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HPLC Separation of Thirty Perfluorinated Compounds and Isomers Using a Pentafluorophenyl Reverse Phase Column

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Key Words

- Hypersil GOLD PFP
- Perfluorinated Compounds (PFCs)
- Selectivity
- Speed

Introduction

Perfluorinated compounds (PFC) are considered as emerging contaminants that attract high interest from the international scientific community. These substances have been synthesized for half a century for their particular physicochemical properties. They are present in a large amount of commercialized products mainly as an anti-sticking or amphoteric material (carpet, clothes, fire-fighting-foams, detergents, etc.). They can be consequently released into the environment during any part of their life cycle, from production to final use. Some toxicological effects have been reported for PFCs (for instance hepatotoxicity and endocrine disruption), but there are still no regulations regarding these compounds in Europe. PFCs can be synthesized by two different processes, namely electrochemical fluoration or telomerisation. The latter process leads to either linear or branched isomers. Thus, to quantify the linear isomers of perfluorooctanesulfonic acid (PFOS) and perfluorooctanoic acid (PFOA), which are the main degradation products of PFC, their efficient separation is required. The aim of this work is to separate thirty perfluorinated compounds, including isomers of PFOA and PFOS, in a short time analysis with the Thermo Scientific Hypersil GOLD PFP column.

Experimental Conditions

PFCs analysis was performed using a binary HPLC pump coupled with a triple quadrupole MS system.

Columns	Part Number
Hypersil™ GOLD 1.9 µm, 100 mm × 2.1 mm	25002-102130
Hypersil GOLD PFP 1.9 µm, 100 mm × 2.1 mm	25402-102130

LC Conditions

Mobile Phase:	A: Ammonium acetate 20 mM B: MeOH	
Gradient:	Time (min)	%B
	0	20
	0.9	20
	1.8	70
	5.3	80
	7.1	80
	8	100
	12	100
Flow Rate:	0.45 mL/min	
Temperature:	55 °C	
Injection Volume:	10 µL	
Sample Concentration:	0.02 ng/µL in A/B (20:80, v/v)	
Detection:	MS (MRM)	

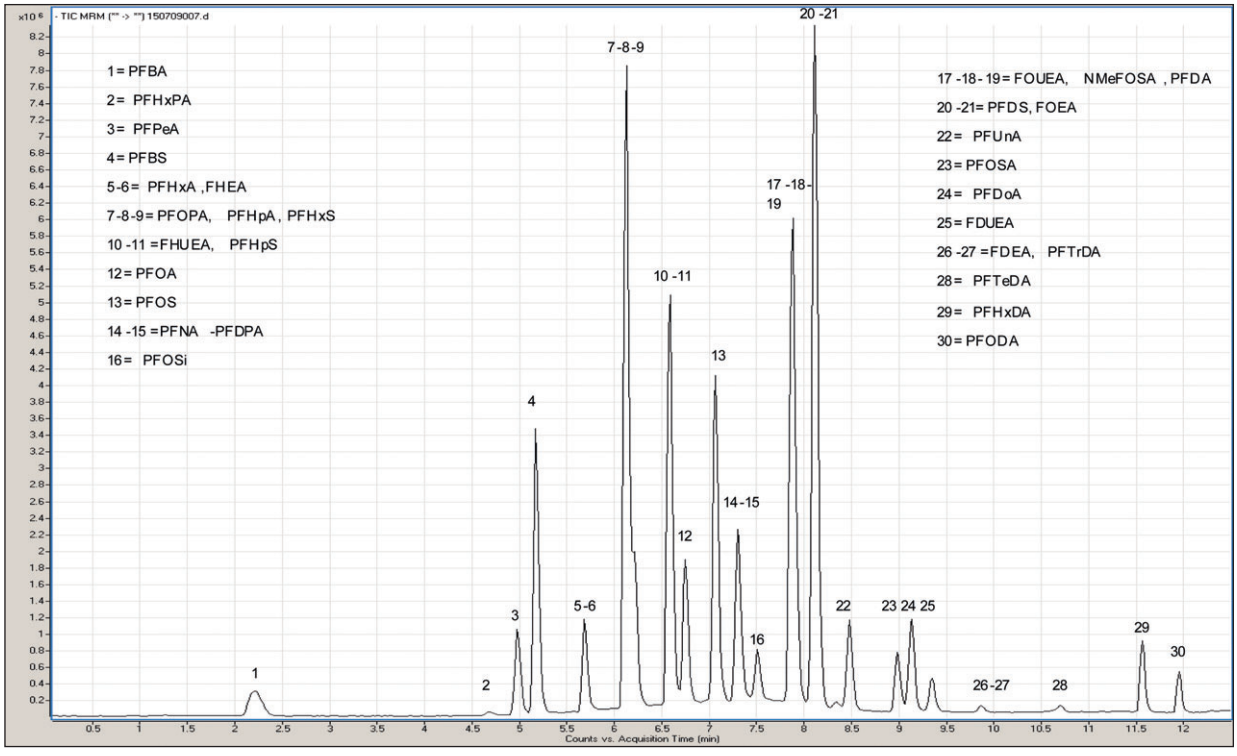

Figure 1: Separation of thirty perfluorinated compounds on the Hypersil GOLD PFP column

Results

Elution using the Hypersil GOLD PFP column was optimized with an isocratic plateau from 5.3 to 7 minutes to help separate the majority of the compounds (Figure 1). Moreover, the addition of ammonium acetate in the mobile phase allows for improved chromatographic separation. The effect of the concentration of ammonium acetate on the separation was investigated in the concentration range 0 to 20 mM. The concentration of 20 mM gave the best chromatographic resolution and was therefore used in the method.

Using the same gradient, a better separation was observed on the pentafluorophenyl phase compared to the alkyl chain phase (Figure 2). The superior selectivity of the perfluorinated phase can be explained by the capability of the pentafluorophenyl phase to provide to dipole-dipole, π- π, charge transfer, and ion-exchange interactions with the analytes.

Furthermore, PFOA isomers were found to be equally separated on both columns (Figure 3).

Conclusions

A method has been developed for the separation and LC-MS/MS analysis of thirty PFCs in twelve minutes. Two phases were compared, a classical reversed-phase chemistry and a perfluorinated phase. On the basis of the same elution gradient, fewer coelutions were observed with the perfluorinated phase. The selectivity of this phase can be explained by the wider range of interactions. However, only small selectivity and sensitivity differences were observed between the separation of isomers on the two columns. Finally, this method will be applied to analyze perfluorinated compounds in fish.

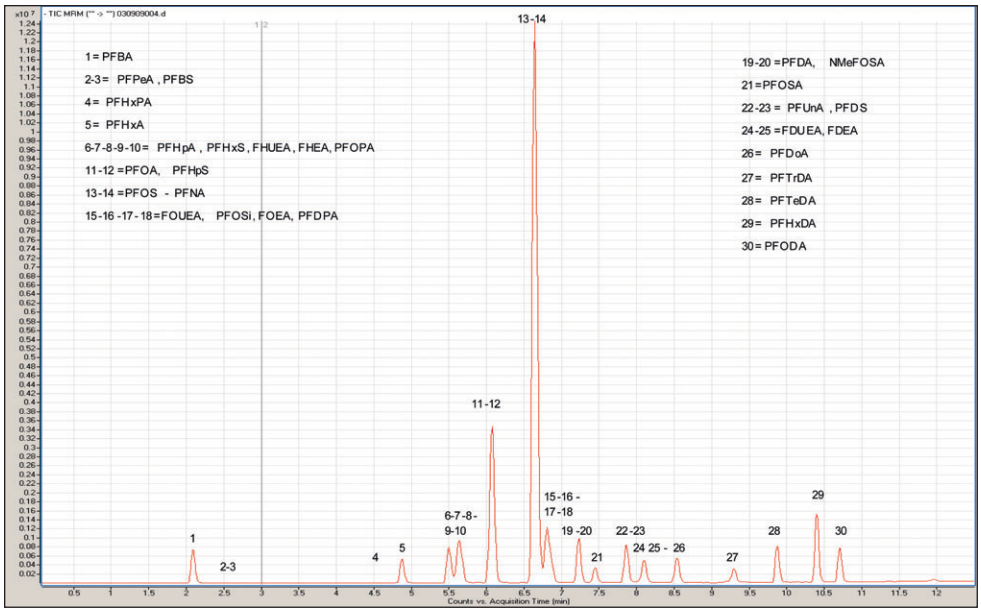

Figure 2: Separation of thirty perfluorinated compounds on the Hypersil GOLD column

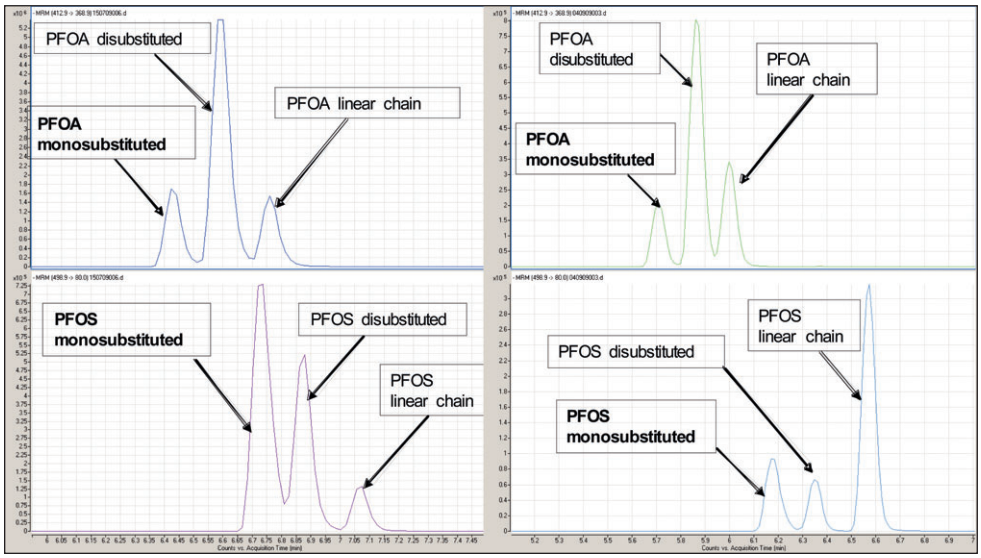

Figure 3: Comparison of the separation of isomeric species on Hypersil GOLD PFP phase on the left and on Hypersil GOLD phase on the right

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