APPLICATION OF CAPILLARY ELECTROPHORESIS IN ANALYSIS OF PERFLUORINATED CARBOXYLIC ACIDS

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Introduction

Perfluorinated carboxylic acids (PFCAs) belong to a broader class of compounds named perfluoroalkyl substances (PFAS). The PFAS are of anthropogenic origin and have been extensively monitored in the last decade and detected in the natural environment and organisms of humans and animals [1]. Although these compounds, including perfluorinated surfactants, are known and used on an industrial scale since 1950s, a large increase of interest in these compounds has been caused by finding these compounds in the biota in remote, arctic regions and humans of general, non-occupationally exposed population. . Numerous physiological and toxicological studies has been carried out on perfluorinated compounds. It has been shown in animal studies that perfluorinated surfactants such as *e.g.* perfluoro-*n*-decanoic acid (PFDA) may affect metabolism of lipids. Perfluorinated surfactants have also effects on hormone balance, ion-channels and transmission of intercellular signals due to interaction with lipid cell membranes, and some cancerogenic effects were reported for rats.

Analytical determination of perfluorinated surfactants at the ppb and sub-ppb level in complex environmental samples and biological materials is a difficult analytical task. It requires application of high-performance separation techniques, detection methods with very low detection limits, development of suitable clean-up methods and often additional sample preconcentration. Liquid chromatography with mass spectrometry is most commonly used for determination of these compounds due to its high sensitivity and selectivity, but high-performance liquid-chromatography (HPLC) with other detection techniques such as conductivity or fluorescence with derivatization have also been reported [2].

A very high resolution of capillary electrophoretic (CE) methods and availability of commercial CE instruments with various detectors is a reason for considering the application of CE methods for determination of perfluorinated carboxylic acids. The aim of this work was to develop a capillary zone electrophoretic method for separation of PFCAs with the alkyl chain length from 6 to 12 carbon atoms. The major effort was put into optimization of background electrolyte (BGE) composition, including additives that provided satisfactory separation and detection in the direct and indirect UV mode of PFCAs.

Material and Methods

A Beckman P/ACE MDQ capillary electrophoresis system with a diode array detector (Beckman Instruments, Fullerton, CA, USA) was used. Fused silica capillaries 75-μm i.d. were used in all measurements. All measurements were carried out at 25°C. Spectrophotometric measurements were carried with spectrophotometer model UV-2401 from Shimadzu (Japan).

A poly (methyl methacrylate) column–coupling (CC) type electrophoretic chip was kindly provided by Merck KGaA (Darmstadt, Germany) and they were manufactured by a procedure described earlier with two conductivity detectors. CE chip is equipped with two channels that can be used for independent ITP and capillary electrophoretic separations. The CE measurements with CC electrophoretic chip were carried out with a home-made CE setup . It is composed of two units. One is functioning as an electrolyte and sample management unit and is equipped with peristaltic micro-pumps and membrane driving electrodes. The other is a source of driving current in the chip channels and includes electronics for conductivity detectors. Whole system was run and controlled by the laboratory – written software Micro CE Win on a PC interfaced to both units.

Perfluorohexanoic acid (C6-PFCA) was obtained from Oakwood Product Co. whereas perfluoroheptanoic acid (C7-PFCA), perfluorooctanoic acid (C8-PFCA), perfluorononanoic acid (C9-PFCA), perfluorodecanoic acid (C10-PFCA), perfluoroundecanoic acid (C11-PFCA) and perfluorododecanoic acid (C12-PFCA) were obtained from Aldrich. Barium pentafluorobenzenesulfonate (PFBS), sodium salt of benzenesulfonic acid (BS), 2,6-naphthalenedicarboxylic acid (NDC) 2,4-dinitrobenzoic acid (2,4-DNBA), and 3,5-dinitrobenzoic acid (3,5-DNBA) were also purchased from Aldrich. Disodium hydrogen phosphate used for background electrolyte (BGE) preparation was obtained from P.O.Ch (Gliwice, Poland).

Results and Discussion

Efficient separation of perfluorocarboxylic acids by capillary zone electrophoresis is more difficult to achieve than that reported in the literature for hydrogen-containing carboxylic acids. Replacement of hydrogen by fluorine in a molecule of a long-chain alkyl carboxylic acid changes its properties significantly, making the existing information about separation of carboxylic acids by capillary electrophoresis inadequate. Optimization of the type and concentration of buffer used in BGE, and especially of the type and content of organic modifier in the BGE are crucial for successful separation of PFCAs. A CE method with direct UV detection at 190 nm for separation and quantitation of PFCAs from C6 to C12-PFCA has been developed using a phosphate buffer with 40% isopropanol as BGEwith limits of detection from 2 to 33 μg/ml.

For the indirect UV detection the best results were obtained for BGE containing TRIS, 50% methanol and 2,4-dinitrobenzoic acid, with limits of detection in the range from 0.5 to 2.3 μg/ml for PFCAs from C6 to C12-PFCA. The detection limits achieved are significantly lower than those obtained for CE withthe direct UV detection. Inclusion of additional preconcentration step in sample preparation will allow analysis of analytes at levels observed in the environmental samples. Comparison of methods is shown in Table 1, and example electropherogram obtained in the optimized conditions with indirect UV detection is shown in Fig.1.

Table 1. Comparison of efficiency expressed by theoretical plate number N and limits of detection for CE determination of PFCAs with direct and indirect UV detections in optimized conditions

Analyte	Direct detection *)			Indirect detection		
	N	Limit of detection		N	Limit of detection	
		mg/L	μM		mg/L	μM
C12-PFCA	23 100	33	54	160 600	2.4	4
C11-PFCA	16 800	24	44	152 200	1.7	3
C10-PFCA	18 800	19	37	186 400	1.5	3
C9-PFCA	27 800	16	36	210 000	1.3	3
C8-PFCA	47 900	13	33	196 400	0.8	2
C7-PFCA	74 700	2	7	200 000	0.7	2
C6-PFCA	59 800	2	6	220 000	0.6	2

From general analytical point of view a reliable environmental monitoring of the presence of these compounds still requires solution of several other problems such as development of standard reference materials, development of separation of isomeric species e.g. of PFOA and PFOS and studies of blank problems and matrix effects in complex,real matrices. The developed methods have been applied for monitoring of PFCAs from radiolytic degradation of PFCAs using γ irradiation.

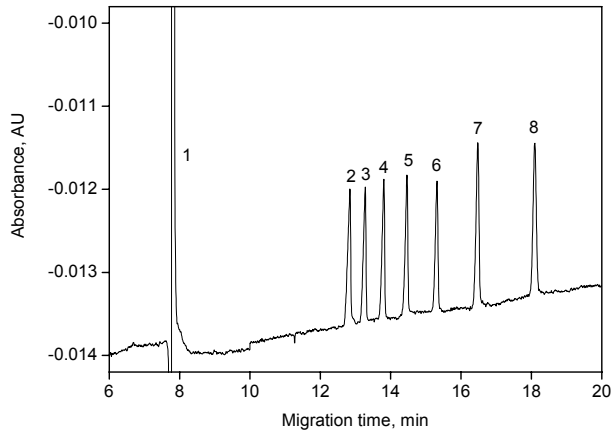

Fig. 1. Electropherogram recorded for a mixture of 0.1 mM PFCAs each in the optimized conditions: BGE: 50 mM Tris, 7 mM 2,4-DNBA, pH 9, 50% methanol. Voltage +25 kV. Hydrostatic injection 3 s at 0.5 psi, capillary 60/50 cm. Signal assignment: 1 – 5 mM DMSO (EOF marker), 2-C12, 3-C11, 4-C10, 5-C9, 6-C8, 7-C7, and 8-C6 PFCA.

Preliminary studies on the development of capillary electrophoresis microfluidic chip made of poly(methyl-methacrylate) with integrated conductivity detection for determination of PFCAs have also been carried out. Although the separation efficiency in this miniaturized device is poorer than that observed in the conventional CE system with fused silica capillary and UV detection, using a borate BGE containing 20% ethanolsatisfactory separation of C6 to C10 PFCAs with detection limits from 0.3 for C6 and 6.7 μM for C10 was obtained. Further optimization of BGE might be helpful in obtaining better separation efficiency, including also separation of C10 or C12 PFCAs. The improvement of detection limits can be expected by the use of the same chip by means of on-line ITP preconcentration.

References

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