

**Surface Water  
Quality Criterion for  
Perfluorooctane Sulfonic Acid**

Minnesota Pollution Control Agency  
St. Paul, Minnesota

STS Project 200604796

August, 2007

# **SURFACE WATER QUALITY CRITERION FOR PERFLUOROOCTANE SULFONIC ACID**

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## 1.0 INTRODUCTION

In January 2006, the Minnesota Pollution Control Agency (MPCA) contracted with STS Consultants, Ltd. (STS) to develop an ambient surface water quality criterion for perfluorooctane sulfonic acid (PFOS). The method that was to be used to develop this criterion is that published in Minnesota Rules, Chapter 7050.0218 Methods for Protection of Surface Waters from Toxic Pollutants for Which Numerical Standards Not Promulgated. As detailed in Subpart 10 Applicable Criteria, this effort entails the development of the following values for a chemical:

- a Chronic Criterion (CC) and Maximum Criterion (MC), based on toxicity to aquatic organisms;
- a Chronic Criterion (CC), based on toxicity to aquatic plants;
- a Drinking Water plus Fish Consumption Criterion (dfCC) and a Fish Consumption Criterion (fCC), based on human health risk;
- a Wildlife Chronic Criteria (WCC), based on toxicity to mammalian and avian species; and
- a concentration that will prevent unacceptable taste and/or odor in water, fish or other edible aquatic organisms.

The Minnesota Department of Health (MDH) developed the human health risk values for this project. These values were then incorporated into the algorithms used to derive the dfCC and fCC criteria. STS developed the CC and MC criteria, and calculated the dfCC and fCC criteria using the MDH toxicity values and a project-derived bioaccumulation factor for PFOS. No WCC was calculated due to a lack of sufficient toxicological data on this chemical. Also, no organoleptic criteria were developed because this chemical does not exhibit these chemical properties.

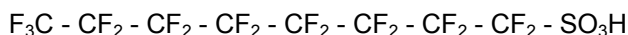
The entire chemical database for this document is contained in the accompanying four project binders. Appendix A to this document provides the Table of Contents for these binders.

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## 2.0 CHEMICAL IDENTITY AND CHEMICAL PROPERTIES

### 2.1 Chemical Identity

Perfluorooctane sulfonic acid has the molecular formula:  $C_8HF_{17}O_3S$ . The structure of the n-octane isomer is:



It can exist in the acid form or as various salts. The most common forms and their CAS numbers are:

acid	1763-23-1
potassium salt	2795-39-3
lithium salt	29457-72-5
ammonium salt	29081-56-9
diethanolamine salt	70225-14-8
didecyldimethylammonium salt	251099-16-8

3M had produced fluorochemicals commercially for over 40 years. They produced fluorochemicals by combining anhydrous hydrogen fluoride with hydrocarbon stock in the presence of electrical energy. This fluorination process yields about 35-40% straight-chain chemical, along with higher or lower straight-chain homologues (7%), branched chain perfluoroalkyl products (18-20%), other branched and cyclic perfluoroalkanes and ethers (20-25%) and tars (high molecular weight fluorocarbon byproducts; 10-15%) (226-0620). The commercial products were mixtures of approximately 70% linear derivatives with 30% branched derivative impurities. PFOS was not a primary commercial fluorocarbon product of 3M; it was an important intermediate in the synthesis of higher molecular weight fluorocarbon products. Small amounts of PFOS were present as chemical residuals in the commercial products, however. A number of the manufactured fluorocarbons have also been shown to breakdown in the environment to PFOS, leading to its global presence. POSF has been shown to breakdown into PFOS in the environment.

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## 2.2 Relevant Chemical Properties

### 2.2.1 Water Solubility

#### Pure Water Studies

In 1980 and in 1982, 3M Environmental Laboratory conducted two non-GLP water solubility studies on potassium PFOS (n-octane isomer) (STS-308; STS-309). Deionized water was used in both tests. In the first test, 2.5 grams of PFOS was stirred with one liter of deionized water for 24 hours. The suspension was then allowed to settle at room temperature for one hour, then filtered through 0.45 µm membrane filters or 0.7 µm glass fiber filters or centrifuged at 10,000 kg for 10 minutes. TOC measurements confirmed the same result regardless of the separation procedure utilized. The second test was run in a similar manner at the same PFOS concentration. The results of these two studies were S = 1080 mg/L and 1090 mg/L at 20-22.5°C.

In 1999 Wildlife International, Ltd. conducted a water solubility test on perfluorooctane sulfonic acid, potassium salt (AR 226-0052). According to the 3M reference sheet (AR 226-0054), this was the n-octyl isomer of PFOS. This was a GLP test following the following guidelines: OECD105; OPPTS 830.7840 and 40 CFR 796.1840. The procedure is called the shake flask method. Triplicate samples were run in NANO pure® water at 30°C, followed by 24-hour equilibrium at 20°C. PFOS was quantified by LCMS. The reported PFOS solubility in this test system was 519 mg/L. In 1999, an addendum to this original study was issued by Wildlife International, Ltd. that corrected the initially reported results for chemical purity (AR 226-1768). The revised solubility value at 20°C is 498 mg/L.

In 2001, 3M Environmental Laboratory conducted a GLP water solubility study on potassium PFOS using the shake flask method (OPPTS 830.7840 and OECD 105) (STS-129). This was a well conducted study, that involved sequential sampling (every 24 hours) throughout the 72-hour test. The analytical methodology for quantifying PFOS

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was HPLC/ES/MS. The result of this study was a reported solubility of 680 mg/L at 24-25°C.

#### Fresh Water Study

In 1999 Wildlife International, Ltd. conducted a water solubility test on perfluorooctane sulfonic acid, potassium salt (AR 226-1767) for 3M. This was a non-GLP shake flask study, but was performed in an equivalent manner to a GLP study. In this study, the water used in the test was laboratory fresh water, as utilized in Algal toxicity tests. PFOS was quantified by LCMS. The mean solubility from triplicate samples of PFOS was 326 mg/L at 20°C.

#### Salt Water Studies

Wildlife International, Ltd. also conducted in 1999 a water solubility test on perfluorooctane sulfonic acid, potassium salt in sea water (AR 226-1767). In this shake flask study, the mean solubility of triplicate samples of PFOS was reported as 21.8 mg/L at 20°C. This was a GLP-equivalent study. PFOS was quantified by LCMS.

In 2001, 3M Environmental Laboratory conducted a GLP water solubility study on potassium PFOS in sea water and in an aqueous solution of 3.5% sodium chloride (STS-130) using the shake flask method (OPPTS 830.7840; OECD 105). This was a 72-hour study, with aliquots taken for analysis every 24 hours. PFOS was quantified by HPLC/ES/MS. The mean results of this study from triplicate samples were:

S = 12.4 mg/L in sea water

S = 20.0 mg/L in 3.5% sodium chloride

#### Other

3M Environmental Laboratory in 1982 also performed a hand calculation of a water solubility value for PFOS using a regression equation and a PFOS  $K_{ow}$  value. It is



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unknown where the  $K_{ow}$  value came from. This calculation resulted in a water solubility value of 250 g/L, which is not in agreement with the various experimental test results.

PFOS is considered a moderately soluble chemical, dissolving in fresh water at >300 mg/L. It is much less soluble in salt water (12.4 - 21.8 mg/L). This large differential in solubility certainly would influence its water distribution in the environment.

### 2.2.2 Vapor Pressure

In 1993, 3M Environmental Laboratory conducted an impinger study to determine the volatility of PFOS (STS-402). The test consisted of dissolving PFOS in a number of aqueous salt solutions and water/isopropanol solutions and then bubbling air through them. The results indicated that with this procedure an average of 11% of the dissolved PFOS could be released from the aqueous solutions, but only 5% on average was released from the water/isopropanol solutions. In a second experiment, PFOS was dissolved in a solution of 500 ppm ammonium acetate in 1-propanol:water (50:50) and allowed to evaporate at room temperature. As a function of time, impingers were sampled for the compound. No detectable amounts were recorded, indicating that the PFOS vapor pressure from this solution is less than  $1 \times 10^{-7}$  torr ( $< 1.3 \times 10^{-5}$  Pa), or essentially non-volatile.

In 1999, 3M contracted with Wildlife International, Ltd. to determine the vapor pressure of PFOS (AR 226-0048). According to 3M's fact sheet (AR 226-0047) this test was made with the potassium salt of the n-octane isomer of PFOS. The test followed GLP and the test method used was OECD Guideline 104 Vapor Pressure Curve and OPPTS 830.7950 Vapor Pressure. The procedure is called the Spinning Rotor Gauge method. The result of this test (three measurements) was a mean steady-state vapor pressure of  $3.31 \times 10^{-4}$  Pa at 20°C, indicating a very low potential for evaporation.

PFOS exhibits a very low vapor pressure, which indicates that environmentally it would evaporate from surface waters very slowly.

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### 2.2.3 Air-Water Partition Coefficient

In 1999, 3M, along with an outside consultant (Don Mackay), designed a non-GLP test to determine the air-water partition coefficient (H) for potassium PFOS (226-0051). According to 3M's fact sheet, this chemical was the n-octane isomer of PFOS. Wildlife International, Ltd. conducted the test. The test consisted of dissolving PFOS in distilled water at a concentration of 50 mg/L. Then, the solution was incrementally evaporated using a hot plate. According to the outside consultant, since no evaporation of PFOS occurred, the unitless H-value for PFOS is less than  $2 \times 10^{-6}$ ; so PFOS will not equilibrate between surface water and air to any significant extent. Air-water equilibration of PFOS is therefore not likely to occur in the environment.

### 2.2.4 Octanol-Water Partition Coefficient

In 1999, 3M contracted with Wildlife International, Ltd. to determine the octanol-water partition coefficient for potassium PFOS. According to 3M's fact sheet (AR 226-0050), this was the n-octane isomer of PFOS. The test was conducted at 50 mg/L PFOS. A feasibility test was conducted to determine if the physical properties of the compound were compatible with the shake flask method, as published in OECD 107. Since a definitive partition interface could not be obtained in the test flasks upon mixing (a beige white emulsion formed), this test system was unable to determine a  $K_{ow}$  for PFOS.

In a subsequent report from Wildlife International, Ltd., a  $K_{ow}$  value was calculated mathematically from the chemical's individually measured solubility in both water and n-octanol following the solubility test method detailed in OPPTS 834.7840 and OECD 105. PFOS solubility in fresh water at 22-25°C was determined to be 680 mg/L; its solubility in n-octanol was measured at 56 mg/L. The log  $K_{ow}$  was calculated to be -1.08 ( $K_{ow} = 0.083$ ). PFOS, therefore, is not considered a lipophilic compound and would not be expected to partition to lipid matrices in the environment, as do the organochlorine compounds. Its bio-accumulative potential in the lipid portion of biota would also not be expected to occur to any extent.

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### 2.2.5 Adsorption-Desorption Studies

In 1978, 3M Environmental Laboratory conducted a non-GLP soil adsorption-desorption study (AR 226-0055) on potassium PFOS (n-octane isomer) using the U.S. EPA method for pesticides. Radiolabeled ( $^{14}\text{C}$ ) chemical was mixed with a sandy loam soil, pH 6.5, containing 1.5% organic carbon at 16-19°C. The test consisted of shaking 25 ml of various concentrations of PFOS (28, 51, 90, 158, 282 mg/L in deionized water) with 5 grams of soil for 24 hours, followed by scintillation counting of both fractions. The result was a  $K_{oc}$  (water-organic carbon partition coefficient) of 66 L/kg, indicating that PFOS does bind to but not tightly to organic matter.

3M Environmental Laboratory in 2000 conducted a GLP soil adsorption study at 19-30°C on potassium PFOS (n-octane isomer) following the test method detailed in OECD 106 (226-1107). Three different soil types (a clay, a clay loam, a sandy loam), one river sediment and a domestic sludge sample were used in the test, with varying organic carbon contents of 1.3% (sediment) - 2.8% (sandy loam). The test consisted of replicate samples of 0.5 mg/L radiolabeled PFOS equilibrated with soil material for 48 hours. The  $K_{oc}$  values measured for PFOS in this experiment varied from 37.4 L/kg in clay loam to 126 L/kg in sandy loam. The  $K_{oc}$  for river sediment was 57.1 L/kg. These data again indicate that PFOS does not strongly bind to organic carbon material and thus would be considered mobile in the environment.

### 2.2.6 Abiotic Degradation

Abiotic degradation of chemical substances can occur via direct hydrolysis (in an aqueous environment) or from photolysis.

#### Hydrolysis

In 2001, 3M Environmental Laboratory conducted a hydrolysis test on potassium PFOS (0.5 mg/L) based on the method detailed in OPPTS 835.2110 (STS-125). Samples were evaluated at six different pH values ranging between 1.5 and 11.0. Test time was a period of 42 days. Incubation occurred at 50°C. Weekly aliquots were taken for analysis

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of PFOS using HPLC/MS. The test was a GLP-equivalent procedure. Based on the data obtained, the experiment could not detect any degradation of PFOS over the 42 day time period in any of the pH solutions.

### Photolysis

In 1978, 3M Environmental Laboratory conducted a photolysis test with  $^{14}\text{C}$ -labeled potassium PFOS (n-octane isomer) using simulated sunlight from a GE F-40BL fluorescent black light (AR 226-0056). The procedure followed the U.S. EPA method described in the Federal Register, Vol. 23 No. 132-Monday July 10, 1978. This test also was a GLP-equivalent methodology. The test substance (50 mg/L) was irradiated (300-600 nm) at 23°C for up to 30 days. Samples were taken for analysis by TLC-autoradiography throughout the test period. No photodegradation products were detected in the study.

In 2001, 3M Environmental Laboratory conducted a GLP-equivalent photodegradation study with the potassium salt of PFOS (n-octane isomer) (STS-126). The procedure that was followed was the method detailed in OPPTS 835.5270 and OFCD Draft Document "Phototransformation of Chemicals in Water - Direct and Indirect Photolysis" (Aug., 2000). PFOS degradation (10 mg/L) was evaluated in five different test systems at 25°C for up to 167 hours. The test systems included: water, hydrogen peroxide/water (1:1 molar equivalent);  $\text{Fe}^{3+}$ /water ( $\text{Fe}^{3+}$  at 24X molar excess),  $\text{Fe}^{3+}$ /water/ $\text{H}_2\text{O}_2$ , as well as commercial humic material. The  $\text{Fe}/\text{H}_2\text{O}_2$  solutions were utilized to evaluate the potential for hydroxyl radical-radiated breakdown of PFOS. Light (290-800 nm) irradiated the samples from either of two lamp types, Suntest CPS+ or Suntest XLS+. The results of these tests indicated no direct or indirect (radical-mediated) photolysis of PFOS occurred, as assessed by GC/MS. Based on the sensitivity of this test system, a photodegradation half-life of >3.7 years was calculated.

Because the carbon-fluorine bond is one of the strongest in nature, with high bond energies, its cleavage requires large amounts of energy. Chemical and physical processes naturally occurring in the environment lack the required energy. It is not

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surprising therefore that the fully fluorinated chemical, PFOS, was not shown to be degraded by abiotic mechanisms.

### **2.2.7 Biodegradation**

There have been a number of studies that have addressed the biodegradation of PFOS. None of these studies have definitively shown that this chemical can degrade under either aerobic or anaerobic conditions.

In 1976, 3M Environmental Laboratory conducted a room temperature aerobic biodegradation test on the potassium salt of PFOS using microorganisms from a wastewater treatment plant sludge (AR 226-0057). Two PFOS concentrations were evaluated, 400 mg/L and 4000 mg/L. No increase in O<sub>2</sub> uptake was observed in the Warburg respirometer upon the addition of PFOS to the incubate. No toxicity was reported either. Although not a GLP test, a positive control substance (glucose) was used to show the methodology worked, and other fluorocarbons were shown to be degraded in the test, indicating that the test system was sufficiently sensitive to detect fluorocarbon biodegradation. One drawback of this test, however, was that it only lasted for three hours.

In 1978, 3M Environmental Laboratory conducted a 2.5 month room temperature shake flask biodegradation test on the potassium salt of PFOS, using microorganisms from a wastewater treatment plant sludge (AR 226-0052). The PFOS concentration in this study was 50 mg/L. No toxicity to the microorganisms was observed. In this test, no biodegradation was found when measured by the formation of fluoride ion during the test or by the formation of organic metabolites from radiolabeled (<sup>14</sup>C)-PFOS by thin layer chromatography, measured at the end of the test. A positive control substance (phenol) was used to verify that the test system was operational throughout the procedure.

In 1979, 3M Environmental Laboratory conducted a 20-day BOD study with the diethanolamine (DEA) salt of PFOS (AR 226-0059). Test concentrations of PFOS were: 20, 40, 80, 160 and 500 mg/L. Microorganisms from a wastewater treatment plant sludge were again used. In this test some metabolism occurred, as measured by oxygen

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utilization, but it may have been due to the breakdown of DEA. No analytical data were obtained during the test to confirm this, however. This study documentation lacks sufficient information from which to draw conclusions.

In 2000, Pace Analytical conducted two biodegradation studies on the potassium salt of PFOS (STS-156; STS-300) for 3M. In the first test, PFOS, along with a number of other fluorocarbon compounds, was incubated at 25°C for 18 days in a microorganism culture derived from wastewater treatment plant sludge. The PFOS concentration in the test mixture was 2.5 mg/L. HPLC/MS was used to evaluate biodegradation. PFOS was not measurably biodegraded in this test; however, it was shown that other compounds degraded to PFOS. This was a GLP-equivalent study. The second test was conducted in an identical manner to the first except it lasted twice as long, 35 days. PFOS concentration in this test was 2.6 mg/L. Again, degradation of PFOS was not measured either in the culture system or in the abiotic controls, as assessed by HPLC/MS. This also was a GLP-equivalent study.

Springborn Laboratories, Ltd. conducted four separate GLP-equivalent biodegradation tests on the potassium salt of PFOS for 3M in 2000. The first two tests (STS-301) were conducted using a sewage treatment plant sludge/river sediment mixture. The first test involved a 14-week aerobic shake flask method at a temperature of 22°C. The PFOS concentration was 20.8 mg/L, and was verified by weekly measurements using HPLC/MS. No toxicity was observed during the test, as judged by decreased respiration. The data indicated some loss of PFOS from the culture media, but the investigators noted that a white precipitate had formed. Analysis of the precipitate was not performed, however. The second study was an anaerobic biodegradation test, wherein PFOS was evaluated at two concentrations -- 0.2 mg/L and 105 mg/L. This was a 63 day test, and biodegradation was followed by CO<sub>2</sub> formation in the vial headspace and by HPLC/MS identification of the parent compound throughout the study. No CO<sub>2</sub> evolution occurred during the study. A decrease in recoverable PFOS was seen (20-30%) at both test concentrations at the end of the test, but no organic degradation products were reported. Due to these technical issues, definite conclusions concerning the biodegradation of PFOS cannot be drawn from either of these studies.

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The third test (STS-302) consisted of incubating 21.2 mg/kg potassium PFOS in potting soil for 63 days at 22°C. Aerobic conditions were present throughout the test. In this test, no biodegradation of PFOS occurred, as measured by loss of parent compound by LC/MS throughout the incubation time period.

In the fourth test (STS-303), PFOS (20.8 mg/L) was incubated with a digester sludge mixture under aerobic conditions for 56 days at 35°C. No degradation was seen, as measured by loss of parent compound throughout the study by LC/MS.

Two studies addressing the biodegradation potential of PFOS have been published. Key et al. (1998; STS-124) incubated the potassium salt of PFOS (concentration and isomer(s) not specified) with Pseudomonas sp. (Strain D2) under aerobic conditions for 24 hours at 30°C. No biodegradation occurred, as measured by fluoride ion formation, either in the test mixture or in the abiotic control. Other test fluorocarbons were shown to be degraded in this test system.

Meesters and Schröder (2004; STS-123) incubated PFOS (5 mg/L) with wastewater treatment plant effluent microorganisms under both aerobic and anaerobic conditions. No degradation was observed under aerobic conditions, as measured by loss of PFOS from the incubate, using LC/MS. Under anaerobic conditions, a rapid loss of PFOS was measured in the incubate; however, no organic metabolites were detected by LC/MS and no fluoride ion formation occurred. PFOS was also not found to adsorb to the glass walls of the flask. The authors thus could not explain their findings and could not conclude that PFOS was degraded.

As discussed above, a number of biodegradation studies of varying time periods have been conducted on PFOS to date. Although some are unacceptable from a technical standpoint, other GLP-equivalent studies have clearly indicated that PFOS was not biodegradable in the test systems used -- soil, sludge, sediment, under both anaerobic and aerobic conditions. Whether or not environmental microorganisms can ultimately mutate in the long-term presence (years) of this chemical to use it as a nutrient is not known at this time.

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### 2.3 Environmental Fate and Transport Summary

Based on its various chemical fate properties detailed above, PFOS would be expected to be quite stable in the environment. In a fresh water surface water system, PFOS possesses good solubility and is not likely to undergo hydrolysis or photolysis. Its very low vapor pressure and air-water partition coefficient indicate that this chemical would not appreciably transfer to ambient air from a surface water body. Thus, PFOS once discharged to a surface water body would tend to remain there for long periods of time.

Its relatively low  $K_{oc}$  value would indicate that it also would not quantitatively bind to suspended solids or sediments in a surface water body. Due to its surface active properties, a direct measurement of its  $K_{ow}$  could not be performed; however, based on its individual solubilities in water and n-octanol, a log  $K_{ow}$  value of -1.08 was calculated. Thus, PFOS also is not lipophilic, as are the organochlorine compounds. It thus would not be expected to preferentially chemically partition to the hydrophobic components in the environment including biota.



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### **3.0 BIOCONCENTRATION/BIOACCUMULATION/BIOMAGNIFICATION**

#### **3.1 Laboratory Tests**

Two unpublished PFOS bio-concentration tests have been conducted with bluegill sunfish. In the first study, conducted by 3M in 1978 (226-0061), bluegills were placed in effluent water from 3M's Decatur, AL plant for 22 days. Control fish were placed in Tennessee River water. Water concentrations of PFOS were not measured, nor were the fish tissue concentrations. 3M qualitatively identified PFOS in whole fish homogenates of the exposed fish by TLC. No PFOS was detected in the tissues of unexposed fish. Of the exposed fish, 12/30 died during the test; none of the control sample fish died. The effluent-exposed fish were also of smaller average weight (25.7 vs. 33.0 grams). Based on the information provided, this study cannot be utilized to develop a BCF for PFOS.

The second study (STS-328) was a GLP flow-through bioconcentration study on the potassium salt of PFOS conducted by Wildlife International Ltd. in 2001. According to 3M's summary sheet (STS-329), this was the n-octane isomer of PFOS. Juvenile bluegills were used in the test. This was a flow-through exposure test following the method described in OPPTS 850.1730 and OECD 305. The test involved fish exposure to two concentrations of PFOS, 0.086 mg/L and 0.87 mg/L. PFOS measurements in the water and in the fish were taken at various times during a 62 day uptake phase and during a 56 day depuration phase with the lower concentration, using LC/MS. The higher concentration was toxic to the fish after 35 days, so this part of the study was stopped at that time. The study reported the following steady-state bioconcentration factors using the lower PFOS concentration:

edible portion of fish	1124 L/kg
whole fish	2796 L/kg

The test report indicated a depuration half-life of 86 days for the edible portions of the fish and 112 days for whole fish. Due to the level of variability in the concentration of PFOS measured at the various time points in the study and the fact that the exposure period

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was less than the depuration half-life, the reported BCFs were calculated from averaged time point data using the procedure outlined in OPPTS 850.1730 Guidance Document. Since these values are extrapolations, they should be considered as first approximation values.

Ankley et al. (2005) (STS-318) exposed fathead minnows to PFOS for 21 days at concentrations of 0.03, 0.1 and 0.3 mg/L in a flow-through test methodology. Weekly sampling of the water was performed to verify exposure conditions. Tissue samples (blood, liver, gonads) were analyzed for PFOS only at the end of the experiment. Blood had higher levels of PFOS than liver, which had higher levels than the gonads. Based on data presented in figure form, female tissues had higher PFOS levels than males at each of the three concentrations. For plasma, BCF ranged 1167-1300 L/kg (males) and 1600-1750 L/kg (females). For liver, BCF ranged 250-400 L/kg (males) and 900-1333 L/kg (females). For gonads, BCF ranged 167-367 L/kg (males) and 800-1000 L/kg (females). This study shows bioconcentration of PFOS into this fish, but is not adequate to calculate appropriate BCFs for an ambient water quality standard; it is not known if the steady-state conditions existed at the end of the experiment, and PFOS was not quantified in either whole fish or edible portions.

Martin et al. (2003) (STS-160) exposed juvenile rainbow trout to PFOS at a single water concentration in a mixture of perfluorinated chemicals in a flow-through experimental design. Animals were exposed for 12 days, followed by a 33 day depuration phase. The carcass half-life for PFOS was reported at 15 days. No toxicity to the fish was observed. Measurements of both water and fish tissue concentrations of PFOS were taken throughout both phases. Kinetic modeling of the data yielded the following reported steady-state BCFs for PFOS:

Carcass	1100 ± 150 L/kg
Blood	4300 ± 570 L/kg
Liver	5400 ± 860 L/kg

Since the carcass equates to the edible portion of these fish, this BCF represents an approximation of the bioconcentration potential of this chemical. However, since the fish

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were exposed to multiple fluorinated chemicals simultaneously, no definitive BCF for PFOS can be resolved. In addition, no whole fish data were obtained by the investigators. The experimental depuration data suggests a biphasic elimination phenomenon; however the authors did not assume so. This decision therefore could have over-estimated the elimination rate constant and thus under-estimated the steady-state BCFs.

## 3.2 Environmental Studies

### 3.2.1 General Surveys

There have been a number of environmental survey studies conducted recently that have documented that PFOS is detectable in the tissues of a variety of animal species at various trophic levels. Since PFOS has been found to concentrate in the liver, these studies have focused on this tissue. All data are presented in this section of the report as PFOS tissue concentrations on a wet weight basis. Each study has added to the environmental pattern of PFOS, but most in general do not contain sufficient information to estimate BCF/BAF/BMF. Tables A (Freshwater Ecosystems) and B (Marine Ecosystems) summarize these data.

Kannan *et al.* (2001) (STS-106) studied the PFOS concentrations in oysters from 77 locations in the Gulf of Mexico and Chesapeake Bay. PFOS was detected in 51 of the 77 locations sampled. PFOS concentrations ranged from <42 to 1,225 ng/g. Oysters from the mouth of the Lavaca River, Texas had the highest concentrations. Only one oyster sample collected from Chesapeake Bay had an identified concentration of PFOS. Water samples at the collection points were not analyzed for PFOS, however.

Kannan *et al.* (2001) (STS-132) reported that PFOS was detected in the livers of 15 species of marine mammals collected off the coasts of Florida, California and Alaska, from the Northern Baltic Sea, the Arctic and Sable Island (Canada). The pattern of concentration reflected the feeding habit of the animal; the bottlenose dolphin, who feeds near shore, had the highest tissue levels (liver - 1520 ng/g), whereas the pygmy sperm whale which feeds exclusively off-shore had the lowest liver levels (14.8 ng/g). The seals

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and sea lions from California had lower tissue concentrations of PFOS than the dolphins and whales of Florida. River otters contained greater liver concentrations (329 ng/g) than ocean otters (8.9 ng/g) which could be attributed to their closer location to PFOS sources and/or the fact that PFOS is much more soluble in fresh water than in sea water. In their study in Alaska, the polar bear livers contained greater amounts of PFOS (350 ng/g) than had its prey (northern fur seal, ringed seal, gray seal; <10-122 ng/g)), suggesting food chain bio-magnification. No age-dependent increase in PFOS concentrations was observed.

Kannan et al. (2002a) (STS-133) reported that sea birds from both Japan and Korea contained PFOS in their livers. The common cormorant had the highest concentrations in the Japan study (mean of 380 ng/g), while the black-headed gull had the highest concentration in the Korean study (mean = 296 ng/g).

Kannan et al. (2002b) (STS-134) in this study evaluated the levels of PFOS in the livers of mink and river otters from the United States. Mink from Illinois (47-5140 ng/g) and South Carolina (650-3110 ng/g) had higher PFOS concentrations than animals from Massachusetts (20-1100 ng/g) and Louisiana (40-320 ng/g). No differences were noted with the PFOS tissue levels with respect to age or sex of the animals. River otters from Oregon (34-994 ng/g) and Washington (25-442 ng/g) had liver PFOS concentrations similar to the mink levels. In a controlled feeding experiment, mink were fed carp containing 270 ng/g PFOS vs. their normal diet that contained 87 ng/g. The concentration of PFOS in the animal's livers increased proportionally to the percentage of carp in their diet. Based on the data reported, a BMF of 18 (average; range 11-23) was calculated by these investigators. The authors noted, however, that other fluorocarbons were present in these studies; ones that are metabolized to PFOS, so the BMFs calculated may be over-stated.

Kannan et al. (2002c) (STS-135) surveyed a number of marine animals from both the Mediterranean and Baltic Seas in this study. PFOS was measured in both the liver (<1.4-110 ng/g) and muscle tissue (77 ng/g) of Mediterranean dolphins. PFOS was also detected in the livers of cormorants (32-150 ng/g), tuna (21-87 ng/g) and swordfish (<1-13 ng/g). In the Baltic sea, PFOS was present in the livers of gray seals (mean = 243

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ng/g in males and 190 ng/g in females) and ringed seals (mean = 490 ng/g in males and 430 ng/g in females). No differences in the levels were found with respect to animal age. PFOS was also detected in the white-tailed eagle (<3.9-127 ng/g), but at levels less than bald eagles in the continental United States.

Van De Vijver et al. (2003a) (STS-136) reported on PFOS concentrations in the livers of marine animals in Western Europe. Harbor seals, gray seals and white-beaked dolphins fell into the highest trophic levels in the study and also had the highest reported liver PFOS concentrations (<10-532 ng/g, 11-233 ng/g, 14-443 ng/g, respectively). The off-shore feeders (sperm whales, fin whales, striped dolphins, white-sided dolphins) had lower tissue PFOS concentrations (<10-52 ng/g) than the inshore species.

Taniyasu et al. (2003) (STS-137) reported the PFOS concentrations in the livers of various aquatic fish and birds along the shore of Japan. They also reported water concentrations of PFOS at the locations of the animal samples. The salt water concentrations of PFOS varied 8-59 ng/L (mean = 26 ng/L) in Tokyo Bay. The Tama River (fresh water) had the highest measured concentration, at 157 ng/L. Based on the PFOS levels in the livers of the various fishes, BCF values for PFOS varied 274-41,600 (mean = 8,540). BCFs from the ocean locations were less than those from the fresh water locations.

Hoff et al. (2003) (STS-140) studied PFOS levels in the livers and muscle tissue in two fish species (bib and plaice) off the Belgian continental shelf (salt water) and the Western Scheldt (fresh water). For both species, the estuary fish contained greater levels of PFOS than did the ocean fish (mean of 180 ng/g vs. mean of 45 ng/g for bib; mean = 2750 ng/g vs. mean of 900 ng/g for plaice). Liver enzymes were elevated in the blood of the bib but not in the plaice suggesting liver damage in the one species of animal.

Van De Vijver et al. (2003b) (STS-141) evaluated the PFOS concentrations in various aquatic invertebrates in the Western Scheldt estuary and the southern North Sea. In this study, whole body data were reported. The PFOS levels in starfish ranged 9-176 ng/g. The levels in shrimp ranged 19-520 ng/g, and the levels in crabs ranged 24-877 ng/g. A

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PFOS pollution gradient was observed in these organisms along the estuary, with the highest concentrations near Antwerp.

Bossi et al. (2005) (STS-204) reported on the PFOS concentrations in the livers of various marine mammals, fish and birds from Greenland and the Faroe Islands. The PFOS concentration in the short-horn sculpin (13-18 ng/g) was less than the levels detected in the ringed seal (10-67 ng/g) which was less than the level in the polar bear (1245-1325 ng/g) from Greenland. These data suggest food chain biomagnification of this chemical. The Faroe Islands data included only the pilot whale and fulmar. The pilot whale livers contained 28-65 ng/g PFOS, while the fulmar livers contained 24-29 ng/g.

Houde et al. (2005) (STS-205) reported on the liver tissue levels of PFOS in free-range dolphins off the east coast of the United States and off of Florida's west coast. In general, the tissue concentrations corresponded with the general pollution levels in the waters where the animals were caught. Animals caught off of Charleston, S.C. had the highest tissue levels, at a mean of 1315 ng/g. This was followed by mean levels of 781 ng/g and 751 in Sarasota Bay and Delaware Bay, respectively. Animals caught by Bermuda had the lowest mean levels (49 ng/g). No sex difference was observed in this study either, which is interesting in that these authors reported that a calf had about ten times the levels of PFOS of its lactating mother dolphin.

Smithwick et al. (2005) (STS-206) reported PFOS levels in the livers and plasma of polar bears from five locations in the North American Arctic and two locations in the European Arctic. Animal populations in South Hudson Bay (mean = 2730 ng/g) had significantly higher PFOS levels than western populations (mean = 729 ng/g for Chukchi Sea; mean = 1320 for Northwest Territories). No sex difference was observed in this study.

Holmstrom et al. (2005) (STS-207) reported that the PFOS concentrations in Baltic Sea guillemot eggs showed an upward temporal trend from 1968 to 2003. An almost 30-fold increase was noted to 1997. A significant drop in the PFOS concentration (~40%) was seen from 1997 to 2003. No explanation was given, and this timeline does not correlate with the stoppage of fluorocarbon production.

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Van De Vijver et al. (2005) (STS-310) monitored the tissue levels of PFOS in harbor seals in the Dutch Wadden Sea. The reported mean concentrations of PFOS in various tissues were:

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kidney - 378 ng/g	liver - 175 ng/g
blubber - 100 ng/g	muscle - 59 ng/g

In general, older animals contained higher levels in liver tissue. No sex differences were found; however, a trend toward higher female liver and kidney levels vs. males was mentioned.

Bossi et al. (2005) (STS-311) measured the PFOS concentrations in the livers of adult Ringed Seals from Greenland. The east coast animals contained higher levels than the west coast animals. It was noted that the average age of the seals from the east coast was 1-3 years greater than the animals from the west coast. The mean PFOS liver concentration in the east coast animals was reported 25.5-95.6 ng/g (several populations), whereas the mean PFOS liver concentrations in the west coast animals was 12.5-27.9 ng/g (single population). A definite upward trend in tissue levels in both sets of animals was seen from 1985-2003. No sex differences in PFOS concentration were observed.

Verreault et al. (2005) (STS-312) examined PFOS levels in various tissues of adult Glaucous Gulls from the Norwegian Arctic. These gulls are a top avian scavenger-predator in this area. Plasma levels of PFOS (mean = 134 ng/g) were similar to the levels reported in livers and eggs (mean = 104 ng/g), which were much higher than in brain tissue (mean = 2 ng/g). No sex differentiation of the data was performed.

Olivero-Verbel et al. (2006) (STS-313) examined PFOS concentrations in various tissues of a piscivorous bird (pelican) and a bottom feeder fish (mullet) residing off the coast of Columbia, South America (Cartegena Bay and Covenas). PFOS levels in the bile of the fish were reported as 0.75-3.6 ng/ml. In the pelican, the spleen and liver exhibited the highest PFOS concentrations (59.8 and 36.7 ng/g, respectively), which were much higher levels than the mean values found in the other tissues (lung - 7.5 ng/g; kidney - 4.3 ng/g; brain - 3.5 ng/g; heart - 2.1 ng/g; muscle - 0.8 ng/g).

Hoff et al. (2005) (STS-116) quantified the PFOS concentrations in the livers of three fresh water fish species in the Flanders region of Belgium. The animals studied were the



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gibel carp, carp and eel. The liver PFOS concentrations in the two carp species were reported at 11.3-1822 ng/g (carp) and 11.2-781 ng/g (gibel carp), whereas eel liver was reported at 17.3-9031 ng/g. In carp, the liver PFOS concentration was positively related to the serum ALT activity (liver damage) and negatively related to serum protein levels and electrolytes,  $\text{Cl}^-$ ,  $\text{Na}^+$  and  $\text{Ca}^+$  concentrations. In eel, the PFOS levels were positively correlated with serum ALT activity and negatively correlated with serum protein level. Visual examination of the reported graphical data suggest tissue PFOS thresholds of 500 ng/g for all three measured toxicological endpoints.

Morikawa et al. (2006) (STS-314) studied the tissue levels of PFOS in two species of wild turtles caught along the Ai river in Japan. Surface water samples were taken at the times of turtle capture to determine approximate bioconcentration factors. Along the river, PFOS concentrations varied from 2.9 to 12.3 ng/L. The mean serum PFOS concentrations of the animals caught in the river ranged 89.5 to 283.3  $\mu\text{g/L}$ . These data led to reported water-serum BCF values of 10,000 - 38,000. Since the data were not normally distributed, a geometric mean BCF value of 10,964 was calculated.

Smithwick et al. (2005) (STS-315) reported on the PFOS concentrations in liver tissue of polar bears from east Greenland. Polar bears are the apex predator, feeding primarily on ringed seals and bearded seals. The mean liver tissue level of PFOS was reported as 2470 ng/g, which is similar to the levels in bears from Hudson Bay, Canada (3100 ng/g), but higher than what was found in bears from Alaska (350 ng/g) by other investigators. No sex or age differences in the PFOS levels were found in this study.

So et al. (2006) (STS-316) studied the PFOS concentrations in mussels and oysters from the coasts of South China and Japan. PFOS concentrations (whole animal) ranged from 0.11 to 0.59 ng/g. Oysters from Tokyo Bay had the highest concentrations.

Dai et al. (2006) (STS-331) reported on the concentration of PFOS in the serum of both the Giant Panda and the Red Panda in China. PFOS levels measured 0.8-73.8  $\mu\text{g/L}$  (mean = 20.4  $\mu\text{g/L}$ ) in the Red Panda and 0.8-19.0  $\mu\text{g/L}$  (mean = 11.1  $\mu\text{g/L}$ ) for the Giant Panda. No age- or sex-differences were observed. Higher concentrations were seen in samples collected from animals from zoos near urbanized or industrialized areas.

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Furdui *et al.* (2007) (STS-330) analyzed lake trout whole tissue homogenates for a variety of PFCs. The fish were collected in 2001 from each of the five Great Lakes. The lowest average total PFC level was found in the fish from Lake Superior (13 ng/g); the highest average total PFC level was found in fish from Lake Erie (152 ng/g). As with other water bodies, PFOS was detected at the highest concentrations of any of the PFCs tested. The PFOS data from this study and the calculated BAFs by the authors are provided below:

<u>Lake</u>	<u>Average PFOA Concentration (ng/g)</u>	<u>Whole Fish BAF (L/kg)</u>
Superior	4.8 +/- 0.4	20,000
Michigan	16.0 +/- 3.0	6,300
Huron	39.0 +/- 10.0	16,000
Erie	121.0 +/- 14.0	25,000
Ontario	46.0 +/- 5.0	<u>8,000</u>
		Mean = 12,500

### 3.2.2 Regional Food Web Studies

There were three food web studies that have been published on PFOS, two fresh water studies in the Great Lakes area and one marine study.

#### Freshwater

In the first study, Martin *et al.* (2004) (STS-142) quantified the concentrations of PFOS from benthic macroinvertebrates through a top predator fish, lake trout, in Lake Ontario. The mean PFOS concentration results (whole fish data) indicated the following:

benthic invertebrate : Diporeia - 280 ng/g  
benthic invertebrate : Mysis - 13 ng/g  
forage fish : alewife - 46 ng/g  
forage fish : rainbow smelt - 110 ng/g  
forage fish : slimy sculpin - 450 ng/g  
predator fish : lake trout - 170 ng/g

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One striking finding in this study was that the second highest mean concentration for PFOS was detected in the benthic macroinvertebrate *Diporeia*, which occupies the lowest trophic level of all of the organisms analyzed. Another interesting finding is that the sculpin are more heavily contaminated than lake trout. Food web data on Lake Ontario indicate that lake trout consume only about 2% sculpin in their diet, with the balance made up of alewife (90%) and smelt (7-8%). Sculpin feed on Mysis and *Diporeia*. Based on the tissue data, it would suggest that their major diet in this lake is *Diporeia*. For PFOS, a statistically significant trend was observed whereby concentrations increased with increasing trophic levels, suggesting biomagnification in the "mainly Pelagic" food web.

These authors calculated a diet weighted prey/lake trout BMF of 0.38 kg/kg (sculpin) and 3.7 kg/kg (alewife). They also reported an upward temporal trend in lake trout (PFOS) levels from years 1980 - 2000. No surface water data were collected in this study, however, from which to calculate BCFs/BAFs.

Kannan et al. (2005) (226-8000) studied PFOS levels in a Great Lakes benthic food web at three separate sites - Raisin River, St. Clair River and Calumet River. The summary results in this study were:

	<u>Apparent BCF/BAF (L/kg)</u>
surface water -- 1.9-3.9 ng/L	---
algae -- 2.4-3.1 ng/g	1000
amphipods -- <2-2.9 ng/g	1000
zebra mussel -- <2-3.1 ng/g	1000
round goby -- 4.1-19.1 ng/g (liver)	2000 - 4000
smallmouth bass -- <2-41 ng/g (liver)	1000 - 3000

These authors also reported that Chinook salmon and lake whitefish livers contained PFOS levels (mean = 67-100 ng/g) 10-20 times their prey fish, and that bald eagles and mink livers (1740 and 18,000 ng/g, respectively) contained PFOS levels up to 10-100 times salmon, carp and turtle levels, respectively. In turtles, they found that the males

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(137 ng/g) contained much higher levels than females (6 ng/g), suggesting that PFOS can be transferred out of the female body by egg laying.

#### Salt Water

In the Marine food web study, Tomy et al. (2004) (STS-317) examined PFOS levels in the livers of animals in a variety of trophic levels from an eastern arctic marine food web. Their mean data are presented below:

zooplankton -- 1.8 ng/g  
clams -- 0.28 ng/g  
glaucous gulls -- 20.2 ng/g (liver)  
shrimp -- 0.35 ng/g  
cod -- 1.3 ng/g (liver)  
beluga whale -- 12.6 ng/g (liver)  
walrus -- 2.4 ng/g (liver)  
narwhal -- 10.9 ng/g (liver)  
black-legged kittiwake -- 10.0 ng/g (liver)

They calculated the following trophic level BMFs (kg/kg):

<u>Predator : Prey</u>			<u>Predator : Prey</u>		
walrus	: clam	4.6	kittiwake	: cod	5.1
narwhal	: cod	7.2	glaucous gull	: cod	9.0
beluga	: cod	8.4	cod	: zooplankton	0.4

The authors contend that significant amounts of PFOS precursor compounds were found that could contribute to the body burden in the higher trophic level animals, thus skewing the calculated BMFs that were calculated.

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### 3.2.3 Minnesota Site-Specific Studies

In 2006, the Minnesota Pollution Control Agency (MPCA) quantified perfluorinated compound levels in Lake Calhoun in Minneapolis, Minnesota, the St. Croix River along the Minnesota/Wisconsin border, and in several stretches of the Mississippi River (Pools 3, 4, 5 and 5a). Surface water samples were collected and submitted for analysis of PFC concentrations from Lake Calhoun, the St. Croix River and Mississippi River Pool 3. The summary results of the surface water samples submitted for PFOS in this study ranged as follows:

Lake Calhoun	0.10400 - 0.11700 ng/mL
St. Croix River	<0.00560 - <0.00930 ng/mL
Mississippi River Pool 3	0.01900 – 0.03780 ng/mL

Fish tissue samples including fillets and whole animal were submitted by the MPCA for PFC analysis from Lake Calhoun, the St. Croix River and the Mississippi River (Pools 3, 4, 5 and 5a). The fillets for this study were defined as the edible (by humans) portion of the fish tissue; the samples were scaled but the skin remained at the time of laboratory analysis. The remaining portions of some of the fish (minus the scales) were processed by the analytical laboratory and added to the fillet data in order to obtain whole animal PFC concentrations. This methodology (scaling the larger fish prior to analysis) may have underestimated the animal's PFOS burden that wildlife would encounter because it is possible, given the propensity of this chemical to adhere to materials, that the surface scales of the fish could have had PFOS "stuck" on their surfaces, thus presenting an additional dose to piscivorous animals.

PFOS was identified in all of the fishes submitted for analysis from Lake Calhoun and from the Mississippi River (Pools 3, 4, 5 and 5a). No or trace concentrations of PFOS were identified in the fishes submitted from the St. Croix River. The detailed results of the fish tissue analysis are summarized in Table C. The complete analytical reports prepared by Axys are included in Appendix B.

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### Lake Calhoun

With respect to the Lake Calhoun study, two species of fish were caught -- female bluegills, and male and juvenile white suckers. Five bluegills were caught. The range of fillet concentrations of PFOS was 181-373 ng/g. The data were normally distributed, with an arithmetic mean of 319 ng/g. Based on an average water concentration of 0.11 ng/ml (n=3; 104-117 ng/ml), the calculated BAFs for these fish were:

maximum	3390 L/kg
arithmetic average	2896 L/kg
minimum	1645 L/kg

The single male white sucker that was caught had no detectable PFOS in its fillet (detection limit = 3.5 ng/g). Of the four juvenile white suckers caught, only one had detectable PFOS levels, 49.1 ng/g. Based on this tissue level, a BAF of 446 L/kg was calculated.

### Mississippi River - Pool 3

Two species of fish were also caught in this stretch of the Mississippi River -- male bluegills; and juvenile white bass. Three male bluegill fillets were analyzed for PFOS. The data ranged from 108 to 440 ng/g, and was lognormally distributed. The geometric mean value was calculated as 180 ng/g. Based on an average water concentration of 0.0266 ng/ml (n=3; 0.0190-0.0378 ng/ml), the calculated BAFs for these fish were:

maximum	16,541 L/kg
geometric mean	6763 L/kg
minimum	4624 L/kg

Five juvenile white bass were caught and their fillets analyzed for PFOS. The range of tissue levels of PFOS was 86.7 -154 ng/g, normally distributed. The arithmetic mean value was calculated as 132 ng/g. Based on the water concentration of PFOS detected in Pool 3, 0.0266 ng/ml, the following BAFs were calculated:

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maximum	5789 L/kg
arithmetic average	4967 L/kg
minimum	3259 L/kg

#### Mississippi River - Pool 4

Only one fish species was caught in this Pool from which the fillets were analyzed for PFOS -- male and female bluegills. The PFOS concentration in the three male fish fillets varied 28.1-152 ng/g. The geometric mean was calculated as 75 ng/g; the arithmetic mean was 94 ng/g. The PFOS concentration in the two female fish fillets varied 45.5-98.3 ng/g, with the arithmetic mean of 72 ng/g. No water samples were taken in this Mississippi River pool, however.

#### Mississippi River - Pool 5

Six species of fish were caught in Pool 5 of the Mississippi River and had their fillets analyzed for PFOS -- male, female and juvenile bluegill; male, female and juvenile smallmouth bass; male and female largemouth bass; male and juvenile walleye; female and juvenile northern pike; and male and juvenile channel catfish. No water samples were taken in this Mississippi River pool, however.

Three female bluegill fillets were assayed for PFOS. The normally distributed data varied 40.3-77.2 ng/g, with an arithmetic mean of 62.4 ng/g. The one male fillet contained 42.7 ng/g PFOS, while the one juvenile fish fillet contained 94.7 ng/g, considerably greater than any of the other mature fish.

The PFOS concentration in the two female smallmouth bass fillets varied 93.5-150.0 ng/g, with an arithmetic mean of 121.8 ng/g. The PFOS concentration in the two male fillets ranged 47.7-104.0 ng/g, with an arithmetic mean of 75.9 ng/g. The one juvenile fish fillet had a PFOS concentration of 83.5 ng/g.

The two female largemouth bass fillets had PFOS levels of 82.9-91.1 ng/g; arithmetic mean of 87.0 ng/g. The three male largemouth bass fillets had PFOS levels of 74.3-107.0 ng/g, with an arithmetic mean of 85.3 ng/g.

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The two male walleye fillets had PFOS levels of 27.1-30.4 ng/g; arithmetic mean of 28.8 ng/g. The two juvenile fish fillets had PFOS levels of 60.6-93.2 ng/g (mean of 76.9 ng/g), which were considerably greater than the mature fish levels.

The three female northern pike fillet PFOS levels varied 91.2-230.0 ng/g, with an arithmetic mean of 140 ng/g. The two juvenile fish fillets had PFOS levels of 12.2-130.0 ng/g (mean of 71.1 ng/g), considerably less than the mature fish.

Finally, the one juvenile catfish that was caught had a PFOS level of 9.6 ng/g in its fillet, compared to a non-detected level in the male fish fillet (detection limit of 3.3 ng/g).

#### Mississippi River - Pool 5a

Four species of fish were caught in Pool 5a -- male and female bluegills; male and female smallmouth bass; female and juvenile walleye; and male and female channel catfish. No water samples were taken in this Mississippi River pool, however.

Four male bluegill had PFOS levels in their fillets ranging 34.0-99.2 ng/g. The mean value was calculated at 62.6 ng/g. The one female bluegill had a fillet PFOS concentration of 55.5 ng/g.

The four male smallmouth bass had fillet PFOS levels ranging 45.0-116.0 ng/g. The geometric mean value was 69.4 ng/g. The one female smallmouth bass had a fillet PFOS level of 67.1 ng/g.

The four female walleye had fillet PFOS levels ranging 41.0-103.0 ng/g. The geometric mean value was 58.5 ng/g. The one juvenile walleye had a fillet PFOS level of 75.4 ng/g.

The single male catfish had undetected amounts of PFOS in its fillet (detection limit of 3.26 ng/g). The three female catfish had fillet PFOS levels ranging 9.6-18.6 ng/g. The mean value was 13.9 ng/g.



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### 3.3 Summary Hypothesis and Proposed BAF

As discussed earlier in this section, there have been a number of environmental surveys conducted, both in freshwater and marine systems, that document that PFOS does bioaccumulate in food webs. A summary of the relevant data from these studies is presented on Table A (Freshwater Ecosystem) and Table B (Marine Ecosystems). The majority of these data, although supporting the concept that PFOS does bioaccumulate in food webs, is insufficient to quantitatively calculate accurate bioconcentration, bioaccumulation or biomagnifications factors due to:

- lack of water quality data in area(s) where the aquatic life were sampled,
- only select species were sampled, sometimes only one species,
- lack of food chain/food web information relating to various trophic level animals that were tested.

Two freshwater food web studies have been completed (Martin et al., 2004; Kannan et al., 2006) wherein more complete datasets have been obtained. In the Martin et al. study, no water quality data were collected, so bioconcentration/bioaccumulation factors could not be calculated. However, since multiple trophic level animals were sampled in a single region of Lake Ontario, biomagnification factors could be approximated. Based on the data provided, the following BMFs are calculated (assuming single food source):

mysis - alewife	3.5 kg/kg
diporeia - alewife	0.16 kg/kg
diporeia - smelt	0.39 kg/kg
mysis - smelt	8.5 kg/kg
mysis- sculpin	34.6 kg/kg
diporeia - sculpin	1.6 kg/kg
sculpin - trout	0.38 kg/kg
alewife - trout	3.7 kg/kg

In nature all of the higher trophic animals would have a mixed diet, so the above values can only be viewed as approximate. In general, however, with the one exception of the

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mysis-sculpin food chain, the data indicate that PFOS either biomagnifies a small amount (BMF <10) or not at all (BMF <1) in upper trophic levels.

In the study by Kannan et al. surface water data were collected so a better idea can be obtained concerning the food chain behavior of PFOS. Based on their data, it is apparent that the lower trophic level animals - algae, amphipods and mussels - are able to bioconcentrate PFOS to a considerable extent (~1000 fold over the water concentration). Then, as was observed earlier in the Martin et al. study, further biomagnification is relatively small (<5).

These data suggest that PFOS undergoes bioaccumulation in nature primarily from the lowest trophic level biota. These animals/plants have shown the largest bioconcentration potential (~1000), but whether this is true chemical absorption into their cells or simple chemical adsorption onto their surfaces is unknown at this time. Also unknown is their true potential to bioconcentrate PFOS. It is possible that these lowest trophic level organisms could simply continue to adsorb/absorb PFOS until they die; and therefore, what is observed in nature are those biota that are still in the process.

The other interesting phenomenon that seems to be occurring with PFOS has to do with the higher trophic levels (non-aquatic life animals). Based on the liver data presented in Table A, it is apparent the PFOS biomagnification takes another large jump in mink and eagles. Whether this phenomenon occurs due to their specific metabolism of PFOS (or lack thereof) is not known. Controlled studies addressing this issue are needed. In the marine ecosystem, a similar jump in PFOS biomagnifications is seen with the polar bear but not with birds.

There have been three controlled laboratory bioconcentration tests with PFOS. Wildlife, Intl. (2001) conducted a flow-through PFOS bioconcentration test using juvenile bluegills. Exposure occurred for 62 days, which was then followed by a 56-day depuration period. Based on the data obtained, the following BCFs were calculated for PFOS: whole fish – 2796 L/kg; edible portion of fish - 1124 L/kg. This GLP study was conducted in a technically correct manner, however the results are not ideal from which to develop a definitive BCF for PFOS. The reasons for this shortcoming include:

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- The fact that only one PFOS concentration was used to calculate BCFs.
- Since the estimated depuration half-life was calculated at 86 days, the exposure period (56 days) was relatively short. Given the variability in the data during this phase, an accurate kinetic extrapolation cannot be made.

Ankley et al. (2005) exposed fathead minnows to PFOS for 21 days at multiple water concentrations of the chemical. Tissue samples (plasma, liver, gonads) were obtained and analyzed for PFOS only at the end of this exposure period. These data are not appropriate for calculating a BCF for PFOS because:

- Whole animal and edible portions were not quantified with respect to PFOS concentration.
- Since no depuration phase was included in the study and no time point data for PFOS in the fish were taken during the exposure, there is no way of knowing if steady-state conditions existed at the time of the tissue analysis.

Martin et al. (2003) exposed juvenile rainbow trout to PFOS at a single water concentration (as a component of a mixture of perfluorinated chemicals) in a 12-day exposure, 33 day depuration phase flow-through test. The carcass (edible portions of fish) depuration half-life was calculated at 15 days. Therefore, as with the Wildlife, Intl. study, steady-state BCFs were calculated using kinetic extrapolation. The BCFs reported in this study for PFOS were:

carcass	1100 L/kg
liver	5400 L/kg
blood	4300 L/kg

Whole fish data were not obtained, so a BCF for ecological risk analysis cannot be developed from their data. The carcass BCF is likely a somewhat accurate value for the PFOS BCF. However, since the study involved the simultaneous exposure to multiple perfluorinated compounds, some of which can be metabolized to PFOS and others that

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may competitively compete with PFOS for absorption, metabolism, etc., the chemical-chemical interactions could significantly affect the test results that were obtained.

The site-specific analytical data collected from water samples in bluegill fillets from Lake Calhoun and Pool 3 of the Mississippi River can also be used to estimate a BCF/BAF for PFOS. Based on the data obtained (Table C), the following range of BCF/BAF values were calculated:

		<u>Range</u>	<u>Geo. Mean</u>
Lake Calhoun	bluegill	1645 - 3390	2802
Mississippi River	bluegill	4624-16,541	6771
Mississippi River	white bass	3259-5789	4861

The geometric mean for the Mississippi River fish at Pool 3 is 5737.

These calculated values are higher than the BCFs calculated from the laboratory studies, but are in general agreement with the data reported by other investigators conducting natural food web investigations (Martin *et al.*, 2004; Kannan *et al.*, 2005). This inconsistency may likely be due to the fact that with this chemical, the higher trophic level species in the environment may obtain a significant portion of their daily doses from their food chains, as compared to the water column.

Because of the statistically significant differences between the BAFs for the two water bodies, site-specific values are proposed. The Lake Calhoun final BAF is based upon the bluegill data. The MPCA staff determined that using the juvenile white sucker data in Lake Calhoun will artificially lower the final BAF for the Lake Calhoun associated PFOS water quality criteria. The final BAF for Lake Calhoun was calculated as the geometric mean of the data. This BAF is 2802 L/kg. The Mississippi River final BAF was calculated as the geometric mean from both the bluegill and the white bass data. This value is 5737 L/kg.

**TABLE A: Freshwater Ecosystems**

<b>Animal</b>	<b>Location</b>	<b>PFOS Conc.</b>	<b>Tissue</b>
Algae	3 Gr. Lake rivers	2.4 - 3.1 ng/g	whole animal
Amphipods	3 Gr. Lake rivers	<2 - 2.9 ng/g	whole animal
Diporeia	Lake Ontario	280 ng/g	whole animal
Mysis	Lake Ontario	13 ng/g	whole animal
Zebra mussel	3 Gr. Lake rivers	<2 - 3.1 ng/g	whole animal
Round Goby	3 Gr. Lake rivers	4.1 - 19.1 ng/g	liver
Smallmouth Bass	3 Gr. Lake rivers	<2 - 41 ng/g	liver
Alewife	Lake Ontario	46 ng/g	liver
Rainbow Smelt	Lake Ontario	110 ng/g	liver
Slimy Sculpin	Lake Ontario	450 ng/g	liver
Lake Trout	Lake Ontario	170 ng/g	liver
Lake Trout	Gr. Lakes	4.8-121 ng/g	whole animal
Lake Whitefish	Gr. Lakes	67-100 ng/g	liver
Gibel Carp	Belgium	11.2 - 781 ng/g	liver
Carp	Belgium	11.3 - 1822 ng/g	liver
Eel	Belgium	17.3 - 9031 ng/g	liver
Turtles	Gr. Lakes	6 (♀) - 137 (♂) ng/g	liver
Mink	Gr. Lakes	18,000 ng/g (x)	liver
Mink	Illinois	47 - 5140 ng/g	liver
Mink	S. Carolina	650 - 3110 ng/g	liver
Mink	Massachusetts	20 - 1100 ng/g	liver
Mink	Louisiana	40 - 320 ng/g	liver
Otter	Oregon	34 - 994 ng/g	liver
Otter	Washington	25 - 442 ng/g	liver
Bald Eagles	Gr. Lakes	1740 ng/g	liver
Bluegill sunfish	Lake Calhoun (Minneapolis, MN)	181-373 ng/g	fillet
Bluegill sunfish	Lake Calhoun (Minneapolis, MN)	280-590 ng/g	whole animal
White sucker	Lake Calhoun (Minneapolis)	<3.26-49.1 ng/g	fillet

**TABLE A: Freshwater Ecosystems (continued)**

<b>Animal</b>	<b>Location</b>	<b>PFOS Conc.</b>	<b>Tissue</b>
White sucker	Lake Calhoun (Minneapolis)	1.96-77 ng/g	whole animal
Bluegill sunfish	St. Croix River (MN/WI)	<3.37-<4.21	fillet
Bluegill sunfish	St. Croix River (MN/WI)	<3.38-4.22	whole animal
Smallmouth bass	St. Croix River (MN/WI)	<3.41-<3.52 ng/g	fillet
Smallmouth bass	St. Croix River (MN/WI)	<3.32-1.52 ng/g	whole animal
Walleye	St. Croix River (MN/WI)	<3.29-<3.57 ng/g	fillet
Northern pike	St. Croix River (MN/WI)	<3.17-<3.48 ng/g	fillet
White sucker	St. Croix River (MN/WI)	<3.21-<3.33 ng/g	fillet
Bluegill sunfish	Mississippi River Pool 5a	34-99.2 ng/g	fillet
Smallmouth bass	Mississippi River Pool 5a	45-116 ng/g	fillet
Walleye	Mississippi River Pool 5a	41-103 ng/g	fillet
Channel catfish	Mississippi River Pool 5a	<3.26-18.3	fillet
Bluegill sunfish	Mississippi River Pool 3	87.5-440 ng/g	fillet
Bluegill sunfish	Mississippi River Pool 3	186-815 ng/g	whole animal
White bass	Mississippi River Pool 3	86.7-154 ng/g	fillet
White bass	Mississippi River Pool 3	114-161 ng/g	whole animal
Emerald shiner	Mississippi River Pool 3	84.2 ng/g	whole animal (composite of 35 fish)
Gizzard shad	Mississippi River Pool 3	17.9 ng/g	whole animal (composite of 35 fish)

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**TABLE A: Freshwater Ecosystems (continued)**

<b>Animal</b>	<b>Location</b>	<b>PFOS Conc.</b>	<b>Tissue</b>
Bluegill sunfish	Mississippi River Pool 4	28.1-152 ng/g	fillet
Bluegill sunfish	Mississippi River Pool 5	40.3-94.7 ng/g	fillet
Smallmouth bass	Mississippi River Pool 5	47.7-150 ng/g	fillet
Largemouth bass	Mississippi River Pool 5	74.3-107 ng/g	fillet
Walleye	Mississippi River Pool 5	26.5-93.2 ng/g	fillet
Northern pike	Mississippi River Pool 5	12.2-235 ng/g	fillet
Channel catfish	Mississippi River Pool 5	<3.32-9.59 ng/g	fillet
Gizzard shad	Mississippi River Pool 5	20.1 ng/g	whole animal (composite of 40 fish)

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**TABLE B: Marine Ecosystems**

<b>Animal</b>	<b>Location</b>	<b>PFOS Conc.</b>	<b>Tissue</b>
Zooplankton	E. Arctic	1.8 ng/g	whole animal
Clam	E. Arctic	0.28 ng/g	whole animal
Mussels/Oysters	S. China/Japan	0.11 - 0.59 ng/g	whole animal
Starfish	North Sea	9 - 176 ng/g	whole animal
Shrimp	North Sea	19 - 520 ng/g	whole animal
Shrimp	E. Arctic	0.35 ng/g	whole animal
Crabs	North Sea	24 - 877 ng/g	whole animal
Bib	Belgium	45 ng/g	liver
Plaice	Belgium	900 ng/g	liver
Short-horn Sculpin	Greenland	13 - 18 ng/g	liver
Cod	E. Arctic	1.3 ng/g	liver
Dolphin	S. Carolina Coast	1315 ng/g	liver
Dolphin	Sarasota (Fla.) Bay	781 ng/g	liver
Dolphin	Delaware Bay	751 ng/g	liver
Dolphin	Bermuda	49 ng/g	liver
Dolphin	Coastal U.S.	1520 ng/g	liver
Dolphin	Mediterranean Sea	<1.4 - 110 ng/g	liver
Dolphin	W. Europe	14 - 443 ng/g	liver
Dolphin	W. Europe	<10 - 52 ng/g	liver
Tuna	Mediterranean Sea	21 - 87 ng/g	liver
Swordfish	Mediterranean Sea	<1 - 13 ng/g	liver
Whale	Faroe Islands	28 - 65 ng/g	liver
Whale	W. Europe	<10 - 52 ng/g	liver
Whale	Coastal U.S.	14.8 ng/g	liver
Whale	E. Arctic	12.6 ng/g	liver
Narwhal	E. Arctic	10.9 ng/g	liver
Seal	Greenland	10 - 67 ng/g	liver
Seal	Dutch Wadden Sea	175 ng/g	liver
Seal	E. Greenland	25.5 - 95.6 ng/g	liver
Seal	W. Greenland	12.5 - 27.9 ng/g	liver
Seal	Alaska	<10 - 122 ng/g	liver
Seal	Baltic Sea	190 - 490 ng/g	liver
Seal	W. Europe	<10 - 532 ng/g	liver
Walrus	E. Arctic	2.4 ng/g	liver
Polar Bear	Greenland	1245 - 1325 ng/g	liver
Polar Bear	S. Hudson Bay	2730 ng/g (x)	liver
Polar Bear	Chukchi Sea	729 ng/g (x)	liver
Polar Bear	Northwest Territories	1320 ng/g (x)	liver
Polar Bear	E. Greenland	2470 ng/g (x)	liver
Polar Bear	Alaska	350 ng/g (x)	liver
Glaucous Gull	E. Arctic	20.2 ng/g	liver
Black-headed Gull	Korea	296 ng/g	liver
Kittiwake	E. Arctic	10 ng/g	liver
Fulmar	Faroe Islands	24 - 29 ng/g	liver
Cormorant	Japan	380 ng/g	liver
Cormorant	Mediterranean Sea	32 - 150 ng/g	liver
Pelican	S. America (Columbia)	36.7 ng/g	liver
White-tailed Eagle	Baltic Sea	<3.9 - 127 ng/g	liver



**TABLE C**  
**Lake Calhoun**  
**Aqueous and Fish Tissue Analytical Results - PFOS**  
**Tissue Concentrations in ng/g**  
**Surface Water Concentrations in ng/mL**

<b>Compound</b>				<b>PFOS</b>
<b>Sample</b>	<b>Age/sex (yrs/m-f-j)</b>	<b>Length (cm)</b>	<b>Medium</b>	
Bluegill 1	3/F	15.5	Tissue - Fillet	<b>373</b>
Bluegill 1	3/F	15.5	Tissue - Whole Body	<b>493</b>
Bluegill 2	2/F	13	Tissue - Fillet	<b>356</b>
Bluegill 2	2/F	13	Tissue - Whole Body	<b>438</b>
Bluegill 3	3/F	15	Tissue - Fillet	<b>181</b>
Bluegill 3	3/F	15	Tissue - Whole Body	<b>280</b>
Bluegill 4	3/F	16	Tissue - Fillet	<b>311</b>
Bluegill 4	3/F	16	Tissue - Whole Body	<b>590</b>
Bluegill 5	3/F	16	Tissue - Fillet	<b>373</b>
Bluegill 5	3/F	16	Tissue - Whole Body	<b>528</b>
White Sucker 1	2/M	29	Tissue - Fillet	<3.52
White Sucker 1	2/M	29	Tissue - Whole Body	<b>2.91</b>
White Sucker 1 (Duplicate)	2/M	29	Tissue - Whole Body	<b>1.96</b>
White Sucker 2	2/J	31	Tissue - Fillet	<3.40
White Sucker 2 (Duplicate)	2/J	31	Tissue - Fillet	<3.37
White Sucker 2	2/J	31	Tissue - Whole Body	<3.23
White Sucker 3	2/J	27	Tissue - Fillet	<3.54
White Sucker 3	2/J	27	Tissue - Whole Body	<3.24
White Sucker 4	3/J	35	Tissue - Fillet	<b>49.1</b>
White Sucker 4	3/J	35	Tissue - Whole Body	<b>77</b>
White Sucker 5	2/J	29	Tissue - Fillet	<3.25
White Sucker 5	2/J	29	Tissue - Whole Body	<3.40
Water 1				<b>0.10500</b>
Water 1 (Duplicate)				<b>0.11700</b>
Water 2				<b>0.11500</b>
Water 3				<b>0.10400</b>

F = female, M = male, J = juvenile



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**TABLE C**  
**St. Croix River**  
**Aqueous and Fish Tissue Analytical Results - PFOS**  
**Tissue Concentrations in ng/g**  
**Surface Water Concentrations in ng/mL**

<b>Compound</b>				<b>PFOS</b>
<b>Sample</b>	<b>Age/sex (yrs/m-f-j)</b>	<b>Length (cm)</b>	<b>Medium</b>	
Bluegill 1	3/F	16.5	Tissue - Fillet	<3.57
Bluegill 1 (Duplicate)	3/F	16.5	Tissue - Fillet	<3.48
Bluegill 1	3/F	16.5	Tissue - Whole Body	<3.38
Bluegill 2	2/M	13	Tissue - Fillet	<4.21
Bluegill 2	2/M	13	Tissue - Whole Body	<b>4.22</b>
Bluegill 3	2/F	14.5	Tissue - Fillet	<3.38
Bluegill 3	2/F	14.5	Tissue - Whole Body	<b>3.4</b>
Bluegill 4	2/M	14	Tissue - Fillet	<3.37
Bluegill 4	2/M	14	Tissue - Whole Body	<b>4.07</b>
Bluegill 5	2/J	15	Tissue - Fillet	<3.48
Bluegill 5	2/J	15	Tissue - Whole Body	<3.54
Smallmouth Bass 1	6/M	37	Tissue - Fillet	<3.41
Smallmouth Bass 1	6/M	37	Tissue - Whole Body	<b>1.52</b>
Smallmouth Bass 2	4/M	29	Tissue - Fillet	<3.50
Smallmouth Bass 2	4/M	29	Tissue - Whole Body	<b>1.16</b>
Smallmouth Bass 3	4/F	30	Tissue - Fillet	<3.41
Smallmouth Bass 3	4/F	30	Tissue - Whole Body	<3.32
Smallmouth Bass 4	4/F	27	Tissue - Fillet	<3.48
Smallmouth Bass 5	4/M	28	Tissue - Fillet	<3.52
Walleye 1	5/M	40	Tissue - Fillet	<3.48
Walleye 2	6/F	46	Tissue - Fillet	<3.57
Walleye 3	3/J	33	Tissue - Fillet	<3.37
Walleye 4	3/J	28	Tissue - Fillet	<3.40
Walleye 5	3/J	32	Tissue - Fillet	<3.29
Northern Pike 1	6/J	43	Tissue - Fillet	<3.33
Northern Pike 2	6/F	42	Tissue - Fillet	<3.17
Northern Pike 3	7?/M	48	Tissue - Fillet	<3.30
Northern Pike 3 (Duplicate)	7?/M	48	Tissue - Fillet	<3.27
Northern Pike 4	8/M	58	Tissue - Fillet	<3.27
Northern Pike 5	6/F	43	Tissue - Fillet	<3.48
White Sucker 1	3/J	31	Tissue - Fillet	<3.33
White Sucker 2	3/F	36	Tissue - Fillet	<3.21
White Sucker 3	3/F	33	Tissue - Fillet	<3.33
Water 1			Surface Water	<0.00560
Water 2			Surface Water	<0.00930
Water 3			Surface Water	<0.00557



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**TABLE C**  
**Mississippi River Pool 3**  
**Aqueous and Fish Tissue Analytical Results - PFOS**  
**Tissue Concentrations in ng/g**  
**Surface Water Concentrations in ng/mL**

<b>Compound</b>				<b>PFOS</b>
<b>Sample</b>	<b>Age/sex (yrs/m-f-j)</b>	<b>Length (cm)</b>	<b>Medium</b>	
Bluegill 1	3/M	17	Tissue - Fillet	<b>440</b>
Bluegill 1	3/M	17	Tissue - Whole Body	<b>815</b>
Bluegill 2	3/M	17	Tissue - Fillet	<b>108</b>
Bluegill 2	3/M	17	Tissue - Whole Body	<b>187</b>
Bluegill 3	3/M	18	Tissue - Fillet	<b>123</b>
Bluegill 3	3/M	18	Tissue - Whole Body	<b>186</b>
White Bass 1	1/J	12.5	Tissue - Fillet	<b>122</b>
White Bass 1	1/J	12.5	Tissue - Whole Body	<b>134</b>
White Bass 2	1/J	13	Tissue - Fillet	<b>154</b>
White Bass 2	1/J	13	Tissue - Whole Body	<b>161</b>
White Bass 3	1/J	13	Tissue - Fillet	<b>150</b>
White Bass 3	1/J	13	Tissue - Whole Body	<b>148</b>
White Bass 4	1/J	13	Tissue - Fillet	<b>148</b>
White Bass 4	1/J	13	Tissue - Whole Body	<b>153</b>
White Bass 5	1/J	14.5	Tissue - Fillet	<b>86.7</b>
White Bass 5	1/J	14.5	Tissue - Whole Body	<b>114</b>
Emerald Shiner		~4.5	Whole Body - Composite of 35 Fish	<b>84.2</b>
Gizzard Shad		~9	Whole Body - Composite of 35 Fish	<b>17.9</b>
Water 1			Surface Water	<b>0.01900</b>
Water 2			Surface Water	<b>0.02310</b>
Water 3			Surface Water	<b>0.03780</b>



**TABLE C**  
**Mississippi River Pool 4**  
**Aqueous and Fish Tissue Analytical Results - PFOS**  
**Tissue Concentrations in ng/g**

<b>Compound</b>				<b>PFOS</b>
<b>Sample (Lab Sample ID)</b>	<b>Age/sex (yrs/m-f-j)</b>	<b>Length (cm)</b>	<b>Medium</b>	
Bluegill 1	4/F	20	Tissue - Fillet	<b>98.3</b>
Bluegill 2	3/M	17.5	Tissue - Fillet	<b>28.1</b>
Bluegill 3	3/F	18	Tissue - Fillet	<b>45.5</b>
Bluegill 4	3/M	16	Tissue - Fillet	<b>152</b>
Bluegill 5	3/M	18	Tissue - Fillet	<b>101</b>

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**TABLE C**  
**Mississippi River Pool 5**  
**Aqueous and Fish Tissue Analytical Results - PFOS**  
**Tissue Concentrations in ng/g**

<b>Compound</b>				<b>PFOS</b>
<b>Sample (Lab Sample ID)</b>	<b>Age/sex (yrs/m-f-j)</b>	<b>Length (cm)</b>	<b>Medium</b>	
Bluegill 1	4/F	172	Tissue - Fillet	<b>40.3</b>
Bluegill 2	4/J	252	Tissue - Fillet	<b>94.7</b>
Bluegill 3	4/M	199	Tissue - Fillet	<b>42.7</b>
Bluegill 4	4/F	189	Tissue - Fillet	<b>69.6</b>
Bluegill 5	3/F	114	Tissue - Fillet	<b>77.2</b>
Smallmouth Bass 1	8/F	1512	Tissue - Fillet	<b>150</b>
Smallmouth Bass 2	2/J	131	Tissue - Fillet	<b>83.5</b>
Smallmouth Bass 3	4/M	449	Tissue - Fillet	<b>47.7</b>
Smallmouth Bass 4	3/F	262	Tissue - Fillet	<b>93.5</b>
Smallmouth Bass 5	5/M	565	Tissue - Fillet	<b>104</b>
Largemouth Bass 1	6/F	456	Tissue - Fillet	<b>82.9</b>
Largemouth Bass 2	7/M	1043	Tissue - Fillet	<b>74.3</b>
Largemouth Bass 3	6/F	689	Tissue - Fillet	<b>85.8</b>
Largemouth Bass 3 (Duplicate)	6/F	689	Tissue - Fillet	<b>96.5</b>
Largemouth Bass 4	4/M	455	Tissue - Fillet	<b>107</b>
Largemouth Bass 5	5/M	502	Tissue - Fillet	<b>74.6</b>
Walleye 1	7/M	47	Tissue - Fillet	<b>34.3</b>
Walleye 1 (Duplicate)	7/M	47	Tissue - Fillet	<b>26.5</b>
Walleye 2	3/J	31	Tissue - Fillet	<b>60.6</b>
Walleye 3	3/J	33	Tissue - Fillet	<b>93.2</b>
Walleye 4	5/M	43	Tissue - Fillet	<b>27.1</b>
Northern Pike 1	8/F	58	Tissue - Fillet	<b>91.2</b>
Northern Pike 2	8/F	64	Tissue - Fillet	<b>224</b>
Northern Pike 2 (Duplicate)	8/F	64	Tissue - Fillet	<b>235</b>
Northern Pike 3	2/J	28	Tissue - Fillet	<b>130</b>
Northern Pike 4	6/J	45	Tissue - Fillet	<b>12.2</b>
Northern Pike 5	6/F	42	Tissue - Fillet	<b>97.5</b>
Channel Catfish 1	?/J	31	Tissue - Fillet	<b>9.59</b>
Channel Catfish 2	5/M	52	Tissue - Fillet	<b>&lt;3.32</b>
Gizzard Shad		~13	Whole Body - Composite of 40 Fish	<b>20.1</b>



**TABLE C**  
**Mississippi River Pool 5a**  
**Aqueous and Fish Tissue Analytical Results - PFOS**  
**Tissue Concentrations in ng/g**

<b>Compound</b>				<b>PFOS</b>
<b>Sample (Lab Sample ID)</b>	<b>Age/sex (yrs/m-f-j)</b>	<b>Length (cm)</b>	<b>Medium</b>	
Bluegill 1	3/M	17	Tissue - Fillet	<b>34.6</b>
Bluegill 2	3/M	18	Tissue - Fillet	<b>99.2</b>
Bluegill 3	4/M	20	Tissue - Fillet	<b>82.9</b>
Bluegill 4	4/M	20	Tissue - Fillet	<b>34</b>
Bluegill 5	4/F	19	Tissue - Fillet	<b>55.5</b>
Smallmouth Bass 1	7/M	36	Tissue - Fillet	<b>52.3</b>
Smallmouth Bass 2	7/M	36	Tissue - Fillet	<b>116</b>
Smallmouth Bass 3	6/F	35	Tissue - Fillet	<b>67.1</b>
Smallmouth Bass 4	4/M	28	Tissue - Fillet	<b>84.6</b>
Smallmouth Bass 5	6/M	34	Tissue - Fillet	<b>45</b>
Walleye 1	5/F	41	Tissue - Fillet	<b>56.4</b>
Walleye 2	6/F	49	Tissue - Fillet	<b>49.3</b>
Walleye 3	6/F	47	Tissue - Fillet	<b>41</b>
Walleye 4	2/J	25.5	Tissue - Fillet	<b>75.4</b>
Walleye 5	9/F	55	Tissue - Fillet	<b>103</b>
Channel Catfish 1	6/M	57	Tissue - Fillet	<b>&lt;3.26</b>
Channel Catfish 2	4/F	46	Tissue - Fillet	<b>18.3</b>
Channel Catfish 3	3/F	41	Tissue - Fillet	<b>9.55</b>
Channel Catfish 4	2/F	39	Tissue - Fillet	<b>13.4</b>

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#### 4.0 DIRECT TOXICITY TESTING EVALUATION

In this section of the report, the toxicity studies on PFOS with a variety of aquatic organisms are evaluated. Appendix C to this document contains a summary of this information in tabular form, in the format utilized by the MPCA for water quality criteria development.

Table 1:	Summary of all useful and non-useful acute and chronic toxicity studies on aquatic animals
Table 2a:	Acceptable chronic toxicity data
Table 2b:	Acceptable acute-to-chronic ratio (ACR) data
Table 3a:	Acceptable acute toxicity data
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Table 4a:	Summary of all useful and non-useful toxicity studies on aquatic plants
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Table 5a:	All BCF and BAF information
Table 5b:	Useable BCF/BAF Data
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Table 6:	Wildlife acute and chronic toxicity data (not included)
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Table 8:	Behavioral effects data (not included)

#### 4.1 Acceptable Toxicity Bioassay Summaries

##### 4.1.1 Aquatic Plants

###### Lemna gibba

A published bioassay (STS-104) on the effects of PFOS (potassium salt) on *Lemna gibba* (duckweed) was reviewed by STS. In this study the acute toxicity of PFOS to *L. gibba* was observed over a 7-day exposure period at the University of Guelph in Ontario, Canada and was completed in 2002. The test followed ASTM E 1415-91. A control and five nominal concentrations of PFOS were tested: 10, 20, 40, 80 and 160 mg/L. The

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results of the test identified a 7-day EC50 (based on wet weight of 31.1 mg/L and a 7-day NOEC (based on wet weight) of 6.6 mg/L PFOS.

#### Myriophyllum spp.

One toxicity test on the effects of PFOS (potassium salt) on *Myriophyllum spp.* (milfoil) was reviewed by STS (STS-108). In this study toxicity of PFOS (potassium salt) to *Myriophyllum spicatum* and *M. sibiricum* was observed using 12,000 L outdoor microcosms at the University of Guelph Microcosm Facility located in Ontario, Canada. Endpoints of toxicity that were monitored on days 1, 14, 28 and 42 of the study included growth in length, biomass, root number, primary root length, number of nodes and pigment concentrations. A control and four nominal concentrations of PFOS were tested: 0.3, 3.0, 10.0 and 30.0 mg/L. *M. sibiricum* was found to be more sensitive to PFOS than *M. spicatum* in this study. The results identified a 42-day EC50 of 12 mg/L (lowest recorded EC50) and a 42-day NOEC of 11.4 mg/L for *M. spicatum*. We note that *M. spicatum* (Eurasian milfoil) is a non-native species, and that data cannot be used for calculation of the water quality standard for the State of Minnesota. The results identified a 42-day EC50 of 1.6 mg/L based on longest root (lowest recorded EC50) and a 42-day NOEC of 0.3 mg/L based on longest root, plant length, root length, chlorophyll a and carotenoids for *M. sibiricum*.

#### Navicula pelliculosa

A bioassay (STS-118) on the effects of PFOS (potassium salt) on *Navicula pelliculosa* (freshwater diatom) was reviewed by STS. In this GLP study the toxicity of PFOS to *N. pelliculosa* was observed under static conditions over a 96-hour exposure period at the Wildlife International, Ltd. Facility in Easton, Maryland in February, 2000. The test followed U.S. EPA OPPTS Number 850.5400 (1996). Seven measured concentrations of PFOS were tested from one control and seven nominal values: 61.5, 81.3, 110, 147, 198, 264 and 347 mg/L. The lowest recorded 96-hour EC50 value from the test was 252 mg/L based on area under the growth curve. The lowest recorded NOEC value obtained from the test was 62.3 mg/L based on area under the growth curve.



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#### Chlorella vulgaris

A bioassay (STS-104) on the effects of PFOS (potassium salt) on *Chlorella vulgaris* (freshwater green alga) was reviewed by STS. In this study the toxicity of PFOS to *C. vulgaris* was observed under static conditions over a 96-hour exposure period. The test followed ASTM (1999). A control and six nominal concentrations of PFOS were tested: 12.5, 25.0, 50.0, 100.0, 200.0 and 400.0 mg/L. The results of the test identified a 96-hour EC50 (based on cell density) of 81.6 mg/L and a 96-hour NOEC (based on cell density) of 8.2 mg/L PFOS.

#### Selenastrum capricornutum

A bioassay (AR226-0084/AR226-0085/AR226-1760) on the effects of PFOS (potassium salt) on *Selenastrum capricornutum* (freshwater alga) was reviewed by STS. In this GLP study the toxicity of PFOS to *S. capricornutum* was observed under static conditions over a 96-hour exposure period at the Wildlife International, Ltd. Facility in Easton, Maryland in April, 1999. The test followed OECD Guideline 201 (1984) and U.S. OPPTS Number 850.5400 (1996). Measured concentrations of PFOS were tested from one control and six nominal values: 5.7, 11.0, 23.0, 46.0, 91.0 and 183.0 mg/L. The results of the test identified a 96-hour EC50 (based on cell density and area under the growth curve) of 68 mg/L and a 96-hour NOEC (based on cell density, area under the growth curve and growth rate) of 42 mg/L PFOS.

A bioassay (STS-104) on the effects of PFOS (potassium salt) on *Selenastrum capricornutum* (freshwater alga) was reviewed by STS. In this study the acute toxicity of PFOS to *S. capricornutum* was observed under static conditions over a 96-hour exposure period. The results of the test identified a 96-hour EC50 (based on cell density) of 48.2 mg/L and a 96-hour NOEC (based on cell density) of 5.3 mg/L PFOS.

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#### 4.1.2 Aquatic Invertebrates

##### Hyalella azteca

A bioassay (STS-332) on the effects of PFOS (potassium salt) on *Hyalella azteca* (freshwater amphipod) was reviewed by STS. In this GLP study the toxicity of PFOS to *H. azteca* was observed under static renewal conditions over a 96-hour exposure period in 2007 by Wildlife International, Ltd. in Easton, Maryland. A control and five measured concentrations of PFOS were tested: 13, 25, 50, 100 and 200 mg/L. The results of the test identified a 96-hour LC50 of 15 mg/L PFOS and a NOEC of <13 mg/L PFOS.

##### Daphnia magna

A bioassay (AR226-0112) on the effects of PFOS (lithium salt) on *Daphnia magna* (water flea) was reviewed by STS. The test method was not noted. In this study the acute toxicity of PFOS to *D. magna* was observed under static conditions over a 48-hour exposure period in February, 1994. *D. magna* used for this test were less than 24-hours old at test initiation. A control plus five nominal concentrations of PFOS were tested: 100, 180, 320, 560 and 1,000 mg/L. The results of the test identified a 48-hour EC50 of 210 mg/L PFOS and a 48-hour NOEC of 100 mg/L PFOS. Based on a sample content of 24.5% PFOS, these data translate to an EC50 of 51.4 mg/L and a NOEC of 24.5 mg/L PFOS.

A bioassay (AR226-0086/AR226-0087/AR226-1761) on the effects of PFOS (potassium salt) on *Daphnia magna* (water flea) was reviewed by STS. In this GLP study the toxicity of PFOS to *D. magna* was observed under static conditions over a 48-hour exposure period at the Wildlife International, Ltd. facility located in Easton, Maryland in February, 1999. The test followed OECD 202 (1994) and U.S. EPA OPPTS Number 850.1010 (1996). *D. magna* used for this test were less than 24-hours old at test initiation. Five measured concentrations of PFOS and a control were tested. The results of the test identified a 48-hour EC50 of 59 mg/L PFOS and a NOEC of 32 mg/L.

A bioassay (AR226-0098/AR226-0099/AR226-1766) on the effects of PFOS (potassium salt) on *Daphnia magna* (water flea) was reviewed by STS. In this GLP study the life-cycle toxicity of PFOS to *D. magna* was observed under semi-static conditions over a 21-

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day exposure period at the Wildlife International, Ltd. facility located in Easton, Maryland in February, 2000. The test followed OECD 211 (1997) and U.S. EPA OPPTS Number 850.1300 (1996). *D. magna* used for this test were less than 24-hours old at test initiation. Six measured concentrations of PFOS and a control were tested. The results of the test identified a 21-day NOEC of 12 mg/L. A second generation acute survival NOEC was also calculated at 12 mg/L PFOS. A concentration of 24.5 mg/L PFOS led to 100% mortality.

A bioassay (STS-104) on the effects of PFOS (potassium salt) on *Daphnia magna* and *Daphnia pulicaria* (water flea) was reviewed by STS. In this study the acute and chronic toxicity of PFOS to *D. magna* was observed under acute static and chronic renewal conditions. In addition, the acute toxicity of PFOS to *D. pulicaria* was observed under acute static conditions. The acute toxicity testing was observed over a 48-hour period. The chronic toxicity was observed over a 21-day exposure period. The testing followed ASTM (1999). *Daphnia spp.* used for this test were less than 24-hours old at test initiation. A control plus five nominal concentrations were tested for the acute studies: 31, 63, 125, 250 and 450 mg/L. A control plus five nominal concentrations were tested for the chronic study: 6, 13, 25, 50 and 100 mg/L. The results of the acute testing identified a 48-hour EC50 of 67.2 mg/L and a 48-hour NOEC of 0.8 mg/L PFOS for *D. magna*, and a 48-hour EC50 of 134 mg/L and a 48-hour NOEC of 13.6 mg/L PFOS for *D. pulicaria*. The results of the chronic test identified a 21-day NOEC (lethality) of 5.3 mg/L and a 21-day LC50 of 42.3 mg/L PFOS for *D. magna*. The authors concluded that comparative chronic studies on the effects of PFOS to *D. magna* would be beneficial due to the apparent increased toxicity in the chronic study.

#### Chironomus tentans

A bioassay (STS-110) on the effects of PFOS (potassium salt) on *Chironomus tentans* (midge) was reviewed by STS. In this study the acute toxicity of PFOS to *C. tentans* was observed under static renewal conditions for a 10-day exposure period. In addition, the chronic toxicity of PFOS to *C. tentans* was observed under static renewal conditions for a 20-day exposure period. A control and seven nominal concentrations were used for the

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acute testing; 0.001, 0.005, 0.010, 0.020, 0.040, 0.080 and 0.150 mg/L. A control and five nominal concentrations were used for the chronic testing; 0.001, 0.005, 0.010, 0.050 and 0.100 mg/L. The results of the acute test identified a 10-day EC 50 (growth) of 0.0872 mg/L and a 10-day NOEC of 0.0491 mg/L. The maximum animal mortality was 30% in this test. The results of the chronic test identified a 20-day EC 50 (growth) of 0.0938 mg/L and a 20-day NOEC (growth) of 0.0217 mg/L. The MATC (growth) was determined to be 0.04537 mg/L. A 10-day acute EC 50 value was calculated based on an extrapolation, see Section 5.1.

The acute survival assay indicated the EC50 was greater than 0.150 mg/l . STS performed a least squared analysis to extrapolate the EC50 value. The least squared analysis found the project 10 survival EC50 for *Chironomus tentans* is 0.170 mg/l.

#### Lumbriculus variegates

A bioassay (STS-334) on the effects of PFOS (potassium salt) on *L. variegatus* (oligochaete worm) was reviewed by STS. In this GLP study the toxicity of PFOS to *L. variegates* was observed under static renewal conditions over a 96-hour exposure period in 2007 by Wildlife International, Ltd. in Easton, Maryland. A control and five measured concentrations of PFOS were tested: 0.71, 1.4, 2.8, 5.6 and 11 mg/L. The results of the test identified a 96-hour LC50 of 5.6 mg/L PFOS and a NOEC of 2.8 mg/L PFOS.

### **4.1.3 Fishes**

#### Oncorhynchus mykiss

A bioassay (STS-321) on the effects of PFOS (potassium salt) on *Oncorhynchus mykiss* (rainbow trout) was reviewed by STS. In this GLP study the toxicity of PFOS to juvenile *O. mykiss* was observed under static conditions over a 96-hour exposure period at the Wildlife International, Ltd. Facility in Easton, Maryland in June, 2001. This study followed OECD Guideline 203 (1993) and U.S. EPA OPPTS Number 850.1075 (1996). A control and six measured concentrations of PFOS were tested: 3.0, 6.3, 13.0, 25.0, 50.0 and 52.0 mg/L. The results of the test identified an EC50 (based on signs of toxicity or abnormal behavior) of 22 mg/L and a NOEC of 6.3 mg/L.

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#### Pimephales promelas

A bioassay (AR226-0082/AR226-0083/AR226-1759) on the effects of PFOS (potassium salt) on *Pimephales promelas* (fathead minnow) was reviewed by STS. In this GLP study the toxicity of PFOS to *P. promelas* was observed under static conditions over a 96-hour exposure period at the Wildlife International, Ltd. Facility in Easton, Maryland in February, 1999. This study followed OECD Guideline 203 (1993) and U.S. EPA OPPTS 850.1075 (1996). *P. promelas* used for this study were juveniles at test initiation. Five measured concentrations of PFOS and a control sample were tested: 3.3, 5.6, 9.5, 17.0 and 28.0 mg/L PFOS. The results of the test identified a 96-hour LC50 of 9.1 mg/L and a NOEC of 3.2 mg/L PFOS for *P. promelas*.

A bioassay (AR226-0096/AR226-0097/AR226-1765) on the effects of PFOS (potassium salt) on *Pimephales promelas* (fathead minnow) was reviewed by STS. In this GLP study the chronic toxicity of PFOS to *P. promelas* was observed under flow-through conditions over a 47-day exposure period at the Wildlife International, Ltd. Facility in Easton, Maryland in August-September, 1999. This study followed OECD Guideline 210 (1992) and U.S. EPA OPPTS 850.1400 (1996). *P. promelas* used for this study were less than 24-hours old at test initiation. Six measured concentrations of PFOS and a control sample were tested: 0.15, 0.30, 0.60, 1.2, 2.4 and 4.6 mg/L PFOS. The results of the test identified a 42-day NOEC of 0.29 mg/L, a LOEC of 0.58 mg/l and a MATC of 0.41 mg/l PFOS.

#### **4.1.4 Crustaceans**

##### Unio complamatus

A bioassay (AR226-0090/AR226-0091/AR226-1762) on the effects of PFOS (potassium salt) on *Unio complamatus* (freshwater mussel) was reviewed by STS. In this GLP study the toxicity of PFOS to *U. complamatus* was observed under static conditions over a 96-hour exposure period at the Wildlife International, Ltd. facility located in Easton, Maryland completed in August, 1999. The test followed OECD 203 (1993) and U.S. EPA OPPTS Number 850.1075 (1996). A control and five measured concentrations of PFOS and a control were tested: 5.1, 12.0, 19.0, 39.0 and 76.0 mg/L PFOS. The results of the test

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identified a 96-hour LC50 of 57 mg/L and a 96-hour No Mortality Concentration of 19 mg/L PFOS.

#### 4.1.5 Amphibians

A bioassay (STS-333) on the effects of PFOS (potassium salt) on *Pseudacris crucifer* (spring peeper tadpole) was reviewed by STS. In this GLP study the toxicity of PFOS to *P. crucifer* was observed under static renewal conditions over a 96-hour exposure period in 2007 by Wildlife International, Ltd. in Easton, Maryland. A control and five measured concentrations of PFOS were tested: 3.6, 7.0, 14, 27 and 51 mg/L. The results of the test identified a 96-hour LC50 of 38 mg/L PFOS and a NOEC of 3.6 mg/L PFOS.

### 4.2 Unacceptable Toxicity Bioassay Summaries

#### 4.2.1 Aquatic Plants

##### Selenastrum capricornutum (freshwater green alga)

STS-319 – GLP was not followed. Some water quality parameters were slightly out of range (e.g. pH). The toxicity results for this study may be due to a mixture of compounds. The reported 96-hour toxicity value (EC50) was 71 mg/L PFOS (based on growth rate). The reported NOEC was 35 mg/L (based on growth rate). The adjusted EC50 was 71 mg/L and the adjusted NOEC was 35 mg/L.

AR226-0536 – GLP was not followed. The toxicity results for this study may be due to a mixture of compounds. In addition, the cation salt may be toxic. The reported toxicity value (EC50) was <51.8 mg/L PFOS (based on cell density). The reported NOEL was 7.7 mg/L (based on cell density).

AR226-0106 - The pH was out of range relative to the referenced standard. The results of the 96-hour definitive test identified an EC50 (based on cell count) of 82 mg/L PFOS. The results of the 7-day definitive test identified an EC50 (based on cell count) of 99 mg/L PFOS. The results of the 10-day definitive test

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identified an EC50 (based on cell count) of 98 mg/L PFOS. The results of the 14-day definitive test identified an EC50 (based on cell count) of 95 mg/L PFOS.

#### 4.2.2 Aquatic Invertebrates

##### Daphnia magna

AR226-0123 – This study had questionable methodology and lacked detail. In addition, the sample purity was unknown. The reported toxicity values (LC50) were 49.2 and 50 mg/L PFOS.

STS-101 – This was a non-GLP study. The available test summary for STS-101 was abbreviated because the test was superseded by a more recent test. The reported toxicity values (EC50) were 58 and 67 mg/L PFOS.

STS-201The neonates used at the beginning of the test were too old in age. The results of the test identified a 21-day NOEC of 6 mg/L and a 21-day LOEC of 13 mg/L PFOS for *D. pulicaria*. The results of the test identified a 21-day NOEC of 25 mg/L and a 21-day LOEC of 50 mg/L PFOS for *D. magna*.

STS-322 – The toxicity data obtained from this study is from a mixture and accurate toxicity could not be determined. The reported toxicity value (EC50) was 14 mg/L PFOS (estimated based on no mixture).

STS-323 – The concentration of PFOS could not be determined from the information presented in the study. The reported toxicity values (LC50) were 76 and 73 mg/L PFOS.

STS-324 – GLP was not followed. No information regarding sample purity (liquid) was presented in this study. In addition, the cation salt may be toxic. The reported toxicity value (EC50) was 11.3 mg/L PFOS. The reported NOEL was 6.25 mg/L PFOS.

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AR226-0110 - Not an acceptable acute or chronic test. The test temperatures were out of line with accepted methods. The results of the test identified a 48-hour EC50 of 27 mg/L, a 14-day EC50 (reproduction) of 14.7 mg/L, a 21-day EC50 (reproduction) of 12.4 mg/L and a 28-day EC50 (reproduction) of 11.4 mg/L PFOS. The 28-day NOEC was identified at a concentration of 7 mg/L PFOS. The 14, 21 and 28-day MATC was identified at a concentration of 11.2 mg/L PFOS. We note that no statement was made regarding the purity of the powder (PFOS) in test AR226-0110.

#### 4.2.3 Fishes

##### Oncorhynchus mykiss (Rainbow Trout)

STS-320 – No GLP was followed. Mixture concerns were identified in this study. In addition, few water quality parameters were noted. The reported 4-day toxicity value (LC50) was 2500 mg/L PFOS.

AR226-0123 – This study used questionable methodology and lacked detail. The reported 4-day toxicity value (LC50) was 11 mg/L PFOS.

STS-112 - No standard test method was referenced and the methodology lacked detail.



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Pimephales promelas (Fathead Minnow)

STS-112 – No standard test method was referenced. Some water quality parameters (e.g. temperature) were out of range relative to most recent EPA Guidelines. The reported 28-day toxicity value (LC50) was 7.2 mg/L PFOS.

AR226-0123 – Summary report of old data. The reported 4-day toxicity values (LC50) were 37.6 and 51.0 mg/L PFOS.

AR226-0534 – No GLP was performed. The dilution water source was questionable. Some water quality parameters were out of range referenced to most current EPA test method. Only four test concentrations (including control) were used. In addition, the data obtained for this study is for a mixture. The cation salt used may be toxic in addition to PFOS. The reported 4-day toxicity value (LC50) was 562 mg/L PFOS. The reported NOEL was >490 mg/L PFOS.

AR226-0107/AR226-0108 – GLP was not followed. Questionable methodology was used for this study which has been superseded by a more recent test. The reported 30-day NOEC was 1 mg/L PFOS. The reported LOEC was 1.9 mg/L PFOS.

AR226-0358 – GLP was not followed. Some water quality parameters (e.g. hardness) were out of range in reference to most recent EPA method. In addition, not enough detail on methodology was provided. The reported 30-day MTC was >20 mg/L PFOS.

STS-326 – This study was an adaptation to an unknown method. The reported 21-day toxicity value (EC50) was 0.23 mg/L PFOS.

AR226-0111 - The temperature and pH were out of range relative to current methodology. The results of the test identified a 96-hour LC50 of 4.7 mg/L PFOS (adjusted from 19 mg/L based on a test substance concentration of 24.5% in water).

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Lepomis macrochirus (Bluegill sunfish)

AR226-1755/AR226-1756 – This study lacks documentation. The species loading exceeded the most recent EPA method. In addition, some water quality parameters (e.g. temperature) were out of range in reference to most recent EPA method. The reported 4-day toxicity value (LC50) was 68 mg/L PFOS. The reported NOEC was <56 mg/L PFOS.

AR226-0115 – The DEA salt of PFOS may be toxic. In addition, only one replicate was performed for this study. The reported 4-day toxicity value (LC50) was 31 mg/L PFOS. The reported NOEC was 18 mg/L PFOS.

AR226-0123 – Summary information only. The reported 4-day toxicity value (LC50) was 68 mg/L PFOS.

Cyprinus carpio (Common carp)

STS-115 – Non-standard toxicity test

STS-116 – Non-standard toxicity test

**4.2.4 Amphibians (Frog)**

Rana pipiens

STS-114 – Elevated mortality of control animals was identified during the study after six weeks. The reported EC50 values during tadpole stage were >12.5 mg/L after week 1, 11 mg/L after week 2, 7.71 mg/L after week 3, 6.59 mg/L after week 4 and 6.21 mg/L after week 5.

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### 4.3 Microcosm Toxicity Testing

A total of three 35 day microcosm studies of exposure of zooplankton to PFOS were reviewed by STS (STS-202, STS-107 and STS-201). Community structures in the three studies over the 35 day study periods shifted from larger zooplankton dominated communities to communities dominated by smaller zooplankton species (presumably due to reduced competition). It was noted that the toxicity results in these microcosm tests were less than those in referenced laboratory toxicity bioassays, highlighting the possibility that PFOS is accumulated from food more than concentrated from water. Summaries of the microcosm tests are presented below:

#### STS-202

Toxicity of PFOS (potassium salt) to zooplankton communities was observed over a 35 day exposure period using 30 L outdoor aquaria in a static conditions test. Zooplankton microcosm communities for this study consisted of *Cyclops canthocamptus staphylinus*, *C. strenuous*, *C. diaptomus*, *Daphnia magna*, *Keratella quadrata*, *Phyllopora* sp., *Echninorhynchus* sp., *Ostracoda* sp., and total *Rotifera* sp. Other macrophytes and invertebrates were also present. A 90 – 100% reduction of total zooplankton population was found after one week of exposure to 30 mg/L and similar reduction after two weeks at 10 mg/L. No NOEC (community) could be calculated from this study. The study identified an overall community shift from a larger zooplankton dominated community to a community dominated by smaller and more robust species. In general, species sensitivity to PFOS was as follows: Copepoda>Cladocera>Rotifera. Although not specifically analyzed as a toxicity endpoint, it was noted the *Ephemeroptera* sp. (mayflies) were completely absent at 10+ mg/L PFOS and significantly decreased at 1 mg/L PFOS.

#### STS-107

Toxicity of PFOS (potassium salt) to zooplankton communities was observed using 12,000 L outdoor aquaria using a 35 day static test period at the Centre for Toxicology Microcosm Research Facility in Ontario, Canada. Natural zooplankton microcosm communities consisting of 92 species of Rotifera, Cladocera, Copepoda, macroinvertebrates and Ostracoda were obtained from a spring-fed irrigation pond and

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acclimated to the aquaria prior to dosing. The aquaria also contained periphyton, phytoplankton, algae and additional plant species. PFOS concentrations in this study were 0, 0.3, 3, 10 and 30 mg/L. The zooplankton community was evaluated on days -1, 0, 1, 2, 4, 7, 14, 21, 28 and 35.

The study identified an overall community shift from a larger zooplankton dominated community to a community dominated by Rotifers. Cladocera and Copepoda were the most sensitive taxonomic groups after 7 days. The 35 day NOEC (community) concentration was 3 mg/L. In addition, a separate 42 day static test was conducted using the floating macrophyte *Lemna gibba* (duckweed). The 42 day inhibition concentration (IC50) for *Lemna gibba* frond number (the most sensitive toxicity endpoint observed) was 19.1 mg/L; the NOEC was 0.2 mg/L.

#### STS-105

Toxicity of PFOS (potassium salt) to zooplankton communities was observed using both indoor and outdoor static aquaria tests. Results were compared to a 21-day static chronic laboratory bioassay performed on *D. magna* and *D. pulicaria*. Zooplankton microcosm communities for this study consisted of *Cyclops canthocamptus staphylinus*, *C. strenuous*, *C. diaptomus*, *D. magna*, *Keratella quadrata*, *Phyllopora* sp., *Echninorhynchus* sp., *Ostracoda* sp., and total *Rotifera* sp. Other macrophytes and invertebrates were also present. The microcosm study identified an overall community shift from a larger zooplankton dominated community to a community dominated by smaller and more robust species, especially Rotifers.

The 21-day static chronic NOEC and LOEC values for PFOS were 6 mg/L and 13 mg/L, respectively for *D. pulicaria*. The 21-day static chronic NOEC and LOEC values for PFOS were 25 mg/L and 50 mg/L, respectively for *D. magna*.

The 35-day indoor microcosm LOEC (community) value for PFOS was 1 - 10 mg/L. The 35-day outdoor microcosm LOEC (community) value for PFOS was 10 - 30 mg/L. The order of sensitivity rank for the 35-day microcosm testing was Cladocera>Copepoda>Rotifera. The LOEC value for *D. magna* was lower in the microcosm test than in the

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reference laboratory chronic bioassay; the results were not apparently due to food scarcity due to phytotoxicity.

#### 4.4 Microtox Toxicity Testing

Two Microtox tests (AR226-0113 and AR226-0538) on the effect of PFOS on *Photobacterium phosphoreum* were reviewed by STS. The Microtox procedure is a screening test that measures the light output of the bacteria over a 5, 15 and 30 minute exposure period while exposed to an agent. The percent light loss after the exposure period is the recorded endpoint. The Microtox procedure is not an EPA approved procedure for incorporation into aquatic life standards at this time. *P. phosphoreum* are saltwater bacteria. The toxicity data obtained from these tests were not used by STS in the calculation of water quality standards for PFOS. The tests are discussed in detail below.

##### AR226-0113

A Microtox test (AR226-0113) of exposure of PFOS (lithium salt) to *Photobacterium phosphoreum* was reviewed by STS. For this study completed in February, 1994 a control and four nominal concentrations of PFOS were tested: 125, 250, 500 and 1,000 mg/L. The exposure of PFOS (lithium salt) to *P. phosphoreum* resulted in a 30-minute EC50 value of >1000 mg/L. We note that no calculation was made to adjust for the actual concentration of the test substance in the test sample.

##### AR226-0538

A Microtox test (AR226-0538) of exposure of PFOS (didecyldimethylammonium salt) to *Photobacterium phosphoreum* was reviewed by STS. For this study completed in May, 1997 a control and four nominal concentrations of PFOS were tested: 125, 250, 500 and 1,000 mg/L. The exposure of PFOS (didecyldimethylammonium salt) to *P. phosphoreum* resulted in a 30-minute EC50 value of 350 mg/L. We note that no calculation was made to adjust for the actual concentration of the test substance in the test sample. In addition, we note that the cation salt may contribute to the reported toxicity results presented in this study.

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## 4.5 Other Unused Toxicity Testing

### 4.5.1 Saltwater Species

Toxicity tests of PFOS on the following saltwater organisms were obtained by STS. In addition we note that the Microtox testing of PFOS on *Photobacterium phosphoreum* (saltwater bacteria) is discussed in Section 4.4. The values obtained from the saltwater tests were not used in the calculations for water quality standards for PFOS.

#### Mysidopsis bahia

One acute toxicity test (AR226-0094, AR226-0095 and AR226-1758) of the effects of PFOS (unknown salt) on *Mysidopsis bahia* was reviewed by STS. The reported toxicity value (LC50) was 3.5 mg/L PFOS. The reported NOEC was 1.1 mg/L PFOS. In addition, one chronic test of the effects of PFOS (potassium salt) on *M. bahia* (AR226-0100 and AR226-0101) was reviewed by STS. The reported NOECs were 0.25 mg/L PFOS. The reported LOECs were 0.55 mg/L PFOS. Generally, this test appeared to be acceptable in terms of methodology.

#### Skeletonema costatum

One acute toxicity test (STS-117) on the effects of PFOS (potassium salt) on *Skeletonema costatum* (marine diatom) was reviewed by STS. The reported 4-day toxicity value (EC50) was >3.20 mg/L PFOS. The reported NOEC was 3.20 mg/L PFOS.

#### Crassostrea virginica

One acute toxicity test (AR226-0088, AR226-0089 and AR226-1763) on the effects of PFOS (potassium salt) on *Crassostrea virginica* (eastern oyster) was reviewed by STS. The reported 4-day toxicity value (EC50) was >2.9 mg/L PFOS. The reported NOEC and LOEC were 1.8 and 2.9 mg/L PFOS, respectively. Generally, this test appeared to be acceptable in terms of methodology.

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#### Palaemonetes vulgaris

One acute toxicity test (STS-325) on the effects of PFOS (potassium salt) on the grass shrimp *Palaemonetes vulgaris* was reviewed by STS. The reported 4-day toxicity value (TL50) was 280 mg/L PFOS. The reported NOEC was 180 mg/L PFOS.

#### Artemia

One acute toxicity test (STS-109) on the effects of PFOS (potassium salt) on brine shrimp *Artemia* was reviewed by STS. The reported 2-day toxicity values (EC50) ranged from 8.9 to 9.4 mg/L PFOS.

#### Fundulus heteroclitus

One acute toxicity test (STS-325) on the effects of PFOS (unknown salt) on *F. heteroclitus* (mummichog) was reviewed by STS. The reported 96-hour toxicity value (TL50) was 1820 mg/L PFOS. The NOEC was 1400 mg/L PFOS.

#### Uca pugnator

One acute toxicity test (STS-325) on the effects of PFOS (unknown salt) on *U. pugnator* (Fiddler crab) was reviewed by STS. The reported 96-hour toxicity value (TL50) was 3260 mg/L PFOS. The NOEC was 2400 mg/L PFOS.

#### Cyprinodon variegatus

One acute toxicity test (STS-327) on the effects of PFOS (potassium salt) on *Cyprinodon variegatus* (sheepshead minnow) was reviewed by STS. The reported 4-day toxicity value (LC50) was >15 mg/L PFOS. The reported NOEC was >15 mg/L PFOS.

### **4.5.2 Non-Native Species**

#### Xenopus laevis

One toxicity test (STS-113) on the effects of PFOS on *Xenopus laevis* (South African clawed frog) was reviewed by STS. The reported 4-day LC50s ranged from 13.8 to 17.6 mg/L PFOS. The reported 4-day MCIGs were 7.97 and 8.26 mg/L PFOS. The reported EC50s ranged from 12.1 to 17.6 mg/L PFOS. STS-113 was generally observed as an

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acceptable test in terms of methodology and data quality. STS-113 was not used because the South African clawed frog is not native to Minnesota or North America.

Myriophyllum spicatum

One toxicity test (STS-108) on the effects of PFOS on *Myriophyllum spp.* (milfoil) was reviewed by STS and was discussed previously (see Section 4.1.1). STS-108 was generally observed as an acceptable test in terms of methodology and data quality. The toxicity data for *M. spicatum* (Eurasian milfoil) was not used because it is not native to Minnesota or North America.



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## 5.0 SURFACE WATER QUALITY CRITERIA DEVELOPMENT

As described in the Introduction to this document (Section 1.0), a variety of water quality criteria are developed for a chemical to be protective of both aquatic life and human health.

### 5.1 Aquatic Life Criteria

The first step in developing toxicity-based aquatic life criteria is to determine if there are sufficient data available from which to calculate the criteria using the U.S. EPA national method -- Tier 1. According to this method, a minimum of eight acute toxicity studies representing each of the following groups must be available:

- salmonid
- osteichthyes
- chordate (fish, amphibian)
- phylum other than Arthropoda or Chordata
- planktonic crustacean
- benthic crustacean
- aquatic insect
- second insect or a phylum not already represented

In addition, there is a requirement for a minimum of three chronic toxicity studies for the chemical (for species which acute toxicity data exist), from which the Acute Chronic Ratio (ACR) is to be developed.

Table 2a (Appendix C) provides the acceptable chronic toxicity data for PFOS on various aquatic species. As can be seen, the above minimum requirement of three species studies has not been met.

Table 3a (Appendix C) provides the acceptable acute toxicity data for PFOS on various aquatic species. As can be seen, the above minimum requirement of eight species studies has not been met; there are no acceptable data for an aquatic insect. Thus, it is concluded that there are insufficient toxicological data on PFOS from which to develop Tier 1 direct toxicity criteria. Therefore, for PFOS the Tier 2 method, as specified in Minnesota Rules Chapter 7050 Subp. 5 G, will be used to calculate the criteria.

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According to the Tier 2 method, a minimum of two acute values in the following groups must be available:

- Osteichthyes (fish)
- Daphnidae

As shown in Table 3a, this criterion is met for PFOS. Acceptable test data for seven aquatic organisms are available. Table 3b ranks the GMAVs for these organisms, as well as presents the final FAV, as calculated from these data. The Tier 2 final FAV of 1.302mg/L, however, was amended based on an exposure study with the midge (STS-110). In this study a maximum PFOS concentration of 150 mg/L produced an acute toxicity (lethality) in 30% of the test animals. STS performed acute curved fitting extrapolation of the published data to determine an acute LC50 of 170 ug/L. According to the MN 7050 rules, this value becomes the final FAV because it is lower than the above calculated final FAV.

Table 2b provides the acute to chronic ratio data (ACR), as calculated from the PFOS databases. There were two acceptable ACR data points. The generic ACR of 18 was used for the third ACR. The final ACR is 9.12.

#### **5.1.1 Chronic Criterion (CC), Based on Toxicity to Aquatic Organisms**

Per Minnesota Rules 7050 Subp. 5, the chronic criterion (CC) is calculated by dividing the final FAV by the appropriate ACR. As discussed above, the Tier 2 FAV of 1302 ug/L was lowered to 170 ug/L based upon an evaluation of an acute midge study (STS-110). The toxicity-based chronic criterion for PFOS is therefore the final FAV (170 ug/L) divided by the final ACR of 9.12. The toxicity-based chronic criterion is 18.6 ug/L (rounded to 19 ug/L).

In a recent study with *Rana pipiens* (Northern leopard frog), Ankley *et al.* (2004; STS-114) presented chronic exposure data which indicated that PFOS can alter morphogenesis. At the lowest PFOS concentration tested (30 ug/L) abnormal foot development was observed relative to the control animals. Since a NOEC was not reported in this study, it is unknown at this time whether the above calculated chronic value for PFOS of 19 ug/L will be protective for native amphibian development.

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### **5.1.2 Chronic Criterion (CC), Based on Toxicity to Aquatic Plants**

The final plant value is based on the Northern milfoil (*Myriophyllum sibiricum*). Northern milfoil had the most sensitive chronic endpoint of all acceptable plant data. The northern milfoil had a NOEC of 300 ug/L. The animal derived toxicity-based chronic criterion of 19 ug/L will thus protect aquatic plants.

### **5.1.3 Maximum Criterion (MC)**

Per Minnesota Rules Chapter 7050 Subp. 5, the maximum criterion (MC) is calculated by dividing the final FAV by 2. The final FAV for PFOS is 170 ug/L, as presented in Table 3b. Thus, the MC is 85 ug/L.

## **5.2 Human Health-based Criteria**

In order to develop both of the human health-based surface water criteria, a risk-based toxicity criterion is required, along with a chemical-specific bioconcentration/bioaccumulation factor (BCF/BAF). Appendix D to this document provides the risk-based criterion for PFOS, as developed by the Minnesota Department of Health. Tables 5a and 5b (Appendix C) provide all of the BCF/BAF data obtained to date on PFOS. Two different PFOS human health-based criteria were developed. One was completed for Lake Calhoun; the second was completed for the Mississippi River (Pool 3). The two human health-based criteria were developed as a result of the differences in the BAFs found at Lake Calhoun and the Mississippi River (Pool 3).

### **5.2.1 Drinking Water Plus Fish Consumption Criterion (dfCC)**

According to Minnesota Rules 7050 Supb. 6. A., a dfCC criterion is calculated for a non-carcinogen using the following equation:

$$\text{dfCC} = \frac{\text{RfD (mg/kg - d)} \times 70 \text{ kg} \times K}{\text{K}}$$

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$$(\text{mg/L}) \quad 2\text{L/d} + [0.030 \text{ kg/d} \times \text{BAF}]$$

Where: dfCC = drinking water plus fish consumption criterion (mg/L)  
 RfD = reference dose = 0.000075 (see Appendix D)  
 70 kg = standard body weight of an adult  
 K = exposure fraction attributed to drinking water and fish consumption (0.2)  
 2 L/d = amount of water assumed to be consumed per day  
 0.030 kg/d = amount of fish assumed to be consumed per day  
 BAF = final BAF (see Section 3.3 of this report) = 2802 L/kg for Lake Calhoun and 5737 L/kg for Mississippi River (Pool 3)

$$\text{dfCC}_{\text{lake}} = \frac{(0.000075) (70) (0.2)}{2 + [(0.030) (2802)]} = 0.0000122 \text{ mg/L} = 12 \text{ ng/L}$$

$$\text{dfCC}_{\text{river}} = \frac{(0.000075) (70) (0.2)}{2 + [(0.030) (5737)]} = 0.000006 \text{ mg/L} = 6 \text{ ng/L}$$

### 5.2.2 Fish Consumption Criterion (fCC)

According to Minnesota Rules 7050 Subp. 6.B., a fCC is calculated for a non-carcinogen using the following equation:

$$\begin{aligned} \text{fCC} &= \frac{\text{RfD (mg/kg-d)} \times 70 \text{ kg} \times K}{(\text{mg/L}) \quad 0.01 \text{ L/d} + [0.030 \text{ kg/d} \times \text{BAF}]} \end{aligned}$$

Where: fCC = fish consumption criterion (mg/L)  
 RfD = reference dose = 0.000075 (see Appendix D)  
 70 kg = standard body weight of an adult  
 K = exposure fraction attributed to water and fish consumption (0.2)  
 0.01 L/d = assumes incidental ingestion at water

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0.030 kg/d = amount of fish assumed to be consumed per day  
 BAF – final BAF (see Section 3.3 of this report) = 2802 L/kg for  
 Lake Calhoun and 5737 L/kg for Mississippi River (Pool 3)

$$fCC_{\text{lake}} = \frac{(0.000075) (70) (0.2)}{0.01 + [(0.030) (2802)]} = 0.00001248 \text{ mg/L} = 12 \text{ ng/L}$$

$$fCC_{\text{river}} = \frac{(0.000075) (70) (0.2)}{0.01 + [(0.030) (5737)]} = 0.0000061 \text{ mg/L} = 6 \text{ ng/L}$$

## **APPENDIX A**

### Table of Contents for Accompanying Database Binders

## **APPENDIX B**

Analytical Report - PFOS Data in Minnesota Waters and Fish (on CDs)

## APPENDIX C

Toxicity-based Summary Tables on PFOS



## APPENDIX D

### Health Risk-based Criteria for PFOS