EXTRACTABLE ORGANIC FLUORINE AND PERFLUORINATED COMPOUNDS IN CHINESE BLOOD SAMPLES

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Abstract

The ubiquitous occurrence of perfluorinated compounds (PFCs) in environmental samples has drawn much attention. Pefluorooctane sulfonate (PFOS) and perfluorooctanoate (PFOA) are the dominant PFCs that have been found in environmental samples, but it is also known that these compounds only account for a portion of the PFCs that are present in the environmental and biological matrices. A refined combustion ion chromatography for fluorine (CIC-F) that allows for determination of concentrations of total fluorine (TF) as little as 6 ng/mL in blood samples was developed. In this study, the concentrations of both extractable organic fluorine (EOF), and individual PFCs in human whole blood from four cities (Shenyang, Beijing, Guizhou, and Jintan) in China were determined. Whole blood from individuals living in Shenyang had the greatest total PFC concentration, whereas whole blood from people in Jintan had the least total PFC concentration. However, the samples from Jintan and Shenyang had similar fluorine concentrations, whereas those from Beijing and Guizhou had comparatively lesser fluorine concentrations in the EOF. Although the samples from Jintan had the least total PFC concentrations, approximately 60% of the total EOF was comprised of unknown compounds. Thus, the issue is what those compounds might be and if they pose a risk to humans.

Introduction

The ubiquitous occurrence of perfluorinated compounds (PFCs) in the environmental has drawn much attention. Recent studies have shown that more than eighteen kinds of perfluorinated compounds are found in seawater and drinking water, animal tissues, and human samples. Perfluorooctane sulfonate (PFOS) and perfluorooctanoate (PFOA) are the dominant PFCs, but it is also known that these compounds only account for a portion of the total mass of PFCs present in abiotic and biotic environmental matrices. Other fluorochemicals, such as fluorotelomer alcohols and shorter- and longer-chain perfluorocarboxylates (PFCAs) are now included in monitoring surveys and risk evaluations. With the help of advanced analytical instrumentation and greater availability of authentic and isotopically-labeled analytical standards, increasing numbers of PFCs can now be determined, but

despite these advances it is difficult, if not impossible, to identify and quantify all of the individual PFCs that compose the extractable organic fluorine (EOF) of samples.

A refined method that combines combustion followed by ion chromatography has been developed to allow quantification of the total organic fluorine (OF) that is applicable to trace analyses at the parts per trillion level in water[1] and the parts per billion level in blood samples.[2] Previous studies have found that PFCs can account for >80% of the EOF fraction in human blood samples from the general population, but that in bulk human blood samples, inorganic fluoride (IF) and non-extractable organofluorine compounds accounted for >70% of the total fluorine (TF). Measurement of TF, EOF, and IF allows a better understanding of the magnitude of PFC contamination, and can help to identify other unidentified fluorochemicals and in conjunction with other methods be used to determine potential sources.

Concentrations of PFCs in human whole blood have been previously reported for nine cities in China[3]. PFOS concentrations in whole blood of people from these cities were greater than those in other countries. Similar profiles of relative concentrations of PFC among the populations of the cities suggested similar sources and pathways of exposure. However, concentrations of EOF and other PFCs were still unknown. Here we report the results of a study in which concentrations of both EOF and individual PFCs in human whole blood from four Chinese cities (Shenyang, Beijing, Guizhou, and Jintan) were measured and the results were compared with those of the previous study[3].

Materials and Methods

Samples of human whole blood were collected from volunteer donors at local universities or hospitals in China during 2004. The whole blood samples were stored in polypropylene (PP) containers or vacutainers® at -20 ˚C until analysis. Individual PFCs were extracted using an ion-pairing method, and were then reduced to 1.0 mL. To quantify EOF, 0.5 mL of the sample extract was subjected to combustion ion chromatography (CIC), and 0.5mL of the sample extract underwent envicarb® and solid phase extraction (SPE) cleanup. Concentrations of perfluorinated sulfonates (PFOS, perfluorohexane sulfonate – PFHxS), perfluorooctanesulfonamide (PFOSA), and perfluorinated carboxylates (Perfluorohexanoic acid – PFHxA, perfluoroheptanoic acid – PFHpA, PFOA, perfluorononanoic acid – PFNA, perfluorodecanoic acid – PFDA, perfluoroundecanoic acid – PFUnDA, and perfluorododecanoic acid – PFDoDA) as well as other PFCs were determined by using HPLC-MS/MS. Individual PFCs were separated by use of an Agilent HP1100 liquid chromatograph (Agilent, Palo Alto, CA) that was interfaced with a Micromass Quattro Ultima Pt tandem mass spectrometer (Waters Corp., Milford, MA) and operated in an electro-spray negative mode. A 10-µL of the extract was injected onto a Keystone Betasil C18 column (2.1 mm i.d. x 50 mm length, 5 µm, 100Å pore size, endcapped) with 2 mM of ammonium acetate and methanol as the mobile phases. The details of the LC-MS/MS are reported elsewhere[4]. EOF concentrations were determined using CIC with a

method that involves the modification of the traditional CIC by the combination of an automated combustion unit (AQF-100 (type AIST), Dia Instruments Co. Ltd. Japan) and an ion chromatography system (ICS-000 (type AIST), Dionex Co. Ltd., Japan). Sample extracts in silicon boats were placed in a furnace at 900–1000°C. Organically-bound as well as inorganic fluoride were combusted and converted into hydrogen fluoride (HF), which was then absorbed into sodium hydroxide (0.2 mmol/L). Concentrations of F^- in solutions were determined by ion chromatography. Sodium fluoride (99% purity; Wako Pure Chemical Industries, Tokyo, Japan) was used as the standard for quantification. More detailed descriptions of the analytical procedures for ion chromatography are described elsewhere[1,2]. All of the solutions were prepared in Milli-Q® water with a total fluoride concentration of <0.025 μg/L.

Results and Discussion

Concentrations of PFOS, PFHxS, PFOSA, PFDoDA, PFUnDA, PFDA, PFNA, PFOA, PFHpA, and PFHxA in procedural blanks were determined. Concentrations of PFBS and PFHxA, FTUCA, and FTCA concentrations were all less than the limit of quantification (LOQ). The blank and recovery test results and the PFC concentrations are summarized (Table 1).

The results are comparable with those previously reported,[3] although some of the concentrations are different. These differences may be due to the fact that the samples that were measured in this study were from different individuals from those measured in the previous study, or because the sample size in this study was small. Modified clean-up procedures (i.e., Envicarb® and SPE) were applied to lessen the background interference so that the PFHpA could be identified and quantified.

Concentrations of total PFC were greatest in whole blood from individuals in Shenyang, while the least concentrations of total PFCs were observed in whole blood from Jintan. However, whole blood from Jintan and Shenyang had similar total fluorine concentrations. Total PFCs concentrations in whole blood from Beijing and Guizhou had lesser EOF (Table 2). Although whole blood from Jintan contained the least total concentration PFC, this location also had the greatest mass of EOF. The ratio of the percentage of fluorine from known individual PFCs to the percentage of fluorine from extractable organic fluorine showed that known individual PFCs accounted for most of the extractable organic fluorine (70-80%) in the samples from Beijing, Shenyang and Guizhou, which is similar to the finding of ~80% in another study[2]. In contrast, known individual PFCs only accounted for 40% in the samples from Jintan. The remaining portion of the EOF could be comprised of fluoro-telomer alcohols, shorter chain (C2-3) or longer chain (C14-18) PFCs, and some unknown organic fluorine. The extraction and clean-up methods applied in this study were not suitable to efficiently remove some of these compounds. The PFC/EOF ratios in livers of dolphin from Hong Kong (10-15%) were less, which suggests that other organofluorines might bioaccumulate to a different degree in various biological samples[5].

Reference

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Table 1. Concentrations of individual PFCs in Chinese blood samples (ng/mL)

City	N		PFOS	PFHxS	PFOSA	PFDoDA	PFUnDA	PFDA	PFNA	PFOA	PFHpA	PFHxA	Total PFC
Blank	8		<0.068	<0.01	<0.02	<0.01	<0.05	<0.05	<0.05	<0.05	<0.01	<0.01	
Recovery	8	**mean**	97	92	65	93	90	93	86	100	100	89	
S.D.		**S.D.**	4	8	17	20	22	16	19	13	20	14	
Matrix-spike recovery	6	**mean**	87	97	57	101	97	100	98	89	110	117	
S.D.		**S.D.**	1	5	10	7	3	2	4	5	11	6	
Beijing	5	**mean**	13.8	0.507	0.601		0.141	0.116	0.199			0	15.4
		S.D.	6.09	0.266	0.538		0.0993	0.107	0.125			0	1.89
		Range	4.04 - 21.2	0.13 - 0.857	0.136 - 1.46	<0.01 - 0.0413	0.0597 - 0.356	0.0510 - 0.380	0.0859 - 0.499	<0.05 - 0.376	<0.01 - 0.0204	<0.01 - 0.0295	
Guizhou	5	**mean**	20.7	0.958	0.657				0.231	0.325		0.0513	22.9
		S.D.	14.6	0.627	0.431				0.157	0.264		0.0558	4.57
		Range	7.78 - 46.2	0.383 - 1.94	0.335 - 1.41	<0.01 - 0.085	<0.01 - 0.122	<0.05 - 0.077	<0.1 - 0.616	<0.05 - 0.870	<0.01 - 0.0906	<0.01 - 0.18	
Jintan	5	**mean**	5.04	0.18	0.351	0.0499	0.504	0.601	0.766	1.39	0.0326	0.0748	8.99
		S.D.	3.30	0.106	0.201	0.0149	0.186	0.288	0.388	0.542	0.0165	0.0338	0.99
		Range	1.54 - 12.8	0.112 - 0.471	0.0994 - 0.565	0.0276 - 0.0704	0.301 - 0.811	0.221 - 1.08	0.258 - 1.34	0.532 - 2.19	0.0202 - 0.0722	0.026 - 0.112	
Shenyang	5	**mean**	56.3	1.87	1.65	0.0186	0.124		0.303			0	60.3
		S.D.	17.7	0.52	0.809	0.0106	0.104		0.0480			0	5.55
		Range	35.7 - 83.1	1.20 - 2.52	0.691 - 3.10	<0.01 - 0.042	<0.01 - 0.319	<0.05 - 0.102	0.247 - 0.412	<0.05 - 0.4226	<0.01 - 0.0308	<0.01 - 0.067	

Table 2. Fluorine concentration (ng-F/mL) and percentage of F contributed by identified PFCs and extractable organic fluorine (EOF; %)

		Fluorine concentration (ng-F/mL)		
		Known individual PFC	Extractable organic fluorine (EOF) after ion pairing extraction	PFC-F/EOF-F (%)
Beijing	Mean	10.1	18	69
n=5	S.D.	4.41	3	11
Shenyang	Mean	54.3	60	79
n=4	S.D.	24.	33	20
Jintan	Mean	11.9	47	36
n=4	S.D.	4.83	14	12
Guizhou	Mean	20.1	22	88
n=5	S.D.	10.2	10	25