PERFLUORINATED CHEMICALS IN SURFACE WATERS AND SEDIMENTS FROM THE CONASAUGA, OOSTANAULA, AND COOSA RIVERS AND THEIR BIOACCUMULATION IN THE AQUATIC OLIGOCHAETE, *Lumbriculus variegatus*

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REFERENCE: *Proceedings of the 2011 Georgia Water Resources Conference*, held April 11–13, 2011, at the University of Georgia.

Abstract. Concentrations of perfluorinated chemicals (PFCs) were measured in surface waters and sediments from the Coosa River watershed to examine their distribution downstream of a suspected source. Samples from eight sites were analyzed using liquid chromatography-tandem mass spectrometry. To examine PFC bioaccumulation, sediments were also utilized in 28-d exposures with the aquatic oligochaete, *Lumbriculus variegatus*. Concentrations increased significantly below a land-application site (LAS) of municipal/industrial wastewater and were further elevated by unknown sources downstream. Comparisons with past surveys indicate improving conditions, but additional information is needed. Perfluorinated carboxylic acids (PFCAs) with eight or less carbons were the most prominent in surface waters but those with 10 or more carbons predominated sediment and tissue samples. Perfluorooctane sulfonate (PFOS) was the major homologue in contaminated sediments and tissues followed by perfluorodecanoate, perfluoroundecanoate, perfluorododecanoate, perfluorotridecanoate, and perfluorotetradecanoate. This pattern of sediment PFCs was consistent among sites and reflects homologue concentrations emanating from the LAS. Tissue PFC concentrations repeated patterns observed in the respective sediments but were 4 to 46 times greater depending on the homologue. The tendency of PFCs to bioaccumulate increased with PFCA chain length and the presence of the sulfonate moiety. Bioaccumulation factors indicate that short-chain PFCAs with less than seven carbons may be environmentally benign alternatives in aquatic ecosystems, but sulfonates with four to seven carbons may be as likely to bioaccumulate as PFOS.

INTRODUCTION

Perfluorinated chemicals (PFCs) are a class of compounds widely used in commercial and industrial applications because of properties that include water, soil, and oil repellency. A variety of PFC homologues have been utilized that are differentiated by the number of carbons in the structure and by either a carboxylate, sulfonate, or phosphonate moiety attached to one end. Perfluorooctane sulfonate (PFOS) and perfluorooctanoate (PFOA), each containing eight carbons, tend to be the most effective surfactants and are the most common homologues found in environmental samples (Lau et al. 2007). Perfluorinated chemicals are slow to decompose, persisting in the environment and biomagnifying through the food chain (Giesy and Kannan 2002). Contamination of remote regions occurs by atmospheric and oceanic transport (Prevedouros et al. 2005, Young et al. 2007). However, occurrences of significant localized contamination have been the result of chemical spills (Moody et al. 2002), emissions and discharges from PFC-production facilities (Giesy and Newsted 2001, Davis et al. 2007), and surface runoff from fields amended with waste-treatment sludge (Skutlarek et al. 2006). Manufacturing facilities producing steel, electronics, paper, and carpet discharge effluents containing relatively high concentrations of various PFC homologues (Clara et al. 2008), and waste-treatment facilities can be sources of PFCs to surface waters (Becker et al. 2008).

The carpet industry, which is prevalent in northern Georgia, is suspected of producing wastewaters containing PFCs and PFC-precursor chemicals. A significant use of PFCs is in the manufacture of carpet-protection products (Hekster et al. 2003). Effluents from multiple carpet manufacturers in the Dalton area are processed through a municipal wastewater-treatment plant which, in turn, sprays the treated effluent onto a land-application site (LAS) bordering the Conasauga River. A recent analysis of surface-water samples collected immediately above and below the site revealed high concentrations of selected PFCs downstream of the LAS (Konwick et al. 2008). Analyses of sediment samples collected in 2006 at their downstream site (CR4) indicated significant PFC contamination as well (1 to 10 ng/g,dw). However like many surveys of PFCs in water and sediment samples, concentrations of relatively few homologues in addition to PFOS and PFOA were determined. The present study examined concentrations of fifteen PFC homologues in surface waters and sediments upstream and downstream of the LAS and demonstrated their bioaccumulation from the collected sediments into the freshwater oligochaete, *Lumbriculus variegatus*.

METHODS

Surface waters and sediments from the Conasauga, Oostanaula and Coosa Rivers were sampled during the summer of 2008. Eight sampling locations were selected at road crossings based on river confluences, major tributaries, potential sources of PFCs, and access (Figure 1). Three replicate sediment and surface water samples were collected at each site.

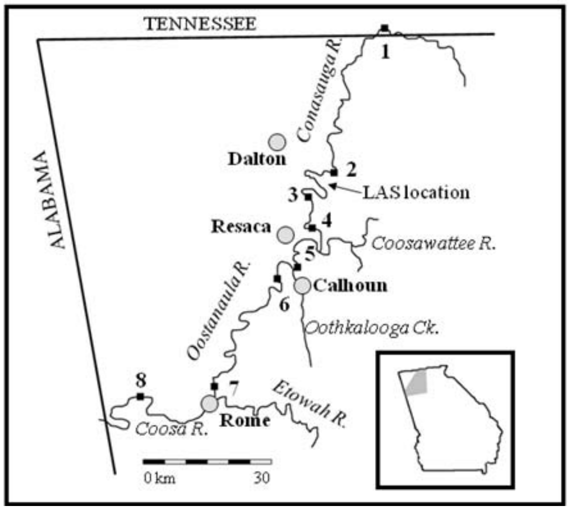

Figure 1. Collection sites for surface waters and sediments in the Coosa River watershed.

Bioaccumulation from sediment exposures was determined using established procedures (USEPA 2000) with *Lumbriculus variegatus* obtained from in-house cultures. Sediment samples used for PFC analyses were moist-sieved through a 2-mm SS sieve and extracted four times with 60:40 acetonitrile:water (v:v). Organic carbon was determined in non-sieved sediments using a Leco CNS-2000 Analyzer. Oligochaete tissue was extracted with an ion-pairing procedure using the method of Henderson et al. (2007). Ultra-high-pressure liquid chromatography with tandem mass-spectrometry was employed to determine PFC concentrations in the prepared water, sediment, and tissue samples. Along with PFOA, PFCAs measured were perfluorohexanoate (PFHxA), perfluorheptanoate (PFHpA), perfluorononanoate (PFNA), perfluorodecanoate (PFDA), perfluoroundecanoate (PFUnDA), perfluorododecanoate (PFDoDA), perfluorotridecanoate (PFTrDA), and perfluorotetradecanoate (PFTeDA). The perfluorosulfonic acids (PFSAs) determined included PFOS, perflourobutane sulfonate (PFBS), perfluorohexane sulfonate (PFHxS), and perfluoroheptane sulfonate (PFHpS). In addition, concentrations of PFCAs, perfluorobutanoate (PFBA) and perfluoropentanoate (PFPA) were determined in water samples. Method detection limits (MDLs) for individual PFC homologues were low ranging from 0.005 to 0.02 ng/g for surface waters, 0.023 to 0.297 ng/g (dw) for sediments, and 0.23 to 0.174 (wet weight, ww) for tissues. Recoveries of internal standards for sediments (71 to 92%) and tissues (65 to 124%) were satisfactory. Use of trade, product, or firm names does not imply endorsement by the U.S. Government.

RESULTS AND DISCUSSION

Several surface-water samples contained total PFC concentrations over 1µg/L and a comparison among sites demonstrated contamination of the Conasauga River downstream of the LAS with subsequent additions and dilutions further downstream (Figure 2). Concentrations increased significantly downstream of the LAS at site 3 and then increased by about10% at site 4 after receiving inputs from several tributaries and wastewater from the carpet-backing plant. Downstream of the confluence of the Conasauga and Coosawattee Rivers (Oostanaula River), concentrations were halved by dilution indicating low PFC contamination emanating from the Coosawattee watershed. Total PFCA concentrations in surface waters were greater than PFSA concentrations at each site, but individually PFOS and PFBS were generally present in the greatest concentrations followed by PFOA. The predominant PFCAs were homologues with 4 to 8 carbons. Despite the increasing discharge of the river, concentrations of PFCAs and PFBS increased and PFOS concentrations remained steady as the river passed the cities of Resaca and Calhoun indicating sources of contamination from this area as well. Most of the homologue concentrations decreased slightly at site 7 after passing through approximately 50 km of agricultural and forest land; although PFOS increased slightly, possibly due to the degradation of precursor chemicals such as sulfonamides that are also used in the carpet industry (Giesy et al. 2010). Sulfonamides were not measured in the present study, but have been a substantial component in effluents processed by the Dalton waste-treatment facility (www.epa.gov/region4/water/PFCdaltonindex.html).

Samples collected at site 8 increased in concentrations of PFBA, PFPA, PFHxA. This site was located 2 km below a paper-manufacturing plant which may discharge wastewaters containing these homologues (Clara et al. 2008). Levels of the other PFCs decreased slightly at site 8, but not to the extent expected by the dilution provided by the Etowah River, which contributes roughly 60% of the Coosa-River discharge. Additional inputs of PFCs could be originating in the Etowah watershed or from sources in the Rome area between sites 7 and 8.

Concentrations of longer-chain PFCAs (PFUnDA, PFDoDA, PFTrDA, PFTeDA) were below detection levels in all water samples. Concentrations of fluorotelomer alcohols, suspected precursors of PFCAs, were also low in these waters. The 6:2 telomer alcohol was detected above the limit of quantitation in samples from two sites (site 2 at 86 ng/L and site 7 at 35 ng/L), but the 8:2 telomer alcohol was not detected above the MDL in any samples.

PFC concentrations in samples from sites 3 – 8 were relatively high compared to most PFC-contaminated waters. The U.S. Environmental Protection Agency recently established provisional health (PHAs) for PFOS and PFOA in drinking waters of 200 ng/L and 400 ng/L, respectively (USEPA 2009). Concentrations of PFOS in surface waters at sites 3 and 4 exceeded this limit and one sample from site 7 contained 199 ng/L, but PFOA concentrations were well below the 400 ng/L PHA. Concentrations of PFCs in surface waters of the Conasauga River may be decreasing. Samples collected for analyses by Konwick et al. (2008) and on behalf of the United Steel Workers union (http://media.timesfreepress.com/docs/2008/03/3_18%20_DALTON%20EPA%20LETTER-%20Final.pdf) generally contained greater concentrations of PFCs than samples collected for this study. However, surface waters sampled at that site in 2009 for Dalton Utilities contained similar PFC concentrations to those reported here (www.epa.gov/region4/water/PFCdaltonindex.html).

Sediments were generally sandy (21 – 93% sand, 7 – 61% silt, 1 – 13% clay) with proportions of organic-C ranging from 0.4 to 2.6%. Perfluorinated carboxylic acids were present in greater mass than PFSAs, but PFOS was the predominant compound with concentrations 1.5 to 4 times that of the highest occurring PFCA (Figure 2). Six PFCs (PFOA, PFNA, PFDA, PFTrDA, PFTeDA, PFBS) were detected at low concentrations in sediments from site 1 with increasing concentrations at site 2. Reflecting the surface-water concentrations, a significant increase in sediment PFC levels occurred at sites 3 and 4. However, the profile of PFCAs in the sediments was dominated by homologues with ten or more carbons despite their low to non-detectable concentrations in the water samples. This suggests that the long-chain PFCAs sorb out of the dissolved phase quickly and are transported downstream as part of the sediment load.

Concentrations of most homologues in sediments from site 4 were significantly greater than those measured in sediments from site 1 or sites 1 and 2, and five homologues (PFDA, PFUnDA, PFDoDA, PFTrDA, PFTeDA) were present in sediments from site 3 at significantly greater concentrations than upstream of the LAS as well (Figure 2). As with the surface waters, PFC concentrations in sediments decreased below the confluence of the Conasauga and Coosawattee Rivers. They subsequently increased downstream of Resaca and Calhoun and then increased again prior to reaching the city of Rome. Sediment concentrations of PFOS increased more than other PFCs (>30%) in this lower, agricultural segment possibly due to the degradation of PFOS precursor chemicals which are components of effluents permitted for discharge upstream (www.epa.gov/region4/water/PFCdaltonindex.html). Sediment PFC concentrations were substantially lower at site 8 below Rome and the confluence with the Etowah River. This implies that most of the long-chained PFCAs were emanating from the Conasauga and Oostanaula Rivers. Concentrations of PFHxA and PFHxS were above the MDL in sediments from two sites (3 and 4); one sample (site 4) contained PFHpS above its MDL; no sediments contained concentrations of the 8:2 and 10:2 fluorotelomer acids above the MDLs.

PFCA concentrations in sediments from all eight sites indicated the same general pattern among homologues where PFDA, PFUnDA, and PFDoDA were most prominent followed by PFTrDA and PFTeDA (Figure 2). This pattern is similar to those determined in sludge and aged compost produced at the Dalton waste-treatment facility when the more soluble homologues are excluded. Sludge and compost samples were dominated by PFDA and PFOA followed by PFBS, PFUnDA, PFDoDA and PFNA with significant amounts of PFCAs with 7 or less carbons and PFTrDA (www.epa.gov/region4/water/PFCdaltonindex.html).

However, PFBS and PFCAs with fewer than 10 carbons appear to have washed out of the sediments leaving PFOS and PFCAs with 10 or more carbons. This observation is supported by the presence of only homologues with fewer than 10 carbons in the surface waters and the corresponding trend observed in the previously-mentioned PFC-water-stability study. Sediment concentrations of PFTeDA appear to be far greater than expected based on sludge and compost concentrations possibly indicating a higher propensity to accumulate in sediments.

Concentrations of PFCs measured in sediments from these sites were relatively high. Sediment concentrations of long-chain PFCAs and PFOS were generally much greater (sites 3 and 4) or equal (sites 6, 7, and 8) to the most contaminated samples in surveys of PFCs in sediments from California, Maryland, and Oregon (Higgins et al. 2005) and Japan (Nakata et al. 2006, Senthilkumar et al. 2007), and demonstrate the relative severity of this contamination and the extent of affected watershed. However, current levels of PFC contamination in these sediments may be decreasing. Sediments collected from the same locations at site 3 for a pilot survey (November,

2006) contained 5 to 9 times the concentrations of PFHxA, PFHpA, PFOA, PFNA, and PFDA than measured in sediments collected 18 months later for the present study. Temporal variability in PFC inputs and sediment movement/washing during flood events could also be responsible for these differences. A more extensive sampling design is required to determine the long-term trends of PFC contamination in this system.

As with the sediments, PFC concentrations in tissues were significantly greater ($\alpha \leq 0.05$) in samples representing sites below the LAS with the greatest concentrations (total PFCs > 1000 ng/g) observed in samples from site 4 (Figure 2). With the exception of samples from site 1, PFOS was accumulated to the greatest concentration in the exposed oligochaetes followed by long-chain PFCAs (PFDA, PFUnDA, PFDoDA, PFTrDA, PFTeDA). Sediments from all sites were contaminated to some degree with PFCs. Homologues that were below detection limits in sediments collected from sites 1 and 2 (PFHpA, PFBS, PFHxS, PFHpS, PFOS) were accumulated by the oligochaetes to concentrations above detection limits, confirming their presence in the sediments. In addition, measurable amounts PFHxS and PFHpS were found in all tissue samples despite non-detectable concentrations in most sediments and surface waters. Even though production and use of PFOS was substantially reduced in the United States after 2002 (Lau et al. 2007), it was the dominant PFC measured in surface waters, sediments, and tissues sampled for this study and continues to pose a potential risk to biota in the watershed.

Accumulation of PFCs at the low trophic level represented by aquatic oligochaetes appears to be directly related to sediment PFC concentrations. Patterns among PFC concentrations were virtually the same for sediments and tissues representing the contaminated sites (Figure 2), and, with the exception of PFBS, positive correlations between sediment and tissue concentrations were highly significant ($\alpha<0.01$). This agrees with previous research using sediments containing PFCs at much lower levels where the rank of homologue concentrations in sediments and *L. variegatus* tissues were nearly the same (Higgins et al. 2007). Contaminated sediments were also suggested as the principal source of PFCs to a Lake-Ontario food web based on concentrations within selected trophic levels (Martin et al. 2004).

CONCLUSIONS

The LAS associated with the Dalton municipal waste treatment plant has contributed significant amounts of PFCs to the Conasauga River and there appear to be additional sources of PFCs downstream. Concentrations of PFCs measured in samples from the Conasauga and Oostanaula Rivers were relatively high compared to other contaminated sites reported in the literature. Low to non-detectable concentrations of fluorotelomer alcohols were observed in these samples indicating that the PFCAs present were components of wastewater discharges. Concentrations of PFCs appear to be decreasing compared to past surveys, but periodic monitoring of surface waters and sediments from these systems will be required to determine long-term trends.

ACKNOWLEDGEMENTS

The United States Geological Survey and Environmental Protection Agency, through its Office of Research and Development, funded the research described here. The work has been subjected to the Agencies' administrative reviews and approved for publication.

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