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EXPOSURE OF GREEK ADULT CONSUMERS TO PFASS FROM FOOD AND DRINKING WATER

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Introduction

PFASs have been manufactured and released in the environment for more than 50 years, and are now acknowledged as widespread, persistent and bioaccumulative pollutants detected in water and sediments, biota and humans[1]. Animal studies have shown that they exhibit hepatotoxicity, neurotoxicity, immunotoxicity, developmental effects and possible carcinogenicity[2]. Although sources of human exposure to PFASs include household dust and drinking water, it has been established that food is the most important source of PFAS intake for non-occupationally exposed humans[3,4]. The European Food Safety Authority (EFSA) published in 2008 a health risk assessment for the two most important PFASs, PFOS (perfluorooctane sulfonate) and PFOA (perfluorooctanoic acid), and assigned a Tolerable Daily Intake (TDI) of 150 ng kg $^{-1}$ b.w. day^{-1} for PFOS and 1500 ng kg^{-1} b.w. day^{-1} for PFOA[5]. Several studies report PFASs levels in food items and provide information on human dietary exposure to PFASs[6-14]. Most of these studies present fish and seafood as the most contaminated food items and PFOS as the dominant compound in most of the cases.
In this paper we summarize several of our findings concerning the levels of PFASs in fish, as well as eggs, another important contributor to human diet. Data from exposure by drinking water are also presented, in order to provide a simple risk assessment on exposure to PFASs in Greece.

Materials and methods

Sample collection

Samples were collected from several locations in Greece and analysed in the Mass Spectrometry and Dioxin Analysis Laboratory in Greece and the RIKILT - Institute of Food Safety, WUR in the Netherlands. The sampling procedure of each sample group is described below:

Fish

In the present study, samples of 7 species of finfish – anchovy, bogue, hake, picarel, sardine, sand smelt and striped mullet – and 3 species of shellfish – Mediterranean mussel, shrimp and squid, were collected during the winter-early spring of 2011 and analysed. These fish are considered very representative of the Greek culinary habits. Finfish, squids and shrimps were purchased from fish markets, while mussels were obtained from a mariculture farm. The fishing locations of the collected samples are presented in Figure 1.

Figure 1: Fishing locations of samples.

Eggs

Chicken egg samples were purchased from various super markets (31 samples) and collected from domestic coops (45 samples) in Greece from August 2013 until August of 2014. Every sample consisted in principle of 20 individual eggs, unless fewer eggs were provided.

Drinking water

43 drinking tap water samples were collected from Greece from August 2013 until January 2014. Polyethylene bottles were used in order to avoid possible leaking and contamination/adsorption. All the water samples were transferred to the laboratory and were directly stored at 4°C until the analysis. The sampling points are illustrated in Figure 2.

Figure 2: Drinking tap water sampling points.

Determination of PFASs

The samples were analysed according to previous published studies[15,16,17]. Briefly, fish samples were extracted by pressurized liquid extraction (PLE), using an ASE Dionex 300 apparatus and MeOH as extraction solvent. Subsequent clean-up was performed by SPE with Florisil and basic alumina[15].
Chicken eggs were manually extracted with MeOH. The extract was further cleaned-up by SPE using weak anion exchange Oasis WAX cartridges[16].
Oasis WAX cartridges were also used for the clean-up of water samples, for which no extraction was needed[17].

Instrumental analysis
Quantification was performed by liquid chromatography combined with tandem mass spectrometry (LC-MS/MS).
Fish samples were injected in a Hypersil GOLD C8 column (150 mm x 2.1 mm i.d, 3 μm, Thermo). The HPLC was connected to a triple quadrupole mass spectrometer (TSQ QUANTUM ULTRA, Thermo).
A Shimadzu LC system equipped either with a Fluorosep analytical column or with an Acquity UPLC BEH C18 column was used for the analysis of chicken eggs and drinking water respectively. The LC system was connected to a triple quadrupole MS (AB SCIEX QTRAP 5500 SYSTEM, Applied Biosystem – Analytical Technologies).
Analysis was performed with a multiple reaction monitoring method (MRM) that monitored two mass transitions (parent ion/product ion) for every analyte. Confirmation of analyte identity was based on retention time, in addition to relative response of the secondary mass transition to the primary mass transition. Quantification of the target compounds was performed by the sum of areas of the two product ions using a response factor calibration curve versus the ^{13}C or ^{18}O-labelled standard.

Calculation of human intake of PFOS and PFOA
The estimated daily intake (EDI) in ng kg^{-1} b.w. of PFOS and PFOA is calculated by the following equation (Figure 3):

Figure 3

where FIR stands for the Food Intake Rate and ABW for Average Body Weight. C is the concentration of PFOS or PFOA (ng g^{-1} ww). Concentrations of zero were assigned when PFOS or PFOA was not detected above the LOD.
Due to lack of statistical data on food consumption in Greece, it was not easy to define the FIR in each case. In the case of fish and eggs, the daily food consumption by adults was according to FAO (Available on line on: http://faostat.fao.org/). It was assumed to be 36 g per person per day for fish, 9.80 g per person per day for cephalopod molluscs and 5.42 g per person per day for crustaceans. This data is in agreement with the daily consumption proposed by EFSA and was used as FIR for the calculation of PFOS and PFOA intake. For eggs it was assumed that each adult consumes 8.9 kg each year, which amounts to a daily intake of 24.4 g of egg. In the case of drinking water, a volume of 2L, which is the suggested volume of water that should be consumed by a healthy adult daily, was assumed as daily consumption. ABW for adults was 70 kg according to EFSA.

Resuls and discussion

Dietary intake by fish

Table 1: Dietary intake of PFOS and PFOA by fish

PFASs above the detection limit were found in all fish samples and in all shellfish except the mussel. The predominant PFAS was PFOS, the highest concentration of which was measured in picarel (20.4 ng g^{-1} fw). PFOS values for the rest of the samples were between <LOD and 5.15 ng g^{-1} fw. The EDI of PFOS and PFOA was calculated separately for each species and the results are presented in Table 1. All calculated values were well below the TDI proposed by EFSA. Several previous studies have shown that fish and seafood are the main contributors to PFAS intake in humans. Taking this into account, we estimate that even with the contribution of other food items to PFAS intake it is highly unlikely that

consumers of these fish species, which are among the most common fish eaten in Greece, exceed the TDI for PFOS and PFOA.

Dietary intake by eggs

Total PFAS levels in eggs were higher in home produced eggs (range <LOQ–15.0 ng g^{-1}) than in eggs purchased in supermarkets. PFOS was the predominant compound, detected in 69% of the home produced samples (range <LOQ–8.9 ng g^{-1}) and in only one supermarket egg sample (0.9 ng g^{-1}). PFOA was not detected above the LOQ in any of the samples. Considering the worst case scenario, in which the most contaminated home produced egg is used for the calculation of the EDI, the daily intake of PFOS by egg consumption is 3.1 ng kg^{-1} bw day^{-1}.

Intake by drinking water

PFASs concentrations above the limit of quantification, LOQ (0.6 ng L^{-1}) were detected in 20.9% of the samples. Total PFAS concentrations ranged between <LOQ and 5.9 ng L^{-1}, while PFOS above the LOQ was not detected in any sample. PFOA above the LOQ was determined in nine samples, and the sample with the highest levels had a value of 3.6 ng L^{-1}.

Conclusions

Table 2 below summarizes the worst possible exposure of consumers to PFASs by the ingestion of the above food items and tap water. Food is the most important contributor while the contribution of water is negligible. It is therefore highly unlikely that average consumers in Greece exceed the TDI proposed by EFSA for PFOS and PFOA.

Table 2. Worst case scenario of exposure of consumers living in Greece based on the analysed sample groups (fish, eggs and drinking water).

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Figure 1

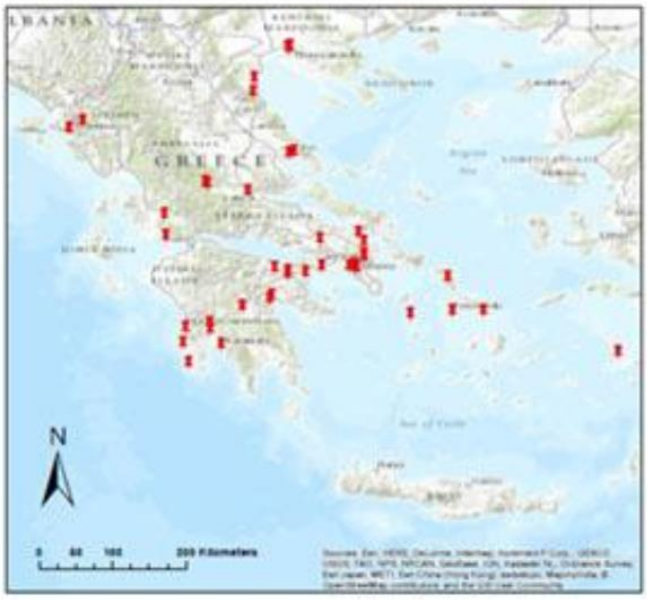

Figure 2

$$EDI = \frac{C \times FIR}{ABW}$$

Figure 3

EDI (ng/kg bw)	PFOS	PFOA
Anchovy	1.57	0.05
Bogue	0.42	-
Hake	0.43	-
Picarel	10.48	-
Sandsmelt	0.60	-
Sardine	-	0.09
Stripped mullet	2.91	0.18
Mussel	-	-
Shrimp	2.65	0.20
Squid	-	-

Table 1

Matrix	Maximum concentration of PFOS	EDI (ng kg^{-1} bw day^{-1}) for PFOS	Maximum concentration of PFOA	EDI (ng kg^{-1} bw day^{-1}) for PFOA
Fish	20.4 ng g^{-1} fw	10.48	0.99 ng g^{-1} fw	0.20
Eggs	8.9 ng g^{-1} fw	3.10	0.5 ng g^{-1} fw	0.17
Drinking water	-	-	3.6 ng L^{-1}	0.10
Sum		13.58		0.47
TDI proposed by EFSA		**150**		**1500**

Table 2