# Chemical purity issues relating to the identity of perfluorooctanesulfonate (PFOS) samples for use as reference standards

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**Abstract**

Samples of potassium perfluorooctanesulfonate (PFOSK) from three suppliers were analyzed by LC-ESI-MS/MS for purity and for the percentage of linear isomer present. Our data indicated that the purity ranged from 80-98% and the percentages of linear isomer from 67-79%. The proportion of branched isomers present in the samples was also estimated using $^{19}$F NMR. These results agreed quite closely with those found by LCMS indicating that there is essentially no difference in overall SIM response factor for the branched isomers *vs* that of the linear isomer.

## 1. Introduction

There has been increased interest regarding the wide spread distribution of fluorinated chemicals, including perfluorooctanesulfonate (PFOS), in the environment and their accumulation in humans.[1-4] The production of PFOS derivatives from linear alkyl precursors using electrochemical fluorination is not a clean process but, instead, gives complex mixtures. Indeed, commercial PFOS from 3M contained[5] approximately 86% PFOS, which is present as a mixture of *ca* 70% linear (**1**; see Fig. 1) and *ca* 30% branched isomers (**2**–**11**; see Fig. 1) as measured by $^{19}$F NMR spectroscopy.[6-7] This process also produced PFOS homologues with 5, 6 and 7 carbon chain lengths, perfluoroalkanoic acids and partially fluorinated compounds.[2,5,8]

Many laboratories are seeking to identify and quantitate PFOS in a variety of samples. In many cases, these laboratories use standards made up from PFOS which has been purchased from one of a number of suppliers. Typically, the PFOS is claimed to be >98% pure. Unfortunately, at least in some cases, the measurements of purity are simply based on an acid/base titration method after ion exchange with a strong acid resin. Clearly, this analysis is not specific to PFOS and therefore it brings uncertainty to the definition of >98%.

The objective of this work was to determine the actual purity of various PFOS standards available on the market. We also summarize the linear to branched isomer ratio in the various PFOS samples, as measured by LCMS and $^{19}$F NMR spectroscopy, and discuss whether or not such differences may have an impact on the "total PFOS" measurement in an environmental sample.

## 2. Materials and methods

### *2.1 Chemicals and standards*

Potassium PFOS (L-PFOSK ; >99% linear) was prepared at Wellington Laboratories using proprietary methods. Samples of potassium perfluorooctanesulfonate (PFOSK) were purchased from Fluka, Matrix, and TCI. A commercial PFOS sample (3M; lot# 217) was supplied by Dr. S.Mabury at the University of Toronto with permission from 3M. NMR solvent methanol-$d_4$ was purchased from CDN Isotopes, the $^{19}$F NMR internal standard, hexafluorobenzene, from Sigma-Aldrich, and HPLC grade methanol, acetonitrile and water from Caledon.

### *2.2 NMR Experiments*

$^{19}$F spectra were recorded at 375.50 MHz on a Bruker DPX 400 NMR spectrometer equipped with a Bruker SEF $^{19}$F/$^{1}$H dual probehead. Chemical shifts are reported relative to hexafluorobenzene using the signal at -169 ppm as internal reference.[9]

*2.3 LCMS Experiments*

LCMS experiments were conducted on a Waters Acquity Ultra Performance LC interfaced to a Micromass Quattro micro API mass spectrometer. Separations were performed on an Acquity UPLC BEH Shield $RP_{18}$ column (1.7 um, 2.1 x 100 mm). It was necessary to employ two different LC gradients in order to achieve the separations required for the variety of experiments carried out in this paper. All chromatographic separation conditions are outlined in Table 1.

For quantitative determination, the Micromass Quattro *micro* atmospheric pressure ionization (API) mass spectrometer was set up in the negative-ion electrospray mode with the following conditions: Capillary Voltage (kV) = 2.60; Cone voltage (V) = 60.00; Cone Gas Flow (L/Hr) = 60; Desolvation Gas Flow (L/Hr) = 650; Desolvation Gas Temperature (°C) = 325; Collision Gas (mbar) ~ 3.50e-3; Collision Voltage (V) = 40. The transition $[M - K]^-$ (m/z = 499) to $[FSO3]^-$ (m/z = 99) was optimized and used for quantification. A dwell time of 0.2 s was used to monitor each transition. The LC conditions are outlined in Table 1, section A.

*2.4 Construction of calibration curves and preparation of test solutions*

All LCMS data were obtained in the SIM mode. Calibration standards, of concentration 100, 75, 50, 25, and 10 ng/mL, were prepared in methanol by serial dilution using in-house 50 µg/mL stock solutions of L-PFOSK. The 50 ng/mL PFOS samples (Fluka, TCI, Matrix and 3M) were also prepared from 50 µg/mL solutions in methanol by serial dilution. $^{13}C_4$-MPFOS (MPFOS; Wellington Laboratories) was added as an internal standard to all solutions (calibration standards and supplier-grade PFOS samples) at a concentration of 1 ng/mL. The 5-point calibration curves obtained were not forced through zero, but were linear over the range of PFOS concentrations utilized ($R^2 > 0.999$, %RSD < 4%). Quantification of PFOS was achieved using relative response factors (RRF) in order to minimize error associated with instrumental drift. The calibration standards were run before and after the sample sets and the percent difference between curves was < 3%.

**3. Results and Discussions**

*3.1 PFOS content in some samples of supplier grade PFOS*

Various chemical suppliers provide PFOS with a claimed purity of >98%. At least in some cases, the purity measurements are based on an acid/base titration method after ion exchange with a strong acid resin. Clearly, this analysis is not specific for PFOS. The presence of impurities[5,8] such as other homologues and perfluorocarboxylates will contribute to the apparent PFOS content when using the titration method and this may result in an over-estimated purity. A better method of determining the actual purity of these supplier grade PFOS samples is by LCMS analysis using a calibration curve derived from a PFOS sample of known purity.

We have undertaken the analysis of three supplier-grade PFOS samples by LCMS using electrospray ionization and SIM. Since supplier-grade PFOS samples exist as potassium salts, calibration curves were prepared based on L-PFOSK. Solutions of the supplier-grade PFOS samples and the PFOS standards were all prepared in 100% methanol. The results for LCMS analysis are summarized in Table 2. In this determination, the linear and branched isomers were integrated as a single peak, assuming that the response factors for branched and linear isomers are equivalent.[10] An evaluation of the veracity of this assumption is included below in section 3.2.

The results in Table 2 do show a marked variability in the purity (from 80% to 98%) of PFOS obtained from three different suppliers. This confirms that PFOS purity measurements based on a titration method after ion exchange is not specific to PFOS and can not properly determine the actual PFOS purity. LCMS analysis using the appropriate reference standards is recommended for determining the chemical purity of PFOS.

We also analyzed the commercial-grade 3M PFOS sample for chemical purity to further validate the usefulness of this LCMS method as a viable means for determining the PFOS content in a technical PFOS sample. The results gave a PFOS content of 85% which is close to the literature value[5] of approximately 86%.

Analytical laboratories using some supplier-grade PFOS samples as reference standards may be overestimating the PFOS present in environmental standards by as much as 20% due to this purity issue. Therefore, it is important for laboratories, not only to state the source of their PFOS standard, but also to find out how the purity of the material was measured. We should note that the PFOS content obtained for each of the three supplier-grade samples in this study is specific only for that particular lot and it is quite possible that different lots will have different PFOS content.

*3.2 Impact of the branched PFOS isomers on the overall LCMS signal intensity of PFOS*

In the work discussed above, it was assumed that the response factors for branched and linear isomers were equivalent using SIM. The isomer content in the PFOS samples was measured by $^{19}$F NMR spectroscopy.[6,7] The linear to branched isomer ratio was also measured by LCMS using SIM. The PFOS samples were prepared in 80:20 methanol/water to obtain good chromatographic profiles of PFOS and this enabled base line separation of the linear isomer from the branched isomers (see Fig. 2). A comparison of the linear to branched PFOS isomer ratio, as measured by LCMS and $^{19}$F NMR spectroscopy, is shown in Table 3 for the Fluka sample. These results appear to confirm that the overall SIM response factor for the branched isomers is similar to that of the linear isomer.

## 4. Conclusions

It has been shown that supplier grade potassium perfluorooctanesulfonate (PFOSK) can have a purity ranging from 80% to 98%. Therefore, laboratories must be careful when selecting these types of samples for use as standards in PFOS quantification.

The proportion of branched isomers present in the samples was estimated using $^{19}$F NMR. The results agreed quite closely with those found by LCMS indicating that there is essentially no difference in overall SIM response for the branched isomers versus the linear isomer.

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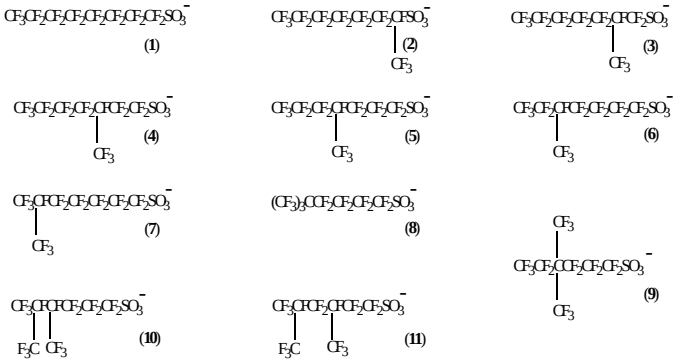

Fig. 1: Structures of the 11 major PFOS isomers.

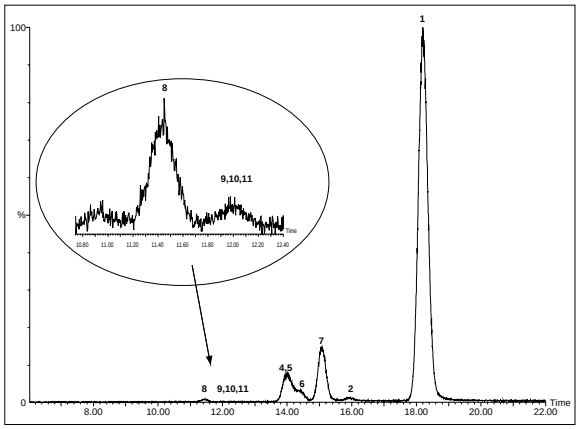


Fig. 2: A chromatogram displaying the separation of PFOS isomers obtained in SIM mode (m/z 499). (LC conditions outlined in Table 1, section B).

Table 1: A summary of chromatographic separation conditions

| | | | |
|---|---|---|---|
| **Instrument** | **Waters Acquity UPLC** | | |
| **Column** | **Acquity UPLC BEH Shield $RP_{18}$ (1.7um, 2.1x100mm)** | | |
| **Mobile Phase** | **A : Water with 10mM $NH_4OAc$** | | |
| | **B : 80:20 MeOH:ACN with 10mM $NH_4OAc$** | | |
| **LC Conditions** | **Description** | **Gradient** | **Flow** |
| **A** | **Quantification of PFOS in technical mixtures** | **0.0min - 67%B** | **0.35mL/min** |
| | | **5.5min - 67%B** | |
| | | **6.0min - 90%B** | |
| | | **9.0min - 90%B** | |
| | | **9.5min - 67%B** | |
| **B** | **Separation of PFOS isomers** | **0.0min - 47%B** | **0.35mL/min** |
| | | **6.0min - 49%B** | |
| | | **23.0min - 49%B** | |
| | | **23.5min - 90%B** | |
| | | **25.5min - 90%B** | |
| | | **26.0min - 47%B** | |

Table 2. PFOS content as measured by LCMS in various technical grade PFOS samples

| **PFOS (lot#)** | **% PFOS (chemical purity)** | **% Linear** |
|---|---|---|
| **Fluka (436098/1)** | **98** | **79** |
| **TCI (GJ01)** | **97** | **67** |
| **Matrix (P15D)** | **80** | **68** |
| **3M (217)** | **85** | **75** |
| **L-PFOSK** | **>98%** | **99** |

Table 3. Determination of the isomer distribution in PFOS-Fluka by LCMS and $^{19}F$ NMR.

| **Isomer** | **PFOS-Fluka by LCMS (SIM)** | **PFOS-Fluka by $^{19}F$ NMR** |
|---|---|---|
| **1** | **82** | **79** |
| **2** | **0.5** | **1.2** |
| **3, 4, 5 & 6** | **7.5** | **9.3** |
| **7** | **9.7** | **10.0** |
| **8** | **0.4** | **0.22** |
| **9, 10 & 11** | **0.08** | **0.50** |