Geographical Distribution of Perfluorinated Compounds in Fish from Minnesota Lakes and Rivers

ARTICLE *in* ENVIRONMENTAL SCIENCE AND TECHNOLOGY · FEBRUARY 2010

Impact Factor: 5.33 · DOI: 10.1021/es903777s · Source: PubMed

7 AUTHORS, INCLUDING:

Mark J Strynar
United States Environmental Protection A…
70 PUBLICATIONS **1,615** CITATIONS
SEE PROFILE

Shoji F Nakayama
National Institute for Environmental Studies
37 PUBLICATIONS **784** CITATIONS
SEE PROFILE

Andrew Lindstrom
United States Environmental Protection A…
46 PUBLICATIONS **1,629** CITATIONS
SEE PROFILE

Environ. Sci. Technol. **2010,** *44,* 2549–2554

Geographical Distribution of Perfluorinated Compounds in Fish from Minnesota Lakes and Rivers

AMY D. DELINSKY,[†] MARK J. STRYNAR,*[,†] PATRICIA J. MCCANN,[‡] JERRY L. VARNS,[§] LARRY MCMILLAN,[§] SHOJI F. NAKAYAMA,[||] AND ANDREW B. LINDSTROM[†]

Human Exposure and Atmospheric Sciences Division, National Exposure Research Laboratory, U.S. Environmental Protection Agency, Research Triangle Park, North Carolina 27711, Health Risk Assessment Unit, Environmental Health Division, Minnesota Department of Health, St. Paul, Minnesota 55164, Senior Environmental Employee Program, National Caucus and Center on Black Aged Inc., Durham, North Carolina 27713, and National Risk Management Research Laboratory, U.S. Environmental Protection Agency, Cincinnati, Ohio 45268

Received December 14, 2009. Revised manuscript received January 28, 2010. Accepted January 29, 2010.

In response to growing interest in human exposure to perfluorinated compounds (PFCs), the state of Minnesota measured and reported PFC concentrations in fish collected from the Minneapolis−St. Paul area. To better determine the geographical distribution of PFC contamination throughout Minnesota, fish were collected from 59 lakes throughout the state and several areas along the Mississippi River. Composite fish samples were analyzed for 10 PFC analytes by solid-phase extraction (SPE) and liquid chromatography−tandem mass spectrometry (LC/MS/MS). PFOS (perfluorooctanesulfonate) was the most commonly detected PFC, occurring in 73% of fish from the Mississippi River but only 22% of fish from lakes. Fish from Mississippi River Pool 2 near the Minneapolis−St. Paul area had the highest levels of PFOS, whereas locations upstream had PFOS concentrations below 40 ng/g, the concentration at which Minnesota issues "one meal per week" fish consumption advice. Fish from most Minnesota lakes tested (88%) had PFOS concentrations below 3 ng/g. Two lakes, McCarrons and Zumbro, contained fish with PFOS levels above 40 ng/g. The results reported here will help researchers to better understand the extent of PFC contamination in Minnesota fish and evaluate potential sources of contamination and will provide a basis for comprehensive fish consumption advice.

Introduction

Perfluorinated compounds (PFCs) have been used in a wide variety of industrial and consumer products due to their inherently useful chemical properties and resistance to degradation (*1*). As a result of their widespread use and persistence, PFCs are now found in environmental, biological, and human samples from areas throughout the world (*2, 3*). Human exposure to the PFCs is of concern because studies have found that perfluorooctanesulfonate (PFOS) and perfluorooctanoic acid (PFOA), the two most commonly studied PFCs, have adverse effects in laboratory animals and their offspring (*4, 5*). In light of mounting concern over potential adverse effects related to the PFCs, the United States Environmental Protection Agency (U.S. EPA) has recently issued provisional health advisories for PFOS and PFOA in drinking water (*6*). Moreover, PFOS has recently been added to the list of restricted use compounds appearing in Annex B of the Stockholm Convention on Persistent Organic Pollutants (POPs) (*7*).

Although the pathways by which humans are exposed to PFCs are still not well characterized, dietary exposure has been hypothesized to be an important human pathway (*8, 9*). In particular, studies have suggested that the ingestion of contaminated fish may be an important human exposure route for some PFCs (*10, 11*). Studies have documented elevated PFC concentrations (particularly PFOS) in the fillets of fish species commonly consumed by humans; however, limited information is available regarding the mechanisms by which fish become contaminated and the geographical distributions of contaminated fish (*12*−*16*). There are many proven health benefits associated with eating fish, but because they may also contain relatively high levels of environmental pollutants (primarily metals and POPs), an evaluation of these potential exposures is helpful in selecting food items.

The state of Minnesota may be in a unique position of both having PFCs present in the environment from known local sources and having a large number of lakes and rivers from which many recreational anglers catch and eat fish (*17*). In response to the growing concerns over potential human exposure to PFCs, Minnesota state agencies collected both water and fish samples for PFC analysis (*17*). Fish fillets from 53 St. Paul−Minneapolis metropolitan area lakes and two Duluth, MN, area lakes have been analyzed for PFCs (*18, 19*). Fillet samples from several lakes in the Minneapolis−St. Paul area, the two Duluth lakes, and some areas of the Mississippi River contained PFOS at concentrations that were high enough to prompt state officials to issue fish consumption advisories for PFOS (*17, 20*). Specifically, Minnesota fish consumption advisories recommend eating no more than one meal of fish per week when PFOS levels in fish are above 40 ng/g and eating no more than one meal per month when PFOS levels are above 200 ng/g (*20*). Minnesota state agencies then collected additional fish samples from a variety of lakes throughout the state and several locations on the Mississippi River in Minnesota in an effort to establish a more complete determination of the extent of PFC contamination.

Ten PFCs were analyzed in the fillets of fish from the Mississippi River and 59 lakes throughout Minnesota by liquid chromatography−tandem mass spectrometry (LC/MS/MS) analysis with a method that has been thoroughly evaluated and was determined to have good performance characteristics (precision, accuracy, recovery) for PFC analysis (*15*). Prior to the work reported here, fish from lakes outside urban areas in Minnesota had not been tested for PFCs. The results of this study provide a better understanding of where contamination may occur, the potential sources of con-

* Corresponding author: Mail Drop D205-05, Environmental Protection Agency, Research Triangle Park, NC 27711. Tel 919-541-3706; fax 919-541-3527; e-mail strynar.mark@epa.gov.
[†] Human Exposure and Atmospheric Sciences Division, National Exposure Research Laboratory, U.S. EPA.
[‡] Minnesota Department of Health.
[§] National Caucus and Center on Black Aged Inc.
[||] National Risk Management Research Laboratory, U.S. EPA.

10.1021/es903777s
Published on Web 02/17/2010

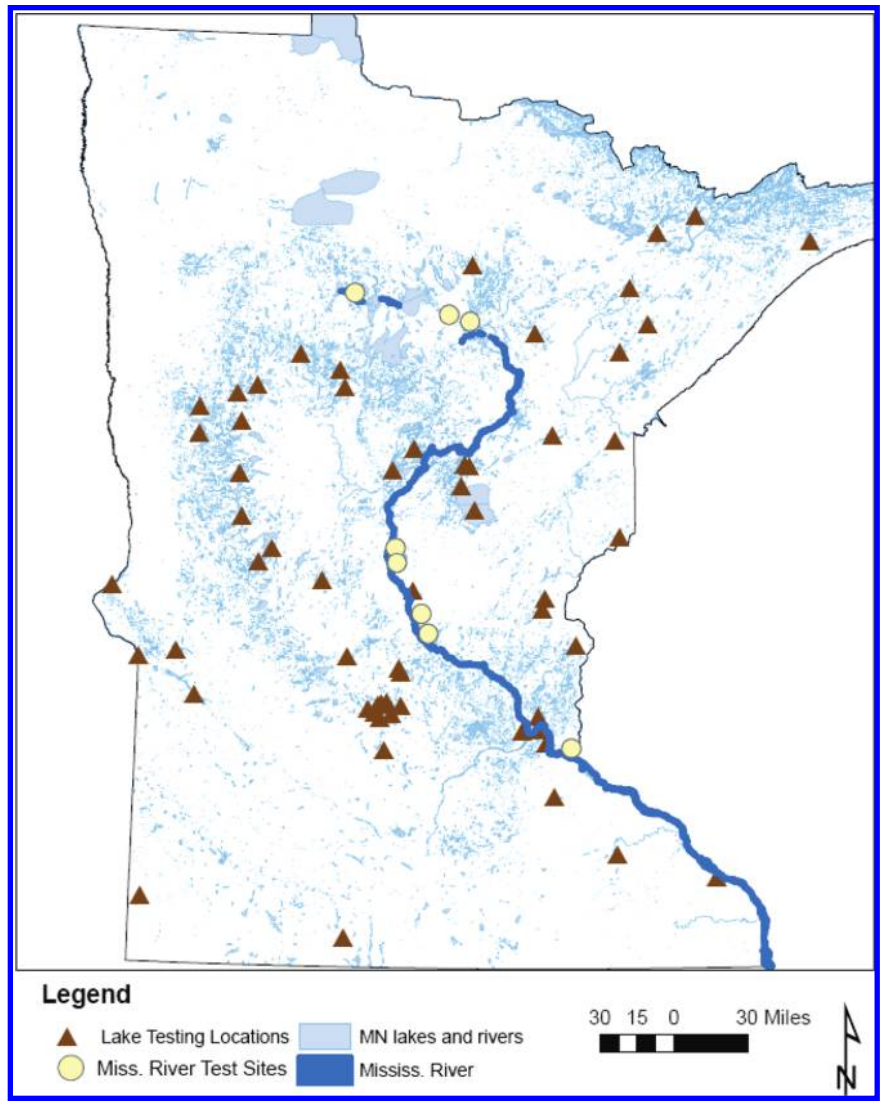

FIGURE 1. Sampling locations in Minnesota.

tamination, and the extent to which consumption of fish may be a pathway by which Minnesota residents are exposed to PFCs.

Materials and Methods

Chemicals and Reagents. Potassium salts of perfluorobutanesulfonate (PFBS, 98% purity) and perfluorohexanesulfonate (PFHS, 93%) were provided by 3M Company (St. Paul, MN). The potassium salt of perfluorooctanesulfonate (PFOS, 98%) was purchased from Fluka (Buchs, Switzerland). Perfluorohexanoic acid (C6, 97%), perfluoroheptanoic acid (C7, 99%), perfluorooctanoic acid (PFOA, 96%), perfluorononanoic acid (C9, 97%), and perfluorodecanoic acid (C10, 98%) were purchased from Sigma−Aldrich (St. Louis, MO). Perfluoroundecanoic acid (C11, 96%) and perfluorododecanoic acid (C12, 96%) were purchased from Oakwood Products (West Columbia, SC). Four isotopically labeled internal standards were used for the quantitation of PFCs. Oxygen-labeled ammonium perfluorooctanesulfonate ($[^{18}O_2]$PFOS) was purchased from RTI International (Research Triangle Park, NC). Isotopically labeled PFOA ($[^{13}C_2]$PFOA) was purchased from Perkin-Elmer Life and Analytical Sciences, Inc. (Waltham, MA). Labeled sodium perfluorohexanesulfonate ($[^{18}O_2]$PFHS) and labeled perfluoroundecanoic acid ($[^{13}C_2]$PFUnA) were purchased from Wellington Laboratories (Ontario, Canada). HPLC-grade methanol was purchased from Burdick−Jackson (Muskegon, MI) and contained no measurable PFCs. Deionized (DI) water was obtained from a Barnstead EASYpure ultraviolet/ultrafiltration (UV/UF) compact reagent-grade water system (Dubuque, IA) and had no detectable amounts of PFCs. Sodium hydroxide (NaOH), sodium acetate, glacial acetic acid, ammonium acetate, and ammonium hydroxide (NH_4OH) were purchased from Sigma−Aldrich (St. Louis, MO).

Fish Collection. Fish were collected by electroshocking and netting from areas of the Upper Mississippi River in Minnesota and from lakes throughout the state by the Minnesota Department of Natural Resources (MDNR) and the Minnesota Pollution Control Agency (MPCA) in 2007 as part of routine collections of fish for contaminant monitoring by the Minnesota Interagency Fish Contaminants Monitoring Program (Figure 1, Figure S1 in Supporting Information). Fish included for PFC analysis were a convenience sample based on availability of fish tissue and lakes selected for scheduled fisheries assessments or other monitoring purposes. Fish from a range of lake sizes, locations, and major watersheds were included in this study (Figure 1, Table S1 in Supporting Information). Lake size ranged from 10 to over 130 000 acres. This screening study included at least one lake from 29 of the 81 major watersheds in Minnesota. Land use in these watersheds ranged from <1% to 86% forested, <1% to 47% developed, and <1% to 87% cultivated crops/hay/pasture lands. Some lakes had no public access while others had multiple public access sites. Lakes within 500 m

of a fire station and those associated with a National Pollutant Discharge Elimination System (NPDES) permit are noted (Table S1 in Supporting Information). PFCs are not regulated under NPDES in Minnesota; however, a permit indicates a point source discharge into the water body.

Species of fish that were included in the present study were bluegill (*Lepomis macrochirus*), black crappie (*Pomoxis nigromaculatus*), and pumpkinseed (*Lepomis gibbosus*). Sunfish and crappie were selected for analysis to allow comparisons to data from previously sampled Minneapolis−St. Paul metro area lakes. Most existing data from these metro lakes are for sunfish, crappie, and bass. Mississippi River Pool 2 fish were collected to serve as a reference because fish from this area have historically had elevated PFC concentrations (*14, 17*).

Fish were wrapped in aluminum foil, kept on ice during transport, and then stored in a −10 °C laboratory freezer. Each fish was thawed and filleted (skin-on), and composite samples from each sampling location were made in a Minnesota state lab by combining fillets of (n = 1−17) fish of the same species. Only three of the 70 samples were from a single fillet. Combined fillets were homogenized in a Minnesota state lab by use of a meat grinder, with aliquots of the ground composites stored in polypropylene Falcon tubes (Becton Dickinson, Franklin Lakes, NJ) at −20 °C until they were shipped to the analytical laboratory at EPA. At the EPA analytical laboratory, samples were stored at −80 °C prior to further processing.

Sample Homogenization and Preparation. Samples were prepared and analyzed at the EPA analytical laboratory following a method previously described by Delinsky et al. (*15*). Briefly, water was added to the ground composite samples at a ratio of 3 mL of DI water/g of fish fillet tissue. Samples were homogenized with a Polytron PT 10/35 homogenizer (Brinkmann Instruments, Westbury, NY) or a Waring blender (Waring Laboratory Science, Torrington, CT). Fish fillet homogenate was stored at −20 °C in 50 mL polypropylene Falcon tubes prior to sample preparation and analysis.

Fillets of tilapia (*Tilapia aurea*) containing no PFCs above the assay limit of detection (LOD) were purchased from a local market (Grand Asia Market, Cary, NC), processed identically as samples, and used as a blank matrix for blank samples and the construction of calibration curves. Quality control (QC) samples were prepared from unspiked bluegill samples that were previously determined to contain naturally occurring high and low PFC levels. All fish homogenates (used in blanks, calibration curves, samples, and QCs) that were frozen after homogenization were thawed and rehomogenized immediately before sample preparation.

The solid-phase extraction (SPE)−LC/MS/MS method used in the present analysis has been thoroughly evaluated for method performance and is shown to have good precision, accuracy, and recovery (*15*). A 2 mL aliquot of either DI water, tilapia blank homogenate, or unknown sample homogenate was placed in a preweighed 15 mL Falcon tube. Aliquots of QC samples (2 mL) had been previously placed into preweighed Falcon tubes. Each tube containing DI water or fish fillet homogenate was reweighed to determine the weight of the homogenate. Appropriate amounts of the 10 PFC analytes were spiked into each of eight tilapia blank samples to generate a calibration curve. Eight milliliters of 0.01 N NaOH in MeOH containing 15 ng each of the four isotopically labeled internal standards was added to each 2 mL sample, and the mixture was vortexed. Each sample was then sonicated in an ultrasonic water bath for 30 min and centrifuged at 16800*g* for 5 min, and 3 mL of the supernatant was placed into a 50 mL Falcon tube. DI water (27 mL) was added to the supernatant (3 mL) of each sample, and the mixture was vortex-mixed. Solid-phase extraction (SPE) was performed on the diluted supernatant by use of Waters Oasis WAX cartridges (60 mg of sorbent, 60 μm particle size). SPE cartridges were conditioned with 4 mL of 0.03% NH_4OH in MeOH, followed by 4 mL of MeOH, and equilibrated with 4 mL of DI water. The entire 30 mL sample was then loaded onto the SPE cartridge. Each cartridge was then washed with 4 mL of 25 mM acetate buffer (pH = 4) followed by 4 mL of MeOH. PFCs were eluted from the cartridge with 4 mL of 0.03% NH_4OH in MeOH. The eluates were evaporated to approximately 0.5 mL in a TurboVap sample concentrator (Caliper Life Sciences, Mountain View, CA) at 35 °C and 10−15 psi. An aliquot of each concentrated sample (280 μL) was added to individual autosampler vials containing 120 μL of 2 mM ammonium acetate buffer for sample analysis.

Instrumental Analysis. Each analytical batch consisted of approximately 20−30 unknown fish composite samples, eight double blanks (consisting of methanol and buffer; three prior to the run and five throughout the run), one method blank (DI water), two matrix blanks (tilapia homogenate), eight calibration curve standards (spiked in tilapia homogenate), and four QC samples (unspiked bluegill homogenate, two low and two high for each analyte). Samples were analyzed on an Agilent 1100 HPLC instrument (Agilent Technologies, Palo Alto, CA) interfaced with a Sciex 3000 triple-quadrupole mass spectrometer (Applied Biosystems/MDS Sciex, Foster City, CA). A 10 min isocratic HPLC run was used with a mobile phase consisting of 25% 2 mM ammonium acetate buffer (component A) and 75% MeOH (component B) at a flow rate of 200 μL/min. The HPLC column was a Phenomenex Luna C18(2) column (3.0 × 50 mm, 5.0 μm) with an injection volume of 10 μL. Electrospray ionization was used in the mass spectrometer source, which was maintained at 400 °C. Analyte-specific mass spectrometer parameters were optimized for each individual compound, and mass transitions for each analyte and internal standard were monitored by multiple reaction monitoring (MRM; Table S2 in Supporting Information). Because the potential for incorrect identification of PFOS and PFHS in biological matrices has been reported (*21*), a subset of samples was reanalyzed by use of quantitation and confirmation ion mass transitions for each analyte in order to ensure correct analyte identification and quantification.

Quantitation. A new eight-point calibration curve was prepared for all 10 analytes with each analytical batch. The calibration curve was run at the beginning and the end of each analytical batch, and the replicate injections were used to construct the calibration curve for sample quantitation. The calibration range was 1−600 ng/g for PFOS and 0.4−50 ng/g for the remaining PFCs. Analyst software (version 1.4.2, Applied Biosystems/MDS Sciex, Foster City, CA) was used for the quantitation of all compounds. Linear calibration curves with $1/x$ weighting were required to have a correlation coefficient (r) of greater than 0.99, with all points except the lowest point being within ±20% of the theoretical concentration (±30% for the lowest calibration curve point). Isotopically labeled PFOS ($[^{18}O_2]$PFOS) was the internal standard used for PFOS quantitation, $[^{18}O_2]$PFHS was used for the quantitation of PFHS and PFBS, $[^{13}C_2]$PFOA ($[^{13}C_2]$C8) was used for quantitation of the C6 to C9 acids, and $[^{13}C_2]$PFUnA ($[^{13}C_2]$-C11) was used for the quantitation of C10−C12 acids. The ratio of the analyte/internal standard area counts was used for quantification. Linear and branched isomers were present in chromatograms for unlabeled PFOS, and all isomers were integrated for quantitation. Chromatograms of $[^{18}O_2]$PFOS had a peak for the linear isomer only, and this isomer was integrated for quantitation. Samples with calculated concentrations outside of the calibration range were diluted with tilapia blank matrix, reprepared, and reanalyzed in a separate analytical batch.

TABLE 1. Method Performance for Sample Analysis

analyte	N[a]	QC[b] pool (avg characterized concn, ng/g)	measd concn (ng/g)	% RSD[c]	avg % accuracy
PFOS	3	low (26.2)	26.1	2.64	99.4
PFOS	3	high (222)	211	4.73	95.2
C10	3	low (6.31)	6.46	6.36	102
C10	3	high (14.7)	14.8	1.21	101
C11	3	low (3.25)	2.86	13.3	88.0
C11	3	high (31.4)	30.1	7.05	95.8
C12	3	low (3.24)	2.89	19.5	87.9
C12	3	high (7.11)	6.34	13.0	89.1

[a] Number of replicates, with each replicate representing the average of duplicate samples. [b] Quality control. [c] Percent relative standard deviation of QC samples run in sample batches in the present study.

Quality Control. Quality control samples were prepared from two pools of unspiked bluegill homogenate that each contained naturally occurring levels of the target PFCs at two distinct levels. Samples from each QC pool were run at the analytical laboratory on multiple days in order to characterize the average concentration for each PFC in each QC pool (Table 1). Two samples from each QC pool (A and B) were run with each sample set, with one replicate run immediately after the calibration curve at the beginning of the run and the other replicate run immediately before the calibration curve at the end of the run. In order for an analytical batch to be considered acceptable for an analyte, the average of the duplicate QC samples had to be within two standard deviations of the characterized average for that analyte.

Previously determined performance characteristics of the current method showed that at three concentration levels (1) limits of quantitation (LOQ) were 0.52 ng/g for PFOS, 5.21 ng/g for C7, and a range from 0.01 to 1.89 ng/g for the remaining eight analytes; (2) interday and intraday precision values were less than 20% RSD; (3) interday and intraday accuracy values were less than 20% different from theoretical values for all analytes except C6 and C7; (4) recovery of the 10 target PFCs from blank tilapia fillet matrix were 76.2−133%; and (5) bluegill samples spiked with 20 ng/g PFOS and 4 ng/g of the remaining nine PFC analytes had accuracies ranging from 74.9% to 136% (*15*).

Repeated analyses of fish samples with this method has shown that (1) PFOS and [$^{18}O_2$]PFOS have retention times within 2% of each other and (2) taurodeoxycholic acid (TDCA), a bile acid found to potentially interfere with PFOS analysis (*21*), has a retention time that is 6% or more different (later retention time) than those of PFOS and [$^{18}O_2$]PFOS. Therefore, unknown samples containing detectable amounts of PFC analytes for which corresponding labeled internal standards were available (such as PFOS and corresponding internal standard [$^{18}O_2$]PFOS) were required to have analyte and internal standard retention times within 2% of each other in order to ensure proper analyte identification. Unknown samples containing detectable levels of analytes with no corresponding labeled internal standard (such as C10 and [$^{13}C_2$]-C11 internal standard) were required to have an analyte/internal standard retention time ratio within 2% of the average retention time ratio in standards. Comparisons of calculated analyte concentrations by quantitation and confirmation ions were also used as an indicator of proper analyte identification, with concentrations less than 25% different indicating proper analyte identification.

Results

Quality Control. In the present study, none of the blank tilapia samples had any quantifiable PFCs above the assay limit of detection (LOD, $n = 6$). QC samples for PFOS, C10, C11, and C12 showed good method performance and met the acceptance criteria for analytical batches containing samples with detectable levels of the PFC analytes (Table 1). Black crappie samples spiked with 20 ng/g PFOS and 4 ng/g of the remaining nine PFCs exhibited accuracies of 88−107% (Table S3 in Supporting Information). All calibration curves had r values greater than 0.99 for all analytes. Calculated concentrations of PFOS, C10, C11, and C12 determined by use of both quantitation and confirmation ions were within 25% of each other for all samples containing detectable levels of PFCs, indicating that all analytes were properly identified and quantitated. For the detectable PFC analytes, the percent difference for duplicate samples ($n = 8$) ranged from 1% to 20%, with much of the variation occurring at the lowest levels of the calibration curve. Percent differences for replicate PFOS analyses ranged from 1% to 5%.

Overview. A summary of all of the analytical data generated in this investigation can be found in Table S4 in Supporting Information. PFOS was the most commonly detected PFC, occurring above the limit of quantitation (LOQ = 1 ng/g, the lowest calibration curve point) in 30% of all fish taken from Minnesota lakes and the Mississippi River. C10 was found above the limit of quantitation (LOQ = 1.11 ng/g) in 7% of samples, and C11 and C12 were each present above the LOQ (1.05 ng/g for C11, 0.72 ng/g for C12) in 3% of samples. PFHS was found above the LOQ (0.40 ng/g = lowest calibration curve point) in one sample. None of the remaining PFC analytes were found above the LOQ.

Mississippi River Samples. Sampling sites along the Mississippi River can be found in Figure 1 and Figure S1 in Supporting Information. PFOS was found above the LOQ of 1 ng/g in 73% of fish collected from the Mississippi River and was the most commonly detected PFC (Table 2). The highest concentrations of PFOS (144 ng/g in pumpkinseed and 2000 ng/g in bluegill fillet) were found in two composite samples from Pool 2, an area of the Mississippi River with historically high PFC concentrations (*14, 17*). Both composite fish samples from Pool 2 contained C10−C12 at concentrations of 2.13−15.0 ng/g of fish. The bluegill from Pool 2 with a PFOS concentration of 2000 ng/g also contained 0.47 ng/g PFHS. Six separate composite samples from fish collected 110−150 miles upstream of Pool 2 all had PFOS concentrations above the LOQ, ranging from 3.06 to 20 ng/g of fillet. The lowest PFOS concentrations (<LOQ) were found in three composite samples from fish collected approximately 400 miles upstream of Pool 2.

Minnesota Lakes. PFOS was the most commonly detected PFC in fish collected from 59 lakes throughout the state, occurring above the LOQ at 22% of the sites, with quantifiable concentrations ranging from 1.08 ng/g (Fall Lake) to 52.4 ng/g in fish from Zumbro Lake (Table 2). Fish collected from 52 of 59 lakes (88% of lakes sampled) had PFOS levels below 3 ng/g. PFOS concentrations in fish from only two lakes (McCarrons and Zumbro) were above 40 ng/g. Fish from the four Minneapolis−St. Paul area lakes included in the study (McCarrons, Nokomis, Pickerel, and Simley) had PFOS concentrations ranging from 4.39 to 47.3 ng/g. C10 was the only other PFC found in fish from Minnesota lakes (1.23−3.24 ng/g), and occurred only in lakes where PFOS concentrations in fish were 10 ng/g or greater (Pickerel, McCarrons, and Zumbro).

Comparison with Previous Studies of Minnesota Fish. Previous studies regarding PFC concentrations in Minnesota fish fillet samples agree with the present study in that (1) PFOS is the predominant PFC present; (2) C10, C11, and C12 acids are found above the LOQ in some samples; and (3) PFHS, PFBS, and the C6−C9 carboxylic acids are found either in very low concentrations or are below the LOQ (*14−18*).

TABLE 2. Concentrations of PFCs in Samples Containing PFOS above the LOQ[a]

	concn (ng/g)				
sampling location[b]	PFOS	C10	C11	C12	PFHS
MR 11	2000	15.0	6.72	3.74	0.47
MR 10	144	2.94	2.13	4.42	nd
Zumbro	52.4	3.24	nd	nd	nd
McCarrons	47.3	1.97	nd	nd	nd
MR 6	20.0	nd	nd	nd	nd
MR 7	17.3	nd	nd	nd	nd
Carlos	12.3	nd	nd	nd	nd
Pickerel	10.0	1.23	nd	nd	nd
MR 9	9.35	nd	nd	nd	nd
MR 4	6.65	nd	nd	nd	nd
MR 8	5.99	nd	nd	nd	nd
Simley	5.13	nd	nd	nd	nd
Winona	4.70	nd	nd	nd	nd
Nokomis	4.39	nd	nd	nd	nd
MR 5	3.06	nd	nd	nd	nd
Whiteface Reservoir	2.29	nd	nd	nd	nd
Tamarack	1.95	nd	nd	nd	nd
Byllesby	1.42	nd	nd	nd	nd
Lac Qui Parle	1.31	nd	nd	nd	nd
Goose	1.25	nd	nd	nd	nd
Fall	1.08	nd	nd	nd	nd

[a] A designation of nd indicates that no determination of the sample concentration was made because the calculated concentration was below the limit of quantitation (<LOQ). [b] Sample locations beginning with MR are Mississippi River samples collected from one of 11 locations along the river; exact locations of these sampling locations can be found in Figure S1 in Supporting Information. The remaining samples in this table were collected from lakes whose locations can also be found in Figure S1 in Supporting Information.

PFOS concentrations in fish taken from Pool 2 of the Mississippi River have historically been high compared to fish taken from other stretches of the Mississippi River in Minnesota (*14, 17*). In two previous studies involving the analysis of fillets from Pool 2 fish, PFOS concentrations ranged from 15 to 90 ng/g in carp and ranged from 25 to 5150 ng/g in the fillets of seven species of fish (*14, 16*). The MPCA reported concentrations of PFOS ranging from 13.1 to 1860 ng/g in the fillets of seven species of fish collected from Mississippi River Pool 2 (*17*). The PFOS levels measured in Mississippi River Pool 2 fish in the present study were lower in pumpkinseed (144 ng/g) compared to bluegill (2000 ng/g), a finding that is consistent with the MPCA study that measured these two species in a Twin Cities Metro Lake (Fish Lake) (*17*). This suggests that species-specific concentration factors may be very different for fish living in the same water and that there is considerable value in determining these relationships. The highest measured value of 2000 ng/g PFOS in Pool 2 bluegill in the present study is 8 times higher than the average bluegill PFOS concentration previously reported (approximately 250 ng/g) but is within the range of Pool 2 PFOS concentrations previously reported in fish fillets (*14*). This finding indicates that there is considerable variation in PFC concentrations within a single species living in one section of the river. Further investigation of the factors that influence this variation would be useful.

Concentrations of PFOS in composite samples from Mississippi River fish taken 110−150 miles upstream of Pool 2 in the present study (3.06−20 ng/g) agree well with previously reported PFOS concentrations in fillets of fish taken from similar stretches of the Mississippi River at the following locations: (1) between Brainerd and St. Cloud, 120 miles upstream of Pool 2 (4.3−19 ng/g in carp); and (2) Brainerd area, approximately 200 miles upstream of Pool 2 (7.38−18 ng/g in four fish species) (*16, 17*). The concentrations of PFOS in Mississippi River fish in the present study taken 110−150 miles upstream of Pool 2 are also lower than previously determined concentrations of PFCs in bluegill from pools 3−5a (approximately 23.5−200 ng/g), which are located downstream of Pool 2 and the Minneapolis−St. Paul area (*14, 16, 17*). In the present study, PFOS concentrations in samples taken approximately 400 miles upstream of the Minneapolis−St. Paul area (<LOQ) are comparable to previously reported values in the St. Croix River (<LOQ), which is an area with minimal known industrial inputs (*14, 17*). Concentrations of C10−C12 acids in Pool 2 of the Mississippi River in the present study (2.13−15.0 ng/g of fish) compared well with previously reported Pool 2 C10−C12 concentrations in the fillets from seven fish species (<LOQ to 17.5 ng/g) (*16, 17*).

The present study contains the first measurements of PFOS concentrations in fish from rural Minnesota lakes. PFOS concentrations in fish fillets (<LOQ to 52.3 ng/g) were generally lower than those previously found in Minneapolis−St. Paul metropolitan area lakes (<LOQ to 345 ng/g in bluegill and <LOQ to 574 ng/g in black crappie) (*14, 17*). Fish from only two out of 59 lakes had PFOS concentrations above the Minnesota fish consumption advisory level of 40 ng/g. One of the two lakes (McCarrons, 47.3 ng/g) is located in the Minneapolis−St. Paul metropolitan area, while the other (Zumbro, 52.4 ng/g) is downstream of the effluent from the Pine Island Wastewater Treatment Plant (WWTP), which had elevated PFOS concentrations and may be a contributing factor to the higher levels of PFOS in the fish (*17*). Fish from 52 out of 59 sampling sites in the present study had PFOS concentrations that were below 3 ng/g, which is generally much lower than PFOS concentrations in urban lakes (*17*). Previously, the release of aqueous firefighting foams (AFFF) to a body of water was shown to result in high concentrations of PFOS in the water to which the AFFF were applied, in the livers of fish living in the waters where AFFF were applied, and in local groundwater (*22, 23*). In the present study, the presence of a fire station within 500 m of the lakes sampled was not an indicator of higher PFC concentrations in fish. There have been no known previous reports of C10 concentrations in fish from Pickerel, McCarrons, and Zumbro Lakes. However, concentrations of C10 found in fish from these lakes (1.23−3.24 ng/g) compare well with levels of C10 reported in other Minneapolis−St. Paul metropolitan area lakes (<LOQ to 8.02 ng/g) (*17*).

In summary, concentrations of 10 PFCs were measured in composite fish fillet samples from the Mississippi River and 59 Minnesota lakes by a previously described LC/MS/MS method in order to provide preliminary information on the geographic distribution of PFCs in Minnesota fish. Results indicate that PFOS contamination in fish collected in Minnesota occurs primarily in watersheds that are in close proximity to the Minneapolis−St. Paul metropolitan area and other water bodies with potential PFC sources. The results also suggest that fish from rural lakes throughout Minnesota and areas of the Mississippi River located 110−450 miles upstream of Pool 2 have PFOS concentrations below the concentration (40 ng/g) at which Minnesota recommends limiting fish consumption. These findings should help researchers better understand the extent of PFOS contamination in Minnesota fish, provide information for fish advisories, and determine areas of focus for future fish collection efforts. Additional studies will be useful in order to better understand the geographical distributions of PFCs in Minnesota fish.

Acknowledgments

This research was supported in part by an appointment to the Research Participation Program at the National Exposure

Research Laboratory administered by the Oak Ridge Institute for Science and Education through an interagency agreement between the U.S. Department of Energy and the U.S. Environmental Protection Agency. We also acknowledge Waters Corporation for the donation of Oasis WAX SPE cartridges (Cooperative Research and Development Agreement 392-06). We thank individuals at the Minnesota Pollution Control Agency and Minnesota Department of Natural Resources for collecting and processing fish samples and for providing information on land use. The U.S. Environmental Protection Agency through its Office of Research and Development funded and managed the research described here. It has been subjected to Agency review and approved for publication. Mention of trade names or commercial products does not constitute endorsement or recommendation for use.

Supporting Information Available

Four tables, describing land use information for Minnesota lakes and fish species collected from each lake, mass transitions monitored for labeled and nonlabeled PFC analytes, accuracy determination in black crappie, and PFC concentrations in all 70 samples, and one figure showing identifications of Minnesota lakes and river sampling locations are provided in the Supporting Information. This information is available free of charge via the Internet at http://pubs.acs.org.

Literature Cited

(1) Prevedouros, K.; Cousins, I. T.; Buck, R. C.; Korzeniowski, S. H. Sources, fate and transport of perfluorocarboxylates. *Environ. Sci. Technol.* **2006**, *40* (1), 32–44.

(2) Calafat, A. M.; Wong, L. Y.; Kuklenyik, Z.; Reidy, J. A.; Needham, L. L. Polyfluoroalkyl chemicals in the U.S. population: data from the National Health and Nutrition Examination Survey (NHANES) 2003−2004 and comparisons with NHANES 1999−2000. *Environ. Health Perspect.* **2007**, *115* (11), 1596–1602.

(3) Kannan, K.; Corsolini, S.; Falandysz, J.; Fillmann, G.; Kumar, K. S.; Loganathan, B. G.; Mohd, M. A.; Olivero, J.; Van Wouwe, N.; Yang, J. H.; Aldoust, K. M. Perfluorooctanesulfonate and related fluorochemicals in human blood from several countries. *Environ. Sci. Technol.* **2004**, *38*, 4489–4495.

(4) Lau, C.; Anitole, K.; Hodes, C.; Lai, D.; Pfahles-Hutchens, A.; Seed, J. Perfluoroalkyl acids: a review of monitoring and toxicological findings. *Toxicol. Sci.* **2007**, *99* (2), 366–394.

(5) Fenton, S. E.; Reiner, J. L.; Nakayama, S. F.; Delinsky, A. D.; Stanko, J. P.; Hines, E. P.; White, S. S.; Lindstrom, A. B.; Strynar, M. J.; Petropoulou, S-S. E. Analysis of PFOA in dosed CD-1 mice. Part 2: Disposition of PFOA in tissues and fluids from pregnant and lactating mice and their pups. *Reprod. Toxicol.* **2009**, *27* (3−4), 365–372.

(6) Provisional health advisories for perfluorooctanoic acid (PFOA) and perfluorooctane sulfonate (PFOS). Available at http://www.epa.gov/waterscience/criteria/drinking/pha-PFOA_PFOS.pdf.

(7) Report of the Conference of the Parties of the Stockholm Convention on Persistent Organic Pollutants on the work of its fourth meeting. Available at http://chm.pops.int/Portals/0/Repository/COP4/UNEP-POPS-COP.4-38.English.pdf.

(8) Weihe, P.; Kato, K.; Calafat, A. M.; Nielsen, F.; Wanigatunga, A. A.; Needham, L. L.; Grandjean, P. Serum concentrations of polyfluoroalkyl compounds in Faroese whale meat consumers. *Environ. Sci. Technol.* **2008**, *42* (16), 6291–6295.

(9) Fromme, H.; Tittlemier, S. A.; Volkel, W.; Wilhelm, M.; Twardella, D. Perfluorinated compounds - exposure assessment for the general population in western countries. *Int. J. Hyg. Environ. Health* **2009**, *212* (3), 239–270.

(10) Berger, U.; Glynn, A.; Holmstrom, K. E.; Berglund, M.; Ankarberg, E. H.; Tornkvist, A. Fish consumption as a source of human exposure to perfluorinated alkyl substances in Sweden - analysis of edible fish from Lake Vattern and the Baltic Sea. *Chemosphere* **2009**, *76* (6), 799–804.

(11) Falandysz, J.; Taniyasu, S.; Gulkowska, A.; Yamashita, N.; Schulte-Oehlmann, U. Is fish a major source of fluorinated surfactants and repellents in humans living on the Baltic Coast? *Environ. Sci. Technol.* **2006**, *40*, 748–751.

(12) Giesy, J. P.; Kannan, K. Global distribution of perfluorooctane sulfonate in wildlife. *Environ. Sci. Technol.* **2001**, *35*, 1339–1342.

(13) Hoff, P.; Van de Vijver, K.; Van Dongen, W.; Esmans, E.; Blust, R.; De Coen, W. Perfluorooctane sulfonic acid in bib (*Trisopterus luscus*) and plaice (*Pleuronectes platessa*) from the Western Scheldt and the Belgian North Sea: distribution and biochemical effects. *Environ. Toxicol. Chem.* **2003**, *22* (3), 604–614.

(14) Perfluorochemicals in fish fillets from Minnesota. Available at http://www.health.state.mn.us/divs/eh/fish/pfcposter.pdf.

(15) Delinsky, A. D.; Strynar, M. J.; Nakayama, S. F.; Varns, J. L.; Ye, X.; McCann, P. J.; Lindstrom, A. B. Determination of ten perfluoronated compounds in bluegill sunfish (*Lepomis macrochirus*) fillets. *Environ. Res.* **2009**, *109*, 975–984.

(16) Ye, X.; Schoenfuss, H. L.; Jahns, N. D.; Delinsky, A. D.; Strynar, M. J.; Varns, J. L.; Nakayama, S. F.; Helfant, L.; Lindstrom, A. B. Perfluorinated compounds in common carp (*Cyprinus carpio*) fillets from the Upper Mississippi River. *Environ. Int.* **2008**, *34* (7), 932–938.

(17) PFCs in Minnesota's ambient environment: 2008 progress report. Available at http://www.pca.state.mn.us/publications/c-pfc1-02.pdf.

(18) 2008 Minnesota Pollution Control Agency fish PFC data summary table. Available at http://www.pca.state.mn.us/cleanup/pfc/index.html#sedimentfish.

(19) Perfluorochemicals. Available at http://www.pca.state.mn.us/cleanup/pfc/index.html#.

(20) Minnesota Department of Health Fish Consumption Advisory Program meal advice categories summary table. Available at http://www.health.state.mn.us/divs/eh/fish/eating/mealadvicetables.pdf.

(21) Benskin, J. P.; Bataineh, M.; Martin, J. W. Simultaneous characterization of perfluoroalkyl carboxylate, sulfonate, and sulfonamide isomers by liquid chromatography-tandem mass spectrometry. *Anal. Chem.* **2007**, *79* (17), 6455–6464.

(22) Moody, C. A.; Martin, J. W.; Kwan, W. C.; Muir, D. C.; Mabury, S. A. Monitoring perfluorinated surfactants in biota and surface water samples following an accidental release of fire-fighting foam into Etobicoke Creek. *Environ. Sci. Technol.* **2002**, *36*, 545–551.

(23) Schultz, M. M.; Barofsky, D. F.; Field, J. A. Quantitative determination of fluorotelomer sulfonates in groundwater by LC MS/MS. *Environ. Sci. Technol.* **2004**, *38*, 1828–1835.

ES903777S