OPTIMIZATION OF DETERMINING METHOD FOR PFOA AND PFOS IN SOIL AND SEDIMENT

Wenlong Yang, Jinling Liu, Bing Du, Shuangxin Shi, Liang Dong, Yeru Huang *

State Environmental Protection Key Laboratory of Dioxin Pollution Control, National Research Center for Environmental Analysis and Measurement, Beijing, China 100029, ywlpops@163.com

Introduction

Because of the excellent stability, surface activity and hydrophobic oil and water, perfluoroalkyl and polyfluoroalkyl substances (PFASs) were kinds of important industrial and commercial raw materials, which was widely used in electroplating, fire protection, coating, carpet, leather, clothing and packaging and other fields. With the increasing application of PFASs, they have been widely detected in environmental media such as atmosphere[1], water[2], soil[3] and sediment[4]. Studies have shown that PFASs have hepatotoxicity, developmental toxicity, immunotoxicity, endocrine disruption and potential carcinogenicity in animals. In 2009, Perfluorooctane sulfonic acid (PFOS) and its salts were listed in Annex B of the Stockholm Convention[5]. In 2019, Perfluorooctanoic acid (PFOA) and its salts are officially listed in Appendix A of the Convention[6,7]. With the continuous production, usage and resource disposal of industrial products containing PFOA and PFOS, the pollution of PFOA and PFOS in environmental media in China can not be ignored. Soil and sediment in particular were important sink of persistent organic pollutants[8,9]. Therefore, it is of great significance to establish accurate monitoring methods for PFOA and PFOS in soil and sediment, and to understand timely the pollution levels of PFOA and PFOS in the environment for the implementation of the Convention, ecological environmental protection and people's health in China.

The extraction methods of PFOA and PFOS in soil and sediment mainly include pressure fluid method, ultrasound-assisted method and mechanical oscillation method[10], and the purification method is mainly solid phase extraction column purification. At present, the most commonly used PFOA and PFOS detection instrument is high performance liquid chromatography triple quadrupole mass spectrometry (HPLC-ESI-MS/MS). The application of high performance liquid chromatography (HPLC) avoids the steps of derivatization, and has good separation degree. Triple quadrupole mass spectrometry (MS/MS) technology can effectively improve the signal-to-noise ratio, and has the advantages of good repeatability and short analysis time. Some scholars have also compared the effects of ion trap mass spectrometry, time of flight and triple quadrupole mass spectrometry on the analysis of trace PFASs. The results showed that compared with triple quadrupole mass spectrometry, ion trap mass spectrometry has a lower sensitivity, but is suitable for the qualitative and structural analysis of PFASs isomers. Although time-of-flight mass spectrometry has high selectivity and sensitivity, its linear range is narrow.

Soil and sediment samples are diverse with complex substrates and long analysis cycle. Furthermore, sample stability, extraction efficiency, purification effect and matrix effect all affect the analysis accuracy. Herein, the conditions of sample preparation, extraction and purification was optimized, and the preservation time range of samples and the effect of matrix was evaluated. An analytical method with high extraction efficiency, good purification effect, accurate qualitative and quantitative analysis, strong applicability and convenient operation was established.

Materials and methods

Chemicals and standards

methanol was HPLC-grade reagents, It was obtained from Honeywell (Morristown, NJ).Acetic acid and ammonium acetate and Ammonium hydroxide were Reagent grade, all were bought from Sigma-Aldrich (St. Louis, Missouri, USA), Ultrapure water was produced by a Milli-Q system (Millipore, USA) in the laboratory. WAX SPE columns (500 mg, 6 mL) were bought from waters Inc (Massachusetts, USA). Native PFOA and PFOS and ^{13}C-labeled surrogate standard solutions ($^{13}C_4$-PFOA, $^{13}C_4$-PFOS, $^{13}C_2$-PFOA) were purchased from Wellington Laboratories (Guelph, Ontario, Canada).

Sample preparation

About 2 g samples was weighed and transferred into a 50 mL centrifuge tube, 10 ng $^{13}C_4$-PFOA and $^{13}C_4$-PFOS and 10 mL methanol water mixed solution (v/v, 1/1) was added, and was swirled for mixing for 1 min. The centrifuge was centrifuged at 4000 rpm for 10 min after shaking of 300 rpm for 2 h. The supernatant was transfer to another centrifuge tube. The above procedure was repeated twice, and the secondary extraction solution was combined. After filtration through a 0.8 μm filter, 80 mL water was added, and pH was adjusted to 6-7 with acetic acid. The SPE column was activated sequentially with 6 mL ammonia and methanol (v/v, 2/98), 6 mL methanol and 6 mL water. During the activation process, the packing in the column should not be exposed to air. The extraction liquid was passed through the SPE column at a flow rate of 3 mL/min to 5 mL/min. After sample

loading, 8 mL ammonium acetate buffer solution (0.025 mol/L, pH=4) was used to elute the solid phase extraction column, and the eluent was discarded. The extraction column was dried with nitrogen purge or vacuum pump of SPE unit for 10 min to remove residual water in the extraction column. An 8 mL methanol elution SPE column was used, and the eluent was discarded. The eluent was collected on a 6 mL ammonia and methanol (v/v, 2/98) elution SPE column. The eluent was concentrated with a nitrogen blower and constant volume of methanol to 1.0 mL. After being filtered by a 0.22 μm filter membrane, 10 ng $^{13}C_2$-PFOA was added. The eluent was mixed and measured.

Determination

High performance liquid chromatography-tandem mass spectrometry (8040, Shimadzu, JP) equipped with an ESI source was used to analyze PFOA and PFOS. Agilent Zorbax Extend C18 (100 mm×3.0 mm i.d.×1.8 μm, Agilent, USA) was used to separate isomers. The mobile phase included methanol and 2 mmol-1 ammonium acetate aqueous solution. The qualitative and quantitative analysis was based on negative ESI mode and multiple reaction monitoring (MRM).

Results and discussion

Sample stability evaluation

The variation trend of PFOA and PFOS concentrations in soil and sediment samples with storage time is shown in Fig. 1a; the variation trend of PFOA and PFOS concentrations in sediment samples with storage time is shown in Fig. 1b; and the variation trend of PFOA and PFOS concentrations in sample extracts with storage time is shown in Fig. 1c. The results showed that the concentrations of PFOA and PFOS in soil and sediment samples ranged from 3.8 μg/kg to 5.9 μg/kg, the recoveries ranged from 76.0% to 118%, and the relative standard deviations ranged from 5.5% to 8.9% within 60 days. The results of PFOA and PFOS in the sample extract ranged from 4.2 μg/kg to 5.7 μg/kg, with the relative standard deviations were from 5.5% to 7.6%. The relative standard deviations of the test results of samples and sample extracts with different preservation time were all less than 30%, and the recoveries of samples were from 70% to 130%, so the PFOA and PFOS in soil and sediment samples and sample extracts were stable within 60 days.

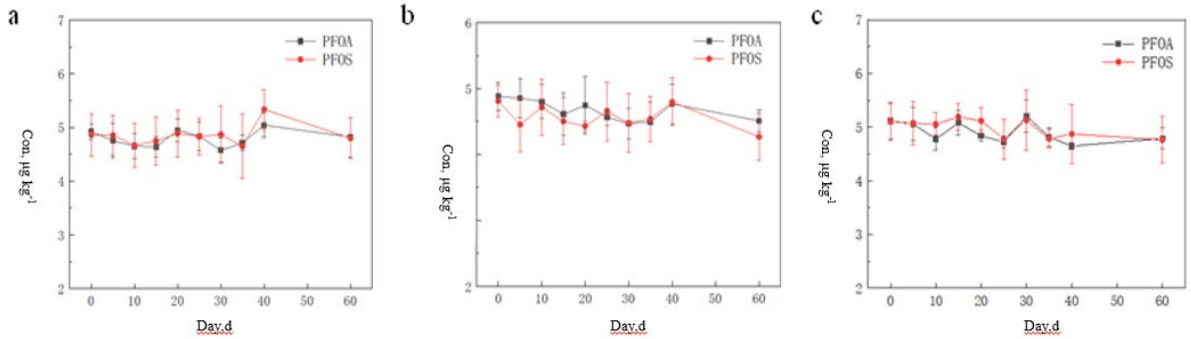

Figure 1. PFOA and PFOS concentrations with storage time. A. Soil samples; B. Sediment samples; C. Sample extract.

Evaluation of sample drying methods

Sample drying results were shown in Table 2. The results showed that the recoveries of the samples were from 91.0% to 97.0% and the relative standard deviations (RSDs) were from 1.9% to 7.9% in PFOA and PFOS. The samples were freeze-dried, and the recoveries of PFOA and PFOS were 101%, and the relative standard deviations were from 5.9% to 7.4%. Under the two drying methods, the test results of the samples meet the requirements of quality control, so the results obtained by using natural drying or freeze-drying to dry the samples are reliable.

Table 2. Sample tests for naturally drying and freeze-drying (concentration unit μg • kg^{-1})

Naturally drying	PFOA	PFOS	Freeze-drying	PFOA	PFOS
Sample 1	4.4	4.9	Sample 1'	5.0	5.7
Sample 2	4.4	5.0	Sample 2'	5.2	5.1
Sample 3	4.6	4.2	Sample 3'	4.5	4.6
Sample 4	4.6	5.2	Sample 4'	5.3	4.8
Sample 5	4.6	5.0	Sample 5'	5.3	5.1
Mean value	4.6	4.9	Mean value	5.1	5.1
Standard deviation	0.1	0.4	Standard deviation	0.3	0.4
Relative standard deviation, %	1.9	7.9	Relative standard deviation, %	5.9	7.4
Recovery rate	91	97	Recovery rate	101	101

Conditions optimization

As can be seen from Table 3, the relative standard deviations of PFOA and PFOS test results were 6.7% ~ 11.7% when the sample extraction time was 0.5 h, 1 h, 2 h, 12 h and 24 h, and the deviation range of test results met the quality control requirements (30%). As shown in Fig. 2, when the single extraction time was longer than 2 h, the extraction efficiency basically reached 100% after two extractions. Considering the convenience and timeliness of the method, the single extraction time in this study was determined as 2 h, and the extraction times were 2.

Table 3. PFOA and PFOS in samples with different extraction times (concentration unit μg • kg^{-1})

Extraction time	PFOA	PFOS
0.5 h	6.8	9.8
1 h	7.5	10.1
2 h	7.6	12.2
12 h	8.1	12.7
24 h	7.9	10.6
Mean value	7.6	11.1
Standard deviation	0.5	1.3
Relative standard deviation, %	6.7	11.7

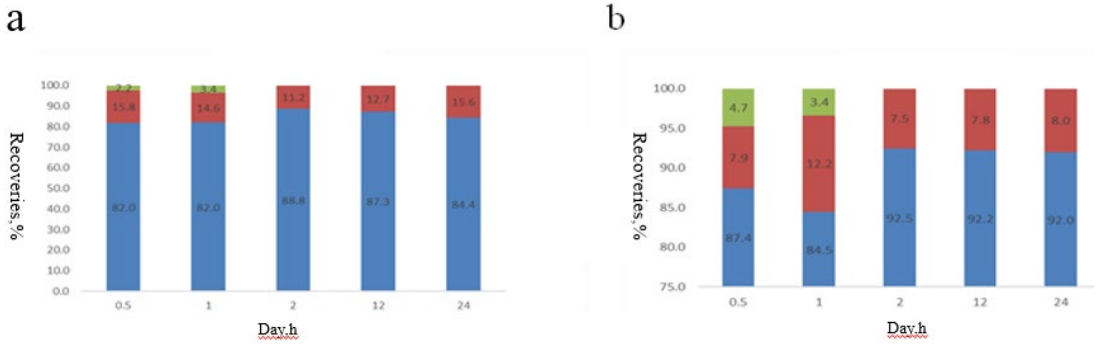

Figure 2. Extraction efficiency of different extraction times. a. PFOA; b. PFOS

Based on the physical properties of PFOA and PFOS, four types of solid phase extraction (SPE) were investigated by this method, including HLB, WAX, PEP and C18, respectively. The results showed that the recoveries of PFOA and PFOS in HLB, WAX, PEP and C18 SPE columns were from 101% to 115%, from 99.0% to 100%, from 90.0% to 110% and from 86.0% to 107%, respectively, which all met the quality control requirements were from 70% to 130%. However, the recovery rates of $^{13}C_4$-PFOA and $^{13}C4$-PFOS in the PEP and C18 SPE columns were from 55.0% to 101% and from 38.0% to 62.0%, respectively. Therefore, PEP and C18 were not recommended as purification columns in this method. The recoveries of $^{13}C4$-PFOA and $^{13}C4$-PFOS in HLB and WAX SPE columns were from 95.0% to 96.0% and from 94.0% to 98.0%, respectively. The WAX extraction column is based on weak anion exchange mechanism, and there were more methanol elution and purification steps in the elution process than HLB extraction column, so the impurity removal effect is better. Therefore, the WAX extraction column was selected as the enrichment and purification column.

Matrix effect evaluation

Soil organic matter content is one of the important factors affecting soil organic matter adsorption capacity. In this method, PFOA and PFOS samples were selected from soils with organic matter content ranging from 0 g/kg to 75.0 g/kg. The variation trend of PFOA and PFOS contents in soil with organic matter content is shown in Figure 3. The results showed that the organic matter content were from 0 to 75.0 g/kg, the concentration of PFOA and PFOS were from 4.5 to 5.9 μg/kg, the recoveries were in the range were from 90.0% to 118%, and the relative standard deviations were from 4.9 to 7.5%. Therefore, the results obtained are reliable when the soil organic matter is less than 75.0 g/kg.

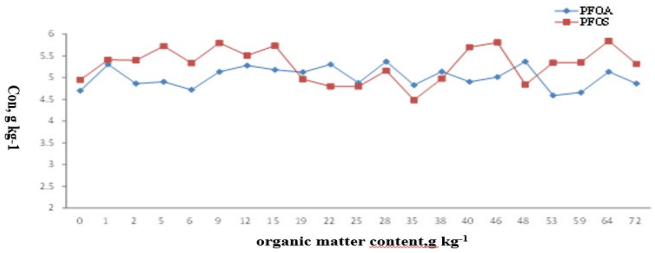

Figure 3. The variation trend of PFOA and PFOS contents in soil with organic matter content

The laboratory selected six different soil types for matrix marker test, and the soil samples covered the southern and northern parts of China and the Central Plains. Guangdong Xuwen basalt laterite respectively (1), shanxi Luochuan loess (2), Inner Mongolia Hanggin HouQi saline soils (3), Jilin Changchun teck whye county chernozem (4), Dalian in Liaoning province king county big arc mountain brown loam (5) and son well bay in hunan changsha red loam (6), six types of soil PFOA and PFOS matrix and. The recoveries of PFOA and PFOS in the six soil types were from 78.3% to 115%, and the recoveries of $^{13}C_4$-PFOA and $^{13}C_4$-PFOS were from 63.0% to 107%, which all met the quality control requirements. Therefore, this method was suitable for the detection of PFOA and PFOS in most soil types in China.

Method accuracy evaluation

The PFOA and PFOS samples in sediments provided by the United Nations Environment Programme were tested by this method when Participating in the Second UNEP Interlaboratory Assessment on the Stockholm Convention POPs, organized by the United Nations Environment Programme, and the test results were satisfactory.

Method characteristic parameter verification

Detection limit and determination limit. According to the requirements of the Technical Guidelines for the Preparation and Revision of the Standard for Environmental Monitoring and Analysis Methods (HJ 168-2010), quartz sand was selected as the blank matrix to analyze the blank labeled sample with a labeled concentration of 1.0 μg/kg, and the detection limit of the method was calculated. The detection limits of PFOA method and PFOS method were 0.4 μg/kg and 0.3 μg/kg respectively when the sample volume was 2.0 g and the constant volume volume was 1.0 mL.

Degree of precision. Low, medium and high concentration markup tests of blank matrix were carried out respectively. The relative standard deviations of the test results were from 10.1% to 10.4%, from 6.8% to 10.2% and from 2.3% to 5.9% at 1.0 μg/kg, 5.0 μg/kg and 25.0 μg/kg supplemented concentrations, respectively.

Degree of accuracy. The environmental background soil of a certain place was selected as the actual labeled sample of low concentration soil, and the soil surrounding a production enterprise was selected as the actual labeled sample of high concentration soil. Sediment of a certain sea area was selected as the actual labeled sample of low concentration sediment, and sediment of a river was selected as the actual labeled sample of high concentration sediment. The RSD of the test results were from 7.5% to 10.7% and the recovery rate were from 97.2% to 107% when the standard concentration of soil was 5.00 μg/kg. The RSD of the test results were from 3.0% to 7.8% and the recovery rate were from 106% to 110% when the spiked concentration of the surrounding soil was 25.0 μg/kg. The relative standard deviations (RSDs) of the test results were from 4.4% to 9.0% and the recoveries were from 93.2% to 96.0% when the standard concentration of seabed was 5.00 μg/kg. The RSD of the test results was 4.8% ~ 6.8% and the recoveries were from 97.4% to 97.5% when the standard concentration of the river bottom was 25.0 μg/kg.

In this study, the preparation, extraction and purification conditions of PFOA and PFOS samples in soil and sediments were optimized, and the preservation time range of samples and the effect of matrix effect were evaluated. A high performance liquid chromatography triple quaternary rod mass spectrometry method for PFOA and PFOS samples in soil and sediments was established. PFOA and PFOS samples and extracts from soil and sediments can be stored for no less than 60 days. Free-drying and natural drying methods can be used for sample drying. WAX SPE column has a better purification effect. The detection limit of this method were from 0.3 μg/kg to 0.4 μg/kg, with good precision, high accuracy and good applicability.

References

1.COSTANZA J., ARSHADI M., ABEIOLA L.M., et al. (2019) *Environmental Science & Technology Letters*. 6(8):487-491.
2.So M.K., Miyake Y., Yeung W.Y., et al. (2007) Lam P.K.S. *Chemosphere.* 68(11): 2085-2095.
3.Houtz E.F., Higgins C.P., Field J.A., Sedlak D. L. (2013) *Environmental Science & Technology.* 47(15): 8187-8195.
4.Lam N.H., Cho C.R., Lee J.S., et al. (2014) *Science of The Total Environment*. 491-492: 154-162.
5.Stockholm convention. The New POPs under the Stockholm Convention [EB/OL]. UNEP, 2009. http://chm.pops.int/TheConvention/ThePOPs/TheNewPOPs/tabid/2511/Default.aspx
6.Stockholm convention. POPRC Recommendations for Listing Chemicals [EB/OL]. UNEP, 2017. http://chm.pops.int/Convention/POPsReviewCommittee/Chemicals/tabid/243/Default.aspx.
7.Stockholm convention. Chemicals listed in Annex A [EB/OL]. UNEP, 2019.
8.YAMASHITA N., TANIYASU S., PETRICK G., et al. (2008) *Chemosphere*. 70(7): 1247-1255.
9.SU Chao, LU Yong long, WANG Tieyu, et al. (2018) *Environmental Pollutio.* 235: 235-244.
10.Schroder H. F. (2003) *Journal of Chromatography A*.1020(1):131-151.