decisions at AFFF sites are currently and will be needed, despite the lack of a perfect and complete knowledge of the ecotoxicology of PFAS. Based on the state-of-the-science review above, ecological risk assessment can be applied to aid in decision making. The current best available ERA approaches outlined above are likely to be more acceptable to stakeholders and decision-makers than basing decisions on an assumption of ecological harm due following the detection of PFAS in environmental samples collected at an AFFF site.

For ecological risk assessments of PFAS to T&E species at AFFF sites, general guidance and observations provided in this section can be summarized into the key points below. Undoubtedly, these key points will be refined in coming years with additional research, guidance and experience, such as more widespread PFAS data collection, interpretation of abiotic and biotic media, and additional Site-specific ERAs for PFAS.

1. **Ecological Risk Assessment of AFFF-derived PFAS to T&E Species is Possible:** Using traditional ecological risk assessment approaches, as well as the best available ecotoxicological information on PFAS, ecological risk assessments can be used to characterize risk and enable risk-based decision-making at AFFF sites.

2. **Off-site Habitats are Most at Risk:** Most AFFF release areas/sites do not generally provide valuable ecological habitat but can lead to contamination of nearby (off-site) habitats.

3. **Aquatic Habitats are Critical to Address:** Because of the relatively high water solubility of PFAS, the ability for PFAS to accumulate in aquatic sediments, and the bioaccumulation of PFAS in the aquatic food web, exposures to aquatic life (e.g., fish, pelagic life, and benthic invertebrates) and vertebrate wildlife that consume aquatic life are critical to include in risk assessments. PFSAs (e.g., PFOS) will likely be primary concerns due to their higher bioaccumulation potential (relative to PFCAs), and longer-chain PFAAs will likely exhibit higher risks than shorter-chain PFAAs.

4. **Terrestrial Habitats may be Important at Some Sites:** AFFF impacts to terrestrial ecosystems are likely to be more concentrated in a smaller area compared to impacted aquatic ecosystems downgradient of AFFF release areas. It is important to note, however, that AFFF release areas are purposely situated in areas of facilities that do not support wildlife populations and therefore often do not provide viable terrestrial habitats. Exposures to wildlife will likely drive concerns at most terrestrial sites given their exposure to the bioaccumulation of PFAS in plants and terrestrial invertebrate diet items. PFSAs will likely drive concerns for consumers of invertebrates due to their higher bioaccumulation potential (relative to PFCAs), and longer-chain PFAAs will likely exhibit higher risks than shorter-chain PFAAs. However, shorter-chain PFAAs will drive concerns for herbivorous wildlife due to their higher bioaccumulation potential (relative to longer-chain PFAAs) in plants.

5. **Risks from Mixtures is Uncertain:** Although ecological receptors will be exposed to a mixture of PFAS, current ecological risk assessment is only possible for the evaluation of single-PFAS effects. Toxicological justification for evaluation of risks from multiple concurrent PFAS exposures in ERAs is needed, and regulatory approaches in the US for human health include addressing mixture and additive exposures for some PFAS. Current SERDP research is underway that is investigating the potential PFAS cumulative mixture effects. Summation of PFAS exposures or risks (i.e., calculation of a PFAS Hazard Index) may be a useful evaluation in the absence of guidance, although it should not necessarily be used as the primary or only basis of decision making.

6. **Effects of Many PFAS are Unknown:** Most of the current ecotoxicological knowledge is based on PFAAs, primarily PFOA and PFOS. For PFAS that can be measured at the site, and exposure estimated, use of effect benchmarks for a compound of similar perfluorinated carbon chain length may be the best available option. This carries high uncertainty, is best performed as a sensitivity analysis, and should not necessarily be used as the primary basis of decision making. Alternatively, site-specific toxicity testing can be used to evaluate the potential for adverse effects from mixtures of PFAS and reduce uncertainty as the test organisms are exposed to the complete, site-specific PFAS mixture to evaluate potential

effects. This approach can be a valuable line of evidence to evaluate mixture effects to directly exposed receptors (i.e., sediment toxicity tests for invertebrates, or plant toxicity tests in soil) but may not address the potential effects of exposure to PFAS mixtures for wildlife. Ecological studies (e.g., benthic invertebrate census) can also be used to evaluate the overall health of the community, a direct measure of the potential effects that could be caused by PFAS and other chemicals. At this time, most assessors, managers, and stakeholders are proceeding under the assumption that decisions and conclusions made with the PFAS for which effect benchmarks are available likely address risks of measurable PFAS that cannot be completely characterized, as well as PFAS that may be present, but are currently unable to be measured in environmental media (PFAS precursors, other PFAS, etc.).

4. UNCERTAINTIES, DATA GAPS, AND RECOMMENDATIONS

4.1 Key Uncertainties

Ecological risks assessments for PFAS, especially for PFAS beyond PFOS and PFOA, are in their infancy, and a high degree of uncertainty remains. This section discusses the uncertainties specific to PFAS-related ERAs. There are a number of uncertainties related to all ERAs, based on the use of assumed parameters for ecological modeling, spatial variation of chemicals in media, and organism habitat use patterns, among other uncertainties. These general ERA uncertainties are not discussed here.

Uncertainty in Exposure and Effects Characterization:

- For the selection of bioaccumulation parameters for fish, values that presented BCFs or BMFs based on whole-body tissue concentrations were preferred; however, these metrics can be calculated on a tissue-specific basis. For example, Martin et al. (2003b) calculated BCFs from concentrations in fish tissue and water for fish carcass (mainly muscle), fish blood, and fish liver. The PFOS carcass BCF was approximately five times lower than the BCF for liver and blood, as these organs accumulate higher levels of some PFAS than muscle tissue. When piscivorous birds consume a whole fish, they consume muscle/carcass, blood, and liver; therefore, the use of the lower BCFs may underestimate uptake. In Larson et al. (2018), the carcass BCFs were selected for use in the model, as muscle represents the highest mass of fish tissue and was considered to be most representative of an avian consumption. Additionally, model predictions showed the best agreement with whole-body fish concentrations for scenarios in which measured concentrations of PFOS were available (Larson et al., 2018).
- For PFAS that do not have specific bioaccumulation parameters or toxicity information, it has been hypothesized that the values for PFOS be applied as a conservative surrogate. Application of bioaccumulation parameters or toxicity information derived from PFAS with a similar perfluorinated carbon chain length may also be evaluated. There is considerable uncertainty in these approaches, and values should be carefully considered before they are applied. The lack of information on PFAS beyond PFOS and PFOA is a clear information gap, but as site-specific decisions are needed at many facilities, this conservative approach can be considered.

Uncertainty in PFAS Characterization:

- Ecological receptors at AFFF-impacted sites will be exposed to multiple PFAS simultaneously. A significant area of uncertainty is the potential for additive or synergistic effects from mixtures of PFAS that may result from exposure to AFFF-impacted media. Currently, the vast majority of studies have focused on exposure of a test mammal to a single PFAS, and there is very limited information on how cumulative or mixture toxicity should be addressed. Various regulatory agencies (e.g., USEPA, and environmental regulatory organizations in several US states, Australia, Germany, and the Netherlands)

have indicated that some PFAS should be summed for a risk-based evaluation (e.g., PFOS + PFOA, PFOS + PFHxS) or that a Toxicity Equivalent Approach may be warranted (Lijzen et al., 2018); however, a robust understanding of relative toxicities is still lacking. One approach to evaluate multiple PFAS exposures is to sum HQ values calculated in an ecological risk assessment and evaluate cumulative risks as a PFAS Hazard Index (HI). Additionally, the sum of detected PFAS exposures can be evaluated against effects benchmarks for PFOS, which tends to be the most toxic PFAS in most reviews. While exceeding the PFOS benchmark (or HI of 1) does not provide a robust understanding of risks, if the exposures are below thresholds, this can be a useful line of evidence to suggest a lack of potential risks. It should be noted that there is no formal guidance or toxicological evidence to support these approaches, and they should not be the sole basis for risk-based decision making at sites.

- Within an AFFF mixture, there is the possibility of the presence of polyfluorinated compounds and other PFAS that are not quantified under standard analytical methods but will degrade over time in the environment to the stable, persistent PFAAs. These compounds, known as PFAA precursors, can be oxidized in environmental samples (via strong oxidative procedures) to transform them rapidly in a laboratory to the PFAAs, which can then be analyzed. Currently, exposure food-web modeling does not incorporate these precursors, which potentially could lead to underestimating PFAA concentrations in diet items and ultimately, PFAA exposures to predators. The mechanism driving the oxidation of these precursors is not well understood, and as a result these precursor compounds are not incorporated into modeling estimates. The analytical method for this oxidation step is known as the Total Oxidable Precursor Assay (TOPA). The TOPA can be an informative tool when an understanding of total mass or source zones is needed (Casson and Chiang, 2018), but this method is not recommended for ERAs. Full oxidation of precursors does not occur naturally in the environment and, therefore, the PFAA concentrations observed following the TOPA do not necessarily reflect concentrations to which a receptor may be exposed. Oxidation occurs over time and therefore oxidation of precursors may vary considerably between sites based on timings of releases. However, it is clear that research to evaluate the presence, transformation, and exposure to precursors in ecological habitats affected by AFFF is needed. At this time, ecological risk assessments are proceeding under the assumption that risk will be driven by the detectable PFAAs, such that risk-based decisions based on the PFAAs will be protective of PFAS precursors. This assumption should be tested.

Uncertainty in Risk Evaluation and Risk Management:

- This guidance reflects a primarily modeling-based approach to evaluate the potential risks to wildlife T&E species, along with a conservative approach for evaluating directly exposed receptors using media-specific benchmarks. The approach herein is conservative, as it aligns with the goals of T&E species management. However, in the cases where conservative estimates and modeling results indicate that there are potential risks to T&E

species, careful consideration of risk management approaches is needed. The majority of active risk management techniques require contaminant mass removal (i.e., excavation, dredging), which can result in considerable damage to habitats and species present, and these losses need to be weighed against the potential benefits. For example, if the T&E species of concern is a benthic invertebrate exposed to PFAS in sediment and dredging of sediment will result in loss of that T&E species or critical habitat, then monitoring and natural recovery would be a more appropriate response, even if risk reduction will be slower. However, if the T&E species of concern is an aquatic bird that would be unimpacted by sediment removal, then active remediation is a more preferable option. These considerations are needed on a site-specific basis when the potential for adverse effects has been identified.

4.2 Research Needs and Critical Data Gaps for Ecological Risk Assessment of PFAS

Compiling the key information required to evaluate potential risks for T&E species at AFFF-impacted sites reveals multiple data gaps, including:

- Toxicity of PFAS to benthic invertebrates.
- Toxicity of PFAS to birds, aside from PFBS and PFOS.
- Toxicity of PFAS to terrestrial plants and terrestrial invertebrates, aside from PFOS and PFOA.
- Additional consideration for ecological risk modeling refinement for terrestrial ecosystems is needed.
- A better understanding of exposure to PFAS precursors, which may be present in diet items or exposure media and which may ultimately degrade to PFAAs, particularly for aquatic and terrestrial invertebrates.
- Further understanding of driving mechanisms for oxidation of PFAA precursors in abiotic media and biological species.
- Consideration of a mechanistic model to better predict food-web modeling in higher trophic levels is needed.
- Development of additional bioaccumulation factors to fill data gaps in higher trophic levels transfer (prey to predator) is needed.
- Measurement of ecologically significant endpoints at a site impacted by PFAS from AFFF would be useful to confirm the predictions offered by the current recommended toxicological benchmarks.

The following sections describe the data gaps analysis in further detail and highlights other important and critical research areas for the evaluation of PFAS in ecological risk assessments.

4.2.1 Data Gaps Analysis

To better understand current data gaps for PFAS in terrestrial and aquatic ecosystems, all available recommended values generated in this guidance were summarized in Table 8 and Table 9 for terrestrial and aquatic ecological risk assessment scenarios, respectively. Along with the confidence in the available data, these tables evaluate the overall potential for risk using occurrence data at AFFF sites, bioaccumulation properties, and toxicity properties. To assess the magnitude of occurrence at AFFF sites, median concentration and detection frequency in surface soil (terrestrial habitats), sediment and surface water (aquatic habitats) from 40 military US AFFF sites reported in Anderson et al. (2016) are included. PFAS median concentrations and detection frequencies were multiplied together to represent "occurrence values", and each was ranked from "low," "moderate," or "high" for each habitat type. Bioaccumulation and toxicity information was compiled from the recommended values discussed in previous sections of this document. Bioaccumulation metrics and toxicity information was included, and color coded from red (for properties indicative of higher risk) to green (for properties indicative of lower risk). For example, green shading was used for lower bioaccumulation measurements and lower toxicity values and red shading was used for higher bioaccumulation measurements and lower toxicity values.

The chemical occurrence categorization along with the availability and technical quality of bioaccumulation metrics and toxicity information of the recommended values was assessed for each PFAS using best professional judgement with the goal of evaluating the data gap importance in relation to the occurrence, bioaccumulation potential and toxicity. Based on the review, the relative importance of the identified data gap for each PFAS was ranked "low," "moderate" or "high," and color-coded for ease of interpretation. Following the compilation of the tables, metrics for PFAS where no values were available were identified as data gaps, and the importance of the data gap was evaluated based on the chemical's occurrence in the environment and by attempting to view the data more holistically. For example, while many PFAS had a data gap of plant toxicity, the terrestrial occurrence of PFBA was lower than many PFAS, therefore this data gap was determined to be a lower importance. This ranking system helps to provide direction for future research efforts and to prioritize research PFAS needs, but the authors note there is a level of subjectivity to this evaluation.

A summary table of data gaps is provided below (Figure 12) and specific PFAS data gaps analyses for each type of habitat (aquatic and terrestrial) are discussed in more detail below.

Ecosystem	Data Gap Level of Importance			
	Low	Moderate	High	Key Data Gaps
Terrestrial	PFBA PFHpA PFTrDA PFTeDA PFOS	PFPeA PFOA PFNA PFUnDA PFDoDA PFBS PFDS PFOSA N-EtFOSAA N-MeFOSAA	PFHxA PFDA PFHxS	1) Avian toxicity 2) Plant/invertebrate toxicity 3) Bioaccumulation metrics
Aquatic	PFBA PFPeA PFBS PFOS	PFHxA PFHpA PFOA PFNA PFUnDA PFTrDA PFTeDA PFDS PFOSA N-EtFOSAA N-MeFOSAA	PFDA PFDoDA PFHxS	1) Avian toxicity

Figure 12: Summary of PFAS Data Gaps

4.2.1.1 Terrestrial Data Gaps

The following chemicals were assigned a level of "low" for relative importance of data gaps for terrestrial ecosystems: PFBA, PFHpA, PFTrDA, PFTeDA, PFOS. For all chemicals except PFOS, this rank was assigned based on the chemical's low occurrence at AFFF sites, and overall low potential for exposure and toxicity to receptors. Many of these chemicals do not have bioaccumulation factors or toxicity factors, and further research is warranted but is considered a lower priority. PFOS was assigned a "low" relative importance level, as there is currently a robust amount of terrestrial information available on toxicity (including toxicity to invertebrates, plants, and wildlife) and bioaccumulation factors (invertebrate, plant). This does not mean that further research for PFOS is unnecessary, but highlights that the current body of literature provides a strong understanding of PFOS fate, transport and bioaccumulation in the terrestrial environment and provides adequate toxicological data for assessing risks to terrestrial organisms and wildlife.

The following chemicals were assigned a level of "moderate" for the relative importance of data gaps: PFPeA, PFOA, PFNA, PFUnDA, PFDoDA, PFBS, PFDS, PFOSA, N-EtFOSAA, and N-

MeFOSAA. These PFAS exhibit a moderate occurrence in the environment, and generally had low to moderate bioaccumulation and potential exposure to terrestrial invertebrate and plants. Other than PFOA, these chemicals lack terrestrial invertebrate and plant toxicity information. All PFAS with the exception of PFBS lack avian toxicity information and half of these "moderate" PFAS do not have mammalian toxicity values.

Three chemicals, PFHxA, PFDA and PFHxS were categorized as a "high" importance. All three PFAS have moderate to high occurrence as well as high potential of exposure to terrestrial receptors. While the toxicity to invertebrates, plants and avian wildlife are unknown, the mammalian toxicity is high for PFHxS, moderate for PFDA, and low for PFHxA, highlighting the potential for toxicity in other terrestrial receptors. Additionally, since the AFFF industry has transitioned to shorter chain PFAS formulations (which may result in potential exposures of PFHxA and shorter PFAAs present as impurities or potential end-transformation products), gaining a better understanding of the toxicity and bioaccumulation of these PFAS is imperative.

4.2.1.2 Aquatic Data Gaps

The following chemicals were assigned a level of "low" for relative importance of data gaps for aquatic ecosystems: PFBA, PFPeA, PFBS and PFOS. For all chemicals except PFOS, this rank was assigned based on the chemical's moderate occurrence in the environment, low bioaccumulation in the aquatic food web and overall low potential exposure to receptors. PFOS was assigned a "low" relative importance level as there is currently a robust amount of aquatic information available on toxicity (invertebrate, fish, wildlife) and bioaccumulation factors (invertebrate, aquatic life). This does not mean that further research for PFOS is unnecessary, but highlights that the current body of literature provides a relatively robust understanding of PFOS fate, transport and bioaccumulation in the aquatic environment and provides adequate toxicological data for aquatic receptors.

The following chemicals were assigned a level of "moderate" for the relative importance of data gaps: PFHxA, PFHpA, PFOA, PFNA, PFUnDA, PFTrDA, PFTeDA, PFDS, PFOSA, N-EtFOSAA, and N-MeFOSAA. These PFAS generally had either low to moderate occurrence in the environment and higher bioaccumulative potential, or high occurrence in the environment and low to moderate bioaccumulative potential. Furthermore, for chemicals such as PFOSA that lack any toxicity data for aquatic receptors there is a high level of uncertainty associated with chemical fate and transport and effects to the aquatic ecosystem, that is was considered a moderate data gap despite the low occurrence of these chemicals. Other than PFOA, these chemicals all lack aquatic life direct toxicity values. All PFAS categorized as "moderate" lack avian toxicity information and half of the PFAS do not have mammalian toxicity values.

Three chemicals PFDA, PFDoDA, and PFHxS were categorized as a "high" importance to fill existing data gaps. All three PFAS have moderate to high occurrence as well as moderate to high bioaccumulative potential to aquatic receptors. While the toxicity to aquatic life and avian receptors are unknown, the mammalian toxicity is high for all three PFAS in this category, highlighting the potential for high toxicity in other aquatic receptors.

4.2.2 Ecological Risk Modeling Data Gaps

Ecological risk modeling for PFAS is still in its infancy and while the Larson et al. (2018) model applied an empirical model for evaluating bioaccumulation in aquatic food webs, there is currently a need to develop and refine a terrestrial bioaccumulation model for ecological risk assessments.

4.2.2.1 Empirical Bioaccumulation Model vs. Mechanistic Bioaccumulation Model

Empirical models for bioaccumulation use literature-derived values combined with site abiotic data that delivers a site-specific risk assessment. This type of model is generally conservative, which is often desirable given the level of uncertainty associated with modeling exposure and uptake in various ecosystems. While empirical models are useful, site-specific factors influence bioavailability and uptake in ways often not considered or accounted for in empirical modeling, unless a site-specific model is developed. A recommended next step for ecological risk assessment modeling at AFFF-impacted sites is to develop a mechanistic model that accounts for the complexity of physiological processes in receptors such as the components of the diet and metabolism of polyfluorinated substances in particular as these may degrade to potentially more toxic PFAAs. A mechanistic model that incorporates a more refined understanding of uptake and excretion rates of PFAS, partitioning between internal compartments (lipids, non-lipid organic matter), and incorporates site-specific parameters and their influence on biological processes would allow for a more refined prediction of bioaccumulation for PFAS.

4.2.2.2 Transfer Factors for Higher Level Predators

There is currently a lack of data for higher trophic level transfer factors, such as prey to higher level predator factors (e.g. rodent to a coyote BMF; fish to bird BMF). This evaluation did not focus on prey to higher trophic level transfer for the following reasons: 1) the data were not available; 2) it was assumed that AFFF sites are generally small and risk at the site is driven by lower trophic level organisms; and 3) data are not available for the relevant ecological spatial scales necessary. However, as Kelly et al. (2009) noted, biomagnification in upper trophic levels can occur when exposure is wide-spread across a landscape or entire water body, such as in the case of widespread aerial deposition of PFAS from point or non-point sources or releases of large masses of PFAS in water bodies. Further research and investigation into transfer factors for higher level predators is necessary and would help fill the current data gaps for evaluating ecological risk of PFAS at AFFF impacted sites.

Overview of Uncertainties and Data Gaps: Key Points

- There is a robust body of literature regarding fate, transport and toxicity of PFOS and PFOA, but far less information on other PFAS.
- In terrestrial ecosystems, data gaps for PFHxA, PFDA, and PFHxS have been identified as most critical based on the occurrence and behavior of these PFAS in terrestrial systems.
- In aquatic ecosystems, data gaps for PFHxA, PFDA, and PFDoDA have been identified as most critical based on the occurrence and behavior of these PFAS in aquatic systems.

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TABLES

Table 1. Focused Analyte List for Guidance for Assessing Ecological Risks to T&E Species

Analyte Name (common alternative)	Acronym (common alternative)	Chemical Formula	# Perfluorinated Carbons	Chemical Abstract Services Registry Number (CASRN)[1]	Included in EPA UCMR3 list[2] / ATSDR 2018	States with standard or guidance different from EPA HA[3]
Perfluoroalkyl carboxylates (PFCAs)						
Perfluorobutanoic acid (Perfluorobutanoate)	PFBA	$F(CF_2)_3C(O)O^-$	3	375-22-4	No / **Yes**	MN, TX
Perfluoropentanoic acid (Perfluoropentanoate)	PFPeA	$F(CF_2)_4C(O)O^-$	4	2706-90-3	No / No	TX
Perfluorohexanoic acid (Perfluorohexanoate)	PFHxA	$F(CF_2)_5C(O)O^-$	5	307-24-4	No / **Yes**	TX
Perfluoroheptanoic acid (Perflouroheptanoate)	PFHpA	$F(CF_2)_6C(O)O^-$	6	375-85-9	**Yes / Yes**	CO, CT, MA, OR, TX
Perfluorooctanoic acid (Perfluroroctanoate)	PFOA	$F(CF_2)_7C(O)O^-$	7	335-67-1	**Yes / Yes**	NJ, MN, TX
Perfluorononanoic acid (Perfluorononanorate)	PFNA	$F(CF_2)_8C(O)O^-$	8	375-95-1	**Yes / Yes**	CT, MA, NJ, OR, TX
Perfluorodecanoic acid (Perfluorodecanoate)	PFDA (PFDeA)	$F(CF_2)_9C(O)O^-$	9	335-76-2	No / **Yes**	TX
Perfluoroundecanoic acid Perfluoroundecanoate)	PFUnDA	$F(CF_2)_{10}C(O)O^-$	10	2058-94-8	No / **Yes**	TX
Perfluorododecanoic acid (Perflurododecanoate)	PFDoDA	$F(CF_2)_{11}C(O)O^-$	11	307-55-1	No / **Yes**	TX
Perfluorotridecanoic acid (Perfluorotridecanoate)	PFTrDA	$F(CF_2)_{12}C(O)O^-$	12	72629-94-8	No / No	TX
Perfluorotetradecanoic acid (Perfluorotetradecanoate)	PFTeDA	$F(CF_2)_{13}C(O)O^-$	13	376-06-7	No / No	TX
Perfluoroalkyl sulfonates (PFSAs)						
Perfluorobutanesulfonic acid (Perfluorobutane sulfonate)	PFBS	$F(CF_2)_4SO_3^-$	4	375-73-5	**Yes / Yes**	DE, MA, MN, NV, TX
Perfluorohexanesulfonic acid (Perfluorohexane sulfonate)	PFHxS	$F(CF_2)_6SO_3^-$	6	355-46-4	**Yes / Yes**	CT, MA, MN, TX
Perfluorooctanesulfonic acid (Perflurooctane sulfonate)	PFOS	$F(CF_2)_8SO_3^-$	8	1763-23-1	**Yes / Yes**	NJ, MN, TX
Perfluorodecane sulfonic acid	PFDS	$F(CF_2)_{10}SO_3^-$	10	335-77-3	No / No	TX

Table 1. Focused Analyte List for Guidance for Assessing Ecological Risks to T&E Species

Analyte Name (common alternative)	Acronym (common alternative)	Chemical Formula	# Perfluorinated Carbons	Chemical Abstract Services Registry Number (CASRN)[1]	Included in EPA UCMR3 list[2] / ATSDR 2018	States with standard or guidance different from EPA HA[3]
Perfluoroalkane sulfonamides (FASAs)						
Perfluorooctane sulfonamide	PFOSA	$F(CF_2)_8SO_2NH_2$	8	754-91-6	No / **Yes**	TX, OR
***N*-Ethyl and *N*-Methyl perfluoroalkane sulfonamidoacetic acids and salts (EtFASAAs and MeFASAAs)**						
2-(N-Ethyl perfluorooctane sulfonamido) acetic acid	N-EtFOSAA	$F(CF_2)_8SO_2N(C_2H_5)CH_2COOH$	8	2991-50-6	No / **Yes**	
2-(N-Methyl perfluorooctane sulfonamido) acetic acid	N-MeFOSAA	$F(CF_2)_8SO_2N(CH_3)CH_2COOH$	8	2355-31-9	No / **Yes**	

[1] CASRN number is for protonated form for sulfonates and carboxylates (e.g., PFOA = $F(CF_2)_7C(O)OH$).

[2] The third Unregulated Contaminant Monitoring Rule (UCMR 3) required monitoring for 30 contaminants (including 6 PFAS) between 2013 and 2015 using analytical methods developed by EPA, consensus organizations, or both to provide a basis for future regulatory actions to protect public health (https://www.epa.gov/dwucmr/third-unregulated-contaminant-monitoring-rule, as of 8/2/2018).

[3] State guidance or standards with PFAS levels that differ from current (2016) EPA health advisory (HA) levels for PFOS and PFOA individual or combined concentrations greater than 70 ppt by concentration and/or additional analytes considered in state guidance or standards.

Acronyms:

T&E = Threatened and endangered

Table 2a: Recommended Koc values for PFAS

PFAS	Carbon Chain Length	Number of Perfluorinated Carbon Atoms	LogKoc	Koc L/Kg OC	Study	Notes
PFCAs						
PFBA	C4	3	1.88	76	Guelfo and Higgins, 2013	A[1]
PFPeA	C5	4	1.37	23	Guelfo and Higgins, 2013	A[1]
PFHxA	C6	5	1.31	20	Guelfo and Higgins, 2013	A[1]
PFHpA	C7	6	1.63	43	Guelfo and Higgins, 2013	A[1]
PFOA	C8	7	1.89	78	Guelfo and Higgins, 2013	A[1]
PFNA	C9	8	2.36	229	Guelfo and Higgins, 2013	A[1]
PFDA	C10	9	2.96	912	Guelfo and Higgins, 2013	A[1]
PFUnDA	C11	10	3.56	3631	Guelfo and Higgins, 2013	A[1]
PFDoDA	C12	11	3.73	5309	Zhao et al., 2012	A[2]
PFTrDA	C13	12	--	--	--	B
PFTeDA	C14	13	--	--	--	B
PFSAs						
PFBS	C4	4	1.79	62	Guelfo and Higgins, 2013	A[1]
PFHxS	C6	6	2.05	112	Guelfo and Higgins, 2013	A[1]
PFOS	C8	8	2.8	631	Guelfo and Higgins, 2013	A[1]
PFDS	C10	10	3.53	3388	Higgins & Luthy, 2006	A[3]
FASAs						
PFOSA	C8	8	4.15	14125	Ahrens et al., 2011	A[4]
EtFASAAs and MeFASAAs						
N-EtFOSAA	C8	8	0.10	1.2	Higgins & Luthy, 2006	A[3]
N-MeFOSAA	C8	8	3.23	1698	Higgins & Luthy, 2006	A[3]

Notes:

A Laboratory study with spiked sediments/soils

B A recommended laboratory-derived value was not identified, although parameters derived from field studies may be available and potentially relevant for consideration at some sites

1 Average value from Table S10

2 Average of samples from Table 2

3 Value from Table 2

4 Average of samples from Table 3

Acronyms:

Koc = organic carbon-water partitioning coefficient

PFAS = Perfluoroalkyl and polyfluoroalkyl substances

PFCA = Perfluoroalkyl carboxylic acids

PFSA = Perfluoroalkyl sulfonic acids

Table 2b: Recommended Bioaccumulation Parameters for Aquatic Invertebrates

PFAS	Carbon Chain Length	Number of Perfluorinated Carbon Atoms	Aquatic Invertebrates											
			Water to Pelagic Invertebrate BCF-PI (lab) (L/kg, ww)				Water to Pelagic Invertebrate BAF-PI (field) (L/kg, ww)				Sediment to Benthic Invertebrate BSAF-BI (g, OC/g, ww)			
			Value	Source	Species	Notes	Value	Source	Species	Notes	Value	Source	Species	Notes
PFCAs														
PFBA	C4	3	--	--	--	F	--	--	--	D	--	--	--	D
PFPeA	C5	4	--	--	--	F	--	--	--	D	--	--	--	D
PFHxA	C6	5	--	--	--	F	--	--	--	D, G	0.040	Lasier et al., 2011	*Lumbriculus variegatus* (blackworm)	B[3]
PFHpA	C7	6	--	--	--	F	--	--	--	D	0.18	Lasier et al., 2011	*Lumbriculus variegatus* (blackworm)	B[3]
PFOA	C8	7	91	Dai et al., 2013	*Daphnia magna (water flea)*	A[1]	--	--	--	E	0.95	Higgins et al., 2007	*Lumbriculus variegatus* (blackworm)	A[4]
PFNA	C9	8	152	Dai et al., 2013	*Daphnia magna (water flea)*	A[1]	--	--	--	E	1.6	Higgins et al., 2007	*Lumbriculus variegatus* (blackworm)	A[4]
PFDA	C10	9	175	Dai et al., 2013	*Daphnia magna (water flea)*	A[1]	--	--	--	E	1.0	Higgins et al., 2007	*Lumbriculus variegatus* (blackworm)	A[4]
PFUnDA	C11	10	270	Dai et al., 2013	*Daphnia magna (water flea)*	A[1]	--	--	--	E	0.62	Higgins et al., 2007	*Lumbriculus variegatus* (blackworm)	A[4]
PFDoDA	C12	11	380	Dai et al., 2013	*Daphnia magna (water flea)*	A[1]	--	--	--	E	0.55	Higgins et al., 2007	*Lumbriculus variegatus* (blackworm)	A[4]
PFTrDA	C13	12	--	--	--	F	--	--	--	D	0.55	Lasier et al., 2011	*Lumbriculus variegatus* (blackworm)	B[3]
PFTeDA	C14	13	--	--	--	F	--	--	--	D	0.55	Lasier et al., 2011	*Lumbriculus variegatus* (blackworm)	B[3]
PFSAs														
PFBS	C4	4	0.0065	Chen et al., 2018	*Caenorhabditis elegans* (round worms)	A[2]	--	--	--	E	0.34	Lasier et al., 2011	*Lumbriculus variegatus* (blackworm)	B[3]
PFHxS	C6	6	--	--	--	F	--	--	--	D, G	0.86	Lasier et al., 2011	*Lumbriculus variegatus* (blackworm)	B[3]
PFOS	C8	8	179	Dai et al., 2013	*Daphnia magna (water flea)*	A[1]	--	--	--	E	1.2	Higgins et al., 2007	*Lumbriculus variegatus* (blackworm)	A[4]
PFDS	C10	10	--	--	--	F	--	--	--	D, G	0.50	Higgins et al., 2007	*Lumbriculus variegatus* (blackworm)	A[4]
FASAs														
PFOSA	C8	8	--	--	--	F	--	--	--	D	0.098	Bertin et al., 2014	*Chironomus riparius* (harlequin fly)	B
EtFASAAs and MeFASAAs														
N-EtFOSAA	C11	8	--	--	--	F	--	--	--	D	0.12	Higgins et al., 2007	*Lumbriculus variegatus* (blackworm)	A[4]
N-MeFOSAA	C12	8	--	--	--	F	--	--	--	D	--	--	--	D

Footnotes:

A Laboratory study with spiked sediments/soils or water

B Laboratory study with field-contaminated sediments/soils or water

C Based on Quantiative Structure Activity Relationship (QSAR) using perfluorinated carbon chain length

D A recommended laboratory-derived value was not identified, although parameters derived from field studies may be available and potentially relevant for consideration at some sites

E BCF value available, use over field based BAF value

F Recommended laboratory or other field-derived parameter unavailable

G Extrapolation from one parameter to another could be used to estimate parameter, but with great uncertainty.

1 Values from Table 2 BAF (L/Kg)

2 Estimated BCF from Figure 2 for PFBS

3 Calculated BSAFs from Table S10 (sediment concentrations), S13 (tissue concentrations). Organic Carbon normalized using Table S3.

4 Values from Table 2; BSAF estimated steady-state (SS) values

Acronyms:

g = gram
L/kg = Liter per kilogram
OC = Organic carbon
ww = wet weight
BI = Benthic Invertebrate
BCF = Bioconcentration factor
BAF = Bioaccumulation factor
BSAF = Biota-sediment accumulation factor
PFAS = Perfluoroalkyl and polyfluoroalkyl substances
PFCA = Perfluoroalkyl carboxylic acids
PFSA = Perfluoroalkyl sulfonic acids

Table 2c: Recommended Bioaccumulation Parameters for Terrestrial Invertebrates

PFAS	Carbon Chain Length	Number of Perfluorinated Carbon Atoms	Terrestrial Invertebrates			
			Soil to Terrestrial Invertebrate BSAF-TI (g, OC/g, ww)			
			Value	Source	Species	Notes
PFCAs						
PFBA	C4	3	--	--	--	--
PFPeA	C5	4	0.021	Zhao et al., 2014	*Eisenia fetida* (earthworm)	A[1]
PFHxA	C6	5	0.071	Zhao et al., 2014	*Eisenia fetida* (earthworm)	A[1]
PFHpA	C7	6	0.075	Zhao et al., 2014	*Eisenia fetida* (earthworm)	A[1]
PFOA	C8	7	0.30	Zhao et al., 2014	*Eisenia fetida* (earthworm)	A[1]
PFNA	C9	8	0.57	Zhao et al., 2014	*Eisenia fetida* (earthworm)	A[1]
PFDA	C10	9	1.6	Zhao et al., 2014	*Eisenia fetida* (earthworm)	A[1]
PFUnDA	C11	10	2.4	Zhao et al., 2014	*Eisenia fetida* (earthworm)	A[1]
PFDoDA	C12	11	3.8	Zhao et al., 2014	*Eisenia fetida* (earthworm)	A[1]
PFTrDA	C13	12	--	--	--	D
PFTeDA	C14	13	--	--	--	D
PFSAs						
PFBS	C4	4	0.58	Zhao et al., 2014	*Eisenia fetida* (earthworm)	A[1]
PFHxS	C6	6	2.1	Zhao et al., 2014	*Eisenia fetida* (earthworm)	A[1]
PFOS	C8	8	3.5	Zhao et al., 2014	*Eisenia fetida* (earthworm)	A[1]
PFDS	C10	10	0.017	Rich et al., 2015	*Eisenia fetida* (earthworm)	A[2]
FASAs						
PFOSA	C8	8	--	--	--	D
EtFASAAs and MeFASAAs						
N-EtFOSAA	C11	8	0.084	Zhao et al., 2016	*Eisenia fetida* (earthworm)	A[3]
N-MeFOSAA	C12	8	--	--	--	E

Footnotes:

A Laboratory study with spiked soils

B Laboratory study with field-contaminated soils

C Based on Quantiative Structure Activity Relationship (QSAR) using perfluorinated carbon chain length

D A recommended laboratory-derived value was not identified, although parameters derived from field studies may be available and potentially relevant for consideration at some sites

E Recommended laboratory or other field-derived parameter unavailable

F Extrapolation from one parameter to another could be used to estimate parameter, but with great uncertainty

1 Geometric mean of BAFs for three exposure levels from Table S3.

2 Measured OC normalized BSAF from Table 2.

3 Estimated OC normalized BSAF from Table 2; converted to wet weight using 84% water content (16% solids) from USEPA 1993.

Acronyms:

g = gram

OC = Organic carbon

ww = wet weight

BCF = Bioconcentration factor

BAF = Bioaccumulation factor

BSAF = Biota-sediment accumulation factor

PFAS = Perfluoroalkyl and polyfluoroalkyl substances

PFCA = Perfluoroalkyl carboxylic acids

PFSA = Perfluoroalkyl sulfonic acids

QSAR = Quantitative-structure activity relationships

TI = Terrestrial invertebrate

Table 2d: Recommended Bioaccumulation Parameters for Fish

PFAS	Carbon Chain Length	Number of Perfluorinated Carbon Atoms	Fish							
			Water to Fish Tissue BCF-Fish (lab) (L/kg, ww)				Diet to Tissue BMF-Fish (lab) (L/kg, ww)			
			Value	Source	Species	Notes	Value	Source	Species	Notes
PFCAs										
PFBA	C4	3	0.60	Wen et al., 2017	*Danio rerio* (zebrafish; muscle)	A[1]	0.0066	Martin et al., 2003a	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	C[5]
PFPeA	C5	4	0.23	Wen et al., 2017	*Danio rerio* (zebrafish; muscle)	A[1]	0.011	Martin et al., 2003a	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	C[5]
PFHxA	C6	5	0.69	Wen et al., 2017	*Danio rerio* (zebrafish; muscle)	A[1]	0.019	Martin et al., 2003a	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	C[5]
PFHpA	C7	6	3.2	Wen et al., 2017	*Danio rerio* (zebrafish; muscle)	A[1]	0.031	Martin et al., 2003a	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	C[5]
PFOA	C8	7	4.0	Martin et al., 2003b	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	A[2]	0.038	Martin et al., 2003a	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	A[6]
PFNA	C9	8	39	Martin et al., 2003b	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	C[3]	0.23	Goeritz et al., 2013	*Oncorhynchus mykiss* (Rainbow Trout; whole body)	A[7]
PFDA	C10	9	450	Martin et al., 2003b	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	A[2]	0.23	Martin et al., 2003a	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	A[6]
PFUnDA	C11	10	2700	Martin et al., 2003b	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	A[2]	0.28	Martin et al., 2003a	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	A[6]
PFDoDA	C12	11	18000	Martin et al., 2003b	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	A[2]	0.43	Martin et al., 2003a	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	A[6]
PFTrDA	C13	12	21627	Chen et al., 2016	*Danio rerio* (zebrafish; whole body)	A[4]	0.71	Martin et al., 2003a	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	C[5]
PFTeDA	C14	13	23000	Martin et al., 2003b	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	A[2]	1.00	Martin et al., 2003a	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	A[6]
PFSAs										
PFBS	C4	4	1.0	Wen et al., 2017	*Danio rerio* (zebrafish; muscle)	A[1]	0.020	Goeritz et al., 2013	*Oncorhynchus mykiss* (Rainbow Trout; whole body)	A[7]
PFHxS	C6	6	9.6	Martin et al., 2003b	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	A[2]	0.14	Martin et al., 2003a	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	A[6]
PFOS	C8	8	1100	Martin et al., 2003b	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	A[2]	0.32	Martin et al., 2003a	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	A[6]
PFDS	C10	10	2630	Martin et al., 2003b	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	C[3]	0.25	Martin et al., 2003a	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	C[5]
FASAs										
PFOSA	C8	8	39	Martin et al., 2003b	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	C[3]	0.023	Brandsma et al., 2011	*Oncorhynchus mykiss* (Rainbow Trout; muscle)	A[8]
EtFASAAs and MeFASAAs										
N-MeFOSAA	C11	8	39	Martin et al., 2003b	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	C[3]	0.089	Martin et al., 2003a	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	C[5]
N-EtFOSAA	C12	8	39	Martin et al., 2003b	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	C[3]	0.089	Martin et al., 2003a	*Oncorhynchus mykiss* (Rainbow Trout; carcass)	C[5]

Footnotes:

A Laboratory study with spiked water/food
B Laboratory study with field-contaminated media
C Based on Quantiative Structure Activity Relationship (QSAR) using perfluorinated carbon chain length
D A recommended laboratory-derived value was not identified, although parameters derived from field studies may be available and potentially relevant for consideration at some sites
E Recommended laboratory or other field-derived parameter unavailable
F Extrapolation from one parameter to another could be used to estimate parameter, but with great uncertainty.

1 Steady-state muscle BCF values from Table 1 (absence of long-chain PFAAs column).
2 Steady-state carcass BCF values from Table 3.
3 Values calculated from regression equation in Figure 5 y=(10^-5.73+0.915x); where x = number of perfluorinated carbons.
4 Steady-state, whole-body LogBCF values from Table 1; average of low and high exposure groups; converted to BCF.
5 Values calculated from regression equation in Figure 5 y=(10^-2.86+0.226x); where x = number of perfluorinated carbons.
6 Steady-state cacass BMF values from Table 3.
7 Values from Pg. 2082 and Figure 3; not guaranteed to be steady-state.
8 Value from Table 3.

Acronyms:

g = gram
L/kg = Liter per kilogram
OC = Organic carbon
ww = wet weight
BCF = Bioconcentration factor
BAF = Bioaccumulation factor
BMF = Biomagnification factor
BSAF = Biota-sediment accumulation factor
PFAS = Perfluoroalkyl and polyfluoroalkyl substances
PFCA = Perfluoroalkyl carboxylic acids
PFSA = Perfluoroalkyl sulfonic acids
QSAR = Quantitative-structure activity relationships

Table 2e: Recommended Bioaccumulation Parameters for Terrestrial Plants

PFAS	Carbon Chain Length	Number of Perfluorinated Carbon Atoms	Soil to Terrestrial Plant BAF-TP (g, OC /g, ww)			
			Value	Source	Species	Notes
PFCAs						
PFBA	C4	3	0.22	Blaine et al., 2013	Lettuce (leaf)	B[1]
PFPeA	C5	4	1.25	Zhao et al., 2014	Wheat (shoot)	A[2]
PFHxA	C6	5	0.81	Zhao et al., 2014	Wheat (shoot)	A[2]
PFHpA	C7	6	0.094	Zhao et al., 2014	Wheat (shoot)	A[2]
PFOA	C8	7	0.017	Zhao et al., 2014	Wheat (shoot)	A[2]
PFNA	C9	8	0.012	Zhao et al., 2014	Wheat (shoot)	A[2]
PFDA	C10	9	0.0084	Zhao et al., 2014	Wheat (shoot)	A[2]
PFUnDA	C11	10	0.0076	Zhao et al., 2014	Wheat (shoot)	A[2]
PFDoDA	C12	11	0.0067	Zhao et al., 2014	Wheat (shoot)	A[2]
PFTrDA	C13	12	--	--	--	F
PFTeDA	C14	13	--	--	--	F
PFSAs						
PFBS	C4	4	0.40	Zhao et al., 2014	Wheat (shoot)	A[2]
PFHxS	C6	6	0.087	Zhao et al., 2014	Wheat (shoot)	A[2]
PFOS	C8	8	0.046	Zhao et al., 2014	Wheat (shoot)	A[2]
PFDS	C10	10	0.0018	Blaine et al., 2013	Lettuce (leaf)	B[1]
FASAs						
PFOSA	C8	8	0.038	Bizkarguenaga et al., 2016	Lettuce (leaves + heart)	A[3]
EtFASAAs and MeFASAAs						
N-EtFOSAA	C11	8	--	--	--	F
N-MeFOSAA	C12	8	--	--	--	F

Footnotes:

A Laboratory study with spiked soils

B Laboratory study with field-contaminated soils

C Based on Quantiative Structure Activity Relationship (QSAR) using perfluorinated carbon chain length

D A recommended laboratory-derived value was not identified, although parameters derived from field studies may be available and potentially relevant for consideration at some sites

E BCF value available, use over field-based BAF value.

F Recommended laboratory or other field-derived parameter unavailable

G Extrapolation from one parameter to another could be used to estimate parameter, but with great uncertainty.

1 Calculated the mean of municipal, industrial, and field BAFs from Table 2. Organic Carbon (OC) normalized using Table S2; converted to wet weight (ww) using assumed moisture content of lettuce of 85%, 15% solids, from Sample et al. 1997.

2 Average OC-normalized BAFs for three exposure levels calculated from concentrations in wheat shoot and soil from Table S2.

3 Mean of Soil 2.4 BAFs and Substrate BAFs from Table 2; OC normalized using Table S1.

Acronyms:

g = gram

OC = Organic carbon

ww = wet weight

BAF = Bioaccumulation factor

BCF = Bioconcentration factor

PFAS = Perfluoroalkyl and polyfluoroalkyl substances

PFCA = Perfluoroalkyl carboxylic acids

PFSA = Perfluoroalkyl sulfonic acids

QSAR = Quantitative-structure activity relationships

TP = Terrestrial Plant

Table 2f: Recommended Bioaccumulation Parameters for Aquatic Plants

PFAS	Carbon Chain Length	Number of Perfluorinated Carbon Atoms	Water to Aquatic Plant BCF-AP (lab) (L/kg, ww)				Water to Aquatic Plant BAF-AP (field) (L/kg, ww)			
			Value	Source	Species	Notes	Value	Source	Species	Notes
PFCAs										
PFBA	C4	3	--	--	--	F	--	--	--	D
PFPeA	C5	4	26	Pi et al., 2017	*E. crassipes* (free-floating macrophyte)	A[1]	--	--	--	D
PFHxA	C6	5	25	Pi et al., 2017	*E. crassipes* (free-floating macrophyte)	A[1]	--	--	--	D
PFHpA	C7	6	25	Pi et al., 2017	*E. crassipes* (free-floating macrophyte)	A[1]	--	--	--	D
PFOA	C8	7	28	Pi et al., 2017	*E. crassipes* (free-floating macrophyte)	A[1]	--	--	--	D
PFNA	C9	8	58	Pi et al., 2017	*E. crassipes* (free-floating macrophyte)	A[1]	--	--	--	D
PFDA	C10	9	110	Pi et al., 2017	*E. crassipes* (free-floating macrophyte)	A[1]	--	--	--	D
PFUnDA	C11	10	315	Pi et al., 2017	*E. crassipes* (free-floating macrophyte)	A[1]	--	--	--	D
PFDoDA	C12	11	581	Pi et al., 2017	*E. crassipes* (free-floating macrophyte)	A[1]	--	--	--	D
PFTrDA	C13	12	1281	Pi et al., 2017	*E. crassipes* (free-floating macrophyte)	A[1]	--	--	--	F
PFTeDA	C14	13	1129	Pi et al., 2017	*E. crassipes* (free-floating macrophyte)	A[1]	--	--	--	F
PFSAs										
PFBS	C4	4	19	Pi et al., 2017	*E. crassipes* (free-floating macrophyte)	A[1]	--	--	--	D
PFHxS	C6	6	28	Pi et al., 2017	*E. crassipes* (free-floating macrophyte)	A[1]	--	--	--	D
PFOS	C8	8	90	Pi et al., 2017	*E. crassipes* (free-floating macrophyte)	A[1]	--	--	--	D
PFDS	C10	10	--	--	--	F	--	--	--	F
FASAs										
PFOSA	C8	8	--	--	--	F	--	--	--	F
EtFASAAs and MeFASAAs										
N-EtFOSAA	C11	8	--	--	--	F	--	--	--	F
N-MeFOSAA	C12	8	--	--	--	F	--	--	--	F

Footnotes:

A Laboratory study with spiked water
B Laboratory study with field-contaminated water
C Based on Quantiative Structure Activity Relationship (QSAR) using perfluorinated carbon chain length
D A recommended laboratory-derived value was not identified, although parameters derived from field studies may be available and potentially relevant for consideration at some sites
E BCF value available, use over field based BAF value.
F Recommended laboratory or other field-derived parameter unavailable
G Extrapolation from one parameter to another could be used to estimate parameter, but with great uncertainty.

1 Whole-plant steady-state BCF values from Table S5.

Acronyms:

L/kg = Liter per kilogram
ww = wet weight
AP = Aquatic plant
BAF = Bioaccumulation factor
BCF = Bioconcentration factor
PFAS = Perfluoroalkyl and polyfluoroalkyl substances
PFCA = Perfluoroalkyl carboxylic acids
PFSA = Perfluoroalkyl sulfonic acids
QSAR = Quantitative-structure activity relationships

Table 3. Scoring System for Wildlife Toxicity Evaluation

Study Attribute	Scoring Value Assigment	
	1	0
Data source	Primary source available publicly for review	Primary source not publicly available for review (e.g., only referenced)
Dose Route	Dosed via spiked food	Dosed via gavage, capsule, liquid, injection, or other method
Test Substance Concentrations	Doses measured or spiking of dose confirmed via measurement	Doses based on nominal values
Contaminant Form	Dose comprised of analytical grade PFAS	Dose contains unverified mixture of PFAS (i.e., AFFF) and/or other chemicals
Dose Quantification	Dose expressed by authors in mass chemical per body mass per unit time	Doses expressed on other basis
Endpoint	Ecologically sensitive and ecologically-relevant effects such as reproduction and growth	Other effects, such as lethality, physiology, behavioral, biochemical, and pathology
Dose Range	Studies with both no-effect and lowest-effect values	Studies with only no-effect or lowest-effect value
Statistical Power	Statistical significance of effects presented by study authors	Statistical significance of effects not presented or analyzed by study authors
Exposure Duration	Chronic duration or multigenerational studies	Sub-chronic and acute studies
Test Conditions	Exposure conditions (temperature, duration, spiking/dosing methods, and effect measurement methods) described	Exposure conditions not described or most information missing

Acronyms:

AFFF = Aqueous film forming foam

PFAS = Perfluoroalkyl and polyfluoroalkyl substances

Table 4: Recommended TRVs for Mammals

PFAS	Carbon Chain Length	Number of Perfluorinated Carbon Atoms	Number of Studies	NOEL (mg/kg-day)	LOEL (mg/kg-day)	LOEL Response (Observed % Decrease from Control)	Study	Test Organism	Test Type	Duration (days)	Ecological Endpoint
PFCAs											
PFBA	C4	3	4	30	--	--	van Otterdiijk, 2007b	Rat	Sub-chronic	90	Growth
PFHxA	C6	5	3	30	200	40%	Klaunig et al., 2015	Rat	Chronic	728	Survival (Growth less sensitive)
PFOA	C8	7	16	1.3	14	10%	Butenhoff et al., 2012b	Rat	Chronic	730	Reproduction
PFNA	C9	8	6	0.83	1.1	46%	Wolf et al., 2010	Mice	Sub-chronic	18	Reproduction
PFDA	C10	9	2	0.3	1	4%	Harris and Birnbaum,1989	Mouse	Sub-chronic	18	Growth
PFUnDA	C11	10	1	0.3	1	13-19%	Takahashi et al., 2014	Rat	Sub-chronic	42	Growth
PFDoA	C12	11	4	0.5	2.5	20-40%	Kato et al., 2015	Rat	Sub-chronic	42	Growth
PFTeDA	C14	13	1	3	10	5-18%	Hirata-Koizumi et al., 2015	Rat	Sub-chronic	42	Growth
PFSAs											
PFBS	C4	4	4	300	1000	8%	Lieder et al., 2009b	Rat	Sub-chronic	120	Growth
PFHxS	C6	6	3	0.3	1	14%	Chang et al., 2018	Mouse	Sub-chronic	77	Reproduction
PFOS	C8	8	14	0.1	0.4	14%	Luebker et al., 2005b	Rat	Sub-chronic	84	Growth

Acronyms:

mg/kg-d = milligram per kilogram body weight per day

LOEL = Lowest observed effect level

NOEL = No observed effect level

PFAS = Perfluoroalkyl and polyfluoroalkyl substances

PFCA = Perfluoroalkyl carboxylic acids

PFSA = Perfluoroalkyl sulfonic acids

Table 5: Recommended TRVs for Avians

PFAS	Carbon Chain Length	Number of Perfluorinated Carbon Atoms	Number of Studies	NOEL (mg/kg-day)	LOEL (mg/kg-day)	LOEL Response (Observed % Decrease from Control)	Study	Test Organism	Duration (days)	Ecological Endpoint	Notes
PFCAs											
PFHxA	C6	5	1	--	--	--	Cassone et al., 2012	--	--	--	The only identified study was internal egg dosing, which is not applicable to oral exposures that occur in wildlife. However, if Site managers have comparable data (internal egg concentrations) for surrogate species of interest at a site, then a value of 9,700 ng/g egg can be applied as a NOEL to infer potential effects to TE avian species.
PFOA	C8	7	1	1	--	--	Yueng et al. 2009 [1]	Chicken	21	Growth	No effects on growth was observed following dosing male 1-day old chicken with a mixture of PFOS/PFOA/PFDA. Due to the use of mixtures and only two doses, this is considered a very high uncertainty no-effect TRV for PFOA or PFDA.
			1	--	--	--	Nordén et al., 2016	--	--	--	The only identified study was internal egg dosing, which is not applicable to oral exposures that occur in wildlife. However, if Site managers have comparable data (internal egg concentrations) for surrogate species of interest at a site, then a value of 0.48 µg/g egg can be applied as a NOEL to infer potential effects to TE avian species.
PFDA	C10	9	1	1	--	--	Yueng et al. 2009 [1]	Chicken	21	Growth	No effects on growth was observed following dosing male 1-day old chicken with a mixture of PFOS/PFOA/PFDA. Due to the use of mixtures and only two doses, this is considered a very high uncertainty no-effect TRV for PFOA or PFDA.
PFSAs											
PFBS	C4	4	3	88	--	--	Newsted et al., 2008/Gallagher et al., 2005	Bobwhite quail	147	Reproduction	No effects to reproduction were observed in the highest chronic test concentration of 900 ppm diet (88 mg/kg/d).
PFHxS	C6	6	1	--	--	--	Cassone et al., 2012	--	--	--	The only identified study was internal egg dosing, which is not applicable to oral exposures that occur in wildlife. However, if Site managers have comparable data (internal egg concentrations) for surrogate species of interest at a site, then a value of 9,300 ng/g egg can be applied as a NOEL to infer potential effects to TE avian species.
PFOS	C8	8	9	--	0.77	< 20%	Newsted et al., 2005; 2007	Bobwhite quail	147	Reproduction	Two studies (Newsted et al. 2006, Newsted et al. 2007) scored equally high (9/10) however 2007 was selected on the basis of being a chronic study over acute. The 10ppm (0.77 mg/kg/d) exposure was a NOEL for multiple endpoints including body weights, feeding rates, egg production. There was a statistically significant but slight (< 20% relative to controls) reduction in 14 day survivorship, therefore this is considered a conservative LOEL.

Footnotes:

1: Not single chemical exposure study; organisms exposed to PFOA/PFDA/PFOS mixture

Acronyms:

mg/kg/d = milligram per kilogram body weight per day
LOEL = Lowest observed effect level
NOEL = No observed effect level
PFAS = Perfluoroalkyl and polyfluoroalkyl substances
PFCA = Perfluoroalkyl carboxylic acids
PFSA = Perfluoroalkyl sulfonic acids
TE = Threatened and endangered
TRV = Toxicity reference values

Table 6: Recommended Aquatic Life Protection Values for PFOS and PFOA

PFAS	Freshwater		Marine	
	HC5 (µg/L)	HC1 (µg/L)	HC5 (µg/L)	HC1 (µg/L)
PFOS	5.85	0.56	7.70	2.57
PFOA	1112	537	NC	NC

Acronyms:

µg/L = microgram per liter

NC = insufficient data to calculate

HC1 = Hazadous Concentration 1%

HC5 = Hazardous Concentration 5%

Table 7: Recommended Toxicity Benchmarks for Terrestrial Plants and Invertebrates

PFAS	Carbon Chain Length	Number of Perfluorinated Carbon Atoms	Terrestrial Invertebrates			Terrestrial Plants		
			NOEC (mg/kg)	Study	Notes	NOEC (mg/kg)	Study	Notes
PFCAs								
PFBA	C4	3	--	--	B	--	--	B
PFPeA	C5	4	--	--	B	--	--	B
PFHxA	C6	5	--	--	B	--	--	B
PFHpA	C7	6	--	--	B	--	--	B
PFOA	C8	7	10	He et al., 2013	A	84	Zhou et al., 2016	A
PFNA	C9	8	--	--	B	--	--	B
PFDA	C10	9	--	--	B	--	--	B
PFUnDA	C11	10	--	--	B	--	--	B
PFDoDA	C12	11	--	--	B	--	--	B
PFTrDA	C13	12	--	--	B	--	--	B
PFTeDA	C14	13	--	--	B	--	--	B
PFSAs								
PFBS	C4	4	--	--	B	--	--	B
PFHxS	C6	6	--	--	B	--	--	B
PFOS	C8	8	80	Xu et al., 2013	A	3.9	Brignole et al., 2003	A
PFDS	C10	10	--	--	B	--	--	B
PFOSA	C8	8	--	--	B	--	--	B
N-EtFOSAA	C8	8	--	--	B	--	--	B
N-MeFOSAA	C8	8	--	--	B	--	--	B

Notes:

A: Selected value from a laboratory toxicity study.

B: No NOEC values identified for this PFAS.

Acronyms:

mg/kg = milligram per kilogram

NOEC = No observable effect concentration

PFAS = Perfluoroalkyl and polyfluoroalkyl substances

PFCA = Perfluoroalkyl carboxylic acids

PFSA = Perfluoroalkyl sulfonic acids

Table 8: Terrestrial PFAS Data Gaps in the Literature

PFAS	Carbon Chain Length	Number of Perfluo-rinated Carbon Atoms	Terrestrial Occurence: Detection Frequency x Median [Surface Soil] (µg/kg)	Terrestrial Invertebrate: Bio-accumulation: BSAF-TI (g, OC/g, ww)	Terrestrial Invertebrate: Toxicity: NOEC (mg/kg)	Terrestrial Plant: Bio-accumulation: BAF (g, OC /g, ww)	Terrestrial Plant: Toxicity: NOEC (mg/kg)	Terrestrial Wildlife: Avian Toxicity: NOEL (mg/kg bw-d)	Terrestrial Wildlife: Mammalian Toxicity: NOEL (mg/kg bw-d)	Summary	Key Data Gaps	Relative Importance of Data Gap
PFBA	C4	3	0.38	NA	NA	0.22	NA	NA	30	Low occurrence, high potential exposure to plants and herbivorous wildlife	Toxicity to plants	Low
PFPeA	C5	4	0.65	0.021	NA	1.25	NA	NA	NA	Moderate occurrence, high potential exposure to plants and herbivorous wildlife	Toxicity to plants/avians/ mammals	Moderate
PFHxA	C6	5	1.2	0.071	NA	0.81	NA	NA	30	High occurrence, high potential exposure to plants and herbivorous wildlife	Toxicity to plants/avians	High
PFHpA	C7	6	0.42	0.075	NA	0.094	NA	NA	NA	Low occurrence, low potential exposure to receptors	Toxicity to plants/avians/ mammals	Low
PFOA	C8	7	1.1	0.30	10	0.017	84	*1.0*	1.3	High occurrence, low bioaccumulation, high toxicity to invertebrates	Toxicity to avians	Moderate
PFNA	C9	8	0.93	0.57	NA	0.012	NA	NA	0.83	High occurrence, moderate bioaccumulation in invertebrates	Toxicity to invertebrates/avians	Moderate
PFDA	C10	9	0.66	1.6	NA	0.0084	NA	*1.0*	0.3	Moderate occurrence, moderate bioaccumulation in invertebrates, high toxicity to mammals	Toxicity to invertebrates/avians	High
PFUnDA	C11	10	0.36	2.4	NA	0.0076	NA	NA	0.3	Low occurrence, high bioaccumulation in invertebrates, high toxicity in mammals	Toxicity to invertebrates/plants/ avians	Moderate
PFDoDA	C12	11	0.43	3.8	NA	0.0067	NA	NA	0.5	Moderate occurrence, high bioaccumulation in invertebrate, high toxicity to mammals	Toxicity to invertebrates/plants/ avians	Moderate
PFTrDA	C13	12	0.10	NA	NA	NA	NA	NA	NA	Low occurrence, no bioaccumulation or toxicity values	Bioaccumulation factors and toxicity values for all receptors	Low
PFTeDA	C14	13	0.12	NA	NA	NA	NA	NA	3	Low occurrence, no bioaccumulation information or toxicity values for most receptors	Bioaccumulation factors and toxicity values for invertebrates, plants, avians	Low
PFBS	C4	4	0.27	0.58	NA	0.40	NA	88	300	Low occurrence, moderate bioaccumulation in invertebrates and plants	Toxicity to invertebrates/plants	Moderate
PFHxS	C6	6	4.4	2.1	NA	0.087	NA	NA	0.3	High occurrence, high bioaccumulation in invertebrates, high toxicity in mammals	Toxicity to invertebrates/avians	High
PFOS	C8	8	52	3.5	80	0.046	3.9	0.77	0.1	High occurrence, high bioaccumulation in invertebrates, high toxicity to plants and mammals	None	Low
PFDS	C10	10	1.8	0.017	NA	0.0018	NA	NA	NA	Moderate occurrence, low bioaccumulation, unknown toxicity	Toxicity to plants/avians/ mammals	Moderate

Table 8: Terrestrial PFAS Data Gaps in the Literature

PFAS	Carbon Chain Length	Number of Perfluo-rinated Carbon Atoms	Terrestrial Occurence: Detection Frequency x Median [Surface Soil] (µg/kg)	Terrestrial Invertebrate: Bio-accumulation: BSAF-TI (g, OC/g, ww)	Terrestrial Invertebrate: Toxicity: NOEC (mg/kg)	Terrestrial Plant: Bio-accumulation: BAF (g, OC /g, ww)	Terrestrial Plant: Toxicity: NOEC (mg/kg)	Terrestrial Wildlife: Avian Toxicity: NOEL (mg/kg bw-d)	Terrestrial Wildlife: Mammalian Toxicity: NOEL (mg/kg bw-d)	Summary	Key Data Gaps	Relative Importance of Data Gap
PFOSA	C8	8	0.78	NA	NA	0.038	NA	NA	NA	Moderate occurrence, moderate bioaccumulation in plants, unknown toxicity	Bioaccumulation factors and toxicity values	Moderate
N-EtFOSAA	C8	8	NA	0.084	NA	NA	NA	NA	NA	Uncertain occurrence, likely low/uncertain bioaccumulation and toxicity	Bioaccumulation factors and toxicity values	Moderate
N-MeFOSAA	C8	8	NA	NA	NA	NA	NA	NA	NA	Uncertain occurrence, likely low/uncertain bioaccumulation and toxicity	Bioaccumulation factors and toxicity values	Moderate

Notes:

Underlined, italicized values are less certain such that the parameter may be relevant for additional investigation.

NA: A reccommended value is Not Available such that the parameter may be relevant for additional investigation.

Acronyms:

g = gram

mg/kg = milligram per kilogram

mg/kg/d = milligram per kilogram body weight per day

OC = Organic carbon

µg/kg = microgram per kilogram

ww = wet weight

Table 9: Aquatic PFAS Data Gaps in the Literature

PFAS	Carbon Chain Length	Number of Perfluor-inated Carbon Atoms	Aquatic Occurence		Pelagic Invertebrate Bio-accumulation	Benthic Invertebrate Bio-accumulation	Aquatic Plant Bio-accumulation	Aquatic Life		Fish		Aquatic Wildlife		Summary	Key Data Gaps	Relative Importance of Data Gap
										Bio-concentration	Bio-magnification	Avian Toxicity	Mammalian Toxicity			
			Detection Frequency x Median [Sediment] (µg/kg)	Detection Frequency x Median [Water] (µg/kg)	BCF-PI (L/kg, ww)	BSAF-BI (g, OC/g, ww)	(L/kg, ww)	Number of Freshwater Studies with a NOEC value	Number of Marine Studies with a NOEC value	BCF-Fish (L/kg, ww)	BMF-Fish (L/kg, ww)	NOEL (mg/kg bw-d)	NOEL (mg/kg bw-d)			
PFBA	C4	3	0.41	0.064	NA	NA	NA	3.00	0.00	*0.60*	*0.0066*	NA	30	Moderate occurrence, low bioaccumulation	Avian Toxicity	Low
PFPeA	C5	4	0.77	0.21	NA	NA	26	0.00	0.00	*0.23*	*0.011*	NA	NA	Moderate occurrence, low bioaccumulation	Avian Toxicity	Low
PFHxA	C6	5	1.1	0.31	NA	*0.040*	25	1.00	0.00	*0.69*	*0.019*	NA	30	High occurrence, low bioaccumulation	Avian Toxicity	Moderate
PFHpA	C7	6	0.52	0.083	NA	*0.18*	25	0.00	0.00	*3.2*	*0.031*	NA	NA	Moderate occurrence, low bioaccumulation	Avian Toxicity	Moderate
PFOA	C8	7	1.6	0.34	91	0.95	28	25	5.0	4.0	0.038	*1.0*	1.3	High occurrence, moderate bioaccumulation in benthic invertebrates	Avian Toxicity	Moderate
PFNA	C9	8	0.13	0.035	152	1.6	58	4.00	0.00	*39*	*0.230*	NA	0.83	Low occurrence, high bioaccumulation in benthic invertebrates, moderate bioaccumulation in pelagic invertebrates and aquatic plants	Avian Toxicity	Moderate
PFDA	C10	9	0.92	0.035	175	1.0	110	2.00	0.00	450	0.23	*1.0*	0.3	Moderate occurrence, high bioaccumulation in pelagic invertebrates, moderate in aquatic plants	Avian Toxicity	High
PFUnDA	C11	10	0.39	0.0042	270	0.62	315	2.00	0.00	2700	0.28	NA	0.3	Low occurrence, high bioaccumulation in pelagic invertebrates and aquatic plants	Avian Toxicity	Moderate
PFDoDA	C12	11	1.3	0.012	380	0.55	581	3.00	0.00	18000	0.43	NA	0.5	Moderate occurrence, high bioaccumulation in pelagic invertebrates, aquatic plants and fish	Avian Toxicity	High
PFTrDA	C13	12	0.40	NA	NA	*0.55*	1281	0.00	0.00	*21627*	*0.71*	NA	NA	Low occurrence, high bioaccumulation in aquatic plants and fish	Avian Toxicity	Moderate
PFTeDA	C14	13	0.25	NA	NA	*0.55*	1129	1.00	0.00	23000	1.0	NA	3	Low occurrence, high bioaccumulation in aquatic plants and fish	Avian Toxicity	Moderate
PFBS	C4	4	0.28	0.085	0.0065	*0.34*	19	7.00	1.00	1.0	0.020	NA	300	Moderate occurrence, low bioaccumulation	Avian Toxicity	Low
PFHxS	C6	6	6.6	0.62	NA	*0.86*	28	0.00	0.00	9.6	0.14	NA	0.3	High occurrence, moderate bioaccumulation	Avian Toxicity	High
PFOS	C8	8	29	2.1	179	1.2	90	63	22	1100	0.32	0.77	0.1	High occurrence, high bioaccumulation in pelagic invertebrates, moderate in aquatic plants	None	Low
PFDS	C10	10	0.67	1.4	NA	0.50	NA	0.00	0.00	*2630*	*0.25*	NA	NA	Moderate occurrence, moderate bioaccumulation in fish	Avian Toxicity	Moderate
PFOSA	C8	8	0.98	0.0073	NA	*0.098*	NA	0.00	0.00	*39*	*0.023*	NA	NA	Moderate occurrence, low bioaccumulation	Avian Toxicity	Moderate
N-EtFOSAA	C8	8	NA	NA	NA	0.12	NA	0.00	0.00	*39*	*0.089*	NA	NA	Uncertain occurrence, likely low/uncertain bioaccumulation	Avian Toxicity	Moderate
N-MeFOSAA	C8	8	NA	NA	NA	NA	NA	0.00	0.00	*39*	*0.089*	NA	NA	Uncertain occurrence, likely low/uncertain bioaccumulation	Avian Toxicity	Moderate

Notes:

Underlined, italicized values are less certain such that the parameter may be relevant for additional investigation.

NA: A reccommended value is Not Available such that the parameter may be relevant for additional investigation.

Acronyms:

g = gram
L/kg = liter per kilogram
mg/kg/d = milligram per kilogram body weight per day
OC = Organic carbon
µg/kg = microgram per kilogram
ww = wet weight

APPENDIX A

Wildlife Exposure Factors for T&E Species

Appendix A-1: Example Wildlife Exposure Factors for Small Terrestrial Mammal T&E species Found in the US

Model Parameter	Value	Units	Source	Note
Buena Vista Lake Ornate Shrew (*Sorex ornatus relictus*)				
Body weight	0.005	kg, ww	NatureServe, 2018 [1]	Average weight
Dietary Ingestion Rate	0.003	kg diet, ww/d	Nagy, 2001	Nagy (2001) for insectivorous mammals based on FMI (fresh matter intake) DIR = 1.130 × [BW (g)^0.622] x 0.001 (kg/g)
Diet Composition, Terrestrial Invertebrates	1.00	kg food, ww/kg diet, ww	NatureServe, 2018 [1]	Primarily feeds on insects
Soil Invertebrate Ingestion Rate	0.003	kg invertebrates, ww/d	Calculated	Calculated using daily dietary ingestion rate
Diet Composition, Soil (dw diet)	0.024	kg soil, dw/kg diet, dw	Value based on meadow vole's diet (USEPA, 1993).	The assumed diet proportion is 2.4% based on the estimated percent soil/sediment in a meadow vole's diet (USEPA, 1993); dry weight diet basis
Soil Invertebrate Moisture Content	0.69	kg water/kg, ww	Terrestrial invertebrates (grasshoppers, crickets) Table 4-1 of USEPA (1993)	Used as basis for moisture content of diet for converting soil/sediment ingestion from dry weight basis to wet weight basis
Diet Composition, Soil (ww diet)	0.007	kg soil, dw/kg diet, ww	Calculated	Normalized to wet weight diet
Soil Ingestion Rate	0.00002	kg soil, dw/d	Calculated	Calculated using daily dietary ingestion rate
Home Range	0.037	ha	Ornate Shrew used a surrogate; NatureServe, 2018 [1]	Average home range in California
Western Pocket Gohpher (*Thomomys mazama)*				
Body weight	0.10	kg, ww	NatureServe, 2018 [1]	Average weight
Dietary Ingestion Rate	0.046	kg diet, ww/d	Nagy, 2001	Allometric Equation for herbivorous mammals based on FMI (fresh matter intake) DIR =2.606 × [BW (g)^0.628] x 0.001 (kg/g)
Diet Composition, Vegetation	1.00	kg food, ww/kg diet, ww	NatureServe, 2018 [1]	Diet consists of roots, tubers, bulbs and some surface vegetation.
Vegetation Ingestion Rate	0.05	kg ww/d	Calculated	Calculated using daily dietary ingestion rate
Diet Composition, Soil (dw diet)	0.02	kg soil, dw/kg diet, dw	Assumed to be a low proportion of diet based on feeding habits	The assumed diet proportion is 2.4% based on the estimated percent soil/sediment in a meadow vole's diet (USEPA, 1993); dry weight diet basis
Plant Moisture Content	0.85	kg water/kg, ww	Terrestrial plants (dicots; leaves, roots, bulbs etc.) Table 4-2 of USEPA (1993)	Used as basis for moisture content of diet for converting soil/sediment ingestion from dry weight basis to wet weight basis
Diet Composition, Soil (ww diet)	0.004	kg soil, dw/kg diet, ww	Calculated	Normalized to wet weight diet
Soil Ingestion Rate	0.00016	kg soil, dw/d	Calculated	Calculated using daily dietary ingestion rate
Home Range	0.025	ha	NatureServe, 2018 [1]	Mean home range for male Botta's Pocket Gopher (*T. Bottae)*

Appendix A-1: Example Wildlife Exposure Factors for Small Terrestrial Mammal T&E species Found in the US

Model Parameter	Value	Units	Source	Note
Anastasia Beach Deermouse (*Peromyscus polionotus phasma)*				
Body weight	0.033	kg, ww	NatureServe, 2018 [1]	Average weight
Dietary Ingestion Rate	0.014	kg, ww/d	Nagy, 2001	Allometric Equation for omnivorous mammals based on FMI (fresh matter intake) DIR =1.346 × [BW (g)^0.678] x 0.001 (kg/g)
Diet Composition, Vegetation	0.80	kg food, ww/kg diet, ww	NatureServe, 2018 [1]	Assumed to eat 80% vegetation (fruits and seeds of dune plants, especially sea oats and sea rocket)
Vegetation Ingestion Rate	0.01	kg ww/d	Calculated	Calculated using daily dietary ingestion rate
Diet Composition, Terrestrial Invertebrates	0.20	kg food, ww/kg diet, ww	NatureServe, 2018 [1]	Assumed to eat 20% invertebrates when seeds scarce
Soil Invertebrate Ingestion Rate	0.003	kg ww/d	Calculated	Calculated using daily dietary ingestion rate
Diet Composition, Soil (dw diet)	0.02	kg soil, dw/kg diet, dw	Table 4-4 of USEPA (1993)	Soil/sediment estimated at <2% for white-footed mouse
Plant Moisture Content	0.09	kg water/kg, ww	Terrestrial plants (seeds) Table 4-2 of USEPA (1993)	Used as basis for moisture content of diet for converting soil/sediment ingestion from dry weight basis to wet weight basis
Soil Invertebrate Moisture Content	0.69	kg water/kg, ww	Terrestrial invertebrates (grasshoppers, crickets) Table 4-1 of USEPA (1993)	Used as basis for moisture content of diet for converting soil/sediment ingestion from dry weight basis to wet weight basis
Weighted Diet Moisture Content	0.21	kg water/kg, ww	Calculated	Calculated based on diet components
Diet Composition, Soil (ww diet)	0.016	kg soil, dw/kg diet, ww	Calculated	Normalized to wet weight diet
Soil Ingestion Rate	0.0002	kg dw/d	Calculated	Calculated using daily dietary ingestion rate
Home Range	0.063	ha	Deer mouse as surrogate; USEPA (1993)	Average of home range size for the deer mouse

Notes:
1: accessed at: http://explorer.natureserve.org

Abbreviations:
bw = body weight
d = day
dw = dry weight
ha = hectare
kg =Kilogram
ww = wet weight

Appendix A-2: Example Wildlife Exposure Factors for Small Terrestrial Avian T&E species Found in the US

Model Parameter	Value	Units	Source	Note
Coastal California Gnatcatcher (*Polioptila californica*)				
Body weight	0.006	kg, ww	NatureServe, 2018 [1]	Average body weight for California Gnatcatcher
Dietary Ingestion Rate	0.006	kg diet, ww/d	Nagy, 2001	Allometric Equation for insectivorous birds. g FMI/d DFI = 1.633 × [BW (g)^0.705] x 0.001 (kg/g).
Diet Composition, Terrestrial Invertebrates	1.00	kg food, ww/kg diet, ww	NatureServe, 2018 [1]	Invertivore; consumes insects and spiders from foliage and twigs
Invertebrate Ingestion Rate	0.006	kg invertebrates, ww/d	Calculated	Calculated using daily dietary ingestion rate
Diet Composition, Soil (ww diet)	0.000	kg soil, dw/kg diet, ww	Assumed to be 0%	Assumed to be negligible based on feeding habits of catching insects from foliage and twigs (Nature Serve [1])
Soil Ingestion Rate	0.000	kg soil, dw/d	Calculated	Calculated using daily dietary ingestion rate
Home Range	3.0	ha	NatureServe, 2018 [1]	Average of range of home ranges (1.6 to 4.4 ha)
Masked Bobwhite Quail (*Colinus virginianus ridgwayi*)				
Body weight	0.18	kg, ww	NatureServe, 2018 [1]	Average for Northern bobwhite (Colinus virginianus)
Dietary Ingestion Rate	0.054	kg diet, ww/d	Nagy, 2001	Allometric Equation for omnivorous birds based on FMI (fresh matter intake) DFI =2.094 × [BW (g)^0.627] x 0.001 (kg/g)
Diet Composition,Vegetation	0.80	kg food, ww/kg diet, ww	The Cornell Lab of Ornithology, 2018 [2]	Bobwhites eat mostly seeds and leaves supplemented with varying amounts of insects during breeding season
Vegetation Ingestion Rate	0.04	kg ww/d	Calculated	Calculated using daily dietary ingestion rate
Diet Composition, Invertebrates	0.20	kg food, ww/kg diet, ww	The Cornell Lab of Ornithology, 2018 [2]	Arthropods can make up 5% of male's diet or 20% of female's diet during breeding season
Invertebrate Ingestion Rate	0.011	kg ww/d	Calculated	Calculated using daily dietary ingestion rate
Diet Composition, Soil (dw diet)	0.10	kg soil, dw/kg diet, dw	Based on surrogate species American woodcock in USEPA (1993; Table 4-4)	Bobwhites are ground foragers that scratch through leaf litter and dead vegetation (NatureServe, 2018 [1], Cornell Lab or Ornithology, 2018 [2])
Plant Moisture Content	0.85	kg water/kg, ww	Terrestrial plants (leaves) Table 4-2 of USEPA (1993)	Used as basis for moisture content of diet for converting soil/sediment ingestion from dry weight basis to wet weight basis
Soil Invertebrate Moisture Content	0.69	kg water/kg, ww	Terrestrial invertebrates (grasshoppers, crickets) Table 4-1 of USEPA (1993)	Used as basis for moisture content of diet for converting soil/sediment ingestion from dry weight basis to wet weight basis
Weighted Diet Moisture Content	0.82	kg water/kg, ww	Calculated	Calculated based on diet components
Diet Composition, Soil (ww diet)	0.018	kg soil, dw/kg diet, ww	Calculated	Normalized to wet weight diet
Soil Ingestion Rate	0.001	kg soil, dw/d	Calculated	Calculated using daily dietary ingestion rate
Home Range	10.3	ha	Northern Bobwhite used as a surrogate from USEPA (1993)	Average from mean home ranges for Northern Bobwhite

Appendix A-2: Example Wildlife Exposure Factors for Small Terrestrial Avian T&E species Found in the US

Model Parameter	Value	Units	Source	Note
Florida Scrub-jay (*Aphelocoma coerulescens*)				
Body weight	0.091	kg, ww	NatureServe, 2018 [1]	Average weight used
Dietary Ingestion Rate	0.039	kg, ww/d	Nagy, 2001	Allometric Equation for Omnivorous birds g FMI/d DFI = 1.633 × [BW (g)^0.705] x 0.001 (kg/g).
Diet Composition, Invertebrates/Vertebrates	0.60	kg food, ww/kg diet, ww	Assumed to be 60%	Eats invertebrates; opportunistic omnivore; lizards and arthropods dominate diet in spring and summer, acorns in fall and winter NatureServe, 2018 [1]
Invertebrate Ingestion Rate	0.02	kg ww/d	Calculated	Calculated using daily dietary ingestion rate
Diet Composition, Other Items	0.40	kg food, ww/kg diet, ww	NatureServe, 2018 [1]	Opportunistic omnivore; lizards and arthropods dominate diet in spring and summer, acorns in fall and winter
Diet Composition, Soil (dw diet)	0.10	kg soil, dw/kg diet, dw	Based on surrogate species American woodcock Table 4-4 of USEPA (1993)	Assumed to be a low proportion of wet weight diet based on feeding habits; hop along the ground eating insects and vertebrates and buries acorns in the ground
Soil Invertebrate Moisture Content	0.69	kg water/kg, ww	Terrestrial invertebrates (grasshoppers, crickets) Table 4-1 of USEPA (1993)	Used as basis for moisture content of diet for converting soil/sediment ingestion from dry weight basis to wet weight basis
Other Moisture Content (acorns)	0.09	kg water/kg, ww	Terrestrial plants (seeds) Table 4-2 of USEPA (1993)	Used as basis for moisture content of diet for converting soil/sediment ingestion from dry weight basis to wet weight basis
Weighted Diet Moisture Content	0.45	kg water/kg, ww	Calculated	Calculated based on diet components
Diet Composition, Soil (ww diet)	0.055	kg soil, dw/kg diet, ww	Calculated	Normalized to wet weight diet
Soil Ingestion Rate	0.0022	kg dw/d	Calculated	Calculated using daily dietary ingestion rate
Home Range	9.0	ha	NatureServe, 2018 [1]	Mean/median home range

Notes:
1: accessed at: http://explorer.natureserve.org
2: accessed at: https://www.allaboutbirds.org/guide

Abbreviations:
bw = body weight
d = day
dw = dry weight
ha = hectare
kg =Kilogram
ww = wet weight

Appendix A-3: Example Wildlife Exposure Factors for Aquatic Mammal T&E species Found in the US

Model Parameter	Value	Units	Source	Note
West Indian manatee (*Trichechus manatus*)				
Body weight	400.0	kg, ww	NatureServe, 2018 [1]	Average weight
Dietary Ingestion Rate	26.0	kg diet, ww/d	US Fish & Wildlife Service [2]	Manatees consume between 4- 9 % of their body weight each day of plants
Diet Composition, Herbivorous	1.0	kg food, ww/kg diet, ww	NatureServe, 2018 [1]	Feed on submergent, floating vegetation
Herbivorous Ingestion Rate	26.0	kg plants, ww/d	Calculated	Calculated using daily dietary ingestion rate
Diet Composition, Sediment (ww diet)	0.0	kg sediment, dw/kg diet, ww	Assumed to be 0% of wet weight diet based on feeding habits	Feed on submergent, floating vegetation
Sediment Ingestion Rate	0.0	kg sediment, dw/d	Calculated	Calculated using daily dietary ingestion rate
Home Range	17000	ha	NatureServe, 2018 [1]	Inferred minimum extent of habitat use with a diameter of 15 km
Southern Sea Otter (*Enhydra lutris nereis*)				
Body weight	35.0	kg, ww	NatureServe, 2018 [1]	Average weight
Dietary Ingestion Rate	8.8	kg diet, ww/d	US Fish & Wildlife Service [3]	Consume about 25% of their body mass per day
Diet Composition, Benthic Invertebrates	0.8	kg food, ww/kg diet, ww	NatureServe, 2018 [1]	Diet often dominated by benthic invertebrates (sea urchins, crabs, variety of molluscs)
Benthic Invertebrate Ingestion Rate	7.0	kg ww/d	Calculated	Calculated using daily dietary ingestion rate
Diet Composition, Fish	0.2	kg food, ww/kg diet, ww	Assumed	Fish are important food items at high population densities
Fish Ingestion Rate	1.8	kg ww/d	Calculated	Calculated using daily dietary ingestion rate
Diet Composition, Sediment (ww diet)	0.0	kg sediment, dw/kg diet, ww	Assumed to be 0% of wet weight diet based on feeding habits	Based on feeding habits does not eat any sediment. NatureServe, 2018 [1]
Sediment Ingestion Rate	0.0	kg sediment, dw/d	Calculated	Calculated using daily dietary ingestion rate
Home Range	200	ha	Use Northern sea otter as a surrogate from NatureServe, 2018 [1]	Generally occur within 2 km of shore

Appendix A-3: Example Wildlife Exposure Factors for Aquatic Mammal T&E species Found in the US

Model Parameter	Value	Units	Source	Note
Stellar sea lion (*Eumetopias jubatus*)				
Body weight	263.0	kg, ww	NatureServe, 2018 [1]	Average weight of females
Dietary Ingestion Rate	15.8	kg, ww/d	Marine Mammal Research Consortium [4]	On average adults consume around 6% of their body weight each day
Diet Composition, Piscivore	0.8	kg food, ww/kg diet, ww	NatureServe, 2018 [1]	Opportunistically feed on fishes
Fish Ingestion Rate	12.6	kg ww/d	Calculated	Calculated using daily dietary ingestion rate
Diet Composition, Invertivore	0.2	kg food, ww/kg diet, ww	NatureServe, 2018 [1]	Feed on cephalopods and sometimes on various other invertebrates
Invertebrate Ingestion Rate	2.5	kg food ww/d	Calculated	Calculated using daily dietary ingestion rate
Diet Composition, Sediment (ww diet)	0.0	kg sediment, dw/kg diet, ww	Assumed to be 0% of wet weight diet based on feeding habits	Based on feeding habits does not eat any sediment
Sediment Ingestion Rate	0.0	kg ww/d	Calculated	Calculated using daily dietary ingestion rate
Home Range	3500	ha	NatureServe, 2018 [1]	Range of habitat most often found

Notes:
1: accessed at: http://explorer.natureserve.org
2: accessed at: https://www.fws.gov/endangered/esa-library/pdf/manatee.pdf
3: accessed at: https://www.fws.gov/ventura/endangered/species/info/sso.html
4: accessed at: http://www.marinemammal.org/biology/steller-sea-lion/

Abbreviations:
bw = body weight
d = day
dw = dry weight
kg =Kilogram
ww = wet weight

Appendix A-4: Example Wildlife Exposure Factors for Small Aquatic Avian T&E species Found in the US

Model Parameter	Value	Units	Source	Note
Mississippi Sandhill Crane (*Antigone canadensis pulla*)				
Body weight	5.83	kg, ww	NatureServe, 2018 [1]	Average weight for whooping crane
Dietary Ingestion Rate	0.973	kg diet, ww/d	Nagy, 2001	Allometric Equation for carnivorous birds based on FMI (fresh matter intake) DIR =3.048 × [BW (g)^0.665] x 0.001 (kg/g)
Diet Composition, Invertebrates	0.500	kg food, ww/kg diet, ww	Estimated based on diet; NatureServe, 2018 [1]	Eats reptiles, amphibians, insects, and aquatic plants; in winter, may feed on grain remaining in fields after harvest
Invertebrate Ingestion Rate	0.5	kg ww/d	Calculated	Calculated using daily dietary ingestion rate
Diet Composition, Vegetation	0.500	kg food, ww/d	Estimated based on diet; NatureServe, 2018 [1]	Eats reptiles, amphibians, insects, and aquatic plants; in winter, may feed on grain remaining in fields after harvest NatureServe, 2018 [1]
Vegetation Ingestion Rate	0.486	kg ww/d	Calculated	Calculated using daily dietary ingestion rate
Diet Composition, Sediment (dw diet)	0.10	kg sediment, dw/kg diet, dw	American woodcock used as a surrogate Table 4-4 of USEPA (1993)	Picks food items from ground surface or probes into substrate NatureServe, 2018 [1]
Vertebrate Moisture Content	0.76	kg water/kg, ww	Reptiles and amphibians (average moisture content) Table 4-2 of USEPA (1993)	Used as basis for moisture content of diet for converting sediment ingestion from dry weight basis to wet weight basis
Invertebrate Moisture Content	0.69	kg water/kg, ww	Terrestrial invertebrates (grasshoppers, crickets) Table 4-1 of USEPA (1993)	Used as basis for moisture content of diet for converting sediment ingestion from dry weight basis to wet weight basis
Vegetation Moisture Content	0.855	kg water/kg, ww	Aquatic plants (average of algae, aquatic macrophytes) Table 4-2 of USEPA (1993)	Used as basis for moisture content of diet for converting sediment ingestion from dry weight basis to wet weight basis
Weighted Diet Moisture Content	0.77	kg water/kg, ww	Calculated	Calculated based on diet components
Diet Composition, Sediment (ww diet)	0.023	kg sediment, dw/kg diet, ww	Calculated	Normalized to wet weight diet
Sediment Ingestion Rate	0.022	kg sediment, dw/d	Calculated	Calculated using daily dietary ingestion rate
Home Range	2100	ha	Home Range for Sandhill Crane; NatureServe, 2018 [1]	Upper estimate of home ranges
California clapper rail (*Rallus obsoletus*)				
Body weight	0.32	kg, ww	NatureServe, 2018 [1]	Average weight for similar species Light-footed Clapper Rail
Dietary Ingestion Rate	0.078	kg diet, ww/d	Nagy, 2001	Allometric Equation for omnivorous birds. g FMI/d DFI = 2.094 × [BW (g)^0.627] x 0.001 (kg/g).
Diet Composition, Invertebrates	1.00	kg food, ww/kg diet, ww	NatureServe, 2018 [1]	Diet mostly mussels, clams, small crabs, and spiders; probes in mud or sand in or near shallow water, or picks items from substrate
Aquatic Invertebrate Ingestion Rate	0.08	kg ww/d	Calculated	Calculated using daily dietary ingestion rate
Diet Composition, Sediment (dw diet)	0.18	kg sediment, dw/kg diet, dw	Western sandpiper used as a surrogate Table 4-4 of USEPA (1993)	Clapper rail probes in mud or sand in or near shallow water, or picks items from substrate (NatureServe, 2018 [1])
Aquatic Invertebrate Moisture Content	0.82	kg water/kg, ww	Aquatic invertebrates (bivalves) Table 4-1 of USEPA (1993)	Used as basis for moisture content of diet for converting sediment ingestion from dry weight basis to wet weight basis

Appendix A-4: Example Wildlife Exposure Factors for Small Aquatic Avian T&E species Found in the US

Model Parameter	Value	Units	Source	Note
Diet Composition, Sediment (ww diet)	0.03	kg sediment, dw/kg diet, ww	Calculated	Normalized to wet weight diet
Sediment Ingestion Rate	0.003	kg sediment, dw/d	Calculated	Calculated using daily dietary ingestion rate
Home Range	0.4	ha	NatureServe, 2018 [1]	Average in California
Hawaiian stilt (*Himantopus mexicanus knudseni*)				
Body weight	0.166	kg, ww	NatureServe, 2018 [1]	Average weight for similar species black-necked stilt
Dietary Ingestion Rate	0.060	kg diet, ww/d	Nagy, 2001	Allometric Equation for insectivorous birds. g FMI/d DFI = 1.633 × [BW (g)^0.705] x 0.001 (kg/g).
Diet Composition, Aquatic Invertebrates	1.00	kg food, ww/kg diet, ww	NatureServe, 2018 [1]	Eats various aquatic organisms--worms, small crabs, insects, small fishes
Aquatic Invertebrate Ingestion Rate	0.06	kg ww/d	Calculated	Calculated using daily dietary ingestion rate
Diet Composition, Sediment (dw diet)	0.17	kg sediment, dw/kg diet, dw	Stilt sandpiper used as a surrogate Table 4-4 of USEPA (1993)	Hawaiian stilt frequent mudflats, feeds in tidal wetlands (NatureServe, 2018 [1])
Aquatic Invertebrate Moisture Content	0.78	kg water/kg, ww	Aquatic invertebrates (average of crabs, isopods/amphipods, cladocerans) Table 4-1 of USEPA (1993)	Used as basis for moisture content of diet for converting sediment ingestion from dry weight basis to wet weight basis
Diet Composition, Sediment (ww diet)	0.04	kg sediment, dw/kg diet, ww	Calculated	Normalized to wet weight diet
Sediment Ingestion Rate	0.002	kg ww/d	Calculated	Calculated using daily dietary ingestion rate
Home Range	300	ha	Black-necked stilt used as a surrogate; NatureServe, 2018 [1]	Upper estimate of breeding home range

Notes:
1: accessed at: http://explorer.natureserve.org

Abbreviations:
bw = body weight
d = day
dw = dry weight
kg =Kilogram
ww = wet weight

Appendix A: References

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APPENDIX B

Compilation of Bioaccumulation Metrics for PFAS

Appendix B-1: Sources of Data for PFAS Sediment and Organic Carbon Partition Coefficients (Kd, KOC)

Study	Setting	Solid Phase	Source of Aqueous Media	PFAS Source	PFAS																		Supporting Data				
					PFBA	PFPeA	PFHxA	PFHpA	PFOA	PFNA	PFDA	PFUnDA	PFDoDA	PFTrDA	PFTeDA	PFBS	PFHxS	PFOS	PFDS	PFOSA	N-EtFOSAA	N-MeFOSAA					
				Carbon Chain Length	C4	C5	C6	C7	C8	C9	C10	C11	C12	C13	C14	C4	C6	C8	C10	C8	C8	C8	Organic Carbon	Grain Size	pH	Cation or Anion Exchange Capacity	Salinity
				Number of Perfluorinated Carbon Atoms	3	4	5	6	7	8	9	10	11	12	13	4	6	8	10	8	8	8					
Ahrens et al., 2009	Field Sampling	Sediment	Field Collected Porewater	Field sediment with PFAS																	X		X				
Ahrens et al., 2011	Lab	Sediment	Laboratory water added to spiked sediment	Spiked sediment					X									X		X			X				
Chen et al., 2013	Lab	Soil	Laboratory water added to spiked soil	Spiked soil					X									X					X			X	
Chen et al., 2015	Field Sampling	Sediment/Suspended Sediment	Field Collected Overlaying Water	Field sediment with PFAS			X	X	X	X	X	X	X			X	X	X		X			X				
Chen et al., 2016	Lab	Sediment	Laboratory water added to spiked sediment	Spiked sediment					X	X	X						X	X					X	X	X		
Guelfo and Higgins, 2013	Lab	Soil	Laboratory water added to spiked soil	Spiked soil	X	X	X	X	X	X	X					X	X	X					X	X	X	X	
Higgins & Luthy, 2006	Lab	Sediment	Laboratory water added to spiked sediment	Spiked sediment					X	X	X	X						X	X		X	X	X	X	X	X	
Hong et al., 2013	Field Sampling	Sediment/Suspended Sediment	Field Collected Overlaying Water	Field sediment with PFAS					X	X	X	X	X					X	X				X		X		X
Joen et al., 2011	Lab	Sediment	Laboratory water added to spiked sediment	Spiked sediment					X									X					X				X
Kwadijk et al., 2010	Field Sampling	Sediment	Field Collected Overlaying Water	Field sediment with PFAS					X	X	X					X	X	X									
Labadie & Chevreuil, 2011	Field Sampling	Sediment	Field Collected Overlaying Water	Field sediment with PFAS		X	X	X	X	X	X	X	X	X	X	X	X	X	X				X				
Lasier et al., 2011	Field Sampling	Sediment	Field Collected Overlaying Water	Field sediment with PFAS			X	X	X	X	X					X	X	X					X				
Li et al., 2012	Lab	Sediment	Laboratory water added to spiked sediment	Spiked sediment					X														X	X	X	X	
Munoz et al., 2015	Field Sampling	Sediment	Field Collected Overlaying Water	Field sediment with PFAS				X	X	X	X	X	X	X			X	X	X	X			X	X			
Pan & You, 2010	Field Sampling	Sediment	Field Collected Overlaying Water	Field sediment with PFAS														X					X	X	X		X
Pico et al., 2012	Field Sampling	Sediment	Field Collected Overlaying Water	Field sediment with PFAS		X	X	X	X	X	X					X		X	X				X		X		X
Xiang et al., 2018	Lab	Soil	Laboratory water added to spiked soil	Spiked soil					X														X	X		X	
Yang et al., 2011	Field Sampling	Sediment	Field Collected Overlaying Water	Field sediment with PFAS					X								X	X					X				
Zhang et al., 2012	Field Sampling	Sediment	Field Collected Overlaying Water	Field sediment with PFAS	X	X	X	X	X	X	X	X	X					X					X				
Zhao et al., 2012	Lab	Sediment	Laboratory water added to spiked sediment	Spiked sediment		X		X	X	X	X		X			X	X	X					X	X			

Appendix B-2: Sources of Data for Uptake Parameters for Aquatic Bioaccumulation of PFAS - Aquatic Invertebrates

Study	Exposure Setting	PFAS Source	Primary Exposure Medium	Biota: Type of organism(s) evaluated in study	PFAS																		Parameters Provided or that can be Derived		
					PFBA	PFPeA	PFHxA	PFHpA	PFOA	PFNA	PFDA	PFUnDA	PFDoDA	PFTrDA	PFTeDA	PFBS	PFHxS	PFOS	PFDS	PFOSA	EtFOSAA	MeFOSAA	BCF-Pl	BAF-Pl	BSAF-Bl [1,2]
				Carbon Chain Length	C4	C5	C6	C7	C8	C9	C10	C11	C12	C13	C14	C4	C6	C8	C10	C8	C12	C11			
				Number of Perfluorinated Carbon Atoms	3	4	5	6	7	8	9	10	11	12	13	4	6	8	10	8	8	8			
Bertin et al., 2014	Lab (w/ field-collected sediments from Rhone River, France)	Fluoropolymer manufacturing plant	Sediment, diet	Midge larvae								X	X	X	X			X		X					X
Bertin et al., 2016	Lab (w/ field-collected sediments from Rhone River, France)	Fluoropolymer manufacturing plant	Sediment	Benethic species (gammarids, fresh water amphipods)						X		X	X	X				X		X	X	X			X
Bertin et al., 2018	Lab (w/ field-collected sediments from Beurre Island, France)	Fluoropolymer manufacturing plant	Sediment	*Chironomus riparius* larvae (midge)										X											X
Chen et al., 2018	Lab	Spiked	Water	Nematodes												X		X					X		
Coffey Environments Australia Pty, Ltd., 2018	Field (RAFF Base Darwin Australia)	Airport: AFFF	Water	Molluscs and crustaceans					X								X	X						X	
Dai et al., 2013	Lab	Single compounds spike	Water	Water flea (*Daphnia magna*)					X	X	X	X	X					X					X		
de Solla et al., 2012	Field (Ontario, Canada)	Airport (potentially AFFF)	Water	Amphipods, shrimp		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X				X	
Fang et al., 2014	Field (Taihu Lake, China)	Unknown	Water	Phytoplankton. zooplankton, invertebrates, white shrimp, freshwater mussel, pearl mussel			X	X	X	X	X	X	X			X	X	X		X				X	
Gomez et al, 2011	Field (Northern Spain)	Urban wastewater, industrial waste	Water, sediment	Mussels					X	X						X	X	X						X	X
Hazelton et al., 2012	Laboratory	Spiked	Water	Mussels														X					X		
Higgens et al., 2007	Lab	spiked	Sediment	*Lumbriculus variegatus* blackworm					X	X	X	X	X					X	X		X				X
Hong et al., 2015	Field (West Coast of S. Korea)	Unknown	Water	Various aquatic invertebrates (shrimp, crab, bivalve, gastropod)	X	X	X	X	X	X	X	X				X	X	X	X					X	
Houde et al., 2006	Field (ocean food web around Charleston Harbor, SC and Sarasota Bay, FL)	WWTP	Water, sediment	Zooplankton					X	X	X	X	X				X	X		X				X	X
Houde et al., 2008	Field (Lake Ontario food web)	Unknown	Water, sedimient	Zooplankton, *Mysis, Diporeia*														X						X	X
Jacobson et al., 2010	Lab (with field collected samples from Lake Mälaren)	Spiked	Water, sediment	Amphipod														X					X		X
Jeon et al., 2010a	Lab (with field collected samples from Korea)	Spiked	Water, diet	Pacific oyster					X		X	X						X					X		
Kannan et al., 2005	Field (Laurentian Great Lakes food web)	Unknown	Water	Amphipods, crayfish, zebra mussel					X								X	X		X				X	
Kobayashi et al., 2018	Field (estuary of Omuta River, Japan)	Unknown	Water	Sea snail		X	X	X	X	X	X	X					X	X						X	
Lam et al., 2014	Field (6 major rivers and lakes in S. Korea)	domestic/industrial WWTP	Water, sediment	Phytoplankton, zooplankton			X	X	X	X	X	X	X				X	X	X					X	X
Lam et al., 2017	Field (major river basins in Vietnam)	Unknown	Water, sediment	Gastropod (golden apple snail), and bivalve (golden freshwater clam), crustaceans (paddle crab, giant prawn)			X	X	X	X	X	X	X				X	X						X	X
Lasier et al., 2011	Field/Lab (Conasauga, Oostanaula and Coosa Rivers, Georgia)	airport/ manufacturing facility for local carpet industry	Water, sediment	Oligochaetes (worms)				X	X	X	X	X	X	X	X	X		X							X
Lescond et al., 2015	Field (Resolute Bay, Nunavut, Canada.)	Airport (potentially AFFF)	Water, sediment	Benthic and pelagic invertebrates			X		X		X						X			X				X	X
Liu et al., 2011	Lab	Single compounds spike	Water	Green mussels					X	X	X							X					X		
Liu et al., 2018	Field (Lake Chaohu, China)	industrial wastewater	Water	Shrimp, snail	X	X	X	X	X	X	X	X	X			X	X	X						X	X
Loi et al., 2011	Field (Mai Po Marshes Nature Reserve, Hong Kong)	Unknown	Water, sediment, diet	Gastropods, marine worms, shrimps, sand prawn, phytoplankton					X	X	X	X	X		X		X	X			X			X	X
Munksgaard et al., 2016	Field (Darwin Harbour, Northern Territory)	Airport, AFFF	Sediment	Red claw yabbies, cockles, oysters and mudmussels	X	X	X	X	X	X	X	X	X			X	X	X							X
Munoz et al., 2017	Field (macrotidal estuary, Gironde, SW France)	Unknown	Diet, intertidal sediment, subtidal water	Benthic food web (shrimp, oyster, copepods mysids, ragworm, gammarids, etc.)				X	X	X	X	X	X	X	X		X	X	X	X	X	X		X	X
Naile et al., 2010	Field (Western Coast of Korea)	Unknown	Water	Surf clam, oyster, asian periwinkle, crab, mussel, gastropod	X		X	X	X	X	X	X	X			X	X	X	X					X	

Appendix B-2: Sources of Data for Uptake Parameters for Aquatic Bioaccumulation of PFAS - Aquatic Invertebrates

Study	Exposure Setting	PFAS Source	Primary Exposure Medium	Biota: Type of organism(s) evaluated in study	PFAS																		Parameters Provided or that can be Derived		
					PFBA	PFPeA	PFHxA	PFHpA	PFOA	PFNA	PFDA	PFUnDA	PFDoDA	PFTrDA	PFTeDA	PFBS	PFHxS	PFOS	PFDS	PFOSA	EtFOSAA	MeFOSAA	BCF-PI	BAF-PI	BSAF-BI [1,2]
				Carbon Chain Length	C4	C5	C6	C7	C8	C9	C10	C11	C12	C13	C14	C4	C6	C8	C10	C8	C12	C11			
				Number of Perfluorinated Carbon Atoms	3	4	5	6	7	8	9	10	11	12	13	4	6	8	10	8	8	8			
Naile et al., 2013	Field (estuarine and coastal areas along west coast, Korea)	Unknown	Water, sediment, soil	Crab, gastropods, and bivalves				X	X	X	X	X				X	X	X						X	X[3]
Prosser et al., 2016	Field (Creek in southwestern Ontario)	Airport and Manufacturing of PFAS	Sedements	Mayflies				X	X	X	X	X	X		X		X	X	X	X					X
Quinete et al., 2009	Field (Rio de Janeiro, Brazil)	Industrial, agricultural and highway runoff, domestic sewage	Water	Mussels				X	X			X						X	X					X	
Wen et al., 2016	Lab	Single compounds spike	Water	*Chironomus plumosus* larvae					X	X	X	X	X					X					X		
Wilkinson et al., 2018	Field	Sewage treatment works	Aquatic sediment, water, periphyton/biofilm	Amphipod crustaceans (*Gammarus pulex*), and aquatic snails					X	X						X		X						X	X
Xu et al., 2014	Field (Taihu Lake, China)	Unknown	Surface water	Zoobenthos, white shrimp, zooplankton, phytoplankton					X	X	X	X	X					X						X	

Notes:
1) BSAFs can be used to calculate BAFs if organic carbon (OC) content is provided. This table can be used to find that information, but this table focuses on BSAFs, as they are the best measure of accumulation.
2) BSAFs are for sediment unless otherwise noted.

Acronyms:
AFFF = Aqueous film forming foam
BAF = Bioaccumulation Factor
BCF = Bioconcentration Factor
BMF = Biomagnification Factor
BSAF = Biota-Sediment/Soil Accumulation Factor
BSAF-BI = BSAF Benthic Invertebrate
PFAS = Per and polyfluoroalkyl substances
WWTP = Wastewater treatment plant

Appendix B-3: Sources of Data for Uptake Parameters for Aquatic Bioaccumulation of PFAS - Fish

Study	Exposure Setting	PFAS Source	Primary Exposure Medium	Biota: Type of organism(s) evaluated in study	PFAS																	Parameters Provided or that can be Derived			
					PFBA	PFPeA	PFHxA	PFHpA	PFOA	PFNA	PFDA	PFUnDA	PFDoDA	PFTrDA	PFTeDA	PFBS	PFHxS	PFOS	PFDS	PFOSA	EtFOSAA	MeFOSAA	BCF	BAF	BMF	BSAF-Fish[4]
				Carbon Chain Length	C4	C5	C6	C7	C8	C9	C10	C11	C12	C13	C14	C4	C6	C8	C10	C8	C12	C11				
				Number of Perfluorinated Carbon Atoms	3	4	5	6	7	8	9	10	11	12	13	4	6	8	10	8	8	8				
3M, 2003	Lab	Spiked	Water	Bluegill Sunfish														X					X			
Babut et al., 2017	Field (Rhone River, France)	Fluoropolymer manufacturing plant	Diet	3 cyprinid species					X	X	X	X	X	X	X		X	X	X	X					X	
Becker et al., 2010	Field (river Upper Franconia, Germany)	Municipal waste water treatment plant	WWTP-impacted water	Fish (chub and river goby)					X									X						X		
Campo et al., 2015	Field (Llobregat River ecosystem, Mediterranean area, NE Spain)	Industrial waste discharge, wastewater discharge	Water, sediment	Fish (*M. salmoides, B. graellsii, C. carpio*)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X					X		X
Campo et al., 2016	Field (Jucar River basin, E. Spain)	Industrial, sewage treatment plants, agriculture	Water, sediment	Fish Species (various - see notes)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X					X		X
Chen et al., 2016	Lab	Spiked	Water	Zebrafish	X		X		X	X	X	X	X	X	X	X	X	X					X			
Coffey Environments Australia Pty, Ltd., 2018	Field (RAFF Base Darwin Australia)	Airport: AFFF	Water	Freshwater fish					X								X	X						X		
Daikin, 2000	Lab	Spiked	Water	Common carp					X														X			
de Solla et al., 2012	Field (Ontario, Canada)	Airport (potentially AFFF)	Water	Sunfish, bullhead		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X				X		
Falk et al., 2015	Lab	Spiked	Diet	Rainbow trout					X	X						X	X	X							X	
Fang et al., 2014	Field (Taihu Lake, China)	Unknown	Water	Fish (minnow, carp, whitebait, crucian, cutler, mud fish, bitterling, and gobies)			X	X	X	X	X	X	X			X	X	X		X				X	X	
Fang et al., 2016	Lab	Spiked	Water/Sediment	Carp (*Cyprinus carpio*)					X	X	X	X	X					X						X		
Furdui et al., 2007	Field (Great Lakes)	Spiked	Water	Fish (lake trout)					X	X	X						X	X		X				X		
Goeritz et al., 2013	Lab	Spiked	Diet	Fish (market-sized raindbow trout)					X	X						X	X	X							X	
Haukås et al., 2007	Field (Barents Sea)	Unknown	Diet (ice amphipod)	Polar cod														X							X	
Hong et al., 2015	Field (West Coast of S. Korea)	Unknown	Water	Various fish species	X	X	X	X	X	X	X	X				X	X	X	X					X		
Houde et al., 2006	Field (ocean food web around Charleston Harbor, SC and Sarasota Bay, FL)	WWTP	Water, sediment	Charleston Harbor area: Atlantic croaker, pinfish, red drum, spotfish, spotted seatrout, striped mullet Sarasota Bay area: sheephead, pigfish, pinfish, striped mullet, spotted seatrout					X	X	X	X	X				X	X		X				X	X	X
Houde et al., 2008	Field (Lake Ontario food web)	Unknown	Water, sediment	Smelt, sculpin, lake trout, alewife														X						X	X	X
Inoue et al., 2012	Lab	Spiked	Water	Carp (*Cyprinus carpio L.*)					X			X	X		X			X					X			
Jeon et al., 2010b	Lab	Spiked	Water	Blackrock fish					X		X	X						X					X			
Kannan et al., 2005	Field (Laurentian Great Lakes food web)	Unknown	Water	Round goby, smallmouth bass					X									X		X				X	X	
Kelly et al., 2009	Field (piscivorous food web in Hudson Bay region of northeastern Canada)	Unknown	Sediments	Fish, seaducks				X	X	X	X	X	X	X				X		X					X	X
Kobayashi et al., 2018	Field (estuary of Omuta River, Japan)	Unknown	Water	Javeline goby, yellowfin goby, grey mullet, sea bass.		X	X	X	X	X	X	X					X	X						X		
Kwadijk et al., 2014	Field (Schiphol Amsterdam Airport)	AFFF release	Water, sediment	Eel, pike and perch													X	X						X		X
Labadie and Chevreuil, 2011	Field (Orge River near Paris, France)	Urban runoff/ sewage discharge	Water, sediment	Fish (European chub)				X	X	X	X	X	X	X	X	X	X	X	X					X		X
Lam et al., 2014	Field (6 major rivers and lakes in S. Korea)	Domestic/industrial WWTP	Water, sediment	Crucian carp and mandarin fish (blood,liver)			X	X	X	X	X	X	X				X	X	X					X		X
Lam et al., 2017	Field (major river basins in Vietnam)	Unknown	Water, sediment	Fish (tilapia, stripped snakehead, dusky sleeper, shark catfish, flying barb)			X	X	X	X	X	X	X				X	X						X		X
Lanza et al., 2017	Field (Barksdale Air Force Base Louisiana; Coopers Bayou and Macks Bayou)	Airport: AFFF	Water	Sunfish													X	X						X		
Lescond et al., 2015	Field (Resolute Bay, Nunavut, Canada.)	Airport (potentially AFFF)	Water, sediment	Canadian Arctic Char			X		X		X						X			X				X	X	X

Appendix B-3: Sources of Data for Uptake Parameters for Aquatic Bioaccumulation of PFAS - Fish

Study	Exposure Setting	PFAS Source	Primary Exposure Medium	Biota: Type of organism(s) evaluated in study	PFAS																	Parameters Provided or that can be Derived			
					PFBA	PFPeA	PFHxA	PFHpA	PFOA	PFNA	PFDA	PFUnDA	PFDoDA	PFTrDA	PFTeDA	PFBS	PFHxS	PFOS	PFDS	PFOSA	EtFOSAA	MeFOSAA	BCF	BAF	BMF	BSAF-Fish[4]
				Carbon Chain Length	C4	C5	C6	C7	C8	C9	C10	C11	C12	C13	C14	C4	C6	C8	C10	C8	C12	C11				
				Number of Perfluorinated Carbon Atoms	3	4	5	6	7	8	9	10	11	12	13	4	6	8	10	8	8	8				
Martin et al., 2003b	Lab	Spiked	Water	Rainbow trout	(2)	(2)	(2)	(2)	X	(2)	X	X	X	(2)	X		X	X	(2)	(2)	(2)	(2)	X			
Martin et al., 2003a	Lab	Spiked	Diet	Juvenile rainbow trout	(1)	(1)	(1)	(1)	X	(1)	X	X	X	(1)	X	(1)	X	X	(1)	(1)	(1)	(1)			X	
Martin et al., 2004	Field (Lake Ontario food web)	Unknown	Water, sediment, diet	Top predator fish, lake trout, forage fish					X	X	X	X	X	X	X			X		X					X	
Munoz et al., 2017	Field (macrotidal estuary, Gironde, SW France)	Unknown	Sediment, Water, Diet	Fish (goby, anchovy, sprat, mullet, meagre, seabass, sole, flounder)				X	X	X	X	X	X	X	X		X	X	X	X	X	X		X	X	X
Naile et al., 2013	Field (estuarine and coastal areas along west coast, Korea)	Unknown	Water, sediment, soil	Fish				X	X	X	X	X				X	X	X						X		X
NICNAS, 2005	Lab	Spiked	Water	Bluegill												X							X			
Pan et al., 2014	Field (Pearl River Delta, China)	Unknown	Water	Tilapia, crucian carp, common carp, leather catfish, snakehead, grass carp, chub, mud carp, and bream					X	X	X	X						X	X	X	X			X		
Powley et al., 2008	Field (western Canadian Arctic)	Unknown	Diet	Arctic cod							X	X	X					X							X	
Prosser et al., 2016	Field (Creek in southwestern Ontario)	Airport and Manufacturing of PFAS	Sediments	Fathead minnow				X	X	X	X	X	X		X		X	X	X	X				X		X
Quinete et al., 2009	Field (Rio de Janeiro, Brazil)	Industrial, agricultural and highway runoff, domestic sewage	Water	Scabbardfish, whitemouth croaker, mullet				X	X			X						X	X					X		
Sakurai et al., 2013	Lab	Spiked	Water, sediment	Marbeled flounder														X					X			X
Shi et al., 2018	Field (China)	Municipal and industrial wastewater; fluoropolymer production facility	Water	Crucian carp	X	X	X	X	X	X	X	X				X	X	X		X				X		
Taniyasu et al., 2003	Field (Japan)	Unknown	Water	Fish												X	X	X						X		
Terechovs et al., 2019	Field (Shoalhaven region, Austrailia)	Reclaimed water	Water	Silver perch					X									X						X		
Thompson et al., 2011	Field (Sydney Harbor)	Urban/industrial area	Water, sediment	Fish (adult sea mullet)			X	X	X	X	X	X	X	X	X	X	X	X	X					X		X
Tomy et al., 2004	Field (eastern Arctic marine food web)	Unknown	Diet	Arctic cod					X									X		X	X				X	
Wang et al., 2013	Field (captive breeding center in Anhui Research Center, China)	Unknown	Water	Whole body homogenates of six kinds of fish					X	X	X	X	X		X			X						X	X	
Wen et al., 2017	Lab	Spiked	Water	Adult female zebrafish	X	X	X	X	X	X	X	X				X		X					X			
Xu et al., 2014	Field (eutrophic freshwater food web, Taihu Lake, China)	Unknown	Surface water, surficial sediment	Carnivorous fish, omnivorous fish, herbivorous fish					X	X	X	X	X					X						X		

Notes:
1) Information for these PFAS can be calculated using a model presented by the study authors in Figure 5 of Martin et al 2003a.
2) Information for these PFAS can be calculated using a model presented by the study authors in Figure 5 of Martin et al 2003b.
3) BSAFs can be used to calculate BAFs if organic carbon (OC) content is provided. This table can be used to find that information.
4) BSAF-Fish is not a recommended parameter as it does not account for bioaccumulation from diet, but for completeness is shown in the table where both concentrations of sediment and fish tissue are presented.

Acronyms:
AFFF = Aqueous film forming foam
BAF = Bioaccumulation Factor
BCF = Bioconcentration Factor
BMF = Biomagnification Factor
BSAF-Fish = Biota-Sediment Accumulation Factor for Fish
PFAS = Per and polyfluoroalkyl substances

Appendix B-4: Sources of Data for Uptake Parameters for Bioaccumulation of PFAS - Terrestrial Invertebrates

Study	Exposure Setting	PFAS Source	Primary Exposure Medium	Biota: Type of organism(s) evaluated in study	PFAS																	Parameters Provided or that can be Derived
					PFBA	PFPeA	PFHxA	PFHpA	PFOA	PFNA	PFDA	PFUnDA	PFDoDA	PFTrDA	PFTeDA	PFBS	PFHxS	PFOS	PFDS	PFOSA	EtFOSAA	MeFOSAA	BSAF-TI [1,2]
				Carbon Chain Length Number of Perfluorinated Carbon Atoms	C4 3	C5 4	C6 5	C7 6	C8 7	C9 8	C10 9	C11 10	C12 11	C13 12	C14 13	C4 4	C6 6	C8 8	C10 10	C8 8	C12 8	C11 8	
Braunig et al., 2018	Field	AFFF from airports	Soil	Earthworms	X	X	X	X	X	X	X	X	X			X	x	x					x
D'Hollander et al., 2014	Field (Flandres, Belgium near fluorochemical plant)	Fluorochemical plant	Soil, surface water	Earthworms, isopods, millipedes, slugs														x					x
Groffen et al., 2019	Field (Antwerp, Belgium)	Fluorochemical plant	Soil	Isopods	X	X	X	X	X	X	X	X	X	X	X	X	x	x	x				x
Karnjanapiboonwong et al., 2018	Lab	Spiked	Soil	Earthworms				X		X						X	x						x
Navarro et al., 2016	Lab (w/field collected soils)	Biosolids-amended	Soil	Earthworms	X	X	X	X	X		X	X	X	X	X	X		x					x
Navarro et al., 2017	Lab	Spiked	Soil	Earthworms												X	x	x					x
Prosser et al., 2016	Field (Creek in southwestern Ontario)	Airport and Manufacturing of PFAS	Sediments	Earthworms				X	X	X	X	X	X		X		x	x	x	x			x
Rich et al., 2015	Lab (w/field collected soils)	Biosolids-amended; AFFF	Soil	Earthworms					X	X	X	X	X				x	x	x				x
Wen et al., 2015	Lab (w/ field-collected soil samples from Changping, Beijing)	Biosolids-amended fields	Soil	Earthworms					X									X					X[1]
Zhao et al, 2013a	Lab	Spiked	Soil	Earthworms			X	X	X	X	X	X	X			X	X	X					X
Zhao et al., 2014	Lab	Spiked	Soil	Earthworms		X	X	X	X	X	X	X	X			X	X	X					X
Zhao et al., 2016	Lab	Spiked	Soil	Earthworms														X		X	X		X
Zhao and Zhu, 2017	Lab	Spiked	Soil	Earthworms					X	X	X												X
Zhu and Kannan, 2019	Field (Ohio)	Fluoropolymer industry	Soil	Earthworms				X	X	X	X	X	X										X

Notes:
1) BSAFs can be used to calculate BAFs if organic carbon (OC) content is provided. This table can be used to find that information, but this table focuses on BSAFs, as they are the best measure of accumulation.
2) BSAFs are for soil unless otherwise noted.

Acronyms:
AFFF = Aqueous film forming foam
BAF = Bioaccumulation Factor
BCF = Bioconcentration Factor
BMF = Biomagnification Factor
BSAF = Biota-Sediment/Soil Accumulation Factor
BSAF-TI = BSAF Terrestrial Invertebrate
PFAS = Per and polyfluoroalkyl substances

Appendix B-5: Sources of Data for Uptake Parameters for Bioaccumulation of PFAS - Plants

Study	Exposure Setting	PFAS Source	Primary Exposure Medium	Biota: Type of organism(s) evaluated in study	PFAS																		Parameters Provided or that can be Derived Terrestrial Plants		Parameters Provided or that can be Derived Aquatic Plants	
					PFBA	PFPeA	PFHxA	PFHpA	PFOA	PFNA	PFDA	PFUnDA	PFDoDA	PFTrDA	PFTeDA	PFBS	PFHxS	PFOS	PFDS	PFOSA	EtFOSAA	MeFOSAA	BCF-TP	BAF-TP	BCF-AP	BAF-AP
				Carbon Chain Length	C4	C5	C6	C7	C8	C9	C10	C11	C12	C13	C14	C4	C6	C8	C10	C8	C12	C11				
				Number of Perfluorinated Carbon Atoms	3	4	5	6	7	8	9	10	11	12	13	4	6	8	10	8	8	8				
Bizkarguenaga et al., 2016	Lab (greenhouse)	Compost-amended	Soil	Carrot (*Chantenay* and *Nantesa* var of *Daucus carota* ssp *sativus*) and lettuce (*Golden Spring* var of *Lactuca sativa*)					X									X		X				X		
Blaine et al., 2013	Lab (greenhouse) / Field	Industrially/ municipal impacted soil	Biosolids-amended soil	Lettuce (*Lactuca sativa*) and tomato (*Lycopersicon lycopersicum*)	X	X	X	X	X	X	X					X	X	X	X					X		
Blaine et al., 2014a	Lab (greenhouse)	Industrially/ municipal impacted soil	Biosolids-amended soil	Radish (*Raphanus sativus*), celery (*Apium graveolens* var. *dulce*), tomato (*Lycopersicon lycopersicum*), and sugar snap pea (*Pisum sativum* var. *macrocarpon*)	X	X	X	X	X	X	X					X	X	X	X					X		
Blaine et al., 2014b	Lab (greenhouse)	Spiked	Reclaimed water	Lettuce (*Lactuca sativa*) and strawberry (*Fragaria ananassa*)	X	X	X	X	X	X						X	X	X						X		
Brignole et al., 2003	Lab	Spiked	Soil	Terrestrial plant species (onion, ryegrass, alfalfa, flax, lettuce, soybean, and tomato)														X						X		
Braunig et al., 2018	Field	AFFF from airports	Soil	Wheatgrass	X	X	X	X	X	X	X	X	X			X	X	X						X		
Chen, Lo, and Lee, 2012	Field (New Taipei City, Taiwan)	Spiked	Water	Aquatic plants (*Hygrophila pagonocalyx Hayata*, *Ipomoea aquatica Forssk*, *Ludwigia (x) taiwanensis*, and *Eleocharis dulcis*)					X									X								X
Felizeter et al., 2012	Lab (greenhouse)	Spiked	Water	Lettuce (*Lactuca sativa*)	X	X	X	X	X	X	X	X	X	X	X	X	X	X					X			
Felizeter et al., 2014	Lab (greenhouse)	Spiked	Water	Three hydroponically-grown crops (tomatoes, cabbage, and zucchinis)	X	X	X	X	X	X	X	X	X	X	X	X	X	X					X			
Gobelius et al., 2017	Field (Stockholm Arlanda airport, Sweden)	AFFF	Soil and groundwater	Roots, trunk/cores, twigs, and leaves/needles of silver birch, Norway spruce, bird cherry, mountain ash, ground elder, long beechfern, and wild strawberry	X	X	X	X	X	X	X	X	X	X		X	X	X	X					X		
Kelly et al., 2009	Field (piscivorous food web in Hudson Bay region of northeastern Canada)	Unknown	Sediments	Macroalgae				X	X	X	X	X	X	X				X		X						X
Krippner et al., 2015	Lab (climate controlled chamber)	Spiked	Soil	Maize (*Zea mays*)	X	X	X	X	X	X	X					X	X	X						X		
Lechner and Knapp, 2011	Lab (greenhouse)	Spiked	Contaminated sewage sludge-amended soil	Carrots, potatoes, and cucumbers					X									X						X		
Liu et al., 2017	Field (China)	Mega-fluorochemical industrial park	Soil, rainwater, irrigation water, river water	Wheat and corn grain	X	X	X	X	X	X	X	X	X			X	X	X						X		
Navarro et al., 2017	Lab	Spiked	Soil	Spinach, tomato, corn	X	X	X	X	X	X	X	X	X			X		X						X		
Pi et al., 2017	Controlled mesocosm experiments	Spiked	Water	Aquatic plants, submerged (*Echinodorus horemanii*) and one free-floating (*Eichhornia crassipes*)		X	X	X	X	X	X	X	X	X	X	X	X	X							X	
Stahl et al., 2009	Lab (controlled pot experiments)	Spiked	Soil	Maize, oats, ryegrass, potatoes, spring wheat					X									X						X		
Stahl et al., 2013	Field	Spiked	Soil (lysimeter experiment)	Plants (winter wheat, winter rye, canola, winter barley)	X	X	X	X	X							X	X	X						X		
Wen et al., 2013	Lab (hydroponic greenhouse experiments)	Spiked	Nutrient solution	Maize (*Zea mays*)					X									X					X			
Wen et al., 2014	Field (controlled field experiments)	Biosolids-amended	Soil	Wheat (*Triticum aestivum* L.)	X	X	X	X	X	X	X	X			X	X	X	X						X		

Appendix B-5: Sources of Data for Uptake Parameters for Bioaccumulation of PFAS - Plants

Study	Exposure Setting	PFAS Source	Primary Exposure Medium	Biota: Type of organism(s) evaluated in study	PFAS																		Parameters Provided or that can be Derived Terrestrial Plants		Parameters Provided or that can be Derived Aquatic Plants	
					PFBA	PFPeA	PFHxA	PFHpA	PFOA	PFNA	PFDA	PFUnDA	PFDoDA	PFTrDA	PFTeDA	PFBS	PFHxS	PFOS	PFDS	PFOSA	EtFOSAA	MeFOSAA	BCF-TP	BAF-TP	BCF-AP	BAF-AP
				Carbon Chain Length	C4	C5	C6	C7	C8	C9	C10	C11	C12	C13	C14	C4	C6	C8	C10	C8	C12	C11				
				Number of Perfluorinated Carbon Atoms	3	4	5	6	7	8	9	10	11	12	13	4	6	8	10	8	8	8				
Wilkinson et al., 2018	Field (Hogsmill, Blackwater and Bourne Rivers in southern England)	Sewage treatment works	Sediment, water, periphyton/biofilm	Aquatic plants (*Callitriche* and *Potamogeton*)					X	X						X		X								X
Yang et al., 2015	Lab	Spiked	Agar-solidified medium	*Arabidopsis thaliana* (mouse-ear cress)					X															X		
Yoo et al., 2011	Field (sludge-applied fields near Decatur, AL)	Biosolids-amended	Soil	Grasses (tall fescue, barley, bermuda grass, Kentucky bluegrass)			X	X	X	X	X	X	X	X	X	X	X	X						X		
Zhao et al., 2013b	Lab (controlled growth environment)	Spiked	Water	Wheat (*Triticum aestivum* L.)														X					X			
Zhao et al., 2014	Lab (controlled growth environment)	Spiked standards	Soil	Wheat (*Triticum aestivum* L.)		X	X	X	X	X	X	X	X			X	X	X						X		
Zhou et al., 2017	Field (Qing River, China); Lab (greenhouse hornwort bioaccumulation study)	Recycle water discharged from WWTPs	Sediments, water	Submerged (reed, *calamus, scirpus tabernaemontani* - whole plant) and emergenct aquatic plants (*stuckenia pectinata, hydrilla*, and hornwort - foliage and roots)	X	X	X	X	X	X	X	X	X			X	X	X								X
Zhou et al., 2016	Lab	Spiked	Soil	Wheat (*Triticum aestivum* L.)					X															X		
Zhao et al., 2018	Lab	Spiked	Quartz sand	Wheat, soybean, pumpkin														X		X	X		X			
Zhu and Kannan, 2019	Field (Ohio)	Fluoropolymer industry	Soil	Canada wildrye and sedges, five sumac species				X	X	X	X	X	X											X		

Acronyms:
AFFF = Aqueous film forming foam
BAF = Bioaccumulation Factor
BCF = Bioconcentration Factor
BMF = Biomagnification Factor
BSAF-Plant = Biota-Soil/Sediment Accumulation Factor for Plant
PFAS = Per and polyfluoroalkyl substances
WWTP = Wastewater Treatment Plant

Appendix B: References

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APPENDIX C

Compilation of Wildlife Toxicity Studies for PFAS

Appendix C-1: Sources of Data for PFAS Toxicity Reference Values (TRVs) for Mammals

Chemical[1]	Carbon Chain Length	Number of Perfluorinated Carbon Atoms	Study	Test Organism	Test Type	Duration (days)[2]	Measurement Endpoint[3]	Ecological Endpoint[4]	Total Study Score[6]
PFCAs									
PFBA	C4	3	Butenhoff, 2008	Rats	Sub-chronic	28	Ocular histology, bilateral pupillary reflex	N/A[5]	N/A
			Das et al., 2008	Mouse	Sub-chronic	294	Material and fetal developmental toxicity; reproduction, development, body weight, heptic	Reproduction, Growth	7
			van Otterdiijk, 2007a	Rats	Sub-chronic	28	Toxic potential (biochemistryistry, liver hypertrophy and thyroid alterations, hematological alterations) hepatic, biochemistry, body weight, respiratory, cardiovascular, immunological, reproduction, muscle/skeleton, endocrine, neurology	Reproduction, Growth	8
			van Otterdiijk, 2007b	Rats	Sub-chronic	90	Toxic potential (biochemistryistry, liver hypertrophy and thyroid alterations, hematological alterations) hepatic, biochemistry, body weight, respiratory, cardiovascular, immunological, reproduction, muscle/skeleton, endocrine, neurology	Reproduction, Growth	8
PFHxA	C6	5	Chengelis et al., 2009	Rat	Sub-chronic	90	Liver histopathology and liver weight;hepatocellular hypertrophy and liver weight increases (in high-dose M)	Growth	7
			Klaunig et al., 2015	Rat	Chronic	728	Death, hepatic, hematological, body weight, neurological, and renal	Survival	8
			NTP, 2018	Rat	Sub-chronic	28	Kidney weight, progressive nephropathy, liver weight, survival, body weight, reproduction	Growth	6
PFOA	C8	7	3M 1983 (See Butenhoff et al., 2012b)	Rat	Chronic	730	Reproduction, body weight, other tox endpoints	Reproduction, Growth	10
			Butenhoff et al., 2002	Monkey	Chronic	182	Increase in liver weight; body weight; hepatic injury	Growth	6
			Butenhoff et al., 2004; York et al., 2010	Rat	Sub-chronic	84	Increase in liver and kidney weight, decrease in body weight	Growth	7
			Butenhoff et al., 2004; York et al., 2010	Rat	Sub-chronic	112	Decrease in body weight and weight gain, increase in liver and kidney weights	Growth	7
			Butenhoff et al., 2004; York et al., 2010	Rat	Sub-chronic	127	No significant effects observed	N/A[5]	7
			Butenhoff et al., 2004; York et al., 2010	Rat	Sub-chronic	70	Delay in sexual materity, decrease in body weight and weight gain	Reproduction, Growth	7
			Butenhoff et al., 2012b (See 3M, 1983 for original)	Rat	Chronic	730	Decrease in body weight gain	Growth	10
			DeWitt et al., 2008	Mouse	Sub-chronic	15	Immune system: decrease IgM, increased IgG, decrease spleen weight	N/A[5]	8
			Goldenthal, 1978b	Monkey	Sub-chronic	90	Increase relative pituitary weight; body weight	Growth	7
			Goldenthal, 1978b	Monkey	Sub-chronic	90	Decrease in heart and brain weight	Growth	7
			Koskela et al., 2016	Mouse	Sub-chronic	119	Skeletal alterations in mice (from *in utero* and lactational exposure)	N/A[5]	7
			Lau et al., 2006	Mouse	Sub-chronic	17	Reduced pup ossification, accelerated male puberty	Growth	7
			Macon et al., 2011	Mouse	Sub-chronic	17	Delayed mammary gland development; pup liver and brain weight	Reproduction	6
			Onishchenko et al., 2011	Mouse	Sub-chronic	112	Neurodevelopmental effects (locomotion, muscle strength, exploratory behavior)	N/A[5]	N/A
			Perkins et al., 2004	Rat	Sub-chronic	91	Increase in liver weight with hepatocellular hypertrophy; body weight	Growth	9
			White et al., 2009; Wolf et al., 2006	Mouse	Sub-chronic	11	Post-natal weight gain of pups; mammary gland development	Growth	6
PFNA	C9	8	Das et al., 2015	Mouse	Sub-chronic	287	Development; hepatic; reproduction; body weight	Reproduction, Growth	6
			Fang et al., 2008	Mouse	Acute	14	Immunotoxicity	N/A[5]	N/A
			Fang et al., 2012a	Rat	Acute	14	Hepatic glycometabolism	N/A[5]	N/A
			Fang et al., 2012b	Rat	Acute	14	Cytotoxicity, hepatotoxicity	N/A[5]	N/A
			Rogers et al., 2014	Rat	Sub-chronic	365	Development; blood pressure; body weight	Growth	6
			Wolf et al., 2010	Mouse	Sub-chronic	18	Development; Reproduction	Reproduction	7
PFDA	C10	9	Harris and Birnbaum, 1989	Mouse	Sub-chronic	18	Maternal and fetal developmental toxicity (i.e., weight gain in pregnant mice, survival/develop of fetuses) body weight, development	Growth	7
			Kawashima et al., 1995	Rats	Acute	7	Hepatic responses; increased liver weight; growth (bd wt), hepatic	Growth	7
PFUnDA	C11	10	Takahashi et al., 2014	Rat	Sub-chronic	46	Repeated dose and reproductive / developmental toxicity	Reproduction	8
PFDoDA	C12	11	Ding et al., 2009	Rat	Chronic	110	Body chemistry, hepatotoxicity, hepatic (injury)	N/A[5]	5
			Kato et al., 2015	Rat	Sub-chronic	42	Reproduction, growth	Reproduction, Growth	8
			Shi et al., 2009	Rat	Sub-chronic	80	Endocrine, development, reproduction (F only)	Reproduction	7
			Shi et al., 2007	Rat	Acute	14	Reproduction (M only)	Reproduction	7
PFTeDA	C14	13	Hirata-Koizumi et al., 2015	Rat	Sub-chronic	42	Reproduction, development	Reproduction	8

Appendix C-1: Sources of Data for PFAS Toxicity Reference Values (TRVs) for Mammals

Chemical[1]	Carbon Chain Length	Number of Perfluorinated Carbon Atoms	Study	Test Organism	Test Type	Duration (days)[2]	Measurement Endpoint[3]	Ecological Endpoint[4]	Total Study Score[6]
PFSAs									
PFBS	C4	4	3M, 2001	Rat	Sub-chronic	28	Reproductive/development/growth; hematological, hepatic, body weight, respiratory, cardiovascular,gastrological, immunological, reproduction, muscle/skeleton, endocrine, neurology	Reproduction, Growth	NS
			Bijland et al., 2011	Mouse	Sub-chronic	42	Lipoprotein metabolism; lipolytic activity, hepatic	N/A[4]	6
			Lieder et al., 2009b	Rat	Chronic	120	Liver function; reproductive and developmental effects; mult-generational study, reproduction, development (pups), body weight, litter outcomes, estrogen, hepatic, gastrological	Reproduction,Growth	7
			Lieder et al., 2009a	Rat	Sub-chronic	90	Development/growth/function; hepatic, hematological, body weight, endocrine, immunological, neurology, reproduction, muscle/skeleton, gastrological, cardiovascular, respiratory.	Growth	7
PFHxS	C6	6	Bijland et al., 2011	Mouse	Sub-chronic	42	Lipoprotein metabolism; lipolytic activity, hepatic	N/A[5]	6
			Butenhoff et al., 2009b	Rat	Sub-chronic	56	Determine potential reproductive, developmental, and neurological responses to treatment, reproduction, development (F), neurological, immunological, endocrine (F) hepatic, gastrological, cardiovascular, respiratory, body weight, hematological (F)	Reproduction, Growth	8
			Chang et al., 2018	Mouse	Sub-chronic	77	Reproductive and developmental toxicity; hepatic, neurological, hemato, body weight	Reproduction, Growth	8
PFOS	C8	8	Butenhoff et al., 2009a	Rat	Sub-chronic	41	Maternal body weight (lactation); developmental neurotoxicity: increase motor activity, decrease habituation	Growth	8
			Case et al., 2001	Rabbit	Acute	20	Birth weight and delayed ossification	Growth	7
			Christian et al., 1999	Rat	Sub-chronic	84	Gestation length and pup viability, reduced birth weight	Growth, Survival, Reproduction	8
			Dong et al., 2009	Mouse	Sub-chronic	60	Increase liver weight, increase splenic NK cell activity, decrease SRBC response	Growth	7
			Goldenthal et al., 1978a	Rat	Sub-chronic	90	Body weight; increase liver weight, hepatocyte hypertrophy	Growth	6
			Goldenthal et al., 1979	Monkey	Sub-chronic	90	Diarrhea, anorexia	N/A[5]	5
			Lau et al., 2003; Thibodeaux et al., 2003	Rat	Sub-chronic	19	Mortality, decreased body weight	Growth, Survival	7
			Long et al., 2013	Mouse	Sub-chronic	90	Neurological, impaired spatial learning and memory; apoptosis in hippocampal cells	N/A[5]	N/A
			Luebker et al., 2005a	Rat	Sub-chronic	63	Decreased maternal body weight, decreased pup weight, decrease gestational length	Growth	7
			Luebker et al., 2005b	Rat	Sub-chronic	84	Decreased pup body weight	Growth	7
			Onishchenko et al., 2011	Mouse	Sub-chronic	112	Neurodevelopmental effects (locomotion, muscle strength, exploratory behavior)	N/A[5]	N/A
			Seacat et al., 2002	Monkey	Sub-chronic	182	Increase in liver weight, decrease in body weight	Growth	8
			Seacat et al., 2003	Rat	Sub-chronic	98	Increase liver weight, decrease cholesterol (M), increase liver hypertrophy	Growth	9
			Thomford 2002; Butenhoff et al., 2012a	Rat	Chronic	735	Decreased body weight, liver function	Growth	9
PFOSA	C8	8	Seacat and Luebker, 2000	Rat	Acute	29	Body weight; hepatic (liver weight); liver and serum analyses	Growth	7

Footnotes:

1. Due to the number of mammalian toxicity studies available in the literature for PFOS and PFOA, only the sub-chronic and chronic studies were included in this bibliography table. For other, less common PFAS all studies were included.
2. The longest test duration was reported.
3. All test measurement endpoints were reported.
4. Only the endpoints considered to be significant ecological endpoints (Survival, Growth, or Reproduction) were reported.
5. The tested and reported endpoint did not represent a signficant ecological endpoint and is not appropriate for deriving a toxicity value for T&E species. However, the study was retained in this bibliography table for informational purposes only and was not used in the selection process.
6. The scoring approach is described in Section 3.4.2 and the scoring rubric is provided in Table 3.

Acronyms:

F = female
M = male
N/A - not applicable
NS- not scored
PFOA - Perfluorooctanoic acid
PFOS - Perfluorooctane sulfonic acid
PFBA - perfluorobutanoic acid
PFBS - perfluorobutane sulfonic acid
PFNA - Perfluorononanoic acid
PFOSA - Perfluorooctane sulfonamide
PFHxS - Perfluorohexane sulfonic acid
PFHxA - Perfluorohexanoic acid
PFUnDA - Perfluoroundecanoic acid
PFDoA - Perfluorodecanoic acid
PFTeDA - Perfluorotetradecanoic acid

Appendix C-2: Sources of Data for PFAS Toxicity Reference Values (TRVs) for Birds

Chemical[1]	Carbon Chain Length	Number of Perfluorinated Carbon Atoms	Study	Test Organism	Test Type	Duration (days)[2]	Measurement Endpoint[3]	Ecological Endpoint[4]	Total Study Score[5]
PFCAs									
PFHxA	C6	5	Cassone et al., 2012	Fertilized White Leghorn chicken (*Gallus gallus domesticus*) eggs	Acute	22	*In ovo* exposure effects (embryo survival/development)	Reproduction	6
PFOA	C8	7	Yueng et al. 2009 [6]	1 day old amle chickens (*Gallus gallus*)	Acute	21	Growth, organ weight, histological and plasma biochemical parameters	Growth	4
			Nordén et al., 2016	White Leghorn chicken (*G. gallus domesticus*); great cormorant *(P. carbo sinensis*); herring gull (*L. argentatus*)	Acute	26	Embryo survival, body weight, and liver/heart weights	Reproduction, Growth	6
PFDA	C10	9	Yueng et al. 2009 [6]	1 day old amle chickens (*Gallus gallus*)	Acute	21	Growth, organ weight, histological and plasma biochemical parameters	Growth	4
PFSAs									
PFBS	C4	4	Gallagher et al., 2005	Northern Bobwhite Quail (*Colinus virginianus*)	Chronic	147	Development (adults) and reproductive	Reproduction	9
			Newsted et al., 2008	Bobwhite quail (Colinus virginianus)	Acute and chronic	17; 147	Dietary acute study (lethal and nonlethal endpoints); reproductive dietary chronic study	Reproduction	9
			Newsted et al., 2008	Mallard (*Anas platyrhynchos*)	Acute	17	Dietary acute study (lethal and nonlethal endpoints)	Survival	9
PFHxS	C6	6	Cassone et al., 2012	Fertilized White Leghorn chicken (*Gallus gallus domesticus*) eggs	Acute	22	*In ovo* exposure effects (embryo survival/development)	Reproduction	7
PFOS	C8	8	Gallagher et al., 2004a	Mallard Duck (juvenile)	Acute	5	Body weight	Growth	8
			Gallagher et al., 2004b	Northern Bobwhite Quail (juvenile)	Acute	5	Body weight	Growth	8
			Gallagher et al., 2003a (original 3M Final report; see Newsted et al., 2007)	Mallard	Chronic	147	Reproductive	Reproduction	9
			Gallagher et al., 2003b (original 3M Final report; see Newsted et al., 2007)	Northern Bobwhite Quail	Chronic	147	Reproductive	Reproduction	9
			McNabb et al., 2005	Quail (bobwhite and Japanese, adults)	Acute	7; 14	Thyroid (hyperthyroidism)	N/A[7]	2
			Molina et al., 2006	Leghorn chicken (*Gallus domesticus*) eggs	Acute	7	Reduced hatching success	Reproduction	5
			Newsted et al., 2005	Bobwhite quail *(Colinus virginianus*); Mallard (*Anas platyrhynchos*)	Acute and chronic	22; 147	Acute: mortality, growth, behavior, feed consum; Chronic: Reproductive	Growth, Reproduction, Survival	6
			Newsted et al., 2006 (See also Gallagher et al., 2004b - original 3M acute study)	Bobwhite quail (Colinus virginianus)	Acute	5	Body weight (also assessed for feed consum, behavior, physical injury, mortality, gross abnormalities, liver weight, and concentrations in blood serum and liver)	Growth	9

Appendix C-2: Sources of Data for PFAS Toxicity Reference Values (TRVs) for Birds

Chemical[1]	Carbon Chain Length	Number of Perfluorinated Carbon Atoms	Study	Test Organism	Test Type	Duration (days)[2]	Measurement Endpoint[3]	Ecological Endpoint[4]	Total Study Score[5]
PFOS (cont'd)	C8	8	Newsted et al., 2006 (See also Gallagher et al., 2004a - original 3M acute study)	Mallard (*Anas platyrhynchos*)	Acute	5	Body weight (also assessed for feed consum, behavior, physical injury, mortality, gross abnormalities, liver weight, and concentrations in blood serum and liver)	Growth	9
			Newsted et al. 2007 (Gallagher et al., 2003a,b 3M reports)	Mallard	Chonic	147	Body/liver weight, feeding, gross morphology and histology, reproduction and development	Growth, Reproduction	9
			Newsted et al., 2007 (Gallagher et al., 2003a,b 3M reports)	Northern Bobwhite Quail	Chronic	147	Body/liver weight, feeding, gross morphology and histology, reproduction and development	Growth, Reproduction	9
			Nordén et al., 2016	White Leghorn chicken (*G. gallus domesticus*); great cormorant (*P. carbo sinensis*); herring gull (*L. argentatus*)	Acute	26	Embryo survival, body weight, and liver/heart weights	Reproduction, Growth	6
			Yueng et al. 2009 [6]	1 day old amle chickens (*Gallus gallus*)	Acute	21	Growth, organ weight, histological and plasma biochemical parameters	Growth	4

Notes

1. All available PFAS avian studies were included.
2. The longest test duration was reported.
3. All test measurement endpoints were reported.
4. Only the endpoints considered to be significant ecological endpoints (Survival, Growth or Reproduction) were reported.
5. The scoring approach is described in Section 3.4.2 and the scoring rubric is provided in Table 3.
6: Not single chemical exposure study; organisms exposed to PFOA/PFDA/PFOS mixture
7. The tested and reported endpoint did not represent a signficant ecological endpoint and is not appropriate for deriving a toxicity value for T&E species. However, the study was retained in this bibliography table for informational purposes only and was not used in the selection process.

Acronyms

N/A - not applicable
PFOA - Perfluorooctanoic acid
PFHxA - Perfluorohexanoic acid
PFOS - Perfluorooctane sulfonic acid
PFBS - Perfluorobutane sulfonic acid
PFHxS - Perfluorohexane sulfonic acid

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APPENDIX D

Aquatic Life

APPENDIX D-1

Compilation of Aquatic Life Studies for PFAS

Appendix D-1: Compilation of Freshwater Aquatic Life Toxicity Studies

Study	Species	Taxon/Common Name	Exposure Duration (days)	Acute or Chronic	PFAS Included in Study	PFBA	PFPeA	PFHxA	PFHpA	PFOA	PFNA	PFDA	PFUnDA	PFDoDA	PFTrDA	PFTeDA	PFBS	PFHxS	PFOS	PFDS	PFOSA	Et-FOSAA	Me-FOSAA
					Number of Carbon Atoms	C4	C5	C6	C7	C8	C9	C10	C11	C12	C13	C14	C4	C6	C8	C10	C8	C12	C11
					Number of Perfluorinated Carbon Atoms	3	4	5	6	7	8	9	10	11	12	13	4	6	8	10	8	12	11
					Ecological Endpoint																		
3M, 2000	*Pseudokirchneriella subcapitata*	Microalga	4	Chronic	Growth														X				
3M, 2000	*Pseudokirchneriella subcapitata*	Microalga	3	Chronic	Growth														X				
3M, 2000	*Pseudokirchneriella subcapitata*	Microalga	14	Chronic	Growth														X				
Boudreau et al., 2003a	*Chlorella vulgaris*	Green microalgae	4	Chronic	Growth														X				
Boudreau et al., 2003a	*Pseudokirchneriella subcapitata*	Microalga	4	Chronic	Growth														X				
Colombo et al., 2008	*Pseudokirchneriella subcapitata*	Microalga	4	Chronic	Growth					X													
Drottar & Krueger, 2000a	*Pseudokirchneriella subcapitata*	Microalga	4	Chronic	Growth														X				
Ding et al., 2012a	*Pseudokirchneriella subcapitata*	Microalga	5	Chronic	Growth	X				X	X	X											
Liu et al., 2008	*Scenedesmus obliquus*	Green algae	3	Chronic	Growth			X		X				X		X	X		X				
Sutherland & Krueger, 2001	*Navicula pelliculosa*	Diatom algae	4	Chronic	Growth														X				
Wildlife International, 2001d	*Pseudokirchneriella subcapitata*	Microalga	4	Chronic	Growth												X						
3M, 2001	*Xenopus laevis*	African clawed frog	4	Acute	Growth														X				
Ankley et al., 2004	*Rana pipiens*	Northern leopard frog	16	Chronic	Survival														X				
Cheng et al., 2011	*Xenopus laevis*	African clawed frog	67	Chronic	Survival														X				
Palmer & Krueger, 2001	*Xenopus laevis*	African clawed frog	4	Acute	Survival														X				
3M, 2000	*Lepomis macrochirus*	Bluegill	4	Acute	Survival														X				
3M, 2000	*Lepomis macrochirus*	Bluegill	30	Chronic	Survival														X				
3M, 2000	*Pimephales promelas*	Fathead minnow	4	Acute	Survival														X				
3M, 2000	*Pimephales promelas*	Fathead minnow	42	Chronic	Survival, Growth														X				
3M, 2000	*Pimephales promelas*	Fathead minnow	5	Acute	Reproduction														X				
Colombo et al., 2008	*Oncorhynchus mykiss*	Rainbow trout	4	Acute	Survival					X													
Drottar and Krueger, 2000h	*Pimephales promelas*	Fathead minnow	4	Acute	Survival														X				
Drottar and Krueger, 2000i	*Pimephales promelas*	Fathead minnow	47	Chronic	Survival														X				
Drottar and Krueger, 2000i	*Pimephales promelas*	Fathead minnow	42	Chronic	Survival														X				
Drottar et al., 2002	*Lepomis macrochirus*	Bluegill	35	Chronic	Survival														X				
Du et al., 2008	*Dario rerio* [1]	Zebrafish	70	Chronic	Reproduction														X				
Du et al., 2009	*Dario rerio* [1]	Zebrafish	40	Chronic	Growth														X				
Environment and Climate Change Canada, 2014	*Oncorhynchus mykiss*	Rainbow trout	21	Chronic	Survival														X				
EG&G Bionomics, 1978	*Pimephales promelas*	Fathead minnow	30	Chronic	Survival					X													
EnviroSystems, Inc., 1990	*Pimephales promelas*	Fathead minnow	4	Acute	Survival					X													
Hagenaars et al., 2011	*Dario rerio* [1]	Zebrafish	5	Chronic	Development	X				X							X		X				
Hagenaars et al., 2014	*Dario rerio* [1]	Zebrafish	6	Acute	Reproduction														X				
Keiter et al., 2012	*Dario rerio* [1]	Zebrafish	180	Chronic	Growth														X				

Appendix D-1: Compilation of Freshwater Aquatic Life Toxicity Studies

Study	Species	Taxon/Common Name	Exposure Duration (days)	Acute or Chronic	PFAS Included in Study	PFBA	PFPeA	PFHxA	PFHpA	PFOA	PFNA	PFDA	PFUnDA	PFDoDA	PFTrDA	PFTeDA	PFBS	PFHxS	PFOS	PFDS	PFOSA	Et-FOSAA	Me-FOSAA
					Number of Carbon Atoms	C4	C5	C6	C7	C8	C9	C10	C11	C12	C13	C14	C4	C6	C8	C10	C8	C12	C11
					Number of Perfluorinated Carbon Atoms	3	4	5	6	7	8	9	10	11	12	13	4	6	8	10	8	12	11
					Ecological Endpoint																		
Huang et al., 2010	*Dario rerio* [1]	Zebrafish	6	Acute	Development														X				
Oakes et al., 2005	*Pimephales promelas*	Fathead minnow	28	Chronic	Survival														X				
Shi et al., 2008	*Dario rerio* [1]	Zebrafish	5.5	Acute	Reproduction														X				
Wang et al., 2017	*Dario rerio* [1]	Zebrafish	4	Chronic	Development														X				
Wang et al., 2011	*Dario rerio* [1]	Zebrafish	150	Chronic	Growth														X				
Wildlife International, 2001b	*Pimephales promelas*	Fathead minnow	4	Acute	Survival												X						
Wildlife International, 2001c	*Lepomis macrochirus*	Bluegill	4	Acute	Survival												X						
Zheng et al., 2012	*Dario rerio* [1]	Zebrafish	5	Chronic	Development					X	X								X				
3M Company, 1984	*Daphnia magna*	Water flea	21	Chronic	Survival					X													
3M, 2000	*Daphnia magna*	Water flea	2	Acute	Survival														X				
3M, 2000	*Unio complamatus*	Freshwater mussels	4	Acute	Survival														X				
Amraoui et al., 2018	*Unio ravoisieri*	Mollusk	4	Acute	Survival														X				
Bots et al., 2010	*Enallagma cyathigerum*	Common blue damselfly	320	Chronic	Survival														X				
Boudreau et al., 2003a	*Daphnia magna*	Water flea	2	Acute	Survival; Immobility														X				
Boudreau et al., 2003a	*Daphnia magna*	Water flea	21	Chronic	Survival														X				
Boudreau et al., 2003a	*Daphnia pulicaria*	Water flea	2	Acute	Survival; Immobility														X				
Boudreau et al., 2003b	Zooplankton	Zooplankton	35	Chronic	Survival														X				
Ding et al., 2012b	*Chydorus sphaericus* [1]	Cladocera	2	Acute	Survival	X				X	X	X	X	X									
Ding et al., 2012b	*Daphnia magna*	Water flea	2	Acute	Survival	X				X	X	X	X	X									
Drottar & Krueger, 2000b	*Daphnia magna*	Water flea	2	Acute	Survival														X				
Drottar & Krueger, 2000f	*Daphnia magna*	Water flea	21	Chronic	Survival														X				
Drottar & Krueger, 2000f	*Daphnia magna*	Water flea	2	Acute	Survival														X				
Drottar and Krueger, 2000c	*Unio complamatus*	Freshwater mussels	4	Acute	Survival														X				
Ji et al., 2008	*Daphnia magna*	Water flea	2	Acute	Survival					X									X				
Ji et al., 2008	*Daphnia magna*	Water flea	21	Chronic	Reproduction					X									X				
Ji et al., 2008	*Moina macrocopa*	Crustacean	2	Acute	Survival					X									X				
Ji et al., 2008	*Moina macrocopa*	Crustacean	7	Acute	Reproduction					X									X				
Ji et al., 2008	*Moina macrocopa*	Crustacean	2	Acute	Survival					X									X				
Ji et al., 2008	*Moina macrocopa*	Crustacean	7	Acute	Reproduction					X									X				
Li, M. H., 2009	*Daphnia magna*	Water flea	2	Acute	Survival					X									X				

Appendix D-1: Compilation of Freshwater Aquatic Life Toxicity Studies

Study	Species	Taxon/Common Name	Exposure Duration (days)	Acute or Chronic	PFAS Included in Study	PFBA	PFPeA	PFHxA	PFHpA	PFOA	PFNA	PFDA	PFUnDA	PFDoDA	PFTrDA	PFTeDA	PFBS	PFHxS	PFOS	PFDS	PFOSA	Et-FOSAA	Me-FOSAA
					Number of Carbon Atoms	C4	C5	C6	C7	C8	C9	C10	C11	C12	C13	C14	C4	C6	C8	C10	C8	C12	C11
					Number of Perfluorinated Carbon Atoms	3	4	5	6	7	8	9	10	11	12	13	4	6	8	10	8	12	11
					Ecological Endpoint																		
Li, M. H., 2009	*Dugesia japonica* [1]	Flatworm	4	Acute	Survival					X									X				
Li, M. H., 2009	*Neocaridina denticulata* [1]	Cherry shrimp	4	Acute	Survival					X													
Li, M. H., 2009	*Physa acuta*	European bladder snail	4	Acute	Survival					X									X				
Li, M. H., 2010	*Daphnia magna*	Water flea	21	Chronic	Survival, Reproduction					X									X				
Lu et al., 2015	*Daphnia magna*	Water flea	21	Chronic	Growth, Reproduction						X								X				
MacDonald et al., 2004	*Chironomus tentans*	Midge	10	Acute	Growth					X									X				
MacDonald et al., 2004	*Chironomus tentans*	Midge	20	Chronic	Growth					X									X				
Sanderson et al., 2004	*Daphnia pulicaria*	Water flea	21	Chronic	Survival														X				
Wildlife International, 2001a	*Daphnia magna*	Water flea	2	Acute	Survival												X						
Wildlife International, 2001f	*Daphnia magna*	Water flea	21	Chronic	Survival												X						
Boudreau et al., 2003a	*Lemna gibba*	Duckweed	7	Acute	Growth														X				
Boudreau et al., 2003a	*Lemna gibba*	Duckweed	7	Acute	Growth														X				
Desjardins et al., 2001	*Lemna gibba*	Duckweed	7	Acute	Growth														X				
Hanson et al., 2005a	*Myriophyllum sibiricum*	Shortspike watermilfoil, northern watermilfoil	42	Chronic	Growth														X				
Hanson et al., 2005a	*Myriophyllum spicatum*	Eurasian water-milfoil	42	Chronic	Growth														X				
Hanson et al., 2005b	*Myriophyllum sibiricum*	Shortspike watermilfoil, northern watermilfoil	35	Chronic	Growth					X													
Hanson et al., 2005b	*Myriophyllum spicatum*	Eurasian water-milfoil	35	Chronic	Growth					X													

Notes:
1: Non-resident species; not included for further consideration in development of aquatic life criteria, but provided for completeness.

Appendix D-1: Compilation of Marine Aquatic Life Toxicity Studies

Study	Species	Taxon/Common Name	Exposure Duration (days)	Acute or Chronic	PFAS Included in Study	PFBA	PFPeA	PFHxA	PFHpA	PFOA	PFNA	PFDA	PFUnDA	PFDoDA	PFTrDA	PFTeDA	PFBS	PFHxS	PFOS	PFDS	PFOSA	Et-FOSAA	Me-FOSAA
					Number of Carbon Atoms	C4	C5	C6	C7	C8	C9	C10	C11	C12	C13	C14	C4	C6	C8	C10	C8	C12	C11
					Number of Perfluorinated Carbon Atoms	3	4	5	6	7	8	9	10	11	12	13	4	6	8	10	8	12	11
					Ecological Endpoint																		
3M, 2000	*Skeletonema costatum*	Diatom	4	Chronic	Growth														X				
3M, 2000	*Crassostrea virginica*	Eastern oyster	4	Acute	Growth														X				
3M, 2000	*Mysidopsis bahia*	Epibenthic Shrimp	4	Acute	Survival														X				
3M, 2000	*Mysidopsis bahia*	Epibenthic Shrimp	35	Chronic	Reproduction, Survival, Growth														X				
3M, 2001	*Anabaena fios-aquae*	Cyanobacteria	4	Chronic	Growth														X				
Desjardins et al., 2001a	*Anabaena fios-aquae*	Cyanobacteria	4	Chronic	Growth														X				
Desjardins et al., 2001b	*Skeletonema costatum*	Diatom	4	Chronic	Growth														X				
Drottar and Krueger, 2000d	*Mysidopsis bahia*	Epibenthic Shrimp	4	Acute	Survival														X				
Drottar and Krueger, 2000e	*Crassostrea virginica*	Eastern oyster	4	Acute	Growth														X				
Drottar and Krueger, 2000g	*Mysidopsis bahia*	Epibenthic Shrimp	35	Chronic	Growth														X				
Drottar and Krueger, 2000g	*Mysidopsis bahia*	Epibenthic Shrimp	4	Acute	Survival														X				
Fabbri et al., 2014	*Mytilus galloprovincialis* [1]	Mediterranean mussel	2	Acute	Development					X									X				
Fang et al., 2013	*Oryzias melastigma* [1]	Ricefish	10	Chronic	Growth														X				
Han et al., 2015	*Tigriopus japonicas* [1]	Copepod	20	Chronic	Growth														X				
Han et al., 2015	*Tigriopus japonicas* [1]	Copepod	20	Chronic	Reproduction														X				
Mhadhbi et al., 2012	*Isochrysis galbana*	Algae-Haptophyta	3	Acute	Growth					X									X				
Mhadhbi et al., 2012	*Psetta maxima*	Turbot (flatfish)	6	Chronic	Development					X									X				
Mhadhbi et al., 2012	*Paracentrotus lividus* [1]	Sea urchin	2	Acute	Growth					X									X				
Mhadhbi et al., 2012	*Siriella armata*	Epibenthic Shrimp	4	Acute	Survival					X									X				
Palmer et al., 2002a	*Oncorhynchus mykiss*	Rainbow trout	4	Acute	Survival														X				
Palmer et al., 2002b	*Cyprinodon variegatus*	Sheepshead minnow	4	Acute	Survival														X				
Wildlife International, 2001f	*Mysidopsis bahia*	Epibenthic Shrimp	4	Acute	Survival												X						

Notes:
1: Non-resident species; not included for further consideration in development of aquatic life criteria, but provided for completeness.

APPENDIX D-2

Aquatic Life Values included in Species Sensitivity Distributions for PFOS and PFOA

Appendix D-2: Freshwater Aquatic Life Values included in Species Sensitivity Distributions for PFOA

Study	Species	Taxon	Exposure Duration	Acute or Chronic	PFOA						
					Effect Concentration (mg/L)	Effect Magnitude	Ecological Endpoint	Acute to Chronic Ratio [1]	Value for SSD (mg/L)	Species Mean Chronic Value [2] (mg/L)	Genus Mean Chronic Value [2] (mg/L)
Colombo et al. 2008	*Pseudokirchneriella subcapitata*	Algae	96 hr.	Chronic	12.5	NOEC	Growth	--	12.5	50	50
Colombo et al. 2008	*Pseudokirchneriella subcapitata*	Algae	72 hr.	Chronic	200	NOEC	Growth	--	200		
Colombo et al. 2008	*Oncorhynchus mykiss*	Fish	96 hr.	Acute	125	NOEC	Survival	18	6.9	6.9	6.9
EnviroSystems, Inc., 1990a	*Pimephales promelas*	Fish	96 hr.	Acute	500	NOEC	Survival	18	27.8	53	53
EG&G Bionomics Aquatic Toxicology Laboratory, 1978	*Pimephales promelas*	Fish	30 day	Chronic	100	NOEC	Survival	--	100		
MacDonald et al. 2004	*Chironomus tentans*	Invertebrate	10 days	Chronic	100	NOEC	Survival, growth	--	100	100	100
Ding et al. 2012	*Chydorus sphaericus*	Invertebrate	48 hr.	Acute	41	NOEC	Survival	18	2.3	2.3	2.3
3M Company, 1984	*Daphnia magna*	Invertebrate	21 day	Chronic	22	NOEC	Survival	--	22.0	16.3	16.3
Ji et al. 2008	*Daphnia magna*	Invertebrate	48 hr.	Acute	250	NOEC	Survival	18	13.9		
Ji et al. 2008	*Daphnia magna*	Invertebrate	21 day	Chronic	12.5	NOEC	Reproduction	--	12.5		
Li, M. H. 2009	*Daphnia magna*	Invertebrate	48 hr.	Acute	125	NOEC	Survival	18	6.9		
Li, M. H. 2010	*Daphnia magna*	Invertebrate	21 day	Chronic	100	NOEC	Survival	--	100		
Li, M. H. 2010	*Daphnia magna*	Invertebrate	21 day	Chronic	10	NOEC	Reproduction	--	10.0		
Ding et al. 2012	*Daphnia magna*	Invertebrate	48 hr.	Acute	207	NOEC	Survival	18	11.5		
Ji et al. 2008	*Moina macrocopa*	Invertebrate	48 hr.	Acute	62.5	NOEC	Survival	18	3.5	3.29	3.29
Ji et al. 2008	*Moina macrocopa*	Invertebrate	7 days	Chronic	3.125	NOEC	Reproduction	--	3.1		
Li, M. H. 2009	*Physa acuta*	Invertebrate	96 hr.	Acute	250	NOEC	Survival	18	13.9	13.9	13.9
Hanson et al. 2005	*Myriophyllum spicatum*	Plant	35 days	Chronic	23.9	NOEC	Growth	--	23.9	23.9	23.9

Notes:
1: Giesy et al., 2010
2: Calculated as geometric means

Acronyms:
NOEC = No effect concentration
SSD = Species sensitivity distribution
mg/L = milligrams per liter

Appendix D-2: Freshwater Aquatic Life Values included in Species Sensitivity Distributions for PFOS

Study	Species	Taxon	Exposure Duration	Acute or Chronic	PFOS						
					Effect Concentration (mg/L)	Effect Magnitude	Ecological Endpoint	Acute to Chronic Ratio [1]	Value for SSD (mg/L)	Species Mean Chronic Value [2] (mg/L)	Genus Mean Chronic Value [2] (mg/L)
Boudreau et al. 2003a	*Chlorella vulgaris*	Algae	96 hr.	Chronic	8.2	NOEC	Growth	--	8.2	8.2	8.2
Sutherland & Krueger 2001	*Navicula pelliculosa*	Algae	96 hr.	Chronic	150	NOEC	Growth	--	150	176	176
Sutherland & Krueger 2001	*Navicula pelliculosa*	Algae	96 hr.	Chronic	206	NOEC	Growth	--	206		
Drottar & Krueger 2000a	*Pseudokirchneriella subcapitata*	Algae	96 hr.	Chronic	42	NOEC	Growth	--	42.0	26	26
Boudreau et al. 2003a	*Pseudokirchneriella subcapitata*	Algae	96 hr.	Chronic	5.3	NOEC	Growth	--	5.3		
Boudreau et al. 2003a	*Pseudokirchneriella subcapitata*	Algae	96 hr.	Chronic	16.6	NOEC	Growth	--	16.6		
3M, 2000	*Pseudokirchneriella subcapitata*	Algae	96 hr.	Chronic	44	NOEC	Growth	--	44.0		
3M, 2000	*Pseudokirchneriella subcapitata*	Algae	72 hr.	Chronic	70	NOEC	Growth	--	70.0		
3M, 2000	*Pseudokirchneriella subcapitata*	Algae	14 day	Chronic	26	NOEC	Growth	--	26.0		
Liu et al. 2008	*Scenedesmus obliquus*	Algae	72 hr.	Chronic	53	EC10	Growth	--	53.0	53	53
Desjardins et al. 2001a	*Anabaena flos-aquae*	Algae	96 hr.	Chronic	93.8	NOEC	Growth	--	93.8	93.8	93.8
Ankley et al. 2004	*Rana pipiens*	Amphibian	16 wk	Chronic	0.3	NOEC	Survival	--	0.3	0.30	0.30
Palmer & Krueger 2001	*Xenopus laevis*	Amphibian	96 hr.	Acute	4.82	NOEC	Survival	8.3	0.6	0.38	0.38
3M, 2001	*Xenopus laevis*	Amphibian	96 hr.	Acute	7.97	NOEC	Growth	8.3	1.0		
Cheng et al. 2011	*Xenopus laevis*	Amphibian	67 day	Chronic	0.1	NOEC	Survival	--	0.1		
3M, 2000	*Lepomis macrochirus*	Fish	96 hr.	Acute	4.5	NOEC	Survival	8.3	0.5	0.5	0.5
3M, 2000	*Lepomis macrochirus*	Fish	62 days	Chronic	0.87	NOEC	Survival	--	0.9		
Drottar et al. 2002	*Lepomis macrochirus*	Fish	35 days	Chronic	0.3	MATC	Survival	--	0.3		
EC 2014	*Oncorhynchus mykiss*	Fish	21 day	Chronic	0.47	EC10	Survival	--	0.5	0.47	0.47
Drottar and Krueger 2000h	*Pimephales promelas*	Fish	96 hr.	Acute	3.2	NOEC	Survival	8.3	0.4	0.59	0.59
Oakes et al. 2005	*Pimephales promelas*	Fish	28 day	Chronic	0.3	NOEC	Survival	--	0.3		
Drottar and Krueger 2000i	*Pimephales promelas*	Fish	47 day	Chronic	0.29	NOEC	Survival	--	0.3		
3M, 2000	*Pimephales promelas*	Fish	96 hr.	Acute	3.3	NOEC	Survival	8.3	0.4		
3M, 2000	*Pimephales promelas*	Fish	96 hr.	Acute	170	NOEC	Survival	8.3	20.5		
3M, 2000	*Pimephales promelas*	Fish	42 days	Chronic	0.3	NOEC	Survival	--	0.3		
3M, 2000	*Pimephales promelas*	Fish	42 days	Chronic	0.3	NOEC	Growth	--	0.3		
3M, 2000	*Pimephales promelas*	Fish	5 days	Acute	4.6	NOEC	Reproduction	8.3	0.6		
3M, 2000	*Pimephales promelas*	Fish	30 days	Chronic	1	NOEC	Survival	--	1.0		
Drottar and Krueger 2000i	*Pimephales promelas*	Fish	42 day	Chronic	0.4	MATC	Survival	--	0.4		
MacDonald et al. 2004	*Chironomus tentans*	Invertebrate	10 day	Acute	0.05	NOEC	Growth	8.3	0.01	0.01	0.01
MacDonald et al. 2004	*Chironomus tentans*	Invertebrate	10 day	Acute	0.05	NOEC	Survival	8.3	0.01		
MacDonald et al. 2004	*Chironomus tentans*	Invertebrate	20 day	Chronic	0.0217	NOEC	Growth	--	0.02		
MacDonald et al. 2004	*Chironomus tentans*	Invertebrate	20 day	Chronic	0.0949	NOEC	Survival	--	0.1		
MacDonald et al. 2004	*Chironomus tentans*	Invertebrate	20 day	Chronic	0.0023	NOEC	Reproduction	--	0.002		

Appendix D-2: Freshwater Aquatic Life Values included in Species Sensitivity Distributions for PFOS

Study	Species	Taxon	Exposure Duration	Acute or Chronic	PFOS						
					Effect Concentration (mg/L)	Effect Magnitude	Ecological Endpoint	Acute to Chronic Ratio [1]	Value for SSD (mg/L)	Species Mean Chronic Value [2] (mg/L)	Genus Mean Chronic Value [2] (mg/L)
Boudreau et al. 2003a	*Daphnia magna*	Invertebrate	48 hr.	Acute	33.1	NOEC	Survival	8.3	4.0		
Boudreau et al. 2003a	*Daphnia magna*	Invertebrate	48 hr.	Acute	0.8	NOEC	Survival	8.3	0.1		
Drottar & Krueger 2000b	*Daphnia magna*	Invertebrate	48 hr.	Acute	32	NOEC	Survival	8.3	3.9		
Drottar & Krueger 2000f	*Daphnia magna*	Invertebrate	21 days	Chronic	12	NOEC	Survival	--	12.0		
Drottar & Krueger 2000f	*Daphnia magna*	Invertebrate	48 hr.	Acute	12	NOEC	Survival	8.3	1.4		
Boudreau et al. 2003a	*Daphnia magna*	Invertebrate	21 days	Chronic	5.3	NOEC	Survival	--	5.3		
3M, 2000	*Daphnia magna*	Invertebrate	48 hr.	Acute	33	NOEC	Survival	8.3	4.0	2.1	
3M, 2000	*Daphnia magna*	Invertebrate	48 hr.	Acute	100	NOEC	Survival	8.3	12.0		
3M, 2000	*Daphnia magna*	Invertebrate	48 hr.	Acute	2.2	NOEC	Survival	8.3	0.3		
Ji et al. 2008	*Daphnia magna*	Invertebrate	48 hr.	Acute	12.5	NOEC	Survival	8.3	1.5		
Ji et al. 2008	*Daphnia magna*	Invertebrate	21 day	Chronic	1.25	NOEC	Reproduction	--	1.3		
Li M.H. 2010	*Daphnia magna*	Invertebrate	21 day	Chronic	5	NOEC	Survival	--	5.0		
Li M.H. 2010	*Daphnia magna*	Invertebrate	21 day	Chronic	1	NOEC	Reproduction	--	1.0		
Boudreau et al. 2003a	*Daphnia pulicaria*	Invertebrate	48 hr.	Acute	46.9	NOEC	Survival	8.3	5.7		2.8
Boudreau et al. 2003a	*Daphnia pulicaria*	Invertebrate	48 hr.	Acute	13.6	NOEC	Survival	8.3	1.6	3.8	
Sanderson et al. 2004	*Daphnia pulicaria*	Invertebrate	21 day	Chronic	6	EC10	Survival	--	6.0		
Bots et al. 2010	*Enallagma cyathigerum*	Invertebrate	320 day	Chronic	0.01	NOEC	Survival	--	0.01	0.01	0.01
Ji et al. 2008	*Moina macrocopa*	Invertebrate	48 hr.	Acute	6.25	NOEC	Survival	8.3	0.8	0.17	0.17
Ji et al. 2008	*Moina macrocopa*	Invertebrate	7 days	Acute	0.3125	NOEC	Reproduction	8.3	0.04		
Li, M. H., 2009	*Physa acuta*	Invertebrate	96 hr.	Acute	100	NOEC	Survival	8.3	12.0	12	12
Drottar and Krueger 2000c	*Unio complamatus*	Invertebrate	96 hr.	Acute	20	NOEC	Survival	8.3	2.4	2.41	2.41
3M, 2000	*Unio complamatus*	Invertebrate	96 hr.	Acute	20	NOEC	Survival	8.3	2.4		
Amraoui et al. 2018	*Uno ravoisieri*	Invertebrate	96 hr.	Acute	10.00	NOEC	Survival	8.3	1.2	1.2	1.2
Boudreau et al. 2003b	Zooplankton	Invertebrate	35 day	Chronic	3	NOEC	Survival	--	3.0	3.0	3.0
Desjardins et al. 2001c	*Lemna gibba*	Macrophyte	7 days	Acute	15	NOEC	Growth	8.3	1.8	1.7	1.7
Boudreau et al. 2003a	*Lemna gibba*	Macrophyte	7 days	Acute	29.2	NOEC	Growth	8.3	3.5		
Boudreau et al. 2003a	*Lemna gibba*	Macrophyte	7 days	Acute	6.6	NOEC	Growth	8.3	0.8		
Hanson et al. 2005	*Myriophyllum sibiricum*	Macrophyte	42 days	Chronic	2.9	NOEC	Growth	--	2.9	2.1	2.1
Hanson et al. 2005	*Myriophyllum sibiricum*	Macrophyte	42 days	Chronic	0.3	NOEC	Growth	--	0.3		
Hanson et al. 2005	*Myriophyllum spicatum*	Macrophyte	42 days	Chronic	11.4	NOEC	Growth	--	11.4		

Notes:

1: Giesy et al., 2010

2: Calculated as geometric means

Acronyms:

NOEC = No effect concentration

EC10 = 10% effect concentration

MATC = Maximum acceptable toxicant concentration

SSD = Species sensitivity distribution

mg/L = milligrams per liter

Appendix D-2: Marine Aquatic Life Values included in Species Sensitivity Distributions for PFOS

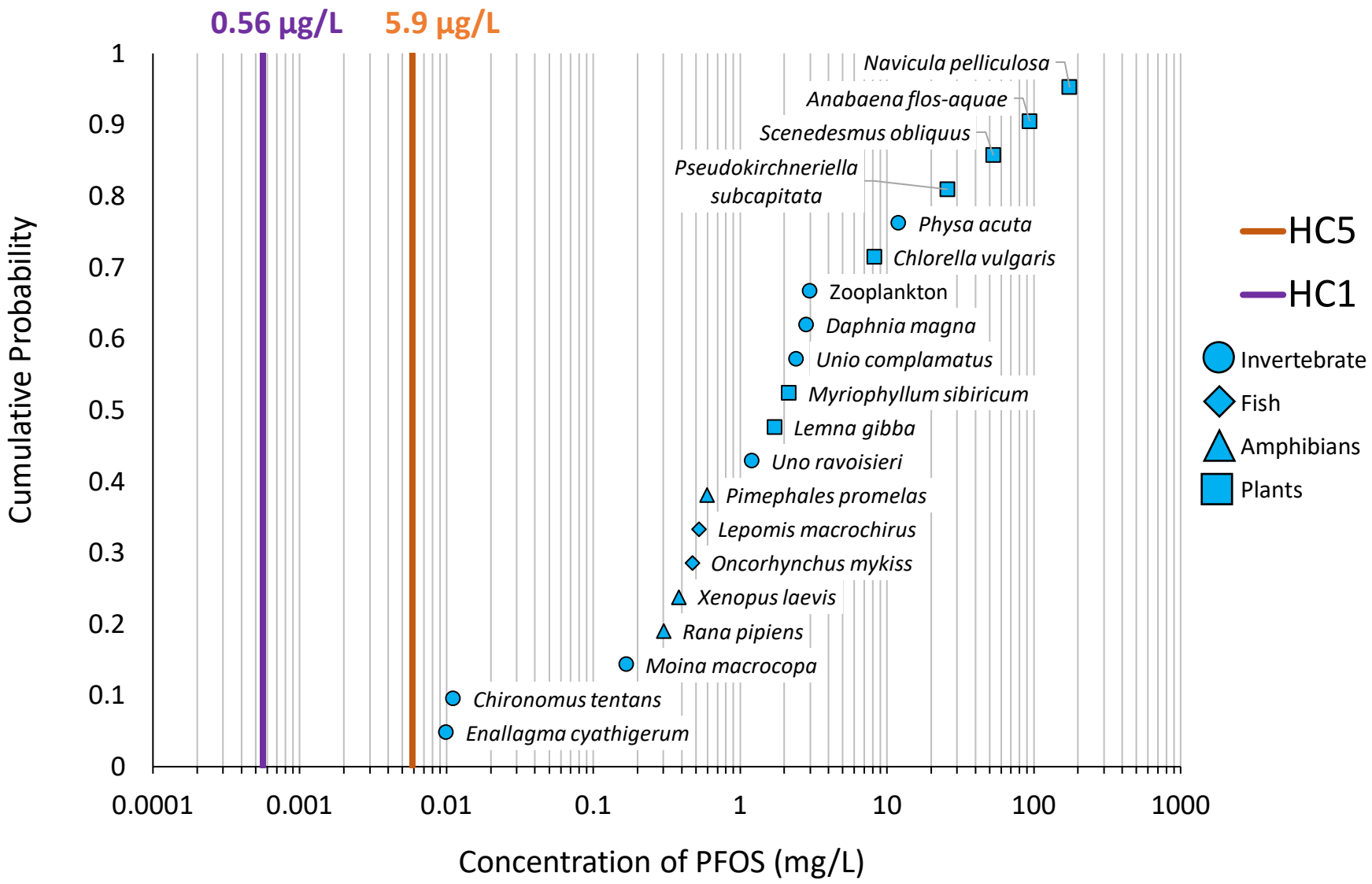
Study	Species	Taxon	Exposure Duration	Acute or Chronic	PFOS						
					Effect Concentration (mg/L)	Effect Magnitude	Ecological Endpoint	Acute to Chronic Ratio [1]	Value for SSD (mg/L)	Species Mean Chronic Value [2] (mg/L)	Genus Mean Chronic Value [2] (mg/L)
Mhadhbi et al. 2012	*Isochrysis galbana*	Algae	72 hr.	Acute	7.5	NOEC	Growth	8.3	0.9	0.90	0.90
3M, 2000	*Skeletonema costatum*	Algae	96 hr.	Chronic	3.2	NOEC	Growth	--	3.2	3.2	3.2
Desjardins et al. 2001b	*Skeletonema costatum*	Algae	96 hr.	Chronic	3.2	NOEC	Growth	--	3.2		
Palmer et al. 2002b	*Cyprinodon variegatus*	Fish	96 hr.	Acute	15	NOEC	Survival	8.3	1.8	2	2
Palmer et al. 2002a	*Oncorhynchus mykiss*	Fish	96 hr.	Acute	6.3	NOEC	Survival	8.3	0.8	0.76	0.76
Mhadhbi et al. 2012	*Psetta maxima*	Fish	144 hr.	Chronic	0.015	NOEC	Development	--	0.02	0.02	0.02
Drottar and Krueger, 2000e	*Crassostrea virginica*	Invertebrate	96 hr.	Acute	1.8	NOEC	Growth	8.3	0.2	0.22	0.22
3M, 2000	*Crassostrea virginica*	Invertebrate	96 hr.	Acute	1.9	NOEC	Growth	8.3	0.2		
Drottar and Krueger, 2000d	*Mysidopsis bahia*	Invertebrate	96 hr.	Acute	1.1	NOEC	Survival	8.3	0.1	0.20	0.20
Drottar and Krueger, 2000g	*Mysidopsis bahia*	Invertebrate	35 day	Chronic	0.25	NOEC	Growth	--	0.3		
Drottar and Krueger, 2000g	*Mysidopsis bahia*	Invertebrate	35 day	Chronic	0.55	NOEC	Survival	--	0.6		
Drottar and Krueger, 2000g	*Mysidopsis bahia*	Invertebrate	35 day	Chronic	0.25	NOEC	Reproduction	--	0.3		
Drottar and Krueger, 2000g	*Mysidopsis bahia*	Invertebrate	96 hr.	Acute	0.55	NOEC	Survival	8.3	0.1		
Mhadhbi et al. 2012	*Siriella armata*	Invertebrate	96 hr.	Acute	1.25	NOEC	Survival	8.3	0.2	0.15	0.15

Notes:
1: Giesy et al., 2010
2: Calculated as geometric means
Acronyms:
NOEC = No effect concentration
SSD = Species sensitivity distribution
mg/L = milligrams per liter

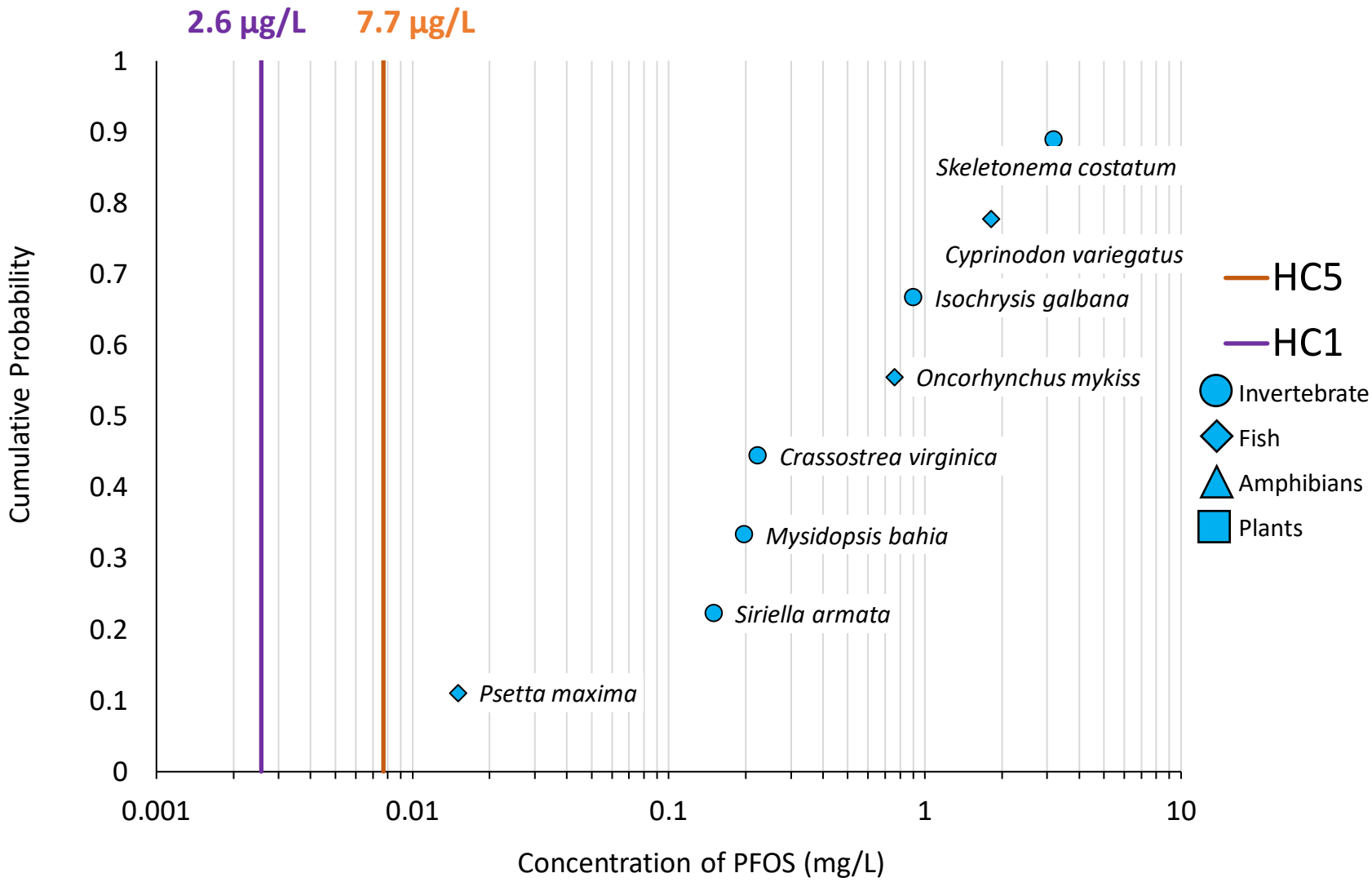
APPENDIX D-3

Species Sensitivity Distributions for PFOS and PFOA

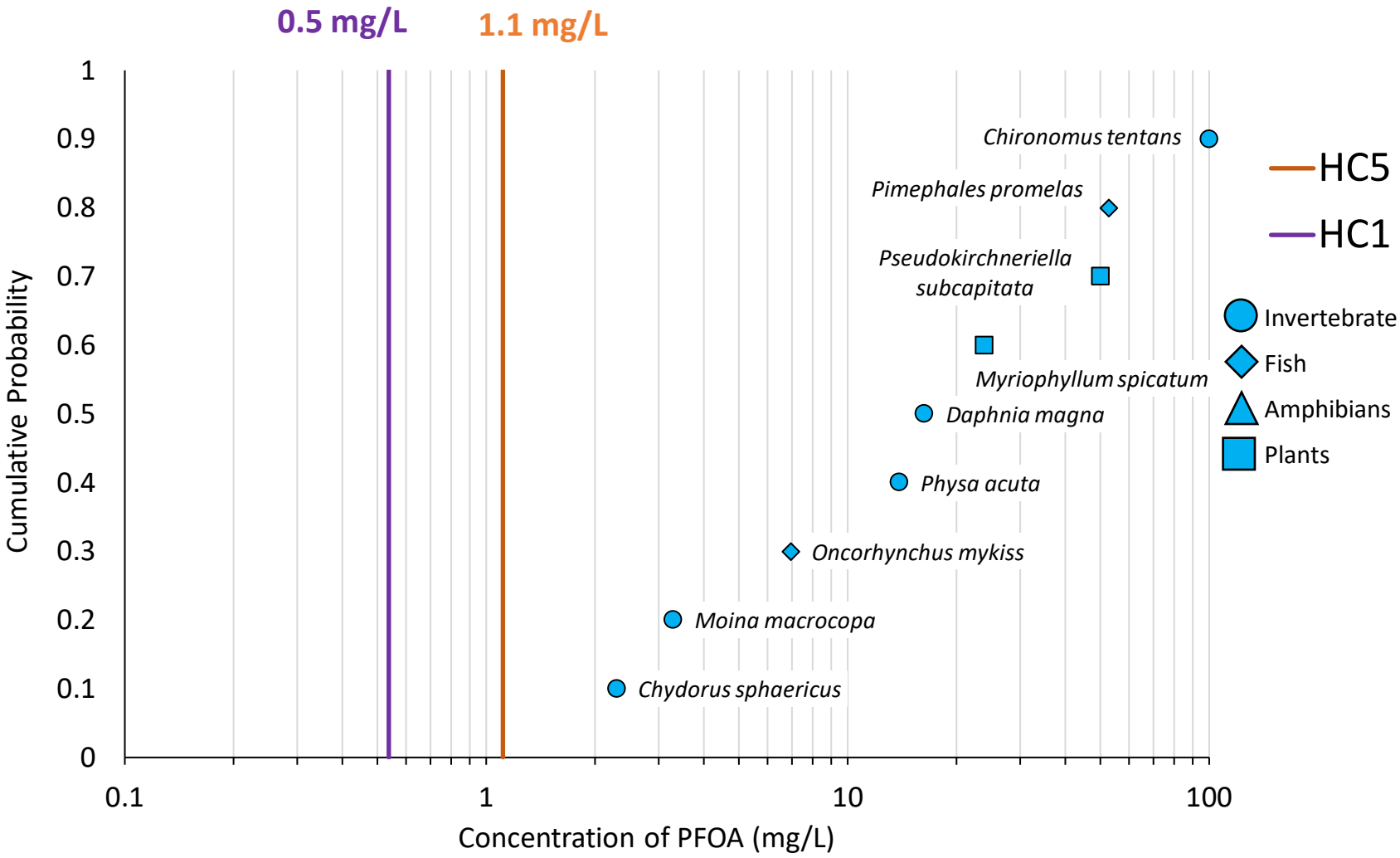
Appendix D-3: SSD for Freshwater Aquatic Life - PFOS



Appendix D-3: SSD for Marine Aquatic Life - PFOS



Appendix D-3: SSD for Freshwater Aquatic Life - PFOA



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APPENDIX E

Compilation of Terrestrial Plant and Invertebrate Toxicity Studies

Appendix E-1: Sources of Data for Terrestrial Invertebrate Toxicity Studies

Study	Species	Taxon/Common Name	Exposure Media	Exposure Duration (days)	Acute or Chronic	PFAS Included in Study	PFBA	PFPeA	PFHxA	PFHpA	PFOA	PFNA	PFDA	PFUnDA	PFDoDA	PFTrDA	PFTeDA	PFBS	PFHxS	PFOS	PFDS	PFOSA	EtFOSAA	MeFOSAA
						Number of Carbon Atoms	C4	C5	C6	C7	C8	C9	C10	C11	C12	C13	C14	C4	C6	C8	C10	C8	C12	C11
						Number of Perfluorinated Carbon Atoms	3	4	5	6	7	8	9	10	11	12	13	4	6	8	10	8	12	11
						Ecological Endpoint																		
Sindermann et al., 2002	*Eisenia fetida*	Earthworm	Soil	7	Acute	Survival														X				
Sindermann et al., 2002	*Eisenia fetida*	Earthworm	Soil	14	Acute	Survival														X				
He et al., 2013	*Eisenia fetida*	Earthworm	Soil	28	Chronic	Growth					X													
Xu et al., 2013	*Eisenia fetida*	Earthworm	Soil	42	Chronic	Growth														X				
Wilkins et al., 2001a	*Apis mellifera*	Western honey bee	Oral	3	Acute	Survival														X				
Wilkins et al., 2001b	*Apis mellifera*	Western honey bee	Contact paper	4	Acute	Survival														X				

Appendix E-2: Sources of Data for Terrestrial Plant Toxicity Studies

Study	Species	Taxon/Common Name	Exposure Duration (days)	Acute or Chronic	PFAS Included in Study	PFBA	PFPeA	PFHxA	PFHpA	PFOA	PFNA	PFDA	PFUnDA	PFDoDA	PFTrDA	PFTeDA	PFBS	PFHxS	PFOS	PFDS	PFOSA	EtFOSAA	MeFOSAA
					Number of Carbon Atoms	C4	C5	C6	C7	C8	C9	C10	C11	C12	C13	C14	C4	C6	C8	C10	C8	C12	C11
					Number of Perfluorinated Carbon Atoms	3	4	5	6	7	8	9	10	11	12	13	4	6	8	10	8	12	11
					Ecological Endpoint																		
Brignole et al., 2003	*Allium cepa, Lolium perenne, Medicago sativa, Linum usitatissimum, Lycopersicon esculentum, Glycine max, Lactuca sativa*	Onion, ryegrass, alfalfa, flax, tomato, soybean, lettuce	21	Chronic	Growth														X				
De Yong et al., 2012	*Brassica rapa pekinensis*	Chinese Cabbage	15	Chronic	Growth														X				
González-Naranjo et al., 2015	*Sorghum bicolor*	Sorghum	15	Chronic	Growth					X													
Li M.H., 2009	*Cucumis sativus, Lactuca sativa, Brassica rapa chinensis*	Cucumber, lettuce, pakchoi	5	Acute	Growth					X									X				
Qu et al., 2010	*Triticum aestivum L.*	Wheat	7	Acute	Growth														X				
Yang et al., 2015	*Arabidopsis thaliana*	Thale cress	21	Chronic	Growth					X													
Zhao et al., 2011	*Brassica chinensis*	Bok Choy	7	Acute	Growth					X									X				
Zhou et al., 2016	*Triticum aestivum L.*	Wheat	28	Chronic	Growth					X													
Zhou et al., 2016	*Triticum aestivum L.*	Wheat	8	Acute	Growth					X													

Appendix E: References

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