

Contribution of polyfluoroalkyl phosphate esters (PAPs) and other precursor compounds to perfluoroalkyl carboxylates (PFCAs) in humans and the environment

If, having endured much, we have at last asserted our "right to know," and if by knowing, we have concluded that we are being asked to take senseless and frightening risks, then we should no longer accept the counsel of those who tell us that we must fill our world with poisonous chemicals; we should look about and see what other course is open to us.

Rachel Carson, Silent Spring

Örebro Studies in Chemistry 18



ULRIKA ERIKSSON

**Contribution of polyfluoroalkyl phosphate esters (PAPs)
and other precursor compounds to perfluoroalkyl
carboxylates (PFCAs) in humans and the environment**

© Ulrika Eriksson, 2016

Title: Contribution of polyfluoroalkyl phosphate esters (PAPs) and other precursor compounds to perfluoroalkyl carboxylates (PFCAs) in humans and the environment

Publisher: Örebro University 2016
www.oru.se/publikationer-avhandlingar

Print: Örebro University, Repro 10/2016

ISSN 1651-4270
ISBN 978-91-7529-164-2

Abstract

Ulrika Eriksson (2016): Contribution of polyfluoroalkyl phosphate esters (PAPs) and other precursor compounds to perfluoroalkyl carboxylates (PFCAs) in humans and the environment. Örebro Studies in Chemistry 18.

Per- and polyfluoroalkyl substances (PFAS) are anthropogenic compounds that have been spread all over the world. The use of fluorotelomer compounds, short-chained homologues, and other PFASs with perfluorinated moieties has emerged recent years. One of these emerging compound classes is polyfluoroalkyl phosphate esters (PAPs), which have the ability to degrade into persistent PFCAs.

The aim of this thesis was to assess the contribution of PAPs and other precursors to the exposure of PFCAs to humans and the environment. The main objective was to analyze a wide range of PFAS in human serum, wild bird eggs, indoor dust, waste water, and sludge. There was a significant contribution from selected precursors to the total amount of PFASs in the abiotic compartments indoor dust, waste water, and sludge. Levels of PAPs found in house dust exceeded those of PFCAs and perfluorosulfonic acids (PFSAs), revealing PAPs as a world-wide important exposure source.

A net increase was during waste water treatment was observed for several PFASs in Swedish waste water treatment plants. Together with presence of precursor compounds and intermediates in the influent water and the sludge, this suggest that degradation of PFCA precursors contributed to the increase of PFCAs. Detection of precursors in human serum, together with slow declining trends of PFCAs, revealed an ongoing exposure of PFCAs to the general population of Australia. The diPAPs and the FTSAs were also detected in raptor bird eggs from Sweden from both the terrestrial and the freshwater environment. The precursors concentrations and patterns observed reveal that current regulatory measures are insufficient for the purpose of protecting humans and the environment from PFASs exposure.

Keywords: PAPs, precursors, PFCA, exposure, indoor dust, human serum, WWTP, bird eggs

Ulrika Eriksson, School of Science and Technology, Örebro University, SE-701-82 Örebro, Sweden, ulrika.eriksson@oru.se

List of papers

This thesis is based on following papers:

Paper I:

Eriksson U, Kärman A. 2015. World-wide indoor exposure to polyfluoroalkyl phosphate esters (PAPs) and other PFASs in household dust. *Environmental Science and Technology* 49 (24), 14503-14511. Reproduced with permission from Environmental Science and Technology. Copyright 2015 American Chemical Society.

Paper II:

Eriksson U, Mueller J, Toms L-M L, Hobson P, Kärman A. 2016. Temporal trends of PFASs, PFCAs and selected precursors in Australian serum from 2002 to 2013. Accepted for publication in *Environmental Pollution*.

Paper III:

Eriksson U, Haglund P, Kärman A. 2016. Per- and polyfluoroalkyl substances (PFASs) in sludge and water from Swedish waste water treatment plants (WWTP). Under review in *Journal of Environmental Science*.

Paper IV:

Eriksson U, Roos A, Lind Y, Hope K, Ekblad A, Kärman A. 2016. Comparison of PFASs contamination in the freshwater and terrestrial environments by analysis of eggs from osprey (*Pandion haliaetus*), tawny owl (*Strix aluco*), and common kestrel (*Falco tinnunculus*). *Environmental Research* 149, 40-47.

Papers not included in this thesis:

Eriksson U, Kärrman A, Rotander A, Mikkelsen B, Dam M. 2013. Perfluoroalkyl substances (PFASs) in food and water from Faroe Islands. *Environmental Science and Pollution Research* 20 (11) 7940-8.

Ekblad A, Mikusinska A, Ågren GI, Menichetti L, Wallander H, Vilgalys R, Bahr A, Eriksson U. 2016. Production and turnover of ectomycorrhizal extramatrical mycelial biomass and necromass under elevated CO₂ and nitrogen fertilization. *New Phytologist* 211(3) 874-885.

Geng D, Ericson Jogsten I, Kukucka P, Eriksson U, Ekblad A, Grahn H, Roos A. 2016. Temporal Trends of Polychlorinated Biphenyls, Organochlorine Pesticides and Polybrominated Diphenyl Ethers in Osprey Eggs in Sweden over the Years 1966 – 2013. Submitted to *Environmental Pollution*.

List of abbreviations

Name	Abbreviation
AFFF	Aqueous film forming foam
diPAP	Polyfluoroalkyl phosphoric acid diester
ECF	Electrochemical fluorination
EtFOSA	Ethyl perfluorooctane sulfonamide
EtFOSE	Ethyl perfluorooctane sulfonamidoethanol
FBSA	Perfluorobutane sulfonamide
FTAC	Fluorotelomer acrylate
FTAL	Fluorotelomer aldehydes
FTCA	Fluorotelomer carboxylic acids
FTMAP	Fluorotelomer mercaptoalkyl phosphate diester
FTOH	Fluorotelomer alcohol
FTSA	Fluorotelomer sulfonic acid
FTUCA	Fluorotelomer unsaturated carboxylic acids
MeFBSA	Methyl perfluorobutane sulfonamide
MeFOSA	Methyl perfluorooctane sulfonamide
MeFOSE	Methyl perfluorooctane sulfonamidoethanol
monoPAP	Polyfluoroalkyl phosphoric acid monoester
PAP	Polyfluoroalkyl phosphate ester
PBSF	Perfluorobutane sulfonyl fluoride
PFAS	Per- and polyfluoroalkyl substance
PFBA	Perfluorobutanoic acid
PFBS	Perfluorobutane sulfonic acid
PFCA	Perfluoroalkyl carboxylic acid
PFDA	Perfluorodecanoic acid
PFDPA	Perfluorodecyl phosphonic acid
PFDS	Perfluorodecasulfonic acid
PFEI	Pentafluoroethyl iodide
PFHpA	Perfluoropentanoic acid
PFHpS	Perfluoroheptasulfonic acid
PFHxA	Perfluorohexanoic acid
PFHxA	Perfluorohexadecanoic acid
PFHxPA	Perfluorohexyl phosphonic acid
PFHxS	Perfluorohexasulfonic acid
PFNA	Perfluorononanoic acid
PFNS	Perfluorononasulfonic acid
PFOA	Perfluorooctanoic acid

PFOcDA	Perfluorooctadecanoic acid
PFOPA	Perfluorooctyl phosphonic acid
PFOS	Perfluorooctasulfonic acid
PFPA	Perfluorophosphonic acid
PFPA	Perfluoroalkyl phosphonic acid
PFPeA	Perfluoropentanoic acid
PFPeS	Perfluoropentane sulfonic acid
PFPiA	Perfluorophosphinic acid
PFPiA	Perfluoroalkyl phosphinic acid
PFSA	Perfluoroalkyl sulfonic acid
PFTDA	Perfluorotetradecanoic acid
PFTTrDA	Perfluorotridecanoic acid
POSF	Perfluorooctane sulfonyl fluoride
PTFE	Polytetrafluoroethylene
PVDF	Polyvinylidene fluoride
SAmPAP	Perfluorooctane sulfonamide phosphate esters
TDI	Tolerable daily intake
TFE	Tetrafluoroethylene
triPAP	Polyfluoroalkyl phosphoric acid triester
WWTP	Waste water treatment plant

Table of Contents

1. INTRODUCTION.....	13
1. 1. Synthesis and use of PFASs	13
1. 2. A historical view	17
1. 3. Direct and indirect sources	20
1. 4. Degradation	21
1. 5. Exposure pathways and sources	24
1. 6. Toxicity.....	28
1. 7. Temporal trends in humans	29
1. 8. Temporal trend in the environment.....	31
2. AIM AND OBJECTIVES	34
3. METHODS.....	35
3. 1. Extraction	36
3. 1. 1. Dust	36
3. 1. 2. Bird eggs	36
3. 1. 3. Serum.....	37
3. 1. 4. Water	37
3. 1. 5. Sludge	38
3. 2. Instrumental analysis and quantification.....	38
3. 2. 1. Daily intake	41
3. 2. 2. Blank contamination.....	42
4. RESULTS AND DISCUSSION.....	43
4. 1. Method development	43
4. 1. 1. Extraction and clean-up.....	43
4. 1. 3. Matrix effects.....	45
4. 1. 4. Composition in extracts.....	49
4. 2. Dust levels of precursors	51
4. 2. 1. Exposure assessment	53
4. 3. Human levels of PFCA precursors.....	56
4. 3. 1. Ongoing exposure.....	57
4. 3. 2. Daily intake	58
4. 4. Waste water treatment plants.....	59
4. 4. 1. Emission to the environment from WWTPs	61
4. 5. Levels of PFCA precursors in wild-life	63
4. 6. Contribution of precursors to PFCA	64
4. 6. 1. Temporal trends.....	72

5. CONCLUSIONS 75

7. ACKNOWLEDGEMENT 78

REFERENCES 80

1. Introduction

The group of per- and polyfluoroalkyl substances (PFASs) consist of a large number of classes, of which most of them have in common that they have an alkyl chain which is partially or fully fluorinated, with a functional head group, typically carboxylate, sulfonate, phosphate, or alcohol (Buck et al. 2011). The high electronegativity of the fluorine provides strong polarity and high energy to the carbon-fluorine-bond (Chambers 2009). The bond strength increase with number of fluorines attached to the carbon atom. The strong carbon-fluorine bond together with effective shielding of the alkyl chain of the fluorine atoms result in high stability of the molecule. Fluorine has a high ionization potential and therefore the inter- and intramolecular interactions in fluorocarbons are low, leading to extremely low surface tension and similar or higher volatility compared to their hydrogen carbon counterparts. A charged moiety attached to the fluorinated alkyl chain enhance the water solubility, resulting in a molecule with both hydrophobic and hydrophilic properties. These superior properties compared to other surfactants make PFASs suitable for oil- and water repellency and high temperature applications, which have led to an extensive use in a wide range of applications such as in wetting and leveling agents, paints, coatings, waxes, chrome plating bath, fire-fighting foams, cosmetics, paper, food packaging, textiles, carpets, cleaning agents, pesticides, photographic emulsifiers. The PFAS group comprises several thousand compounds (KEMI 2015). Only a few of these are usually covered by monitoring and scientific studies. Analysis of total extractable organic fluorine content suggest that a large proportion of the organofluorine in the environment and humans are unknown compounds (Yeung et al. 2013a, Yeung and Mabury 2016). Commonly used commercial PFASs that will eventually be transformed to persistent PFASs, but are always ignored in monitoring programs, may represent a significant portion of the total amount.

1. 1. Synthesis and use of PFASs

There are two main processes for PFAS production; electrochemical fluorination (ECF) and telomerization. In the ECF process, the starting material is octane sulfonyl fluoride ($C_8H_{17}SO_2F$) or a carbonyl fluoride ($C_7H_{15}COF$) (3M Company 1999a, Lehmler 2005, Buck et al. 2011). In the presence of anhydrous fluoride, a current is passed through the solution and all the hydrogen atoms are replaced with fluorine, leading to a perfluorinated

substance (fig. 1). This process can lead to rearrangement and breakage of the carbon chain, yielding a proportion of 20-30% branched isomers. With octane sulfonyl fluoride as starting material, the process yields perfluorooctane sulfonyl fluoride (POSF), which can be further processed to yield perfluorooctane sulfonic acid (PFOS) through base-catalyzed hydrolysis, and with carbonyl fluoride the process yields perfluorooctanoic acid (PFOA). PFOS has been produced using ECF since 1949 and has been widely applied in for example aqueous film forming foam (AFFF), metal plating, and hydraulic fluids (Paul et al. 2009). Further reaction with methyl or ethyl amine yields perfluorooctane sulfonamides (MeFOSA and EtFOSA). FOSA can either be used in commercial applications, for example EtFOSA in pesticides (Gilljam et al. 2016), or be further reacted with ethylene carbonate to perfluoroalkane sulfonamidoethanols (MeFOSE and EtFOSE). MeFOSE has been used in polymeric substances in textiles and carpets (Olsen et al. 2005). EtFOSE was amongst others used as building material for perfluorooctane sulfonamide phosphate esters (SAmPAPs), which has been used for paper treatment (D'eon et al. 2009). POSF-based chemistry has been phased out by the former major manufacturers but homologues with shorter chain length, foremost perfluorobutane sulfonyl fluoride (PBSF), have replaced POSF in the ECF process (OECD 2007, Buck et al. 2011).

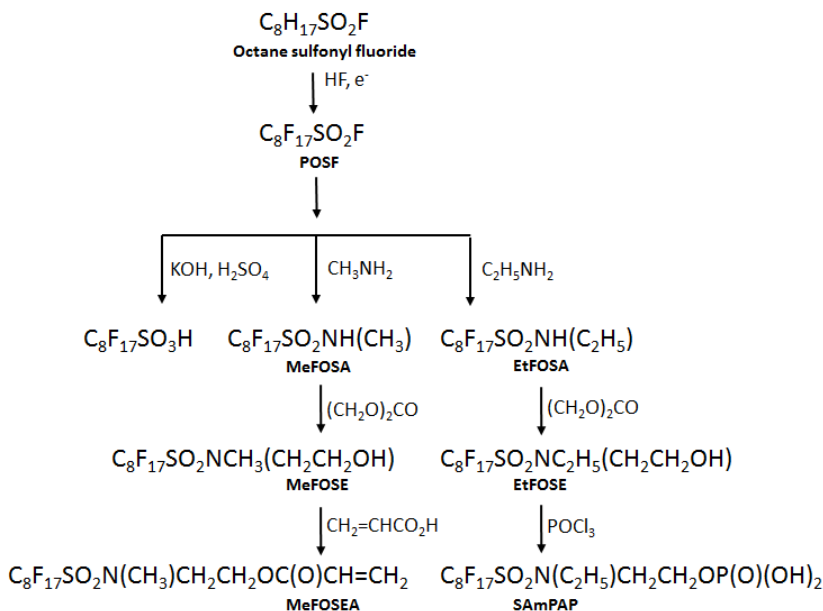


Figure 1. A simplified scheme over the ECF process (Buck et al. 2011)

In the telomerization process, tetrafluoroethylene (TFE) is reacted with a pentafluoroethyl iodide (PFEI) yielding perfluoroalkyl iodides (PFAI) (fig. 2) (Lehmmler 2005, Buck et al. 2011). Almost exclusive linear homologues are produced, in contrast to the ECF process, though synthesis of branched perfluoroalkyl iodides also has been described (Bertocchio et al. 1993). In general, only even-numbered homologues are produced, though there have been some reports about odd-numbered fluorotelomer-based compounds in literature (Ding et al. 2012). The PFAI can be reacted to yield for example PFOA, perfluorononanoic acid (PFNA), fluorotelomer iodide (FTI), and perfluoroalkyl phosphonic acids (PFPA) and phosphinic acids (PFPiA) (Wang et al. 2016a). The major use of PFOA is as processing aid in the synthesis of the fluoropolymer polytetrafluoroethylene (PTFE), and for PFNA as a processing aid in synthesis of polyvinylidene fluoride (PVDF). Mixtures of PFPA and PFPiA have been used in pesticides, wetting and leveling agents, plating, and cleaning products (Dookhith 2001, Pilot Chemical 2016).

FTI can be further reacted to produce various PFASs, fluorotelomer alcohols (FTOH), fluorotelomer olefins (FTO), fluorotelomer sulfonic acids (FTSAs), polyfluoroalkyl phosphate esters (PAPs) and perfluoroalkyl carboxylates (PFCAs). FTSA-based compounds have been used mainly in fire fighting foams as an active ingredient, due to their low surface tension, which enables aqueous film formation on hydrocarbon fuels (Harding-Marjanovic et al. 2015). Other uses have been described as well, for instance in ink to reduce puddling of the ink-jet ink on the nozzle plate (Ma et al. 2002). The FTOHs can be functionalized to fluorotelomer acrylate (FTAC) and further synthesized to fluorotelomer acrylate polymers, for which major use has been in textile, leather, and paper. Alternatively, the FTOHs can be further functionalized to yield PAPs or fluorotelomer mercaptoalkyl phosphate diesters (FTMAPs). Major use for PAPs have been in paper and packaging, including food packaging, but other uses have been described as well, as in personal care products, floor finish, cleaning products, paints, and coatings (Pilot Chemical 2016). The products in the telomerization process are usually mixtures that can have chains of a length between 2 and 18 fluorinated carbons. Distillation is used for separations and purifications. Neither ECF nor telomerization have a 100% yield, and the starting materials can be left as residuals in the final product.

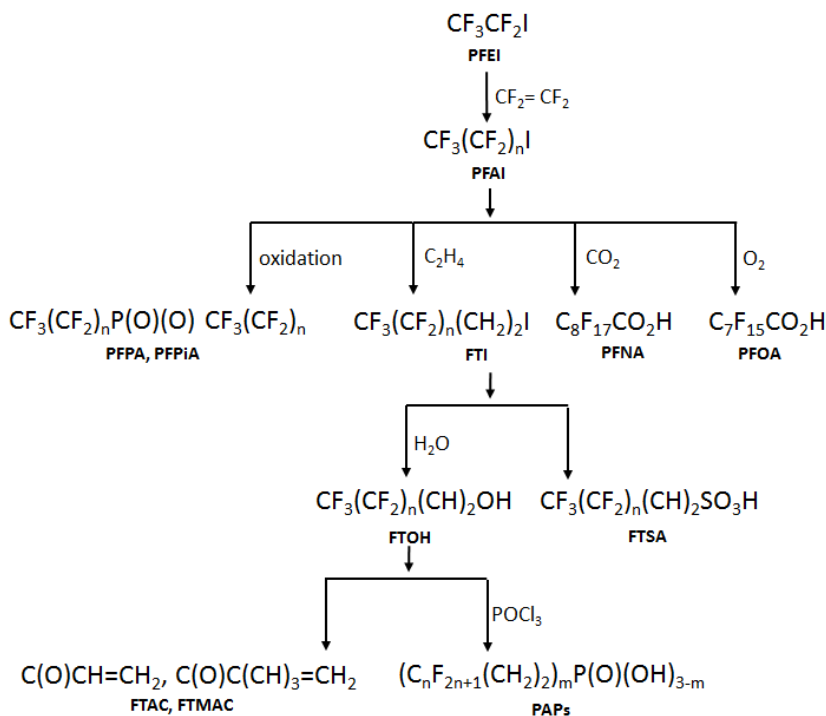


Figure 2. A simplified scheme of the telomerization process (Lehmle 2005, Buck et al. 2011, Wang et al. 2016a).

1. 2. A historical view

PFASs have been produced since the 1950s. Amongst the first applications was the use of ammonium perfluorooctanoate (APFO), a salt of PFOA, as processing aid for polymerization of PTFE (Prevedouros et al. 2006). The fluorinated tail of APFO keep the PTFE particles in dispersion state during the polymerization process (McKeen 2015). Similarly, PFNA was implemented as processing aid for polyvinylidene fluoride (PVDF). Between 1951 and 2002, ECF was the dominating process for manufacturing both PFOS and PFOA (Prevedourus, Buck). The 3M company was the major global POSF producer with manufacturing plants in the US and Europe, though minor quantities were also produced by other companies in Europe, Asia, and South America (Paul et al. 2009). Fluorotelomer production started in 1961 and PFOA production using the telomerization pro-

cess started in the 1970s by DuPont (Wang 2014), though ECF continued for decades to be the major PFOA synthesizing technique (Prevedouros et al. 2006, Wang et al. 2014). In the 1970s, elevated levels of organic fluorine were found in blood of workers at a PFAS production factory, and PFOA was found in their urine (Ubel et al. 1980). This was the first indicator of human exposure to this compound group.

From 1960s to 2000, production volumes of PFASs steadily increased. Annual APFO production has been estimated to increase from 5 – 25 ton in the early 1960s to 200 – 300 ton in the late 1990s (Prevedouros et al. 2006). Production of PFOSF has been estimated to increase from about 500 ton in 1970 to 4 650 ton in 2000 (Paul et al. 2009). Of this amount, 33% was used in paper and packaging, including food packaging, 48% in surface treatment such as textile and leather protection, 3% in AFFF, and 15% in other consumer and industrial applications (3M Company 2000).

In the late 1990s, the analytical technique of electrospray ionization mass spectrometry (ESI-MS) had been largely improved and was applied for the analysis of PFASs, lowering the limit of detection for PFOS to 50 ppb (3M Company 1999b). Attention started to grow around PFAS and in particular PFOS. In 1997, PFOS in serum of the general population was observed by the 3M Company (3M Company 1999b). Previously PFOS and PFOA had been found in occupational exposed workers and monitored for years, but now PFOS was also found in a control group with supposedly contaminant-free serum. Between 1997 and 2000, the 3M Company reduced their waste water discharged by 50% and the air emission by 40% at their facility in Decatur, Alabama, one of their two major POSF plants (3M Company 2000). After increasing pressure from the US EPA, the 3M Company announced their phase-out of the C8-technology in May 2000 and completed it in 2002 (3M Company 2000). This led to several changes in the PFAS production and manufacturing.

As a result of ceased production in North America and Europe, PFOS production expanded in China. The annual production of PFOS in China has increased from <50 ton in 2001 to 250 ton in 2006 and ranged between 220 and 250 ton in 2006 - 2011 (Zhang et al. 2012, Xie et al. 2013). The 3M Company continued their PFAS production, but switched over to PBSF-based chemistry (3M Company 2002). New PFASs with sulfonic acids have also been introduced worldwide, for example perfluoroalkyl ether sulfonic acids (PFESAs) (Wang et al. 2013).

In the years 2006 – 2009, the use of PFOS became further restricted. In the EU, the use and import of PFOS was restricted under REACH in 2006 (REACH 2010). Norway banned the use of PFOS in AFFFs, impregnations, and textiles in 2004 (Norway 2004). Manufacturing, use and sale become prohibited in Canada in 2008 (Canada 2006). PFOS was listed into the Annex B of Stockholm Convention on Persistent Organic Pollutants in 2009 (UNEP 2009). Recently, a project has been initiated by Foreign Economic Cooperation Office (FECO) and World Bank (WB) with the aim to help China reduce their PFOS emission (GEF 2016).

Restrictions of PFCAs have been lagging and implemented later than for PFOS. The 3M Company produced 85% or more of the global production volume of PFOA until 2000, using the ECF technique (OECD 2002). DuPont had previously purchased PFOA from the 3M company, but started producing PFOA through fluorotelomerization at their facility in the US in 2002 (OECD 2007).

In 2006, the Voluntary Stewardship Program was initiated by the US EPA, where eight major PFAS manufacturers participated (DuPont, Arkema, Asahi, BASF Corporation, Clariant, Daikin, 3M/Dyneon, and Solvay Solexis), aiming to reduce PFOA emissions by 95% to 2010 and completely phase-out by 2015 (EPA 2006). Fluorotelomer companies not bound to the Stewardship Program, mainly in China, India, and Russia, have taken over their market share of long-chain PFCAs (Wang et al. 2014). It has been estimated that 31% of the fluorotelomer production was covered by companies not participating in the Stewardship Program in 2013, of which 74% of the production occurred in China (ECHA 2014). The process of regulation of PFOA and long-chain PFCAs in the EU began in 2012-2013 by adding these compounds to the REACH Candidate List of Substances of Very High Concern (SVHC) (REACH EC No. 1907/2006). In 2012, the production volume of PFOA in China was 90 ton, exclusively produced by the ECF process (Li et al. 2015). Emissions of PFOA at one production site in China have been estimated to be 58 ton in 2013 (Wang et al. 2016b). In comparison, 50 ton PFOA was emitted in 2006 from the eight companies participating in the Stewardship program.

A majority, about 80%, of the fluorotelomers were incorporated in polymers in year 2000, while the remaining 20% were used in surfactants (TRP 2002). The information about actual production volumes of

fluorotelomer compounds are however scarce. It has been estimated though that the annual global production was 2 500 ton/year in 1961 – 1979, then increased to 7 500 ton/year in 1980 – 1994, followed by an increase to 20 000 ton/year in 1995 – 2004 (Wang et al. 2014). Current production volume (2004 – 2030) is estimated to be 45 000 ton/year.

Majority of replacement compounds for the long-chained PFASs that have entered the market continues to have a fluorinated structure. One of the major changes is the shift in the industry towards short-chain C4 – C6 PFASs (3M Company 2002, Daikin 2007, DuPont 2008, AGC 2016). The shift towards short-chain PFASs also includes PAPs. For example, the company Daito Kasei has replaced their product PF, a mixture of C8 – C20 mono-, di-, and triPAPs, with 6:2 monoPAP under the trade name EPF (environmental PF) (Kasei 2015). Recently, the use of C8-C18 PAPs in food contact paper and paperboard was banned in the US (FDA 2016). Other replacement compounds are the polyfluorinated ethers (PFPE), for instance ADONA ($\text{CF}_3\text{OCF}_2\text{CF}_2\text{CF}_2\text{OCHF}_2\text{COO-NH}_4^+$) and GenX ($\text{CF}_3\text{CF}_2\text{CF}_2\text{OCF}(\text{CF}_3)\text{COO-NH}_4^+$) from Dyneon and Chemours (formerly DuPont), respectively (Wang et al. 2013). New compounds are constantly entering the market, and it has been estimated that more than 3000 commercial PFAS compounds are circulating on the global market (KEMI 2015). Volumes of PFASs produced are also expected to increase as a consequence of short-chain PFASs having less effective surfactants properties compared to long-chain PFASs. Forecasts predict that the fluorotelomer industry will continue to increase as a consequence of increased living standard and high market demand on products such as textiles, papers, metal plating, and semiconductors (Insights 2016). There are no signs of an instant elimination of PFASs at a global scale within a perspicuous future.

1. 3. Direct and indirect sources

The exposure sources of PFCAs and PFASs are commonly referred to as being of direct or indirect origins. Direct sources are herein described as including both intentionally produced compounds, residuals from the production process, and unintended produced byproducts. Indirect sources are herein described as compounds formed during degradation from precursor compounds. Due to limitations of information on actual production volumes and homologue distribution, and uncertainties in the yield of persistent PFASs from precursors, it has been difficult to determine to

which extent indirect sources contribute to total PFCAs and PFSA's emission. Prevedouros et al. (2006) made the first extensive estimation and concluded that direct emission was the major source for global PFOA and PFOS contamination.

More recently, a global emission inventory showed that emissions of PFOA, PFNA, PFUnDA, and PFTrDA historically (1951 - 2002) came from direct sources, while the majority of the short-chain PFCAs (C4 – C7) were originated from degradation of precursor compounds. Between 2003 and 2015, it was estimated that degradation of precursor compounds and impurities in fluorotelomer-based products were the dominant emission sources for most PFCAs (Wang et al. 2014).

Another possible indirect source that has been heavily debated is the potential contribution from degradation of fluorotelomer polymers (Washington and Jenkins 2015). Their relatively long half-times observed in soil-plant microcosm induce a delay in PFCA release, and bring a possible future dramatic increase of persistent PFAS (Rankin et al. 2014).

1. 4. Degradation

Precursor compounds not having fully fluorinated alkyl chains may have the ability to degrade, in both biotic and abiotic mechanisms, to persistent PFAS. Various pathways for different PFASs in different environmental compartments has been suggested. Biodegradation of PAPs has been demonstrated in rats, sludge, and soil (D'Eon and Mabury 2007, Lee et al. 2010, Lee et al. 2014). It was shown that PAPs degraded to corresponding PFCAs, for example 8:2 diPAP degraded into PFOA.

Biodegradation of n:2 diPAP starts with cleavage of the phosphate ester bond resulting in production of n:2 monoPAP and n:2 FTOH (Lee et al. 2010) (fig. 3). Further cleavage of the phosphate ester bond of n:2 monoPAP will result in n:2 FTOH. Subsequent degradation then follows the FTOH degradation pathway, which has been thoroughly studied in soil, sludge, and microbial culture (Dinglasan et al. 2004, Liu et al. 2010, Kim et al. 2014a). The n:2 FTOH is oxidized to a n:2 fluorotelomer aldehyde, which is further oxidized to n:2 fluorotelomer saturated carboxylic acid (FTCA). Then n:2 fluorotelomer unsaturated carboxylic acid (FTUCA) is formed through microbial defluorination. Degradation then follows two different pathways. Further defluorination of n:2 FTUCA

yield either n-1:2 sFTOH or n-1:3 FTUCA. The terminal product of n-1:2 sFTOH will be C_n PFCA. In the other pathway, C_{n-2} PFCA and C_{n-1} PFCA will be the degradation product from n-1:3 FTUCA. There are differences in the FTOH degradation pathway depending on the chain length. Similar molar yield of PFHxA and PFPeA from 6:2 FTOH has been observed in activated sludge, while in degradation of 8:2 FTOH in soil, PFOA was the major degradation product with only a small fraction of PFHpA (Liu et al. 2007, Wang et al. 2011).

In mammals as rat and mice, and in fish, degradation pathways have similarities with microbial degradation. The only difference is that 8:2 FTOH has shown to also produce PFNA through α -oxidation of 8:2 FTCA in rats, mice, and fish (Martin et al. 2005, Fasano et al. 2006, Butt et al. 2010). Degradation of 8:2 FTOH to PFCAs has been suggested to occur in humans, with PFOA as major metabolite (Nilsson et al. 2013).

Proposed biodegradation pathway of 6:2 FTSA involves desulfonation as an initial step (fig. 3), and a subsequent pathway much in similar with FTOH degradation pathway (Wang et al. 2011). In the degradation pathway suggested by Wang et al., 1-hydroxy 6:2 FTS is formed after desulfonation of 6:2 FTS, followed by rapid conversion to 6:2 fluorotelomer aldehyde, bypassing the formation of 6:2 FTOH. 6:2 FTSA in activated sludge has shown to result in formation of PFPeA and PFHxA. The FTSAs can also be formed from degradation of 6:2 fluorotelomermercaptoalkyl-amido sulfonate (FTSAS), an ingredient in AFFF, which has been demonstrated in sludge biodegradation experiments (Weiner et al. 2013). The proposed pathway involves S-dealkylation of 6:2 FTSAS by P-450 to 6:2 fluorotelomer thiol, which can be further oxidized to 6:2 FTSA. Alternatively, oxygenation of 6:2 FTSAS yields 6:2 FTSAS sulfoxide, and subsequently cleavage of the C-S bond finally yields 6:2 FTOH, which can be further degraded into PFCAs.

PFPiAs have shown to degrade to corresponding PFPA, and PFPA/PFPiAs have also been suggested to be potential PFCA precursors (Lee et al. 2012). However, in a study of PFPA/PFPiA biotransformation in rainbow trout, no PFCAs above LOD were found (Lee et al. 2012).

In atmospheric degradation of n:2 FTOH, oxidation of n:2 FTOH leads to formation of n:2 FTAL, which will be further degraded to equal amounts

of C_n PFCA and C_{n+1} PFCA and to a lesser extent homologues other than C_n PFCA (Ellis et al. 2004). The proportional yield thus differs between biotic and atmospheric degradation, and it has been hypothesized that an odd-even pattern in remote places is a result of atmospheric degradation, followed by increased bioaccumulation with increasing chain length (Martin et al. 2004, Bossi et al. 2005).

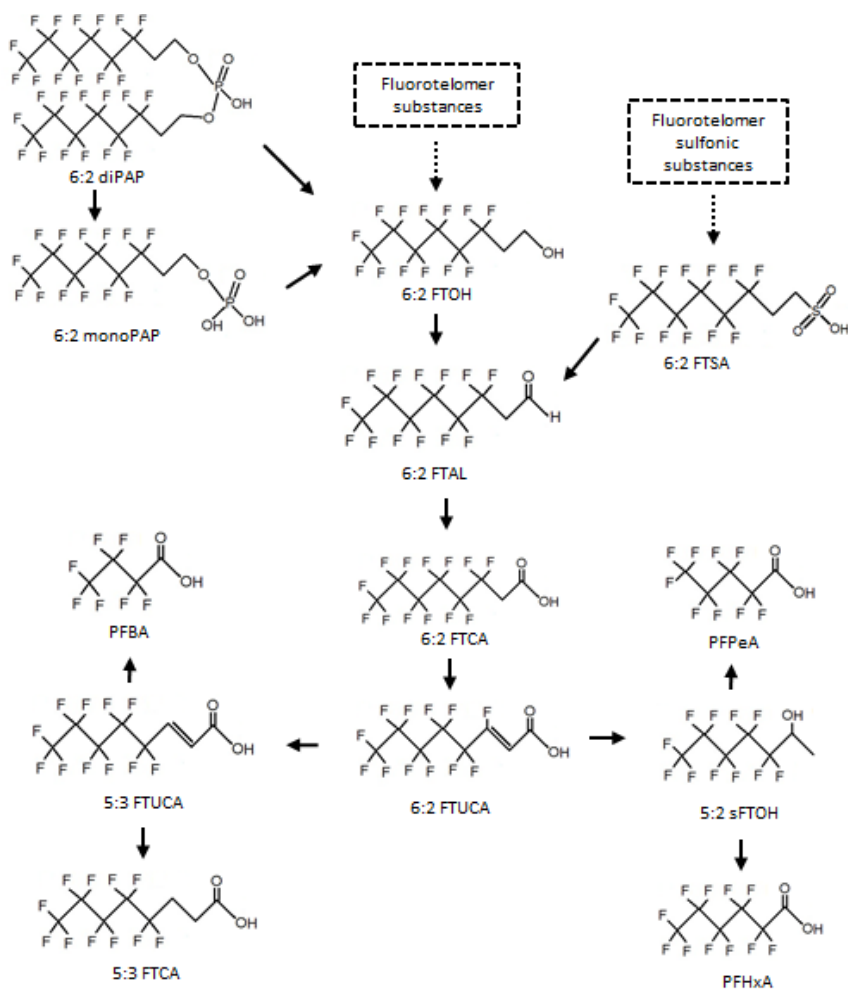


Figure 3. Combined simplified biodegradation pathways for 6:2 diPAP, 6:2 monoPAP, 6:2 FTSA, and 6:2 FTOH (Dinglasan et al. 2004, Lee et al. 2010, Liu et al. 2010, Wang et al. 2011, Kim et al. 2014a)

1. 5. Exposure pathways and sources

PFASs are spread in the environment through point sources and diffuse sources. Point sources are production facilities, industrial facilities that utilize PFASs in their production, domestic waste water treatment plants (WWTPs), landfills, military sites, airports, and firefighting practicing

grounds. Recently, the magnitude of PFAS pollution from such point sources was highlighted in a comprehensive study from the US, where harmful levels of PFOA in drinking water were associated with emissions from industrial sites, military firefighting training areas, airports, and WWTPs, affecting 6 million people (Hu et al. 2016).

A number of studies have reported high levels of PFASs in fish, water, waste water, soil, and sediment in surroundings of manufacturing and production sites (Wang et al. 2010, Bao et al. 2011, Oliaei et al. 2013). For example, groundwater levels of up to 82 µg/L PFOA and 31 µg/L PFOS has been found at a production site in the US (Oliaei et al. 2013), and PFOA levels of up to 48 ng/g and 668 ng/L has been found in river sediment and water close to a fluorochemical plant in China (Bao et al. 2011).

Fire fighting practicing areas are a significant point source, when PFAS-containing AFFF is used as extinguishing. The use of AFFF at fire fighting practicing areas has led to severe contamination of soil and groundwater worldwide (Gewurtz et al. 2014, Houtz et al. 2016). Elevated levels of PFOS, 6:2 FTSA, PFPeA, and PFHxA in waste water have been linked to AFFF sources connected to the WWTPs (Houtz et al. 2016). Studies of former fire fighting practicing areas have revealed that PFASs continue to be released to the surroundings for a long time, even decades (Ahrens et al. 2015, Arias et al. 2015, Filipovic et al. 2015)

Waste water treatment plants are a source for PFASs to water, sludge, and the atmosphere. Increased levels of PFASs in the effluent compared to the influent have been attributed to degradation from precursor compounds, such as PAPs, FTSAs, FTOHs, FOSAs, and FOSEs (Schultz et al. 2006). In the WWTPs, PFASs will be distributed to different environmental compartments depending on their physico-chemical properties. The more water-soluble short-chain PFASs will be distributed mainly to the water phase, while long-chain PFASs are more prone to sorb to the sludge. Volatile PFASs are emitted to the atmosphere, and FTOHs has been recognized as the dominant PFAS class released to the atmosphere from waste water treatment facilities (Ahrens et al. 2011a). However, a recent study reported similar atmospheric emission of PFCA/PFSAs as for FTOHs (Yao et al. 2016). Most WWTPs are not designed for efficient removal of PFASs. Adsorption to activated carbon, nanofiltration membranes, and advanced oxidation processes have shown to be effective for removal of PFOS and

PFOA (Arvaniti and Stasinakis 2015). Other examples of processes that have been evaluated are reverse osmosis and reduction processes. However, short-chain PFASs are not effectively captured by these techniques. While the aquatic environment is affected by the release of waste water, sludge will end up in landfills or is used as bio-solids. Application of sludge in agriculture contributes to further spread of PFASs through plant uptake (Yoo et al. 2011, Lee et al. 2014). Upon disposal of PFASs in landfills, there is a risk of leaching to the environment through migration in soil, reaching the groundwater, and emissions to the atmosphere (Ahrens et al. 2011a).

Except from point source emissions, humans and environment are subjected to diffuse exposure, which are usually related to as inputs from mainly dry- and wet deposition, oceanic currents, but also urban runoff from streets (Ahrens 2011).

The contribution from urban runoff is less studied compared to the release from WWTPs, but emissions have been estimated to contribute equally as WWTPs to PFAS mass load in urban rivers (Zushi and Masunaga 2009).

Currently, there are two hypotheses regarding the transport of PFASs; oceanic transport and atmospheric transport. Oceanic transport is a slow process, where it can take years for pollutants to be transported to remote regions (Armitage et al. 2006). Volatile precursors such as FTOHs and FASAs, can be atmospheric transported long distances and have an atmospheric lifetime of several weeks before degradation (Ellis et al. 2003, Martin et al. 2006). The relative importance between oceanic and atmospheric transport of PFAS to remote areas has been estimated, and PFOA and PFNA have been suggested to be predominantly oceanic transported, while atmospheric transport and precursor degradation were found to be of more significance for PFDA, PFUnDA, PFDoDA and PFTrDA (Armitage et al. 2009). In a global survey of soil from areas with no evident human impact, the homologue pattern of PFASs indicated significant contribution from atmospheric degradation of precursors, based on the ratio of PFOA/PFNA, supporting the hypothesis that atmospheric transport is the dominant pathway for the rural and terrestrial environment (Rankin et al. 2016).

Atmospheric transported PFASs have been found to be a significant source for contamination in the aquatic environment, with similar or higher con-

tribution compared to emissions from waste water (Muller et al. 2011, Kim et al. 2014b). The relative importance of diffuse emissions compared to WWTPs have shown to be related to population density, even at sites with no known PFAS industry activity, and diffuse emissions became more important in less densely populated areas.

In wild-life, animals are subject to PFAS exposure from food and water through point-source emissions, and long-range atmospheric and oceanic transport. Higher levels of PFAS in the terrestrial environment compared to agrarian and close to conurbation environments have been observed in roe deer, suggesting diffuse atmospheric sources to be the dominant exposure pathway for terrestrial mammals (Falk et al. 2012).

Food, drinking water, and indoor environment have been identified as the major exposure pathways to PFASs for the general population (Vestergren and Cousins 2009, Domingo 2012). Of these sources, food has been regarded to be the major contributor, especially fish, seafood and meat (D'Hollander et al. 2010, Domingo 2012, EFSA 2012). As mentioned previously, food items such as vegetables can be contaminated by PFASs through soil and water during growth. Plants have shown to take up PFASs from soil in greenhouse and field experiments, especially short-chain PFASs (Lee et al. 2014). Animals used for food production take up PFASs through ingestion of contaminated food and water, and many PFASs bioaccumulate along the food chain. Contamination can also occur through migration from food packaging material (Begley et al. 2005).

Drinking water is another important exposure pathway. Elevated PFAS concentrations in humans have been linked to contaminated point sources such as firefighting practicing grounds, but there are also reports suggesting that even low water PFAS concentrations could implicate a risk to human health (Post et al. 2012, Weiss et al. 2012, Hu et al. 2016).

Indoor environment has been recognized to be a significant route of exposure (Bjorklund et al. 2009). Route of exposure is mainly through dust for ionic PFASs and through air for neutral, volatile PFASs. The uptake from dust is believed to occur mainly through ingestion, and the dermal uptake is only a small fraction of the total dust intake (Lorber and Egeghy 2011).

Although exposure assessments suggested rather strong positive correlation between PFOS levels in serum and diet, the association between PFOA in serum and food intake was found to be weaker, implying other important exposure pathways for PFOA (Fromme et al. 2007, De Felip et al. 2015). Exposure assessments of relative contribution from different pathways to the total exposure have found that food is of more importance for PFOS compared to PFOA (Gebbink et al. 2015a). Other pathways such as air and dust were of more significance for PFCAs than for PFOS, for example the contribution from precursor compounds in dust was the dominant pathway for PFOA and PFNA in a high exposure scenario.

Exposure and elimination pathways differ among men, women, and children. During pregnancy, PFASs are transferred through the placenta to the fetus, which is a sink for the mother but a source for the fetus (Manzano-Salgado et al. 2015). After giving birth, PFASs continues to be transferred through lactation (Karrman et al. 2007). Women eliminate PFASs through menstruation (Wong et al. 2014). Children spend more time on the floor and are therefore more exposed to dust. Therefore, when conducting exposure assessment, it is important to consider all these factors.

1. 6. Toxicity

PFCAs and PFSAAs are extremely persistent and are well absorbed in the gastrointestinal tract after ingestion and are distributed mainly to the serum, liver and kidney, where they bind to serum albumin and fatty acid proteins. Persistent PFASs are not metabolized, they are excreted mainly through urine and to a lesser extent in feces. The adverse effects of PFASs include immunotoxicity, developmental toxicity, neurotoxicity, hepatotoxicity, tumor induction, weight loss, and endocrine disruption (DeWitt 2015).

Epidemiological studies have linked some PFASs to kidney and testicular cancer (Barry et al. 2013), low birth weight (Darrow et al. 2013), immune dysfunction (Grandjean et al. 2016a), thyroid disease (Melzer et al. 2010), reduced fertility for women (Fei et al. 2009), early menopause (Taylor et al. 2014), and increased cholesterol levels (Nelson et al. 2010).

The toxicity varies among PFASs depending on their structure, such as degree of fluorination, chain length, and active head group. Most toxicological studies have focused on PFOS and PFOA.

The risk of precursor compounds though are not only their potential to degrade into persistent and documented toxic PFASs, but also the harmful effects the precursor themselves may cause, and additionally the potential harmful effects from intermediates formed during degradation. Limited data on toxicity are available for precursor compounds. One study has shown endocrine disruption potential for PAPs in terms of inhibited male sex hormone synthesis (Rosenmai et al. 2013). In a H295R steroidogenesis assay, 8:2 monoPAP and 8:2 diPAP decreased levels of testosterone, dehydroepiandrosterone, androstenedione, and increased levels of estrone. Additionally, aromatase mRNA expression increased with 8:2 monoPAP and 8:2 diPAP, which could be a contribution factor to increased estrogen and decreased androgen levels.

PFPAs have shown to induce changes in apolipoprotein A-IV, related to the fatty acid metabolism, with greater effect than those of PFOS and PFOA in rat hepatoma cells (Jones et al. 2010). PFDPA was the most potent of three PFPAs tested.

The potential toxicity of intermediates and metabolites formed during degradation of precursor compounds have shown to have a reversed relationship with chain length, compared to PFCAs and PFSA. While in general toxicity increase with chain length, FTALs and FTUALs have been reported to have enhanced toxicity for homologues with shorter chain length, where 6:2 FTAL was more toxic for human liver cells than 8:2 FTAL, and 6:2 FTUAL was more toxic than 8:2 FTUAL (Rand et al. 2014).

1. 7. Temporal trends in humans

The use of temporal and spatial monitoring of PFASs in humans and environment are an important tool to elucidate the effects of regulations and changes in production and consumption. After the early report of PFOS in general population, increased attention and concern led to several biomonitoring studies of PFASs (Hansen et al. 2001). Retrospective studies have shown that PFAS levels increased globally in humans since the 1970-80s

until year 2000 (Haug et al. 2009, Sundstrom et al. 2011, Schroter-Kermani et al. 2013, Yeung et al. 2013b, Yeung et al. 2013c).

After year 2000 the trends differ among homologues, and spatial variations can be observed for most PFAS homologues. For PFOS, the levels started to decrease around 2000 in North America, Norway, Sweden, and Australia, which could be linked to the phase-out by the 3M Company (Calafat et al. 2007, Olsen et al. 2008, Haug et al. 2009, Glynn et al. 2012). For instance, the PFOS median level in serum decreased from 30 ng/g to 21 ng/g between 1999/2000 and 2003/2004 in the US (Calafat et al. 2007). In contrast, no change in PFOS levels have been observed in Korea between 1994 and 2008 (Harada et al. 2010). PFOS precursors have been observed in human serum and generally seem to follow the same declining trend as PFOS (Yeung et al. 2013c, Gebbink et al. 2015b).

The levels of PFOA have decreased globally since 2000 at a slower rate than PFOS, whereas an increase has been observed for long-chain (C9 – C11) PFCAs (Calafat et al. 2007, Olsen et al. 2008, Kato et al. 2011, Nost et al. 2014, De Felip et al. 2015). The trend may be attributed to the increased production of fluorotelomer compounds and contribution from fluorotelomer-based precursors. A shift in trends have been noted around 2006. Some studies reported a peak in PFNA concentrations at this time point, and also the increasing trends of PFDA and PFUnDA seem to slow down (Nost et al. 2014, Toms et al. 2014). This could be related to the initiation of the Stewardship program.

The PFSA and PFCA profile in human serum around year 2000 was generally dominated by PFOS, followed by PFOA>PFHxS>PFNA (Nost et al. 2014, Toms et al. 2014, Gribble et al. 2015). This profile has gradually altered, and long-chain PFCAs (>C8) are becoming relatively more important.

Analysis of total fluorine (TF) and extractable organic fluorine (EOF) has revealed that a large proportion of PFAS in human serum cannot be explained by the PFAS compounds generally included in biomonitoring studies (Yeung et al. 2008). A proportion of only 30 – 70% of TF could be explained by PFCAs, PFSAs, and PFOSA in Chinese serum samples from 2004. Precursor compounds could be part of the unknown PFASs. In 2009 diPAPs were detected for the first time in human serum samples from the US (D'eon et al. 2009). More recently, human serum from Germany, Chi-

na, and Sweden have been analyzed for diPAPs (Yeung et al. 2013b, Gebbink et al. 2015b, Yeung and Mabury 2016). While diPAPs were detected in European samples, no diPAPs were found in the Chinese samples, which indicates geographical differences in PFAS exposure patterns. In German samples, temporal trends between 1982 and 2009 were assessed, revealing increasing levels of long-chain PFCAs and decreasing trend for PFOA in recent years (2000 – 2009), and no change in concentration for diPAPs. Another emergent PFAS class, PFPiA, was detected in 2011 in human serum (Lee and Mabury 2011).

The FTSAAs were detected for the first time in human serum in 2002, and have later been detected in serum from US, Germany, and China (Connolly et al. 2002, Lee and Mabury 2011, Yeung and Mabury 2016).

In general, low PFCA precursor levels in human serum have been reported. That does not rule out the possibility that precursors are of significance for human exposure, since they may have been readily biodegraded to corresponding PFCAs.

1. 8. Temporal trend in the environment

Concerns about the impact of PFASs to the environment arose in 2001, when global PFOS contamination in wild-life was reported for the first time (Giesy and Kannan 2001). Environmental contamination can be tracked back to the 1950s. Analysis of sediment cores from Canada has shown that PFOS can be observed with beginning in 1952, whereas FOSA and long-chain PFCAs (C8 – C10) appear first in 1970s. PFAS concentrations continues to increase during the whole study period until 2005 (Yeung et al. 2013b). The patterns observed in the sediment cores are in temporal agreement with appearance of new compounds entering the PFAS market and industry, but are not reflecting the turning point for ceased production of PFOS by the 3M Company.

In wildlife, a number of species of fish, birds, and mammals have been studied, especially in the aquatic environment and in the northern hemisphere. Various PFAS trends have been reported, but a common trend is a significant increase of PFOS since 1970s up to around 2000 (Paul et al. 2009). In the marine environment, increasing trends in polar bears, guillemots, and pilot whales were reported (Smithwick et al. 2006, Holmstrom et al. 2010, Rotander et al. 2012). Geographical variations

where also observed within the same specie; as for herring gull where levels increased in the Baltic Sea but not in the North Sea (Rudel et al. 2011); and for ringed seals where levels increased between 1994 and 2003 in East Greenland, but not in West Greenland (Bossi et al. 2005). In the terrestrial environment, increasing trend has been less apparent compared to the marine environment; for instance in roe deer where levels slightly increased between 1989 and 2001 (Falk et al. 2012).

After the 3M Company's phase-out, there are various trends of PFOS in the environment. For some species and locations, PFOS has been reported to decrease after 2000, as in roe deer (Falk et al. 2012), and sea otter (Hart 2009 temporal trends). A peak around 2000 has been observed for ringed seals and polar bears (Riget et al. 2013). In other cases, little change in PFOS levels are observed, for example in seals, dolphins, and whales, and in golden eagles from the terrestrial environment (Rotander et al. 2012, Herzke et al. 2014). Increasing levels has also been reported, for example in otters from Sweden the PFOS levels continuously increased from 1972 to 2011 (Roos et al. 2013).

The trends of PFCAs are more complicated than that of PFOS. Levels increased between 2000 and 2006 – 2008; after that levels of PFOA and PFNA declined, while other long-chain PFCA levels either stabilized or increased. Increased levels of long-chain PFCAs since 2000 have been observed in the marine environment, for example in seals, polar bears, harbor porpoise, and whales (Routti et al. 2011, Huber et al. 2012, Rotander et al. 2012, Riget et al. 2013). In the terrestrial environment, increasing levels of long-chain PFCAs are also observed, as for tawny owl from Norway and otters from Sweden (Ahrens et al. 2011b, Roos et al. 2013). On the other hand, no apparent trend was observed for PFNA and PFDA in European roe deer after 2001 (Falk et al. 2012).

Only a few studies have reported PFCA precursors in wildlife, therefore temporal trends for these compound classes are lacking. However, PAPs and FTSA's have been found globally in the environment. In the last decade, PAPs have been observed in lake trout from Canada sampled in 2009 (Guo et al. 2012), in mussels from Spain in 2009, in benthic worms from HongKong in 2011 (Loi et al. 2013), in zooplankton from Baltic Sea (Gebbinck et al. 2016a), and in tuna from Indian Ocean in 2013 (Zabaleta et al. 2015). On the other hand, PAPs were not detected in fish from Baltic

Sea. The 6:2 FTSA has been found in ice amphipod sampled in 2004 (Haukas et al. 2007), in benthic worms from HongKong in 2011 (Loi et al. 2013), and in fish from a contaminated area in Norway in 2011 (Karrman et al. 2011).

Global measurements of FTOHs in the atmosphere between 2006 and 2011 have shown increasing levels of 6:2 FTOH (Gawor et al. 2014). While 10:2 FTOH decreased during these studied periods, 8:2 FTOH initially declined but returned to initial level in 2011. Levels of MeFOSA and MeFOSE decreased between 2006 and 2011.

Besides detection of PFCA precursors, product change towards short-chain PFASs is also reflected by recent reports on PFBS and PFBS precursors in the environment. Levels of PFBS in cetacean have increased in the South China Sea between 2002 and 2014 (Lam et al. 2016). PFBS has been found in mammals in Greenland (Gebbink et al. 2016b). Fish from Canada and Europe has been found to have levels of perfluorobutane sulfonamide (FBSA) at up to 80 ng/g (Chu et al. 2016). In addition, methyl perfluorobutane sulfonamidoethanol (MeFBSE) and methyl perfluorobutane sulfonamide (MeFBSA) have been detected in the atmosphere over the North China Sea, at comparable levels as PFOS precursors (Lai et al. 2016).

To summarize, a scarce number of studies reports about emerging PFASs, including precursors, and observations made so far witness of global occurrence and distribution in various types of environmental compartments.

2. Aim and objectives

The aim of the thesis was to assess if polyfluoroalkyl precursor compounds are a significant contributor to PFCA exposure, both environmental and human exposure. Such exposure data of other compounds besides persistent perfluoroalkyl substances is an important basis for risk assessments and not least for regulatory and policy work aiming at reducing hazardous chemicals in the society. The hypothesis of the work was that current monitoring and regulatory efforts are insufficient to protect humans and the environment from PFASs. By a cross-section analysis of the biosphere and technosphere the contribution of precursor compounds in relation to persistent PFASs in humans and the environment can be assessed. The specific objectives were:

- Assess human levels of semi-persistent precursor compounds in relation to persistent PFAS by analysis of sera
- Assess human exposure of semi-persistent precursor compounds in relation to persistent PFAS from indoor environment by analysis of household dust
- Assess the release of semi-persistent precursor compounds in relation to persistent PFAS from households to the environment by analysis of waste water and sewage sludge
- Assess the environmental exposure of semi-persistent precursor compounds in relation to persistent PFAS by analysis of wild bird eggs

3. Methods

The methods used are based on analysis protocols previously described with modifications to suit a broader range of substances (Powley et al. 2005, Taniyasu et al. 2008). The sampling and handling of the different sample matrices and detailed analytical procedures are given in Papers I-IV. Presented here is a schematic overview of the analytical methods used (Table 1) together with brief method descriptions.

Table 1. Method description of sample amount, pretreatment, extraction, clean-up, LC column, solvent extract composition.

	Dust	Eggs	Serum	Water	Sludge
Sample amount	0.1 g	0.25 g	1 mL	500 mL	0.25 g
Pre-treatment	NaOH			Filtration	
Extraction	MeOH	Acetonitrile	Acetonitrile	Oasis SPE-WAX	NaOH in Methanol
Clean-up	Oasis SPE-HLB Oasis SPE-WAX ENVI-Carb	ENVI-Carb			Oasis SPE-WAX
Extract composition	40% methanol	40% acetonitrile	40% acetonitrile	40% methanol	40% methanol
PFCAs, PFSA, FTSAs					
FTCAs, FTUCAs,	40% methanol	40% acetonitrile	-	40% methanol	40% methanol
FOSAs, FOSEs	40% methanol	40% acetonitrile	-	80% methanol	80% methanol
PFPAs, PFPiAs	-	-	-	40% methanol	40% methanol
diPAPs	80% methanol	80% acetonitrile	80% acetonitrile	80% methanol	80% methanol
monoPAPs	100% methanol	-	-	80% methanol	80% methanol
triPAPs	80% methanol-		-	-	-

3. 1. Extraction

3. 1. 1. Dust

Dust was sieved before extraction using 150 μm mesh size and 0.1 g was used in the analysis. The samples were spiked with isotopic labelled standard, followed by addition of 100 μL 0.2 M sodium hydroxide that were left soaking for 30 min; after which extraction with 5 mL of methanol was performed followed by neutralization with an addition of 20 μL 1 M hydrochloric acid. The extracts were ultrasonicated for 15 min and shaken for 30 min. The extraction, sonication and centrifugation steps were repeated once using 2 mL methanol. Clean-up was performed with solid phase extraction (SPE) using Oasis WAX cartridges (Waters, 150 mg, 6 mL, 30 μm) and Oasis HLB cartridges (Waters, 6 mL, 200 mg, 30 μm) coupled in tandem during the whole extraction procedure. Prior to SPE, the sample extracts were adjusted to a 20% methanol content by addition of Milli-Q water. Conditioning of the sorbents were performed with 4 mL of methanol followed by 4 mL of water. The samples were loaded on the SPE cartridges and which were subsequently washed with 4 mL of 25 mM ammonium acetate buffer solution (pH 4) and 4 mL 20% methanol in water, all discarded. The analytes were eluted with 4 mL of methanol followed by 4 mL of 2% ammonium hydroxide in methanol, combining neutral and anionic analytes. Further clean-up was performed with the additions of 50 mg of ENVI-carb and 100 μL glacial acetic acid. The extracts were evaporated under nitrogen to a volume of 200 μL , then filtered through 0.2 μm hydrophilic propylene filters, and split into three fractions for further analysis. The fraction's methanol content was adjusted to 40%, 80%, and 100% with 2mM ammonium acetate (aq), respectively in which the different PFAS classes were analyzed according to Table 1.

3. 1. 2. Bird eggs

In the extraction of bird eggs, 0.25 g sample was spiked with isotopic labelled standards, and then extracted with 4 mL of acetonitrile with ultrasonication for 15 min, shaking for 15 min, and centrifugation. The supernatant was collected and the extraction step was repeated with 4 mL of acetonitrile. The combined supernatants were mixed with 50 mg ENVI-carb and 100 μL glacial acetic acid. The extracts were then evaporated under nitrogen and filtered using 0.2 μm hydrophilic propylene filters. Evaporation to a final volume of 200 μL was performed, and the final extract was split into one fraction with a final composition of 20% 2mM

ammonium acetate (aq) and 80% acetonitrile for analysis of diPAPs, and one fraction with a final composition of 60% 2mM ammonium acetate (aq) and 40% acetonitrile for analysis of PFCAs, PFSA, FTCAs, FTUCAs, FOSAs, FOSEs, and FTSA.

3. 1. 3. Serum

Serum extraction was performed using 1.0 mL serum which was spiked with isotopic labelled standards. An amount of 4 mL acetonitrile was added and the samples were vortexed, ultrasonicated for 15 min, and shaken for 15 min. The extraction step was repeated with 4 mL of acetonitrile, and the supernatant aliquots were combined. The extracts were filtered (0.2 μ m hydrophilic propylene filters) and evaporated under nitrogen to a final volume of 50 μ L. The extracts were split into two fractions; one with a final composition of 20% 2mM ammonium acetate (aq) and 80% acetonitrile for analysis of monoPAPs and diPAPs, and one fraction with a final composition of 60% 2mM ammonium acetate (aq) and 40% acetonitrile for analysis of PFCAs, PFSA, and FTSAs.

3. 1. 4. Water

The water samples (0.5L) were first filtered using GF/B glass fiber filters (1 μ m mesh), and then extracted using SPE after addition of isotopic labelled standards. The sorbent used was Oasis WAX cartridges (Waters, 150 mg, 6 mL, 30 μ m). Before extraction, the sorbents were conditioned with 4 mL of methanol followed by 4 mL of water. The samples were passed through the cartridges at an approximate rate of one drop per second. After that, the sorbents were washed with 4 mL of ammonium acetate buffer solution (pH 4). Finally the analytes were eluted using 4 mL of methanol and collected, followed by 0.1% ammonium solution in methanol and collected separately. The extracts were evaporated to a final volume of 200 μ L, filtered (0.2 μ m hydrophilic propylene filters), and the anionic fraction was split into one fraction with a final composition of 20% water and 80% methanol for analysis of monoPAPs and diPAPs, and one fraction with a final composition of 60% water and 40% methanol for analysis of PFCAs, PFSA, FTCAs, FTUCAs, FTSAs, PFPAs, and PFPiAs. The neutral fraction was adjusted to a final composition of 20% 2mM ammonium acetate (aq) and 80% methanol used for analysis of FOSAs and FOSEs.

3. 1. 5. Sludge

The sludge samples were freeze-dried and an amount of 0.25 g dried material was spiked with isotopic labelled standards. Before extraction, 2 mL 1 M sodium hydroxide in methanol was added to the sample. The samples were ultrasonicated for 15 min, shaken for 15 min, and centrifuged. The extraction was repeated twice with 2 mL of methanol, and the aliquots were combined. Clean-up was performed with Oasis WAX cartridges (Waters, 150 mg, 6 mL, 30 μ m). The sorbents were conditioned with 4 mL methanol followed by 4 mL Milli-Q water, all discarded, and the extracts were thereafter loaded on the sorbents. The sorbents were then washed with 4 mL sodium acetate buffer solution (pH 4), followed by 4 mL of 20% methanol. The neutral analytes were eluted with 4 mL of methanol and collected, followed by elution of anionic analytes with 4 mL 2% ammonium hydroxide in methanol, collected separately. The extracts were filtered (0.2 μ m hydrophilic propylene filters) and evaporated under nitrogen to a final volume of 200 μ L. The anionic fraction was split into two fractions; one with a final composition of 20% 2mM ammonium acetate (aq) and 80% methanol for analysis of monoPAPs and diPAPs, and one fraction with a final composition of 60% 2mM ammonium acetate (aq) and 40% methanol for analysis of PFCAs, PFSAAs, FTCAs, FTUCAs, FTSAAs, PFPAs, and PFPiAs. The neutral fraction was adjusted to a final composition of 20% 2mM ammonium acetate (aq) and 80% methanol used for analysis of FOSAs and FOSEs.

3. 2. Instrumental analysis and quantification

An Acquity UPLC system coupled to a XEVO TQ-S (Waters Corporation, Milford, USA) mass spectrometer was used. A guard column (PFC isolator, Waters Corporation, Milford, USA) was inserted after the solvent mixer and before the injector to prevent contamination from the system. This guard column successfully retained all analyzed PFASs thus led to a contamination free instrumental analysis. The column used was 100 mm Acquity BEH C18 (2.1 mm, 1.7 μ m) except for monoPAP analysis of dust (Paper I) for which a 50 mm column was used. Mobile phases used for diPAPs and triPAPs analysis were water and methanol with addition of 2 mM ammonium acetate and 5 mM 1-methylpiperidine. MonoPAPs were analysed using water and methanol with addition of 5 mM 1-methylpiperidine for dust in Paper I, and water and methanol with addition of 2 mM ammonium acetate and 5 mM 1-methylpiperidine for sludge in Paper III. For PFCAs, PFSAAs, FTCAs, FTUCAs, FOSAs, FOSEs, PFPAs,

and PFPiAs, water and methanol with addition of 2 mM ammonium acetate were used. For each analyte, cone voltage and collision energy were optimized for molecular ion and product ions.

For analysis of the congeners of diPAPs and triPAPs, several transitions were monitored. Since triPAPs were also difficult to ionize in negative electrospray, insource fragmentation was therefore selected for analysis of triPAPs, and transitions for corresponding diPAPs were used for quantification. The transition corresponding to the loss of both alkyl chains was used for quantification and the transitions corresponding to the loss of one alkyl chain were used for qualification. Complete separation of the $[M-H]^- \rightarrow [PO_4H_2]^-$ transition was not achieved for all isomers, for example the peaks of 10:2 diPAP and 8:2/12:2 diPAP overlapped at more than 50% of peak height (fig. 4). Under these circumstances, it could be argued that the transition corresponding to the loss of one alkyl chain would be most suitable for quantification because this transition is unique for each isomer. However, since the loss of one alkyl chain may result in two product ions for $x:2/y:2$ homologues, the response for each product ion is comparable less than the fragment yielded from a $x:2/x:2$ homologue. Using only one of these product ions for quantification would lead to an underestimation in concentrations. The sum of the two product ions yielded from loss of one alkyl chain for $x:2/y:2$ were therefore used as qualification, and the transition corresponding to loss of two alkyl chains were used for quantification. This approach was later validated with the 6:2/8:2 diPAP standard when it became commercially available. Homologues with $x:2/x+2:2$ structures were quantified against their closest homologue where authentic standard was available, for instance 8:2/10:2 diPAP was quantified against 8:2 diPAP. For analysis of other PFASs, at least two transitions were monitored except for PFBA, PFPeA, PFHxPA, PFOPA, and PFDPA, where one transition was monitored.

10_2_diPAP

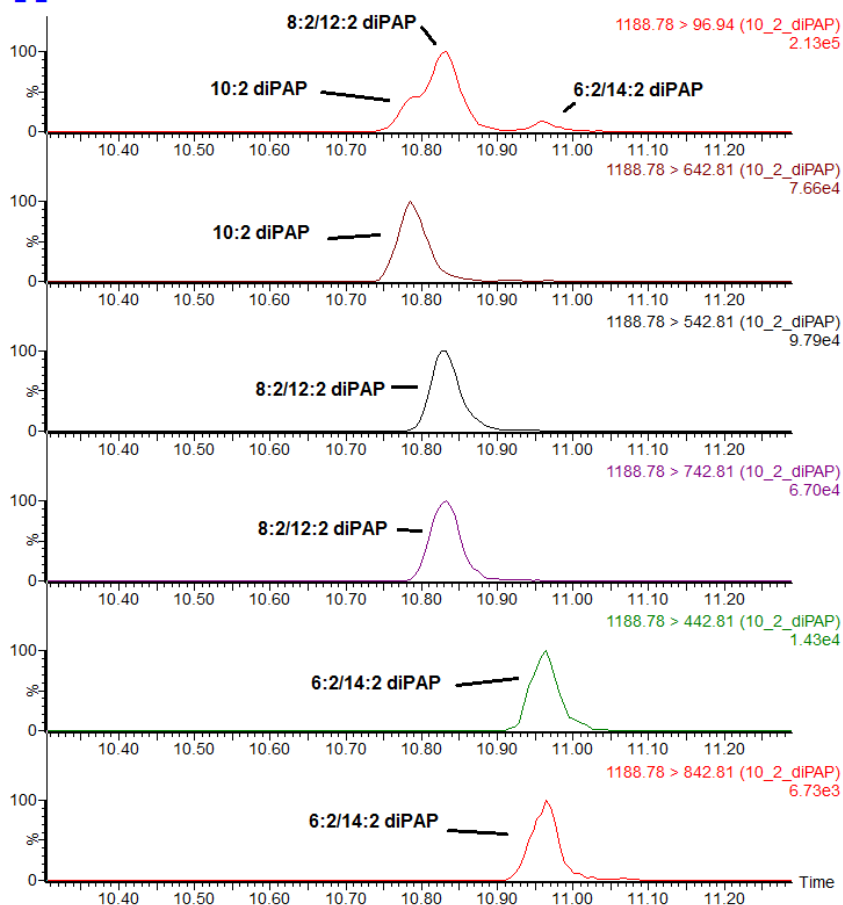


Figure 4. Chromatogram of 10:2 diPAP and its isomers 8:2/12:2 diPAP and 6:2/14:2 diPAP in a dust sample.

Isotopic labelled standards were used and will compensate for matrix effects and losses during extraction. However, labelled standards were not available for all PFASs, therefore matrix effects needs to be taken into consideration when quantifying PFASs using external standards in solvent. Calibration curves within a range of at least five points were run along with the samples. Criteria for limit of detection was a peak of at least 3:1 signal to noise, a concentration within the range of the calibration curve, and a concentration at least three times the blank concentration, or a

mean concentration of the blank levels with an addition of three times the standard deviations. The efficiency of extraction and clean-up were evaluated by comparing matrix effects, process efficiency, matrix recovery, precision, and accuracy. Matrix effects were assessed by the ratio of the area in a sample matrix, spiked with standards after extraction, against the area of standards in pure solvent. Matrix recovery were assessed by the ratio of a spiked sample matrix before extraction against a spiked sample matrix after extraction. Process efficiency were assessed by the ratio of a spiked sample matrix before extraction against standards in pure solvent, thus including matrix effects in the recovery. Accuracy were evaluated by the relative standard deviation of internal standard corrected concentration in spiked sample matrix, and precision as the deviation from the spiked amount.

3. 2. 1. Daily intake

The daily intake of PFAS was calculated from the serum levels in Paper II in a first-order pharmacokinetic (PK) model, using the formula below:

$$VD * (dCs/dt) = Dt - k * VD * Cs$$

VD is volume of distribution (mL/kg b.w.), Cs is the serum concentration (ng/ml), Dt is the total dose (ng/day/kg b.w.), k is the elimination rate constant ($k = \ln(2)/t_{1/2}$). The volume of distribution used was 170 mL/kg b.w. and the value was derived from the study of Thompson et al., which was calibrated from human serum and exposure data (Thompson et al. 2010). Half-time for PFOA used was 2.3 years and was derived from the study of Bartell (2010).

At steady state, $dCs/dt = 0$. The formula can then be resolved as:

$$Dt = k * VD * Cs$$

However, during an increase or decrease in concentration, there will be a non-steady state. The calculated dose from the formula above does not account for changes in exposure. In the serum samples, the contribution from the ongoing exposure is revealed by the discrepancy between the intrinsic half-time and the apparent halving-time. The apparent change over time is described as:

$$dCs/dt = -k_a * Cs$$

where k_a is the apparent elimination rate constant, and the dose is set to 0. The changes in concentration over time using the apparent halving-time can be calculated as follows:

$$D_t = V_D * C_s * (k_i - k_a)$$

This equation is then used to calculate the total dose.

3. 2. 2. Blank contamination

Contamination of PAPs and PFCAs were detected in the procedural blanks during method development. The laboratory environment can have several potential sources for contamination where diPAPs have been detected in a variety of common laboratory consumables, such as transfer pipets, pipet tips, and their containing boxes (Yeung et al. 2013b). Rinsing all lab ware including disposable glass pipettes with methanol, and sonication of propylene filters in 2% NH_4OH in methanol eliminated the diPAPs contamination, while the monoPAPs and the PFCAs weren't completely removed and were occasionally observed in the blanks.

4. Results and discussion

4. 1. Method development

Simultaneous extraction and analysis of different classes of PFASs is a key issue with the increasing number of PFASs that needs to be monitored and analyzed. Measuring a broader range of PFASs in various sample matrices is challenging, since different compound classes have different properties such as ionic strength and polarity, and the matrices differ in composition. To avoid ion signal effects, interferences in the matrix need to be removed during extraction and clean-up steps. Concurrently, the extraction must be efficient enough for quantitative analysis with minimal loss of the analytes.

4. 1. 1. Extraction and clean-up

Different solvents and solvent compositions were evaluated for the extraction of dust. Initially, methanol and acetonitrile were evaluated with satisfactory recoveries for PFCAs and PFSAAs. The process efficiencies were 208% for 6:2 diPAP and 99% for 8:2 diPAP with methanol, while only 13% for 6:2 diPAP and 1.8% for 8:2 diPAP were obtained using acetonitrile. Methanol showed, however, poor recoveries for the monoPAPs. Solvent mixtures have shown to provide more efficient extraction of PFASs (Ballesteros-Gomez et al. 2010). Solubility of a solvent is described by its Hildebrand solubility parameters, which is an overall measure of dispersion (δ_d), dipole-dipole (δ_p), and hydrogen bonding (δ_h) components. A combination of solvents with different solubility parameters allows for a wider range of polar interactions. Methanol, acetonitrile, and isopropanol were chosen due to their different forces of dipole-dipole and hydrogen bonding interactions. Methanol and acetonitrile have δ_h of 22.3 MPa^{1/2} and 6.1 MPa^{1/2}, respectively. Acetonitrile and isopropanol have δ_p of 6.1 and 18.0 MPa^{1/2}, respectively. Two solvent mixtures of methanol:acetonitrile 1:1 and acetonitrile:isopropanol 1:1 were evaluated. No improvement in monoPAP recoveries using these solvent mixtures were found. It was hypothesized that the monoPAPs bound strongly to particles in the dust, and pretreatment step with 100 μ l 0.2 M NaOH in methanol was added to loosen these bond. Additional clean-up step using solid-phase extraction was also added in an attempt to reduce observed suppression by the matrix. Several solid-phase extraction (SPE) sorbents were evaluated for the aim of simultaneously clean-up of a wider range of

PFASs. Initially, a weak anion exchange (WAX, Waters Corporation, Milford US) sorbent was tested in the extraction and clean-up procedure. Satisfactory recoveries were obtained for monoPAPs and diPAPs with the WAX sorbent. However, poor recoveries were observed for triPAPs (<1%). This was unexpected since WAX has previously been used in the extraction of triPAPs in sludge with acceptable recoveries (Liu 2013). The reason for the unexpected low recoveries for triPAPs on WAX remains unclear. Several other SPE sorbents were tested; quaternary amine (CUQAX, United Chemicals, Bristol UK), hydrophilic-lipophilic balanced (HLB, Waters Corporation, Milford US), and C18 (Sep-Pak, Waters Corporation, Milford US). The diPAPs were well recovered on all sorbents (61 - 99%). Recoveries of 6:2 triPAP were 22 - 43% and 8:2 triPAP 71 - 78% on CUQAX, HLB, and C18 sorbents, while triPAPs were not recovered on WAX sorbent. The monoPAPs on the other hand were well recovered on WAX (90 - 97%), but poor recoveries were obtained for other sorbents tested (7 - 17%). Therefore, the strategy applied was the use of both WAX and HLB cartridges coupled in series. Matrix recoveries using this extraction and clean-up method were 23-31% for monoPAPs, 48-86% for diPAPs, 30% for 6:2 triPAP. Recoveries for spiked amount corrected for with internal standards were 96% for 6:2 monoPAP, 123% for 8:2 monoPAP 84% for 6:2 diPAP, and 103% for 8:2 diPAP.

Several extraction solvents were evaluated for analysis of sludge; however the method development was carried out using a sediment sample free from detectable levels of PFASs, since PFAS free sludge sample was not available. Initially, methanol (4 mL) with pretreatment of 100 μ L 0.2 M NaOH was tested. Matrix recoveries were unsatisfactory for the PAPs; 35% for 6:2 diPAP and 11% for 8:2 diPAP, and monoPAPs were not recovered at all. It was hypothesized that the PAPs strongly sorbed to organic and mineral particles in matrices as sludge and sediment. The use of acetic acid in the extraction solvent has been found to increase recoveries of PFASs in sludge (Higgins 2005, Liu 2013). An extraction solvent composition of 1% acetic acid (HAc) in methanol (4 mL) resulted in recoveries of 9.3% for 8:2 monoPAP, 30% for 6:2 diPAP, and 14% for 8:2 diPAP. Still, 6:2 monoPAP was not recovered. An extraction solvent mixture of THF:HAc yielded similar results, with recoveries of 13% for 6:2 diPAP and 14% for 8:2 diPAP, and no recovery of the monoPAPs. Also PFPAs and PFPiAs showed low recoveries of 2.8 to 26% using HAc treatment. In a further attempt to extract the presumed strong bonds of the monoPAPs

to the matrix, the pH was increased in the solvent. Extraction with 2 mL 1 M NaOH in methanol and additionally 2 mL methanol resulted in improved recoveries for the PAPs; 78% for 6:2 monoPAP and 117% for 8:2 monoPAP, 26% for 6:2 diPAP, and 29% for 8:2 diPAP, and was chosen as the final method. This method was also proven to be efficient for PFPAs and PFPiAs, except for PFHxPA, with recoveries of 43 – 117%.

For the analysis of PFCAs and PFSA in serum, the use of protein precipitation and phospholipid removal well plates have proven to be a rapid and robust method (Salihovic et al. 2013), and was initially tested for the diPAPs. This method was found to be less efficient for the diPAPs (recoveries of 62% and 11% for 6:2 diPAP and 8:2 diPAP, respectively). In further method development, acetonitrile extraction was performed for the serum samples with satisfactory recoveries of 93% for 6:2 diPAP and 83% for 8:2 diPAP. The recoveries for PFCAs and PFSAs were all also satisfactory, 63 – 95%.

4. 1. 3. Matrix effects

Ion signal effects are commonly observed in analysis using electrospray ionization. Co-eluting compounds from the matrix affect the number of charges in the droplets formed in electrospray and subsequently affects formation of gas-phase ions from the analyte of interest. When the number of charges decrease, competition of charges in the electrospray droplets leads to suppression of the analyte. Reversely, an excess of charges facilitate ionization and leads to signal enhancement. Formation of gas-phase ions from an analyte in electrospray is also affected by its polarity and pKa (Hirabayashi 2012) and there is a large variety regarding physiochemical properties within the group of PFASs.

Matrix effects were evaluated by spiking samples with native standards after extraction and compare the response with a standard solution made in solvent (fig. 5, 6 and 7). A matrix effect of zero means that the ionization of a compound is not affected by co-extracted substances. In general, matrix effects were more pronounced for the compound classes monoPAPs, diPAPs, triPAPs, PFPAs, PFPiAs compared to PFCAs and PFSAs.

Dust was more affected by matrix effects compared to sludge and serum. Interfering compounds in biological and environmental matrices are main-

ly polar and elutes early, hence relatively polar PFASs are in general most affected by matrix effects (Bonfiglio et al. 1999). This can be seen with the PFCAs, for example PFBA is clearly affected by enhancement in the dust (72%) and serum (56%) samples and suppression in the sludge samples (-44%).

The monoPAPs on the other hand behaved differently, where signal enhancement increased with chain length in both dust and sludge. Severe signal enhancement was observed for monoPAPs in the dust samples; 86% for 6:2 monoPAP, 105% for 8:2 monoPAP, and 175% for 10:2 monoPAP. Also in the sludge samples, enhancement was observed for 8:2 monoPAP (16%) and 10:2 monoPAP (67%).

Matrix effects were less pronounced for the diPAPs compared to the monoPAPs. While moderate effects were observed for 6:2 diPAP and 8:2 diPAP in dust (16% and -9.3%, respectively), sludge (27% and 33%, respectively), and serum (-18% and 37%, respectively), 10:2 diPAP in all matrices and triPAPs in the dust were strongly suppressed.

In sludge, PFCAs and PFSAAs were relatively free from matrix effects, while suppression was observed for most analytes in dust and serum (fig. 5). With exception from early eluting PFBA and PFPeA, there are little variation in matrix effects between homologues of various chain lengths. No pronounced matrix effect was observed for the FTSAAs in sludge (4% - 25%), while 6:2 FTSA was enhanced in dust (219%). Severe suppression was observed for PFPAAs and PFPIAAs, with stronger suppression for PFPIAAs (-73% to -78%) compared to the more polar PFPAAs (-45% to -57%). Also FOSAAs and FOSEAAs suffered from suppression (fig. 5).

The strong matrix effects for phosphate compounds challenge the quantification of these compounds, since even closely eluted homologues can experience totally different ion signal effects. For example, the large discrepancy in matrix effects between 8:2 PAP and 10:2 PAP indicated that ¹³C-labelled 8:2 PAP would not be a suitable surrogate standard for 10:2 PAP quantification. A matrix-matched calibration is an option for isotope dilution when authentic standards are lacking. This is assumed that the sample matrix is homogeneous, free from the targeted analytes. Alternatively, standard additions for each individual sample could be used, given that native standards are available.

Covering a wide range of substances likely leads to quantification using non-authentic standards, for example with the large number of structural isomers of diPAPs and triPAPs. Semi-quantification using standards of other homologues or isomers is an approach by which more analytes can be covered when estimating total amounts of PFASs, though evaluating matrix effects is of outermost importance to assess reliability of these data.

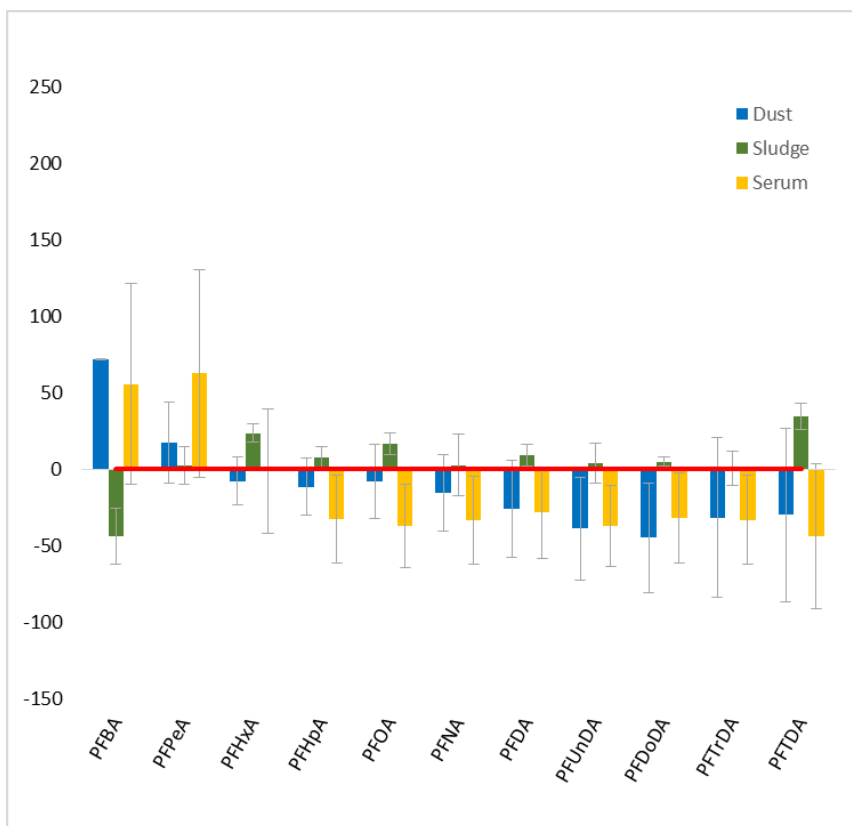


Figure 5. Matrix effects of PFCAs and PFSA in dust ($n=6$), sludge ($n=3$), and serum ($n=2$). Error bars displays standard deviation.

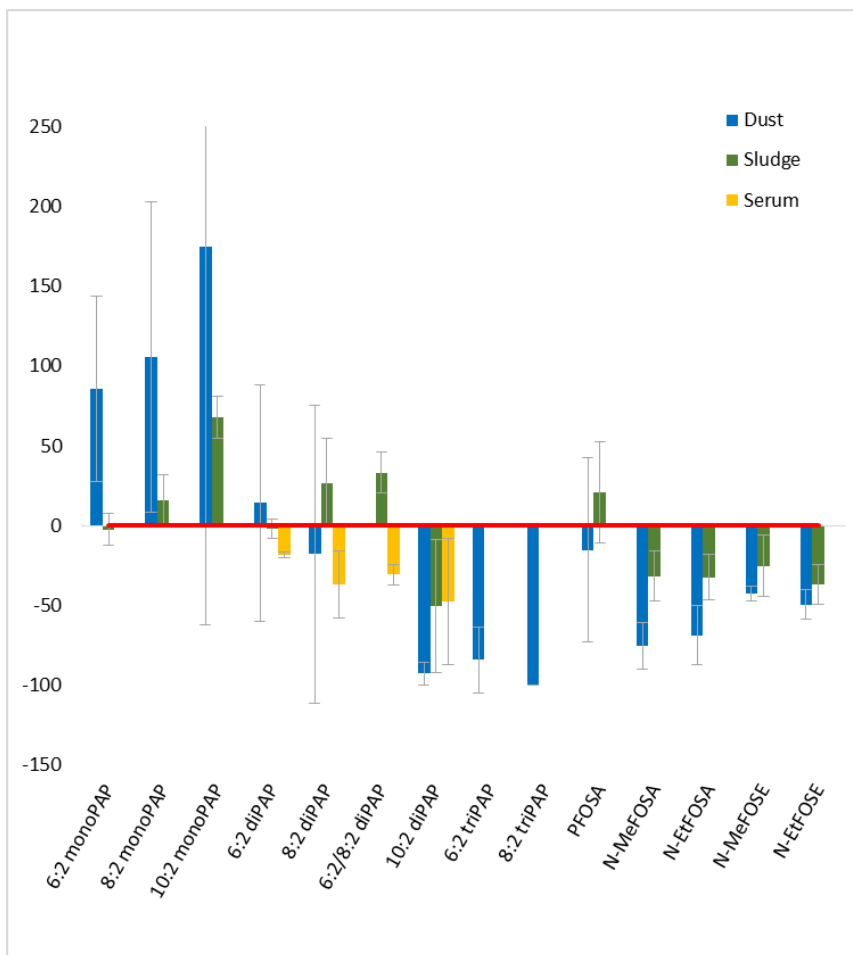


Figure 6. Matrix effects of monoPAPs, diPAPs, triPAPs, and FOFA/FOSEs in dust ($n=6$), sludge ($n=3$), and serum ($n=2$). Error bars displays standard deviation.

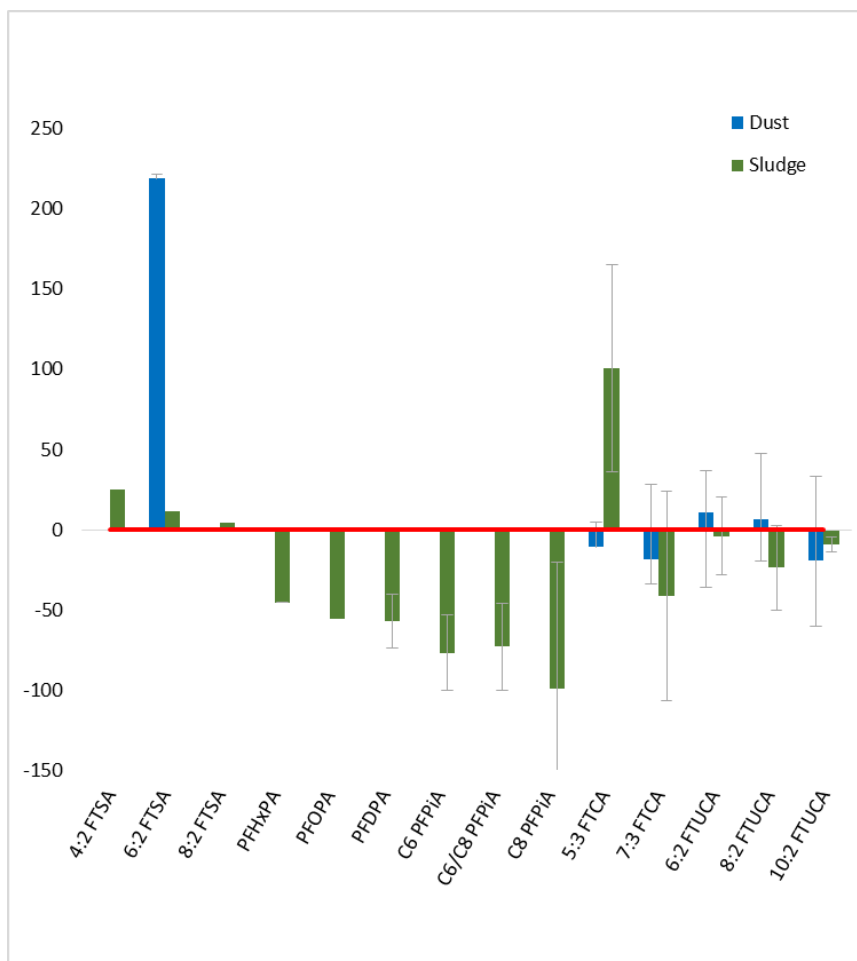


Figure 7. Matrix effects FTSAs, PFPA/PFPiAs, and FTCA/FTUCAs in dust ($n=6$) and sludge ($n=3$). Error bars displays standard deviation.

4. 1. 4. Composition in extracts

The high surface energy of PFASs make them stick to surfaces, as tubings and vial walls. The diPAPs in particular are prone to adsorb to surfaces. In an attempt to force more of the diPAPs out in solution and increase the sensitivity, different composition of organic and water composition were tested in the final extracts. The range of organic solvent tested were between 40 and 100%. A sharp increase was observed above 50% organic

solvent. However, exceeding an amount of 80% organic solvent resulted in poor chromatography with tailing and splitting peaks. A proportion of 80% organic solvent was therefore finally chosen for the extracts. The sensitivity also increased for the monoPAPs with higher proportion of organic solvent, and peak shape could be retained at proportion up to 100% organic solvent.

4. 2. Dust levels of precursors

The importance of different sources for the exposure of persistent PFASs can differ between compound classes and homologues. While food generally is considered to be the major pathway for human exposure, dust has been found to contribute relatively more to PFOA exposure compared to PFOS exposure (Fromme et al. 2007, Jogsten et al. 2012, De Felip et al. 2015).

The contaminant profile in the indoor environment reflects the amount and use of PFAS-containing consumer products in the general population. In Paper I, dust sampled from Canada, Faroe Islands, Sweden, Greece, Spain, Nepal, Japan, and Australia were analyzed for PAPs, PFCAs, PFASs, FTSAs, FOSA/FOSEs, and FTCA/FTUCAs. The results revealed a high contribution from PAPs to the total amount of PFASs in all countries. Including semi-quantified PAPs, the contribution of monoPAPs to Σ PFAS was 8 – 47%, and the contribution of diPAPs to Σ PFAS was 21 – 47%. Several triPAPs were detected, but due to matrix effects and lack of labelled internal standards, these could not be quantified. In total, monoPAPs and diPAPs accounted for 39 – 94% of Σ PFAS. In figure 8, the diPAP profiles in dust from the different countries are shown.

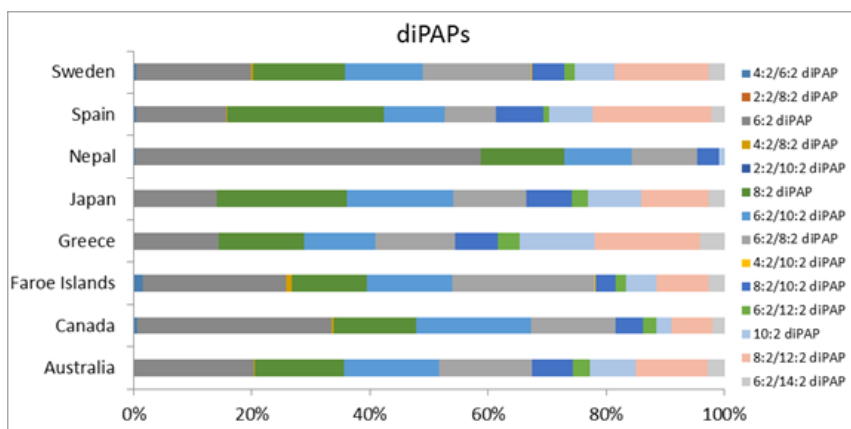


Figure 8. The profiles of diPAPs in indoor dust samples from different countries.

Most abundant diPAPs were 6:2 diPAP, 8:2 diPAP, 6:2/8:2 diPAP, and 6:2/10:2 diPAP. The highest median levels of 6:2 diPAP in the different

countries were 164 ng/g in Canada, 119 ng/g in Japan, and 106 ng/g in Faroe Islands, and the highest median levels of 8:2 diPAP in the different countries were 81 ng/g in Japan, 49 ng/g in Australia, and 47 ng/g in Faroe Islands. Prior to this study, diPAPs in dust have been analyzed in Canada, with levels 3 – 16 times higher compared to this study (De Silva et al. 2012). Recently, 6:2 diPAP and 8:2 diPAP have been found in dust samples from Norway at median levels somewhat higher but similar to the Swedish dust samples in this study (Padilla-Sanchez and Haug 2016).

The diPAPs and triPAPs have a large number of structural isomers. The profiles of the diPAP homologues reveal that a substantial proportion of Σ diPAPs are covered by structural isomers and homologues other than 6:2 diPAP, 6:2/8:2 diPAP, 8:2 diPAP, and 10:2 diPAP, for which authentic standards are available. Quantifying these structural isomers and homologues separately provides important information about total levels and homologue length patterns. For example, 8:2 diPAP has the isomer 6:2/10:2 diPAP. During degradation, 8:2 diPAP will yield PFOA as major PFCA product, while 6:2/10:2 diPAP will yield PFHxA and PFDA.

The PAPs were found in all countries, also those countries without known PFAS production, suggesting that the direct use of commercial products is an important source for human exposure. There was a large variation of PAP levels between countries, while the variation of PFCAs and PFSAs was less pronounced. Highest median level of Σ PAPs was found in Japan (1 720 ng/g), followed by Faroe Islands (900 ng/g), Australia (709 ng/g), Canada (658 ng/g), Sweden (167 ng/g), Greece (31 ng/g), Spain (29 ng/g), and Nepal (7.3 ng/g). These variations can be caused by several reasons. The age of the dust could differ substantially, and lead to different degrees of degradation of PAPs. Also, consumption pattern and PAPs-containing products available can differ between countries.

Second most abundant compound class was the PFCAs, with a contribution of 7 – 50% of Σ PFAS, followed by PFSAs with a contribution of 0.4 – 11%. In general, the levels of PFOA were higher than the levels of PFOS.

Correlations were found both within and between compound classes. When summarizing the compound classes, Σ PFCAs, Σ PFSAs, Σ monoPAPs and Σ diPAPs were all positively significantly correlated to each other. A vast majority of the diPAP homologues, as well as monoPAP homologues,

were correlated with each other, as expected since these compounds usually are produced in mixtures. Most PFCA and PFSA homologues were correlated with each other, especially within the short-chain (<C8) and long-chain (≥C8) homologues. Several positive significant correlations were also found between diPAP and PFCA homologues; as for 6:2 diPAP, 6:2/8:2 diPAP, 6:2/8:2 diPAP, 8:2/10:2 diPAP, 6:2/12:2 diPAP, and PFHxA, PFHpA, and PFNA. For 6:2 diPAP, PFHxA is one of the the major PFCA products formed during degradation. The association between PFOA and diPAPs on the other hand was less clear, significant correlations were only found between PFOA and 6:2/8:2 diPAP and 10:2 diPAP. It could be expected to find co-variance between PFOA and 8:2 diPAP, since PFOA is the major PFCA metabolite formed during degradation of 8:2 diPAP (D'Eon and Mabury 2007, Liu and Liu 2016). It is possible that lack of correlation is due to multiple sources of PFOA in indoor dust. When grouping the data per country, no significant correlations were found between precursor and persistent homologues.

4. 2. 1. Exposure assessment

In Paper I, the daily intake of PFCA via dust was calculated, both from direct and indirect PAPs exposure, in low exposure scenario and high exposure scenario. The limitations of the assessments are partly due to the unknown proportion of the quantified PFCAs that derives from already degraded precursor compounds. This could lead to a possible underestimation of indirect contribution. There are few studies of the biotransformation yield of PFCAs from PAPs which also contributes to the uncertainty in the assessment as well as possible unknown precursor compounds present in the dust which upon degradation could yield PFCAs.

In the high exposure scenario, a majority of the PFCA exposure via dust could be attributed to precursor compounds (fig. 9). The contribution from PAPs to the dust exposure was >71% for PFHxA, PFOA, and PFDA in all countries except for Greece and Spain. There was similar contribution from PFCAs with different chain length to the total intake from dust. The daily intake for a two years old child in the high exposure scenario, including both direct and indirect exposure, was 0.2 – 1.4 ng/day/kg b. w. for PFHxA, 0.2 - 1.8 ng/day/kg b. w. for PFOA, and 0.2 – 2.4 ng/day/kg b. w. for PFDA in Japan, Australia, Canada, the Faroe Islands, and Sweden.

At a lower exposure scenario, there was still a significant contribution from PAP precursors. The contribution from indirect exposure became increasingly important with increasing chain length; indirect exposure dominated for PFDA, and direct exposure dominated for PFHxA. The estimated contribution from diPAPs to daily intake in the low exposure scenario was 5-25% for PFHxA, 20-57% for PFOA, and 54-84% for PFDA, in all countries but Spain, Greece, and Nepal. The daily intake for a two years old child in the low exposure scenario, including both direct and indirect exposure, was 0.02 – 0.04 ng/day/kg b. w. for PFHxA, 0.05 – 0.18 ng/day/kg b. w. for PFOA, and 0.01 – 0.15 ng/day/kg b. w. for PFDA in Japan, Australia, Canada, the Faroe Islands, and Sweden.

Tolerable daily intake (TDI) has been established by European Food Safety Authority (EFSA) for PFOA at an amount of 1 500 ng/day/kg b. w. (EFSA 2008). However, current TDI for PFAS is under debate, and it cannot be ruled out that these levels might be as much as hundreds fold too high. Immunotoxic effects are seen at levels as low as 0.13 ng/mL serum for PFOS and 0.03 ng/mL serum for PFOA (Grandjean et al. 2016b). This corresponds to a daily intake of PFOA as low as 0.004 ng/day/kg b. w. Based on the level derived from the immunotoxicity study, PAPs alone can have an influence on human health of children in all countries but Nepal, and an influence on human health of adults in Faroe Islands and Japan, even in the low exposure scenario.

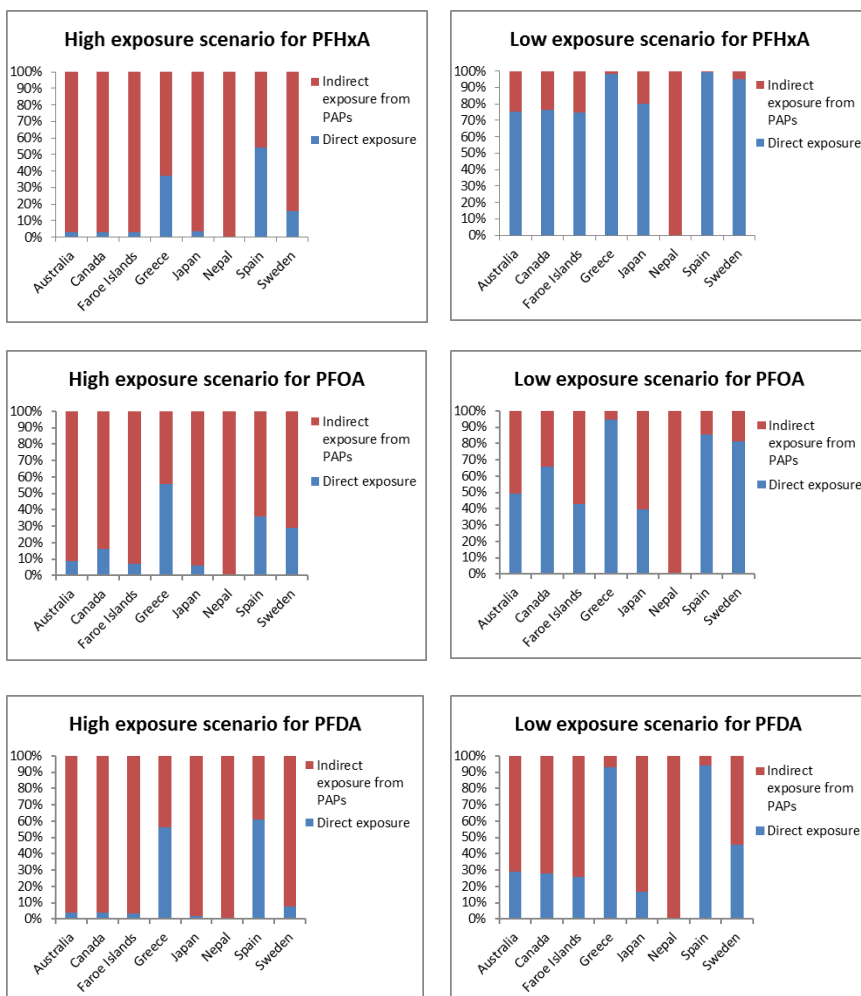


Figure 9. Exposure assessment of proportional contribution from indirect exposure of PAPs and direct exposure from PFCAs to total PFCA exposure via dust intake (%). In low exposure scenario, a biotransformation factor of 10% was used for PAPs \geq C8, 1% for PAPs $<$ C8, and an uptake fraction of 66%. In high exposure scenario, a biotransformation factor of 100% was used for PAPs, and an uptake fraction of 100%.

4. 3. Human levels of PFCA precursors

Human serum levels are suitable indicator for PFAS internal exposure since PFASs mainly distributes to liver, plasma and serum. In Paper II, the PFCA precursors PAPs and FTSA were analyzed together with PFCAs and PFSA in Australian serum stratified by age and gender during the sampling period from 2002 to 2011. The result revealed occurrence of both persistent and precursor PFASs in all age groups, both men and women, during the entire study period. The profile was dominated by PFSA followed by PFCAs, and precursors only made a minor contribution. Overall, precursor compounds were found in the general population at low levels close to LOD. 8:2 FTSA was found in a majority of the samples. 6:2 FTSA, 6:2 diPAP, and 8:2 diPAP were less frequently detected. There is no known production of PAPs in Australia, hence the results indicate exposure from direct use of commercial products to the general Australian population. In 2011, the mean value for adults (>15 years) was 0.09 ng/mL for 6:2 diPAP, and 0.02 ng/mL for 8:2 diPAP, when assigned a value of zero to samples <LOD. These levels are similar or somewhat higher than what previously has been observed in serum samples global (D'eon et al. 2009, Lee and Mabury 2011, Yeung et al. 2013, Gebbink et al. 2015b).

As PAPs are known precursor compounds to PFCAs, long-chain homologues of PAPs are expected to be phased out according to the Voluntary Stewardship program. This infers that 8:2 diPAP is included in the phase-out, but not 6:2 diPAP. It can be noted that 6:2 diPAP was more frequently detected (69%) than 8:2 diPAP (25%) in the Australian adult serum samples from 2011 compared to previous years.

In samples from adults (>15 years) in 2011, 8:2 FTSA was detected in 88% of the samples with a mean value of 0.01 ng/mL, while 6:2 FTSA was detected in only 13% of the samples. This could be due to differences in bioaccumulation however precursor compounds of 6:2 FTSA has become one of the major replacement compounds for PFOS after the phase-out in 2000 (Willson 2010).

It could be presumed that these low levels may imply a negligible contribution from precursor compounds to Σ PFAS in humans. However, precursors have shown to be readily biodegraded, therefore even low levels could be evidence of substantial exposure.

4. 3. 1. Ongoing exposure

The presence of PFCA precursors in the serum samples reported in Paper II indicates an ongoing exposure. Analyzing the precursor compounds in serum provides a qualitative measure of recent exposure, while the quantitative contribution is more difficult to obtain. Precursor compounds are readily degraded in humans and have short half-times. Half-times of 8:2 diPAP in humans have shown to be 0.9 – 1.4 days at high serum concentrations, and then slow down to 5.7 – 15.4 days at low serum concentrations (Trier and Numata 2015). For the persistent PFAS though, with comparable longer half-lives, ongoing exposure can be quantified. While decreasing trends were observed for most PFASs in Paper II, the rate of decline do not equal intrinsic half-times. In order to distinguish between ongoing exposure and historical exposure, halving and doubling times were calculated for compounds with decreasing and increasing temporal trend, respectively, using the data for age group 0-15 years, and compared to their intrinsic half-lives (table 2).

Table 2. Halving time and doubling time of PFASs in serum samples compared to intrinsic half-times.

	Time period	Half-time female (years)	Half-time male (years)	Halving time in this study (years)	Doubling time in this study (years)
PFHpA	2006 – 2013	1.5a	1.2a	7.8	
PFOA	2002 – 2013	1.8 - 2.3a,b	1.7 - 2.3a,b	5.3	
PFNA	2006 – 2013	2.5a	4.3a	13.5	
PFDA	2006 – 2013	4.5a	12a	32.2	
PFUnDA	2006 – 2013	4.5a	12a	29.0	
PFDoDA	2002 – 2013				17.2
PFHxS	2002 – 2013	7.7a	35a	4.6	
PFHpS	2002 – 2013			5.3	
PFOS	2002 – 2013	6.7a	34a	4.2	
8:2 FTSA	2002 – 2013			5.5	

a Zhang, Y. F., et al. (2013).

b Bartell, S. M., et al. (2010)

The halving times for PFCAs were all longer than their intrinsic half-lives, while the halving times for PFSAAs were all shorter than their intrinsic half-lives. This suggests ongoing exposure for PFCAs to humans, from 2006 to 2013. The discrepancy between the apparent halving times and the intrinsic half-time for the PFSAAs could be due to the fact that different individuals were compared in the temporal trend data. The temporal trends were assessed for the age group 0-15 years, and concentrations in infants are affected by mother-child exposure during pregnancy and lactation. Transfer across the placenta of PFASs from mother to fetus are suggested to be dependent on chain-length and active head group (Manzano-Salgado et al. 2015). The more hydrophobic long-chain PFASs have been shown to be transferred at a lower ratio than their short-chain analogues. Additionally, sulfonates have been shown to be transferred at a lower ratio compared to carboxylates.

4. 3. 2. Daily intake

Using the serum data from Paper II as input parameters, and a first-order pharmacokinetic (PK) model, the daily intake of PFOA in Australia was calculated to be 183 pg/day/kg b.w. at a non-steady state. This could be compared to the exposure assessment of ingested dust in Paper I. In a low exposure scenario, the direct PFOA intake from dust for a 3-6 year child was 29 pg/day/kg b.w., and the indirect intake 30 pg/day/kg b.w., in total 58 pg/day/kg b.w.. In a high exposure scenario, intake from dust for a 3-6 year child was 43 pg/day/kg b.w., and the indirect intake 449 pg/day/kg b.w., in total 492 pg/day/kg b.w.. In relation to the results from the Australian serum samples, it seems as the low exposure scenario is more probable for exposure from precursor compounds, i.e. diPAPs. Thus, the estimations show that dust intake in comparison to Australian serum levels in children gives a contribution from diPAPs of 16 -100% to total PFOA exposure.

Exposure from drinking water in Australia has previously been assessed (Thompson et al. 2011). In Queensland, where the Australian serum analyzed in Paper II were sampled, no PFOA above limit of detection was found in the drinking water. Using the limit of detection of 0.5 ng/L as a lower bound, and 9.66 ng/L for the highest PFOA water concentration found in Australia as an upper bound, the intake from water for a 3-6 year child can be calculated to 10 - 184 pg/day/kg b.w. This implies that dust may be an equal or even more important exposure source as drinking

water for children in Australia. There are currently no data available about levels of PFAS in the Australian diet. Food consumption patterns can vary substantially between regions, hence no comparison with food can be made with the PFAS levels in dust.

4. 4. Waste water treatment plants

Municipal waste provides a snapshot of contaminants that are used and released from the urban community, both from households through usage of commercial products and through industrial activities. The composition profile of PFAS can be altered during waste water treatment, due to different properties of PFASs, such as sorption capacity, water solubility, and persistency. Especially, precursor compounds can degrade to their persistent analogues.

In Paper III, PAPs, FTSAs, FOSA/FOSEs, FTCA/FTUCAs, PFCAs, PFSA, and PFPA/PFPiAs were analyzed in water and sludge from three Swedish WWTPs. The influent consisted of municipal waste water but also water from additional sources such as hospitals (Umeå and Henriksdal), industries (Henriksdal) and specific chemical and textile industries (Gässlösa). PAPs, FTSAs, PFPAs, FTCA/FTUCAs, and PFPiAs were present in both sludge and water. The concentrations of PFAS in waste water are shown in table 3 and the concentrations of PFAS in sludge are shown in table 4.

Table 3. Concentrations of detected PFAS classes in waste water (ng/L).

	Umeå		Henriksdal		Gässlösa	
	Influent	Effluent	Influent	Effluent	Influent	Effluent
ΣPFCa	19	22	23	33	18	66
ΣPFSA	4.6	4.8	7.2	7.7	3.9	4.8
ΣFTSA	2.9	2.5	6.0	5.1	2.6	2.0
ΣFTCA/FTUCA	0.5	0.4	1.4	<LOD	3.5	n.q.
ΣmonoPAP	<LOD	n.q.	<LOD	<LOD	n.q.	n.q.
ΣdiPAP	58	n.q.	n.q.	<LOD	n.q.	<LOD
ΣPFPA	11	1.3	1.8	0.6	20	3.0
ΣPFPiA	0.6	0.1	1.6	0.1	0.3	2.1

Table 4. Concentrations of detected PFAS classes in the sludge (ng/g).

	Henriksdal			Gässlösa			Umeå		
	2012	2014	2015	2012	2014	2015	2012	2014	2015
Σ PFCA	6.2	6.2	4.7	13	12	7.3	2.4	1.7	2.5
Σ PFSA	10	5.7	3.9	3.5	3.3	3.6	6.3	3.1	1.9
Σ FTSA	1.8	1.3	0.9	0.9	1.6	1.2	1.0	1.0	0.8
Σ FTCA/ FTUCA	15	13	8.9	65	78	51	4.0	5.5	4.1
Σ mPAP	17	12	11	11	6.2	5.8	18	9.9	11
Σ diPAP	9.9	8.5	7.5	7.7	7.2	8.8	17	3.3 ^a	9.1
Σ PFPIa Σ FOSA/ FOSE	<LOD	<LOD	<LOD	0.03	<LOD	<LOD	<LOD	<LOD	<LOD
	3.8	2.9	<LOD	3.3	3.5	3.3	<LOD	<LOD	1.3

^a8:2 diPAP not quantified.

In the effluent waste water, PFCAs dominated the profile followed by PFSA. Levels of Σ PFCA were in the range 22 – 66 ng/L, and for Σ PFSA 4.8 – 7.7 ng/L. Highest PFCA levels were found in Gässlösa, the WWTP connected to the most industrial activities. A majority of the PFCAs found were the short-chained PFBA, PFPeA, and PFHxA (71 – 87%). The precursor compounds contributed only to a minor portion of all PFASs measured in the waste water. Analysis of PAPs in water was hampered by severe matrix effects and only two effluents could be quantified, with observed levels below limit of detection. The Σ FTSAs were in the range 2.0 – 5.1 ng/L. Most abundant homologue was 6:2 FTSA, which was found in all effluents, while 8:2 FTSA was found in Umeå only. The 4:2 FTSA was not detected in any WWTP effluent. Intermediates, 5:3 FTCA, 6:2 FTUCA, and 8:2 FTUCA, were detected in the effluent of Umeå, at a total level of 0.4 ng/L. The Σ PFPA/PFPiAs were present at lower levels than PFCA/PFSAs, in the range 0.7 – 5.1 ng/L. Industrial activities might contribute to enhanced levels of PFBA, PFPeA, PFHxA, PFDPA, C6 PFPiA, and C6/C8 PFPiA in effluent from Gässlösa, compared to other two WWTPs.

In the sludge samples from 2015, precursors and intermediates were the predominant compound groups with a contribution of 75 – 86% to Σ PFAS. The PAPs were found in all sludge samples. The 8:2 FTSA was the most abundant FTSA in the sludge, detected in all WWTPs at levels of 0.6 – 0.8 ng/g. The 6:2 FTSA was only found in Henriksdal (0.3 ng/g) and

Gässlösa (0.7 ng/g), the two WWTPs with industrial activities. All FTCA/FTUCAs analyzed were detected in the sludge, with highest levels found for 5:3 FTCA (1.3 – 45 ng/g) and 7:3 FTCA (2.0 – 5.0 ng/g). At Gässlösa, the level of 5:3 FTCA was one order of magnitude higher than Umeå and Henriksdal. The 5:3 FTCA is a degradation product of 6:2 fluorotelomer-based compounds, and has shown to further degrade to PFPeA (5.9%) and PFBA (0.8%) in sludge (Wang et al. 2012).

4. 4. 1. Emission from WWTPs to the environment

The emission of PFASs from WWTPs through sludge and effluent water is described in Paper III. Persistent PFASs and selected precursor compounds were found in influent water, effluent water, and sludge. For several PFASs and PFCAs, a net increase was observed in the waste water treatment plants (fig. 11). Highest increase was observed for short-chain and even-numbered homologues. In one of the WWTPs (Umeå), the contribution from the precursor compounds to the net increase of PFCAs, and the contribution from the precursor compounds to the total amount of PFCAs were estimated. Transformation yields from degradation studies of diPAPs and FTSAs in sludge and soil were used as parameters in the calculations of amounts of PFCAs degrading from precursors. It was estimated that 2.1 – 41% of the net increase of PFHxA and 19% of the net increase of PFOA could be attributed to precursor compounds. Though a fairly high proportion of the net increase can be related to degradation of diPAPs and FTSAs, the remaining amount (59 – 99%) is of unknown origin.

Of total amount released, 0.7 – 13% of PFHxA, and 5.5% of PFOA could be attributed to degradation from precursor compounds analyzed in the WWTPs. A large uncertainty factor in these estimations is the degradation yield of precursors to PFCAs. Nevertheless, these values represent a lower bound of precursor contribution, as an unknown proportion of the PFCAs could originate from precursor degraded before entering the WWTP. This can be due to both recent degradation and degradation in the past. A proportion of PFASs in the influent water may originate from tap water, thus recirculated amounts of PFASs can contribute to the total load (Filipovic and Berger 2015).

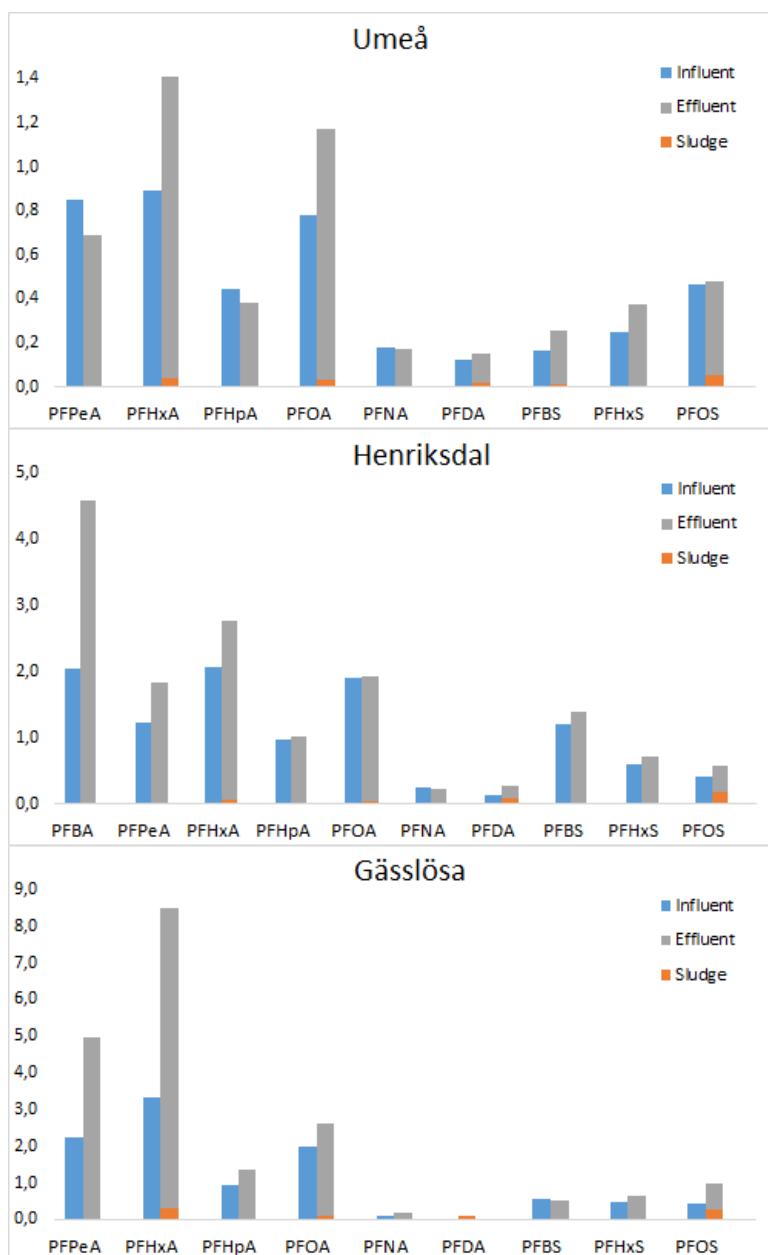


Figure 11. Levels of PFCAs and PFSA in influent, effluent, and sludge in Swedish WWTPs, µg/day/person. Note the different scales on the x-axis.

4. 5. Levels of PFCA precursors in wild-life

Levels and homologue patterns of PFASs can differ substantially between wildlife and humans. Other sources, routes of exposure, bioaccumulation factors, and environmental conditions are factors all have an impact on the levels in the wildlife.

In Paper IV, levels of PFASs in the terrestrial and the freshwater environment were assessed in raptor bird eggs. Three species were examined in the study, osprey (*Pandion haliaetus*), tawny owl (*Strix aluco*), and common kestrel (*Falco tinnunculus*). The tawny owl and common kestrel are terrestrial species, while osprey is an aquatic specie. Stable isotopes of carbon and nitrogen were analyzed as tracers for diet and trophic level. It was revealed by the $\delta^{13}\text{C}$, ranging between -25.2‰ and -30.4‰, that the ospreys in the study fed on a freshwater diet rather than a marine diet. This excludes influence from long-range transport of PFASs with ocean currents. The tawny owl samples were collected in 2014 (n=40), the common kestrel samples were collected in 2014 (n=10), and the osprey egg samples were collected in 1997 – 2001 (n=10), 2007 – 2008 (n=10), and 2013 (n=10). PFCAs, PFSA, FTSA, FTCAs, FTUCAs, and diPAPs were all found in the bird eggs.

The PFAS profiles in the bird eggs were dominated by PFCAs and PFSA, where PFOS was the single most abundant PFAS homologue. The diPAPs were found in eggs from two of the three species, osprey and common kestrel, thus both in the terrestrial and the freshwater environment. In general, they had a low frequency of detection and were found at low ppt levels. Homologues detected were 6:2 diPAP, 6:2/8:2 diPAP, 8:2 diPAP, 6:2/10:2 diPAP, 8:2/10:2 diPAP, 10:2 diPAP, and 8:2/12:2 diPAP. The highest level of 6:2 diPAP (0.4 ng/g) was found in one osprey egg from 1999, and the highest level of 8:2 diPAP (2.4 ng/g) was found in one osprey egg from 2008. PAP levels and homologue distribution in wild life are not well characterized and have only been reported in a limited number of studies. Previously, diPAPs have been detected in fish, zooplankton, benthic worm, and monoPAPs have been detected in mussels (Guo et al. 2012, Loi et al. 2013, Zabaleta et al. 2015, Gebbink et al. 2016a). Generally, levels of PAPs in wildlife have been in the low picogram per gram range, with exception of tuna from Indian Ocean, where levels of 11.4 – 11.5 ng/g were observed (Zabaleta et al. 2015).

The 6:2 FTSA were detected in a majority (63%) of the osprey eggs, while only one bird egg from the terrestrial environment had detectable concentrations. While only a few studies have reported FTSA in wildlife, high levels of 6:2 FTSA have been found in soil, water, and sediment in contaminated areas (Karrman et al. 2011, Boiteux et al. 2016).

4. 6. Contribution of precursors to PFCA

What significance do precursor compounds have to the total amount of PFAS in humans and the environment? The composition profiles of precursor compounds (Σ diPAPs and Σ FTSAs), together with Σ PFCA and Σ PFSA in dust (Paper I), serum (Paper II), sludge (Paper III) and bird eggs (Paper IV) are shown in figure 12.

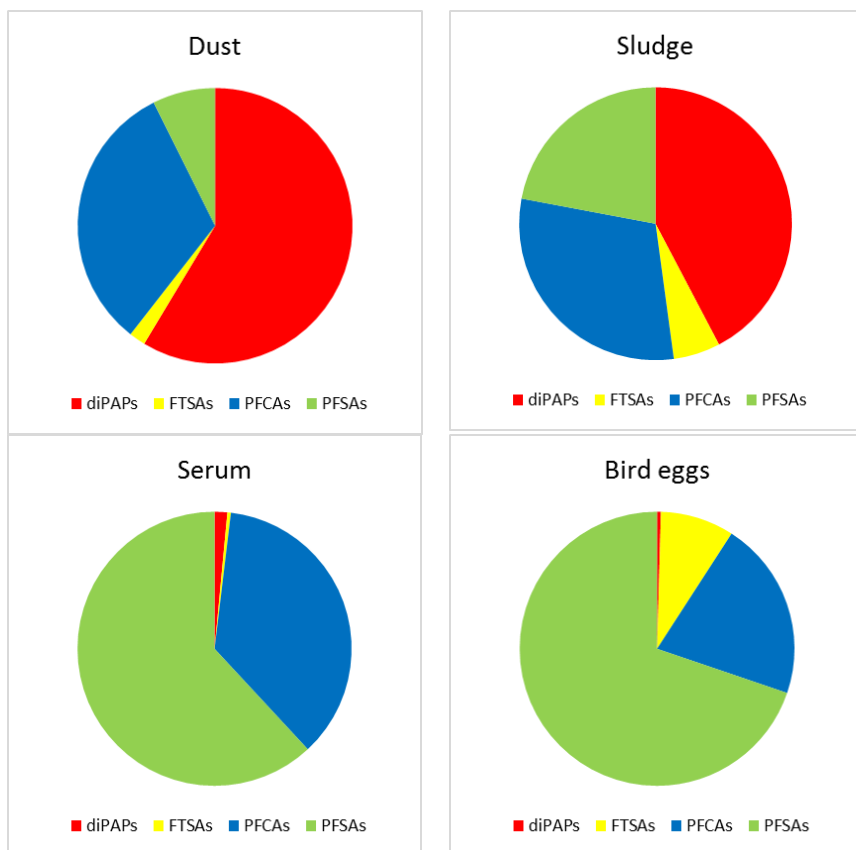


Figure 12. The profiles of Σ diPAPs, Σ FTSAs, Σ PFCAs, and Σ PFASs in different matrices, percentage contribution. The bird eggs were sampled in 1997 – 2014, the sludge samples in 2012 – 2015, the dust samples in 2008 – 2013, and the serum samples in 2002 – 2013.

There are large differences in the profiles between different compartments. The contribution of Σ diPAPs to the total amount of the analyzed PFASs is much higher in abiotic matrices compared to biotic matrices. These differences are a result of the character of the compartments and the various properties of PFAS compounds such as bioaccumulation potential, volatility, stability, and sorption behavior.

In both dust and sludge samples, diPAPs were the major contributor (59% and 42%, respectively). Consumer products that may contain PFASs such as papers, carpets, clothes, furniture, food packaging, cleaning products, and personal care products are widely present in the indoor environment of private households. Therefore, dust can be seen as a fingerprint of contaminants in these products, and represent recent exposure from current industrial PFAS production. The high proportion of diPAPs found in the dust suggest that diPAPs is a significant indirect PFCA source to human exposure. It is also possible that a proportion of the PFCAs detected in the dust is derived from degraded PAPs, or other fluorotelomer precursors.

In the sludge samples, sources from both private household and industrial activities were combined. The sludge PFAS profile is similar to that of the dust, where diPAPs is the dominant PFAS, followed by PFCAs, PFSAAs, and FTSAAs (fig. 12). Different proportion of diPAPs in sludge (42%) and dust (59%) was observed. It is possible that microbial degradation alter the profiles in sludge and dust.

During abiotic conditions, diPAPs are extremely stable and have halftimes of at least decades (D'eon and Mabury 2007). In organisms on the other hand, phosphate esters are dephosphorylated by phosphatase enzymes in intestinal tracts, and are thereafter readily degraded (Jackson and Mabury 2012). This could explain the minor contributions of diPAPs in the human serum and the bird eggs.

The FTSAAs are only a minor contributor to Σ PFASs in dust (2%), while the contribution is comparable higher in sludge (6%). It is possible that FTSAAs are more related to industrial activities than use of commercial products in private households. The contribution of FTSAAs is also much higher in bird eggs (9%) compared to human serum (1%). FTSAAs can be a degradation product from other FTSA-based precursors, and will also further degrade into PFCAs (Weiner et al. 2013). In case of degradation of FTSA-based precursors, this could contribute to the amount of FTSAAs found in sludge and bird eggs.

The contribution from PFSAAs to Σ PFASs is much higher in human serum and bird eggs compared to sludge and dust, of which the majority can be attributed to PFOS. Though the major PFOS production is ceased, the

proportion of PFOS to Σ PFAS continues to be relatively high, due to its long half-life and recirculated levels in the environment.

Overall, the contrasting PFAS profiles in biotic and abiotic compartments implicate that substances included in human and environmental monitoring needs to be carefully chosen. The exclusion of abundant PFAS classes such as diPAPs in monitoring studies, especially abiotic matrices, will give a distorted picture of the relative importance of different exposure sources.

Sources of exposure, emissions and pathways can be tracked by examining the PFCA homologue profiles. Fluorotelomer-based compounds produced are almost exclusive even-numbered, and biodegradation of n:2 fluorotelomer precursors, where n= 8 and higher, will yield preferentially even-numbered homologues (C_n PFCA) (Dinglasan et al. 2004, Liu et al. 2010, Kim et al. 2014a). Atmospheric degradation of n:2 fluorotelomer precursors on the other hand will result in C_n PFCA and C_{n+1} PFCA at similar yield, followed by an increased proportion of C_{n+1} PFCA, due to comparable higher bioaccumulation factor (Ellis et al. 2004, Martin et al. 2004, Bossi et al. 2005). In figure 13, the profiles of PFCAs in different matrices are shown.

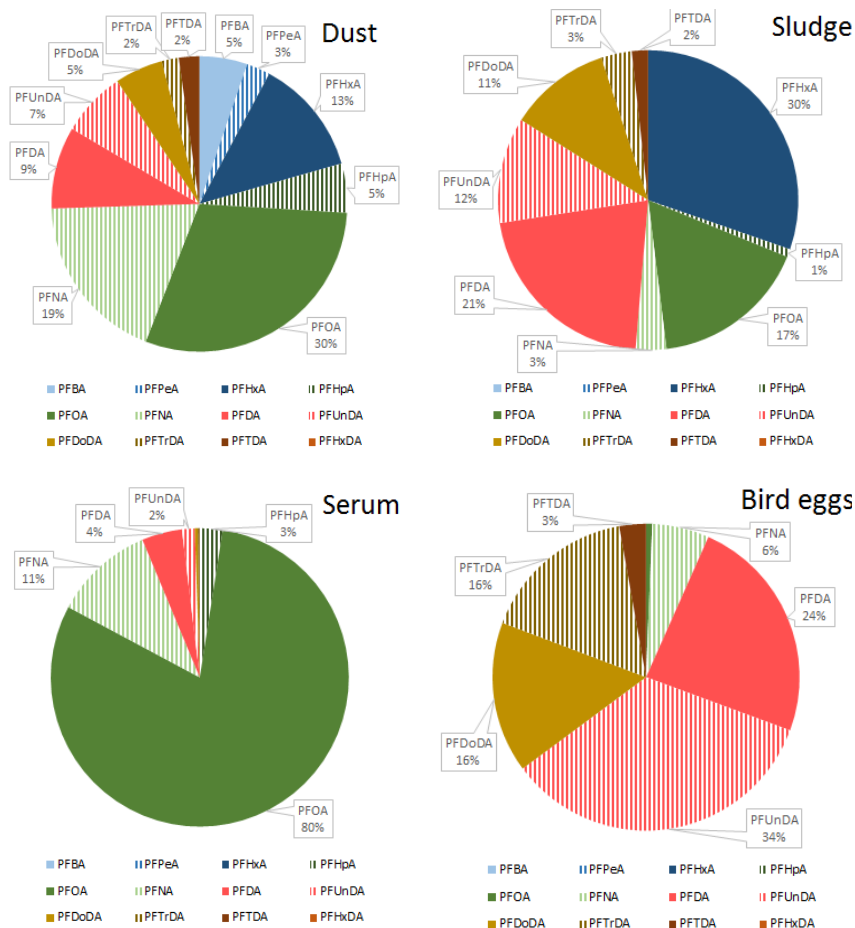


Figure 13. Percentage distribution of PFCAs in dust and sludge. The bird eggs are sampled in 1997 – 2014, the sludge samples in 2012 – 2015, the dust samples in 2008 – 2013, and the serum samples in 2002 – 2013.

The dust and sludge matrices have some similarities in PFCA homologue distribution. However, PFDA, PFUnDA, and PFDaDA are more prevalent in the sludge, which probably is due to increasing sorption capacity with chain length.

Another difference is the higher contribution of PFHxA in the sludge samples compared to the dust. This could possibly be an effect of degrada-

tion of PFHxA precursors in the waste water treatment plants. The non-bioaccumulating short-chain PFCAs are absent in the biotic matrices. Shorter PFCA homologues like PFHxA have a faster elimination rate than those of the long-chain PFCAs in humans. In birds, PFOA and shorter homologues do not bioaccumulate.

The proportion between PFNA and PFDA is similar in dust and serum, while the contribution of PFOA is much higher in serum compared to dust. This indicates that sources other than dust are important for human PFOA exposure. These sources could be from other exposure pathways, such as drinking water or food, or PAPs-intake through dust, with subsequent degradation in the human body.

As discussed in Paper IV, exposure pathways may differ between terrestrial and freshwater environment, where atmospheric sources are suggested to be of higher importance in the terrestrial environment compared to the freshwater environment. In the freshwater environment, other sources such as emissions from waste water treatment plants can be of more importance compared to the atmospheric pathway. The relative importance of exposure pathways has been hypothesized to be reflected by the homologue pattern, where a typical odd-even pattern indicate that the atmospheric transport and degradation of precursors is the dominant pathway for PFASs exposure. In the bird eggs, even-numbered PFCAs contributed more to Σ PFCAs in the freshwater environment (49%), compared to the terrestrial environment (39%) (fig. 14).

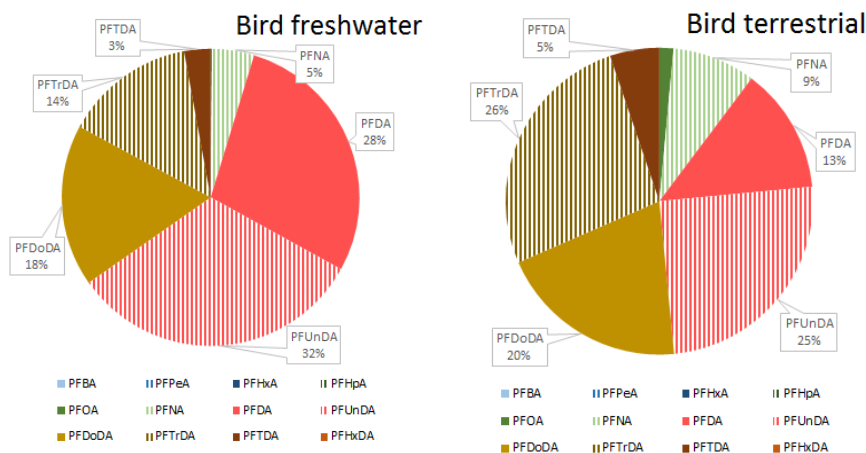


Figure 14. Percentage distribution of PFCAs in raptor bird eggs from the terrestrial and the freshwater environment. The terrestrial bird eggs are sampled in 2014, and the freshwater bird eggs are sampled in 2013.

Even-numbered PFCAs contribute more to Σ PFCAs in the freshwater environment (47%), compared to the terrestrial environment (32%). The hypothesis that major exposure pathway in the terrestrial environment is from degradation of atmospheric transported precursors imply that a typical odd-even pattern can be observed. Atmospheric degradation of $n:2$ fluorotelomer precursors will result in C_n PFCA and C_{n+1} PFCA at similar yield, followed by an increased proportion of C_{n+1} PFCA, due to comparable higher bioaccumulation factor. When other sources such as release from waste water treatment plants are of more importance, PFCAs formed from biodegradation of fluorotelomer precursors will yield mainly even-numbered PFCAs.

The profile of diPAPs in dust and sludge, the matrices where diPAPs predominated the profile, are shown in figure 15.

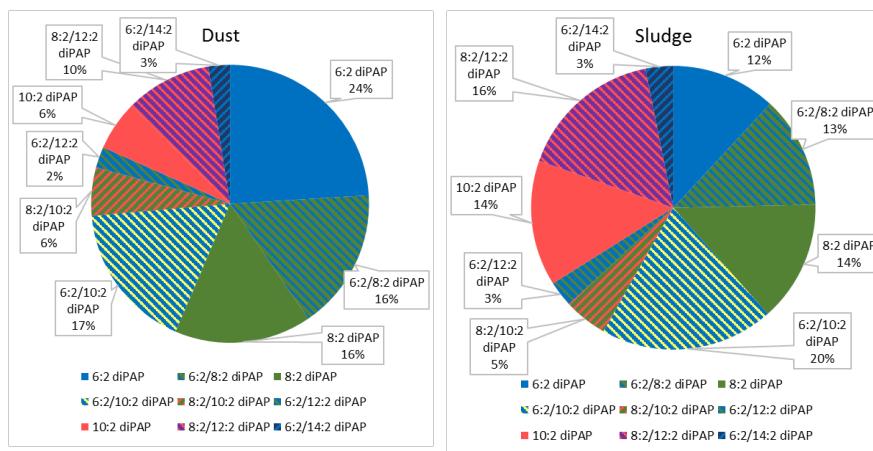


Figure 15. Percentage distribution of 6:2 diPAP, 6:2/8:2 diPAP, 8:2 diPAP, 6:2/10:2 diPAP, 8:2/10:2 diPAP, 6:2/12:2 diPAP, 10:2 diPAP, 8:2/12:2 diPAP, 6:2/14:2 diPAP in dust and sludge. The sludge samples in 2012 – 2015, and the dust samples in 2008 – 2013.

The homologue distributions of diPAPs in dust and sludge are relatively similar. The profiles are dominated by C6 – C10 homologues, with smaller contribution from C12 and C14 homologues. In the sludge samples, there is a tendency of higher proportion of 8:2/10:2 diPAP and 10:2 diPAP and their structural isomers, which could be explained by higher sorption capacity compared to 8:2 diPAP and shorter homologues. This pattern is typical for the fluorotelomer production process, as discussed in Paper I. It seems like long-chain diPAPs still are ubiquitous, either in diPAP production or in consumer products.

The fluorotelomerization process generally generates mixtures of homologues. Therefore, homologues of other chain lengths than the targeted ones can be left as residuals. The PFAS manufacturers are moving towards C6 chemistry (3M Company 2002, Daikin 2007, DuPont 2008, AGC 2016), and it is possible that 8:2 diPAP and longer homologues will be present as residuals in 6:2 diPAP products.

4. 6. 1. Temporal trends

Is regulations and/or voluntary restrictions effective tools to reduce PFAS contamination? Temporal trends is a way to evaluate the effects of implemented regulation.

In Paper II, Australian serum samples from 2002 to 2013 were analyzed for PFASs, stratified into gender and age groups. However, low frequency of detection of precursor compounds hampered the assessments, and sufficient data to evaluate changes over time for precursor exposure was only obtained for 8:2 FTSA. For 8:2 FTSA, a declining trend was observed between 2002 and 2013. It was interesting to note that the highest single levels of 6:2 diPAP were observed more frequently in recent years.

Temporal trends were evaluated for osprey eggs in Paper IV. Similarly to the serum samples, temporal trends of precursor compounds could not be assessed due to low frequency of detection. However, the diPAPs, 6:2 FTSA, and FTCAs could be detected throughout the entire study period from 1997 – 2001 to 2013, suggesting that biota were still exposed to precursor compounds.

As precursor compounds generally are present at low picogram per gram levels in biotic sample, their contribution to PFAS exposure over time is difficult to assess. Evaluating their degradation products can provide an indirect measurement of their impact on exposure. In figure 16, the temporal trends of Σ PFCAs and the proportional contribution of Σ PFCA to Σ PFASs in human serum from Paper II and bird eggs of osprey from Paper IV are shown.

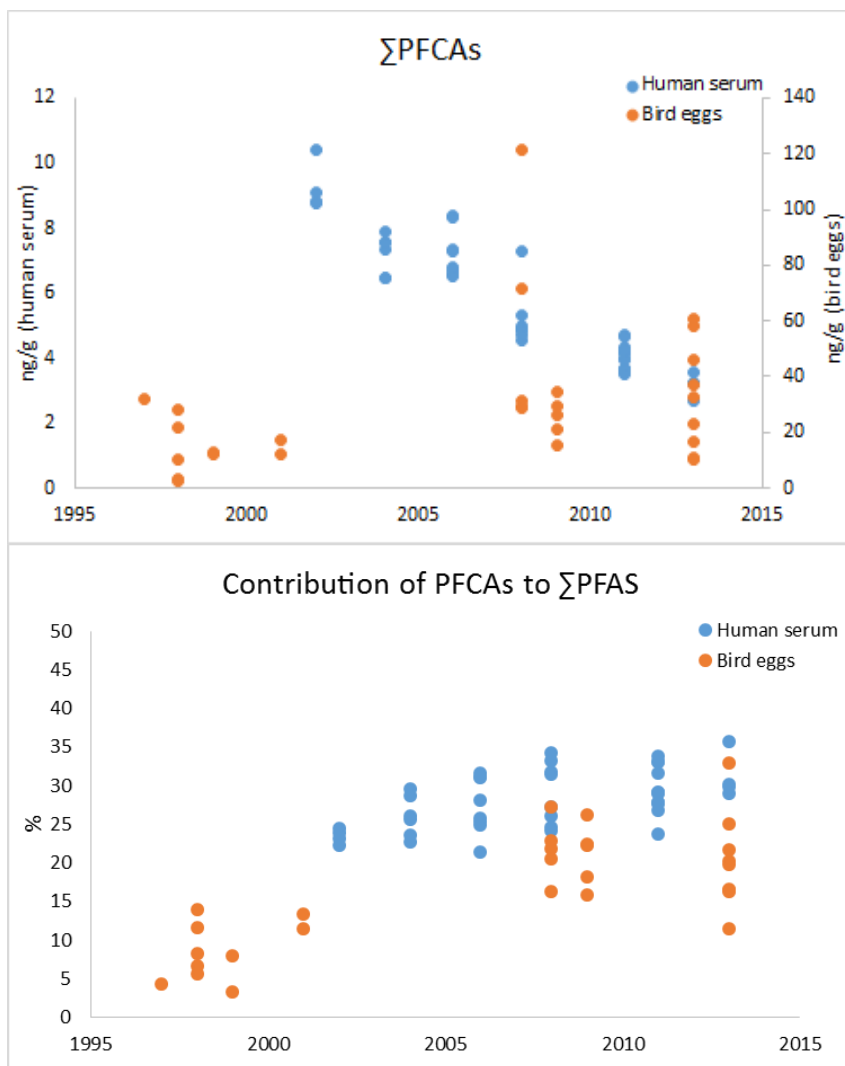


Figure 16. The temporal trend of Σ PFCAs in human serum and bird eggs (ng/g), and the temporal trend of the relative contribution of PFCAs to Σ PFAS in human serum and bird eggs (%).

Contrasting trends are observed in serum and bird eggs. The total amount of PFCAs in serum has decreased from the early 2000s to 2013, while the

PFCA levels have increased in bird eggs. A major contributor of the decrease in human serum is the decreasing PFOA levels. Levels of the long-chain PFCAs (C10 – C14) are of higher importance to Σ PFCAs for bird eggs, however no decrease of these neither in humans nor birds was observed. During the same time span, the contribution of PFCAs to Σ PFASs has become more important. This is both an effect of decreasing PFSA levels, and for birds, increasing PFCA levels.

In the sludge samples, the proportion of 8:2 diPAP compared to 6:2 diPAP was decreased between 2012 and 2015. Additionally, the 8:2 monoPAP levels were lower in 2015 compared to 2012. The tendency of higher proportion of C6 compared to C8 in 2015 compared to previous years could indicate a shift towards more shorter-chain PFASs.

Overall, the switch in industry towards short-chain PFASs is reflected by the patterns and trends in humans and environment. While some PFASs have been phased out of production, replacement compounds have taken their place, as can be seen by the high levels of PAPs in abiotic matrices. Precursor compounds will eventually degrade into persistent PFCAs. It has been argued that the short-chain PFASs do not bioaccumulate and do not fulfil the vPvB (very persistent, very bioaccumulative) criteria set for regulations of chemicals. Though less bioaccumulative than their longer-chained analogues, short-chain PFASs are as persistent as the long-chained PFASs, and do not degrade in humans and environment.

5. Conclusions

In this thesis, the contribution of precursors to the total amount of PFCAs exposure in humans and environment was assessed, by analyzing PAPs together with a wide range of other PFASs in dust, human serum, sludge, water, and raptor bird eggs. The broad approach of this thesis made it possible to characterize the differences between levels and profiles of both PFCAs precursors and other PFAS compound classes in different compartments.

Mono-, di-, and triPAPs were found to be globally abundant in indoor dust, at levels exceeding those of persistent PFAS classes. The high level imply that PAPs in dust might be a significant source for exposure, especially for children.

Detection of low levels of precursors in serum from men, women, and children in Australia revealed direct exposure from consumer products. Slow declining trends further supported the hypothesis of ongoing PFCAs exposure.

During waste water treatment, levels of PFCAs increased in the effluent water compared to the influent water. Precursors were detected in the sludge and in the waste water, and the levels of diPAPs were higher in the influent water compared to the effluent water. One waste water treatment plant showed an estimated contribution of 19% from diPAPs to the net increase of PFOA. This supports the hypothesis that degradation of precursor compounds is a significant contributor to PFAS contamination in the environment. A significant proportion of PFCAs released in the effluent waste water is probably from unknown precursor compounds. Though FTSA are known to degrade in sludge, they were not observed to decrease substantially in the Swedish waste water treatment plants. This suggest that FTSA precursors might be present in the waste water and sludge.

Precursor compounds were detected in raptor wild birds. No decline of PFASs was observed during the years following the regulations and voluntary phase-out of PFOS and PFCAs. This demonstrate the vulnerability of endangered species that are exposed to both historic and recent PFAS sources.

Current regulations have not yet fulfilled their aim to protect humans and the environment from PFAS exposure.

The major conclusions in this thesis can be summarized as:

- PAPs in indoor dust is a global source of human exposure
- There is an ongoing exposure of PFCAs to humans
- Degradation of precursors is a significant contributor to environmental contamination
- PFASs emissions lead to long-term effect in the environment

6. Future perspective

For future monitoring studies, the monoPAPs, diPAPs, and FTSA should be included, especially in abiotic matrices. For regular monitoring 6:2 monoPAP, 8:2 monoPAP, 6:2 diPAP, 6:2/8:2 diPAP, 8:2 diPAP, 6:2 FTSA, and 8:2 FTSA are most important homologues and are suggested to be selected as indicator compounds. Levels of these and ratios between selected PAPs homologues provides information useful for total PAPs exposure estimation and changes in commercial PAPs mixtures compositions. For in-depth evaluation of changes in manufacturing applications, complete analysis of PAPs profile including C4-C18, mono- di- and tri-substituted PAPs is recommended.

The source for FSAs in wild birds needs to be further investigated. Precursors of FSAs need to be analyzed together with FSAs in both wildlife and potential sources, such as in freshwater, sediment, fish, waste water, and sludge.

Little is known about the contribution from polymer degradation and more studies are needed for assessment to what extent they might contribute to the total amount of PFAS.

The number of compounds that need to be monitored is increasing. New PFAS compounds constantly enter the market. Directives such as trade secrets hamper the possibilities for scientists to use straightforward strategies in the analysis of PFASs. The current legal frameworks always leave scientists one step behind, and the scientific community should aim to reduce the gap using the most effective tools available. Both qualitative and quantitative data are needed.

When compositions and formulas of PFAS are constantly changed by the industry, it is important to ensure that the most relevant substances are being monitored. The use of extractable organofluorine (EOF) and total organofluorine combustion ion chromatography (TOF-CIC) are important tools in the assessment of how much of the total PFAS amount is covered in quantitative analysis of individual compounds. Analysis of total EOF content should be included in monitoring studies in order to assess the coverage of identified and quantified PFASs to the total amount of PFASs. The unknown proportion of PFAS needs to be identified, using methods as untargeted screening using liquid chromatography mass spectrometry time-of-flight analysis (LC-MS-TOF) for qualification.

7. Acknowledgement

I would like to express my gratitude to all people that have been around and have been so helpful in any way in the work of this thesis. I am truly happy for all support and encouragement you gave me. It means a lot. There are so many people I would like to thank:

First of all my supervisor Anna Kärrman, for being so wise and patience, for your gently guiding in the jungle of challenges that surrounds analytical chemistry, and for sharing your tremendous knowledge. Always positive, always seeing the possibilities.

Bert van Bavel, my co-supervisor, and Gunilla Lindström. Bert, I hope your next carrier step will be as a politician – you would be a great one! Bert and Gunilla, you created the wonderful spirit of MTM and the Environmental and Health group, and I'm happy that you gave me the opportunity to thrive in it.

Alf Ekblad, though not on paper I consider you also as my supervisor, cause I've learned so much from you. Your enthusiasm and natural curiosity on science is a big source of inspiration.

Anna Roos and Ylva Lind, for all the help with the bird study. I hope I'll get more opportunity in the future to learn about ornithology from you. Jochen Mueller, Leisa-Maree Toms, and Peter Hobson for the help with the serum manuscript. Peter Haglund, for the help with WWTP project. Maria Dam, Takumi Takasuga, and Aris Xenakis, for all the help with the dust sampling.

All colleagues for all good times together. Maria Larsson, you've been so supportive in every way, you spread so much positive energy. Many hugs. Ingrid Jogsten Ericson, you're such a generous a warm persons with so many creative ideas I'll hope will fulfil. Dawei Geng, always so cheerful and encouraging. Hope you still are here next gardening season! Filip Bjurlid, nice, cleaver, fun, all at the same time. Lotta Sartz and Petr Kuckucka (a.k.a. king of the dancefloor), you're such great roommates. And thanks Petr for all your great suggestions. Leo Yeung, all the help from you is most appreciated, and it's so interesting to discuss with you. Victor Sjöberg, and Naeem Saqib, for all nice talks. Steffen Keither, for giving me new inspiration when I needed it at most. Thanh Wang, for great advice and interesting discussion. Jordan Stubbleski, Monika Lam,

and Christine Schönlaui for all fika and nice moments. Josefin Persson, always cheerful, always positive. Irina Kalbina, for all nice talks. Johanna Björklund, a true environmentalist, looking forward to spend more time in the future garden with you. Ulf Hanell, for all interesting cross-disciplinary scientific talks. Marta Szot, for the wonderful spirit you have. Anna Rotander, I'm glad you're back from down under. Samira Salihovic, for being a wonderful roommate, miss you, I'm looking forward to when you're back in Örebro again! Heidi Fiedler, you have such a wonderful laugh. Sumei Li, little sunshine. Jenna Davies, it was great having you here and hope you'll get back some day. Owe Persson, Erik Sjöstedt, and Jörgen Stenlund, for all nice fika moments.

All other supernice old and new MTM people, especially: Melli Sejfic, Sofie Björklund, Charlotte Nilsson, Anna Gradin, Shika Acharaya, Kjell Hope, Tulia Hyötyläinen, Stefan Karlsson, Jessika Hagberg, Magnus Engwall, Kristina Åhlgren.

And many thanks to Anna Mikusinska, for the wonderful time in the isotopic lab and all the fun we had. You'll always have a special place in my heart.

My friends and family, mom, dad, sisters, for being so supportive and understanding, for all wonderful moments together.

Arimande, the best daughter in the world! and Moa, the best bonusdaughter in the world! Keep on rocking girls ☺

Magnus, my love, for everything.

Thank you!

References

- 3M Company. (1999a). Fluorochemical Use, Distribution and Release Overview; U.S. EPA Public Docket AR226-0550.
- 3M Company. (1999b). Perfluorooctane Sulfonate: Current Summary of Human Sera, Health and Toxicology Data. U.S. EPA Administrative Record AR226-0548.
- 3M Company. (2000). Phase-out Plan for POSF-Based Products, U. S. EPA Edministrative Record AR226-0600.
- 3M Company. (2002). 3M Technical Data Bulletin Environmental, Health, Safety, and Regulatory (EHSR) Profile of Perfluorobutane Sulfonate (PFBS).
https://solutions.3m.com/3MContentRetrievalAPI/BlobServlet?locale=en_US&lmd=1120194514000&assetId=1114270648708&assetType=MMM_Image&blobAttribute=ImageFile.
- AGC (2016). <http://www.agcce.com/asahiguard-e-series/>.
- Ahrens, L. (2011). Polyfluoroalkyl compounds in the aquatic environment: a review of their occurrence and fate. *J Environ Monit* 13(1): 20-31.
- Ahrens, L., Norstrom, K., Viktor, T., Cousins, A. P. and Josefsson, S. (2015). Stockholm Arlanda Airport as a source of per- and polyfluoroalkyl substances to water, sediment and fish. *Chemosphere* 129: 33-38.
- Ahrens, L., Shoeib, M., Harner, T., Lee, S. C., Guo, R. and Reiner, E. J. (2011a). Wastewater Treatment Plant and Landfills as Sources of Polyfluoroalkyl Compounds to the Atmosphere. *Environmental Science & Technology* 45(19): 8098-8105.
- Ahrens, L., Herzke, D., Huber, S., Bustnes, J. O., Bangjord, G. and Ebinghaus, R. (2011b). Temporal trends and pattern of polyfluoroalkyl compounds in tawny owl (*Strix aluco*) eggs from Norway, 1986–2009. *Environmental Science & Technology* 45.
- Arias, V. A., Mallavarapu, M. and Naidu, R. (2015). Identification of the source of PFOS and PFOA contamination at a military air base site. *Environmental Monitoring and Assessment* 187(1).

- Armitage, J., Cousins, I. T., Buck, R. C., Prevedouros, K., Russell, M. H., MacLeod, M. and Korzeniowski, S. H. (2006). Modeling global-scale fate and transport of perfluorooctanoate emitted from direct sources. *Environmental Science & Technology* 40(22): 6969-6975.
- Armitage, J. M., MacLeod, M. and Cousins, I. T. (2009). Comparative Assessment of the Global Fate and Transport Pathways of Long-Chain Perfluorocarboxylic Acids (PFCAs) and Perfluorocarboxylates (PFCs) Emitted from Direct Sources. *Environmental Science & Technology* 43(15): 5830-5836.
- Arvaniti, O. S. and Stasinakis, A. S. (2015). Review on the occurrence, fate and removal of perfluorinated compounds during wastewater treatment. *Science of the Total Environment* 524: 81-92.
- Ballesteros-Gomez, A., Rubio, S. and van Leeuwen, S. (2010). Tetrahydrofuran-water extraction, in-line clean-up and selective liquid chromatography/tandem mass spectrometry for the quantitation of perfluorinated compounds in food at the low picogram per gram level. *J Chromatogr A* 1217(38): 5913-5921.
- Bao, J., Liu, W., Liu, L., Jin, Y. H., Dai, J. Y., Ran, X. R., Zhang, Z. X. and Tsuda, S. (2011). Perfluorinated Compounds in the Environment and the Blood of Residents Living near Fluorochemical Plants in Fuxin, China. *Environmental Science & Technology* 45(19): 8075-8080.
- Barry, V., Winkquist, A. and Steenland, K. (2013). Perfluorooctanoic Acid (PFOA) Exposures and Incident Cancers among Adults Living Near a Chemical Plant. *Environmental Health Perspectives* 121(11-12): 1313-1318.
- Begley, T. H., White, K., Honigfort, P., Twaroski, M. L., Neches, R. and Walker, R. A. (2005). Perfluorochemicals: Potential sources of and migration from food packaging. *Food Additives and Contaminants* 22(10): 1023-1031.
- Bertocchio, R., Lacote, G. and Verge, C. (1993). Synthesis of perfluoroalkyl iodides, Google Patents.
- Bjorklund, J. A., Thuresson, K. and De Wit, C. A. (2009). Perfluoroalkyl Compounds (PFCs) in Indoor Dust: Concentrations, Human Exposure Estimates, and Sources. *Environ Sci Technol* 43(7): 2276-2281.

- Boiteux, V., Bach, C., Sagres, V., Hemard, J., Colin, A., Rosin, C., Munoz, J. F. and Dauchy, X. (2016). Analysis of 29 per- and polyfluorinated compounds in water, sediment, soil and sludge by liquid chromatography-tandem mass spectrometry. *International Journal of Environmental Analytical Chemistry* 96(8): 705-728.
- Bonfiglio, R., King, R. C., Olah, T. V. and Merkle, K. (1999). The effects of sample preparation methods on the variability of the electrospray ionization response for model drug compounds. *Rapid Communications in Mass Spectrometry* 13(12): 1175-1185.
- Bossi, R., Riget, F. F. and Dietz, R. (2005). Temporal and spatial trends of perfluorinated compounds in ringed seal (*Phoca hispida*) from Greenland. *Environmental Science & Technology* 39(19): 7416-7422.
- Buck, R. C., Franklin, J., Berger, U., Conder, J. M., Cousins, I. T. and de Voogt, P. (2011). Perfluoroalkyl and polyfluoroalkyl substances in the environment: terminology, classification, and origins. *Integr Environ Assess Manag* 7.
- Butt, C. M., Muir, D. C. and Mabury, S. A. (2010). Elucidating the pathways of poly- and perfluorinated acid formation in rainbow trout. *Environmental Science & Technology* 44(13): 4973-4980.
- Calafat, A. M., Wong, L. Y., Kuklenyik, Z., Reidy, J. A. and Needham, L. L. (2007). Polyfluoroalkyl chemicals in the U.S. population: data from the National Health and Nutrition Examination Survey (NHANES) 2003-2004 and comparisons with NHANES 1999-2000. *Environ Health Perspect* 115(11): 1596-1602.
- Canada (2006). Perfluorooctane Sulfonate and its Salts and Certain Other Compounds Regulations (SOR/2008-178). <http://ec.gc.ca/lcpe-cepa/eng/regulations/detailReg.cfm?intReg=107>.
- Chambers, R. D. (2009). General Discussion of Organic Fluorine Chemistry. *Fluorine in Organic Chemistry*, Blackwell Publishing Ltd.: 1-22.
- Chu, S., Letcher, R. J., McGoldrick, D. J. and Backus, S. M. (2016). A New Fluorinated Surfactant Contaminant in Biota: Perfluorobutane Sulfonamide in Several Fish Species. *Environmental Science & Technology* 50(2): 669-675.

- Connolly, P. D., E., Zhu, X. and Keller, R. (2002). Analysis of Pooled Human Sera and Plasma and Monkey Sera for Fluorocarbons Using Exygen method ExM-023-071; U.S. EPA Administrative Record AR226-1152.
- Cousins, I. T., Vestergren, R., Wang, Z., Scheringer, M. and McLachlan, M. S. (2016). The precautionary principle and chemicals management: The example of perfluoroalkyl acids in groundwater. *Environ Int* 94: 331-340.
- D'eon, J. C., Crozier, P. W., Furdui, V. I., Reiner, E. J., Libelo, E. L. and Mabury, S. A. (2009). Observation of a Commercial Fluorinated Material, the Polyfluoroalkyl Phosphoric Acid Diesters, in Human Sera, Wastewater Treatment Plant Sludge, and Paper Fibers. *Environmental Science & Technology* 43(12): 4589-4594.
- D'Eon, J. C. and Mabury, S. A. (2007). Production of perfluorinated carboxylic acids (PFCAs) from the biotransformation of polyfluoroalkyl phosphate surfactants (PAPS): exploring routes of human contamination. *Environmental Science & Technology* 41(13): 4799-4805.
- D'Hollander, W., de Voogt, P., De Coen, W. and Bervoets, L. (2010). Perfluorinated Substances in Human Food and Other Sources of Human Exposure. *Reviews of Environmental Contamination and Toxicology*, Vol 208 208: 179-215.
- Daikin (2007). <http://www.daikin.com/press/2007/071221/>.
- Darrow, L. A., Stein, C. R. and Steenland, K. (2013). Serum Perfluorooctanoic Acid and Perfluorooctane Sulfonate Concentrations in Relation to Birth Outcomes in the Mid-Ohio Valley, 2005-2010. *Environmental Health Perspectives* 121(10): 1207-1213.
- De Felip, E., Abballe, A., Albano, F. L., Battista, T., Carraro, V., Conversano, M., Franchini, S., Giambanco, L., Iacovella, N., Ingelido, A. M., Maiorana, A., Maneschi, F., Marra, V., Mercurio, A., Nale, R., Nucci, B., Panella, V., Pirola, F., Porpora, M. G., Procopio, E., Suma, N., Valentini, S., Valsenti, L. and Vecchie, V. (2015). Current exposure of Italian women of reproductive age to PFOS and PFOA: A human biomonitoring study. *Chemosphere* 137: 1-8.

- De Silva, A. O., Allard, C. N., Spencer, C., Webster, G. M. and Shoeib, M. (2012). Phosphorus-Containing Fluorinated Organics: Polyfluoroalkyl Phosphoric Acid Diesters (diPAPs), Perfluorophosphonates (PFPA), and Perfluorophosphinates (PFPIAs) in Residential Indoor Dust. *Environmental Science & Technology* 46(22): 12575-12582.
- DeWitt, J. C. (2015). Toxicological Effects of Perfluoroalkyl and Polyfluoroalkyl Substances. DOI 10.1007/978-3-319-15518-0.
- Ding, H. H., Peng, H., Yang, M. and Hu, J. Y. (2012). Simultaneous determination of mono- and disubstituted polyfluoroalkyl phosphates in drinking water by liquid chromatography-electrospray tandem mass spectrometry. *Journal of Chromatography A* 1227: 245-252.
- Dinglasan, M. J., Ye, Y., Edwards, E. A. and Mabury, S. A. (2004). Fluorotelomer alcohol biodegradation yields poly- and perfluorinated acids. *Environmental Science & Technology* 38(10): 2857-2864.
- Domingo, J. L. (2012). Health risks of dietary exposure to perfluorinated compounds. *Environment International* 40: 187-195.
- Dookhith, M. T. (2001). Novel high performance water-soluble antifoam based on perfluoroalkyl-phosphinic and perfluoroalkylphosphonic acids in crop protection. *Pesticide Formulations and Application Systems: Twentieth Volume* 1400: 152-161.
- DuPont (2008). Fact Sheet DuPont™ Capstone™ Products For the Paper Packaging Industry
http://www2.dupont.com/Capstone/en_US/assets/downloads/final_factsheet_paper_packaging_mar_28_2008.pdf.
- ECHA (2014). ANNEX XV Restriction Report Proposal for a restriction. SUBSTANCE NAME: Perfluorooctanoic acid (PFOA), PFOA salts and PFOA-related substances.
<https://echa.europa.eu/documents/10162/e9cddee6-3164-473d-b590-8fcf9caa50e7>.
- EFSA (2008). European Food Safety Authority
<https://www.efsa.europa.eu/en/press/news/contam080721>.
- EFSA (2012). Perfluoroalkylated substances in food: occurrence and dietary exposure. European Food Safety Authority. *EFSA Journal* 2012; 10(6:2743).

- Ellis, D. A., Martin, J. W., De Silva, A. O., Mabury, S. A., Hurley, M. D., Andersen, M. P. S. and Wallington, T. J. (2004). Degradation of fluorotelomer alcohols: A likely atmospheric source of perfluorinated carboxylic acids. *Environmental Science & Technology* 38(12): 3316-3321.
- Ellis, D. A., Martin, J. W., Mabury, S. A., Hurley, M. D., Andersen, M. P. S. and Wallington, T. J. (2003). Atmospheric lifetime of fluorotelomer alcohols. *Environmental Science & Technology* 37(17): 3816-3820.
- EPA, U. (2006). US EPA (United States Environmental Protection Agency) 2010/2015 PFOA Stewardship Program. <http://www.epa.gov/assessing-and-managing-chemicals-under-tsca/20102015-pfoa-stewardship-program>.
- Falk, S., Brunn, H., Schroter-Kermani, C., Failing, K., Georgii, S., Taricone, K. and Stahl, T. (2012). Temporal and spatial trends of perfluoroalkyl substances in liver of roe deer (*Capreolus capreolus*). *Environmental Pollution* 171: 1-8.
- Fasano, W. J., Carpenter, S. C., Gannon, S. A., Snow, T. A., Stadler, J. C., Kennedy, G. L., Buck, R. C., Korzeniowski, S. H., Hinderliter, P. M. and Kemper, R. A. (2006). Absorption, distribution, metabolism, and elimination of 8-2 fluorotelomer alcohol in the rat. *Toxicological Sciences* 91(2): 341-355.
- FDA, U. (2016). US Food and Drug Administration (FDA). <https://www.federalregister.gov/documents/2016/01/04/2015-33026/indirect-food-additives-paper-and-paperboard-components>.
- Fei, C. Y., McLaughlin, J. K., Lipworth, L. and Olsen, J. (2009). Maternal levels of perfluorinated chemicals and subfecundity. *Human Reproduction* 24(5): 1200-1205.
- Filipovic, M., Woldegiorgis, A., Norstrom, K., Bibi, M., Lindberg, M. and Osteras, A. H. (2015). Historical usage of aqueous film forming foam: A case study of the widespread distribution of perfluoroalkyl acids from a military airport to groundwater, lakes, soils and fish. *Chemosphere* 129: 39-45.

- Filipovic, M. and Berger, U. (2015). Are perfluoroalkyl acids in waste water treatment plant effluents the result of primary emissions from the technosphere or of environmental recirculation? *Chemosphere* 129: 74-80.
- Fromme, H., Schlummer, M., Moller, A., Gruber, L., Wolz, G., Ungewiss, J., Bohmer, S., Dekant, W., Mayer, R., Liebl, B. and Twardella, D. (2007). Exposure of an adult population to perfluorinated substances using duplicate diet portions and biomonitoring data. *Environ Sci Technol* 41(22): 7928-7933.
- Gawor, A., Shunthirasingham, C., Hayward, S. J., Lei, Y. D., Gouin, T., Mmereki, B. T., Masamba, W., Ruepert, C., Castillo, L. E., Shoeib, M., Lee, S. C., Harner, T. and Wania, F. (2014). Neutral polyfluoroalkyl substances in the global Atmosphere. *Environmental Science-Processes & Impacts* 16(3): 404-413.
- Gebbink, W. A., Berger, U. and Cousins, I. T. (2015a). Estimating human exposure to PFOS isomers and PFCA homologues: the relative importance of direct and indirect (precursor) exposure. *Environment International* 74: 160-169.
- Gebbink, W. A., Glynn, A. and Berger, U. (2015b). Temporal changes (1997-2012) of perfluoroalkyl acids and selected precursors (including isomers) in Swedish human serum. *Environ Pollut* 199: 166-173.
- Gebbink, W. A., Bignert, A. and Berger, U. (2016a). Perfluoroalkyl Acids (PFAAs) and Selected Precursors in the Baltic Sea Environment: Do Precursors Play a Role in Food Web Accumulation of PFAAs? *Environmental Science & Technology* 50(12): 6354-6362.
- Gebbink, W. A., Bossi, R., Riget, F. F., Rosing-Asvid, A., Sonne, C. and Dietz, R. (2016b). Observation of emerging per- and polyfluoroalkyl substances (PFASs) in Greenland marine mammals. *Chemosphere* 144: 2384-2391.
- GEF (2016). GEF-Reduction and phase-out of PFOS in priority sectors in China.
<http://documents.worldbank.org/curated/en/275251473314607008/pdf/SFG2387-EA-P152959-Box396306B-PUBLIC-Disclosed-9-7-2016.pdf>.

- Gewurtz, S. B., Bhavsar, S. P., Petro, S., Mahon, C. G., Zhao, X. M., Morse, D., Reiner, E. J., Tittlemier, S. A., Braekevelt, E. and Drouillard, K. (2014). High levels of perfluoroalkyl acids in sport fish species downstream of a firefighting training facility at Hamilton International Airport, Ontario, Canada. *Environment International* 67: 1-11.
- Giesy, J. P. and Kannan, K. (2001). Global distribution of perfluorooctane sulfonate in wildlife. *Environmental Science & Technology* 35(7): 1339-1342.
- Gilljam, J. L., Leonel, J., Cousins, I. T. and Benskin, J. P. (2016). Is Ongoing Sulfluramid Use in South America a Significant Source of Perfluorooctanesulfonate (PFOS)? Production Inventories, Environmental Fate, and Local Occurrence. *Environmental Science & Technology* 50(2): 653-659.
- Glynn, A., Berger, U., Bignert, A., Ullah, S., Aune, M. and Lignell, S. (2012). Perfluorinated alkyl acids in blood serum from primiparous women in Sweden: serial sampling during pregnancy and nursing, and temporal trends 1996–2010. *Environmental Science & Technology* 46.
- Grandjean, P., Heilmann, C., Weihe, P. and Budtz-jørgensen, E. (2016a). New Insights Into PFAS Immunotoxicity. Presented at Dioxin 2016 Firenze, 36th International Symposium on Halogenated Persistent Organic Pollutants.
- Grandjean, P., Heilmann, C., Weihe, P., Nielsen, F., Mogensen, U. B. and Budtz-Jorgensen, E. (2016b). Serum Vaccine Antibody Concentrations in Adolescents Exposed to Perfluorinated Compounds. *Environ Health Perspect.*
- Gribble, M. O., Bartell, S. M., Kannan, K., Wu, Q., Faire, P. A. and Kamen, D. L. (2015). Longitudinal measures of perfluoroalkyl substances (PFAS) in serum of Gullah African Americans in South Carolina: 2003-2013. *Environmental Research* 143: 82-88.
- Guo, R., Reiner, E. J., Bhavsar, S. P., Helm, P. A., Mabury, S. A., Braekevelt, E. and Tittlemier, S. A. (2012). Determination of polyfluoroalkyl phosphoric acid diesters, perfluoroalkyl phosphonic acids, perfluoroalkyl phosphinic acids, perfluoroalkyl carboxylic acids, and perfluoroalkane sulfonic acids in lake trout from the Great Lakes region. *Analytical and Bioanalytical Chemistry* 404(9): 2699-2709.

- Hansen, K. J., Clemen, L. A., Ellefson, M. E. and Johnson, H. O. (2001). Compound-specific, quantitative characterization of organic: Fluorochemicals in biological matrices. *Environmental Science & Technology* 35(4): 766-770.
- Harada, K. H., Yang, H. R., Moon, C. S., Hung, N. N., Hitomi, T., Inoue, K., Niisoe, T., Watanabe, T., Kamiyama, S., Takenaka, K., Kim, M. Y., Watanabe, K., Takasuga, T. and Koizumi, A. (2010). Levels of perfluorooctane sulfonate and perfluorooctanoic acid in female serum samples from Japan in 2008, Korea in 1994-2008 and Vietnam in 2007-2008. *Chemosphere* 79(3): 314-319.
- Harding-Marjanovic, K. C., Houtz, E. F., Yi, S., Field, J. A., Sedlak, D. L. and Alvarez-Cohen, L. (2015). Aerobic Biotransformation of Fluorotelomer Thioether Amido Sulfonate (Lodyne) in AFFF-Amended Microcosms. *Environ Sci Technol* 49(13): 7666-7674.
- Haug, L. S., Thomsen, C. and Bechert, G. (2009). Time Trends and the Influence of Age and Gender on Serum Concentrations of Perfluorinated Compounds in Archived Human Samples. *Environmental Science & Technology* 43(6): 2131-2136.
- Haukas, M., Berger, U., Hop, H., Gulliksen, B. and Gabrielsen, G. W. (2007). Bioaccumulation of per- and polyfluorinated alkyl substances (PFAS) in selected species from the Barents Sea food web. *Environ Pollut* 148(1): 360-371.
- Herzke, D., Nygård, T., Heimstad, E. S., Uggerud, H. T., Hanssen, L. and Götsch, A. (2014). Environmental pollutants in the terrestrial and urban environment *Environmental Monitoring* M-261.
- Holmstrom, K. E., Johansson, A. K., Bignert, A., Lindberg, P. and Berger, U. (2010). Temporal Trends of Perfluorinated Surfactants in Swedish Peregrine Falcon Eggs (*Falco peregrinus*), 1974-2007. *Environmental Science & Technology* 44(11): 4083-4088.
- Houtz, E. F., Sutton, R., Park, J. S. and Sedlak, M. (2016). Poly- and perfluoroalkyl substances in wastewater: Significance of unknown precursors, manufacturing shifts, and likely AFFF impacts. *Water Research* 95: 142-149.

- Hu, X. C., Andrews, D. Q., Lindstrom, A. B., Bruton, T. A., Schaider, L. A., Grandjean, P., Lohmann, R., Carignan, C. C., Blum, A., Balan, S. A., Higgins, C. P. and Sunderland, E. M. (2016). Detection of Poly- and Perfluoroalkyl Substances (PFASs) in U.S. Drinking Water Linked to Industrial Sites, Military Fire Training Areas, and Wastewater Treatment Plants. *Environmental Science & Technology Letters*.
- Huber, S., Ahrens, L., Bardsen, B. J., Siebert, U., Bustnes, J. O., Vikingson, G. A., Ebinghaus, R. and Herzke, D. (2012). Temporal trends and spatial differences of perfluoroalkylated substances in livers of harbor porpoise (*Phocoena phocoena*) populations from Northern Europe, 1991-2008. *Science of the Total Environment* 419: 216-224.
- Insights, G. M. (2016). <https://www.gminsights.com/industry-analysis/fluorotelomers-market>.
- Jackson, D. A. and Mabury, S. A. (2012). Enzymatic kinetic parameters for polyfluorinated alkyl phosphate hydrolysis by alkaline phosphatase. *Environmental Toxicology and Chemistry* 31(9): 1966-1971.
- Jogsten, I. E., Nadal, M., van Bavel, B., Lindstrom, G. and Domingo, J. L. (2012). Per- and polyfluorinated compounds (PFCs) in house dust and indoor air in Catalonia, Spain: Implications for human exposure. *Environ Int* 39(1): 172-180.
- Jones, P. D., Naile, J., Wiseman, S. and Giesy, J. P. (2010). Effects of perfluorinated phosphate derivatives on gene expression in the H4IIE rat Hepatome cell line, 2011. Poster from SETAC 2010, available at [www.usask.ca/toxicology/jgiesy/about_prof_giesy/SETAC 2010/Jones et al 2010.pdf](http://www.usask.ca/toxicology/jgiesy/about_prof_giesy/SETAC_2010/Jones_et_al_2010.pdf).
- Karrman, A., Elgh-Dalgren, K., Lafossas, C. and Moskeland, T. (2011). Environmental levels and distribution of structural isomers of perfluoroalkyl acids after aqueous fire-fighting foam (AFFF) contamination. *Environmental Chemistry* 8(4): 372-380.
- Karrman, A., Ericson, I., van Bavel, B., Darnerud, P. O., Aune, M., Glynn, A., Lignell, S. and Lindstrom, G. (2007). Exposure of perfluorinated chemicals through lactation: Levels of matched human milk and serum and a temporal trend, 1996-2004, in Sweden. *Environmental Health Perspectives* 115(2): 226-230.

- Kasei, D. (2015). Surface Treatments.
<http://www.daitokasei.com/english/products/SURFACETREATMENTBROCHURE20151015.pdf>.
- Kato, K., Wong, L. Y., Jia, L. T., Kuklenyik, Z. and Calafat, A. M. (2011). Trends in exposure to polyfluoroalkyl chemicals in the U.S. Population: 1999-2008. *Environ Sci Technol* 45(19): 8037-8045.
- KEMI (2015). Förekomst och användning av högfluorerade ämnen och alternativ (The presence and use of highly fluorinated compounds and alternatives). Report 6/15.
<https://www.kemi.se/global/rapporter/2015/rapport-6-15-forekomst-och-anvandning-av-hogfluorerade-amnen-och-alternativ.pdf>
- Kim, M. H., Wang, N. and Chu, K. H. (2014a). 6:2 Fluorotelomer alcohol (6:2 FTOH) biodegradation by multiple microbial species under different physiological conditions. *Applied Microbiology and Biotechnology* 98(4): 1831-1840.
- Kim, S. K., Li, D. H., Shoeib, M. and Zoh, K. D. (2014b). Contribution of diffuse inputs to the aqueous mass load of perfluoroalkyl acids in river and stream catchments in Korea. *Science of the Total Environment* 470: 1430-1440.
- Lai, S. C., Song, J. W., Song, T. L., Huang, Z. J., Zhang, Y. Y., Zhao, Y., Liu, G. C., Zheng, J. Y., Mi, W. Y., Tang, J. H., Zou, S. C., Ebinghaus, R. and Xie, Z. Y. (2016). Neutral polyfluoroalkyl substances in the atmosphere over the northern South China Sea. *Environ Pollut* 214: 449-455.
- Lam, J. C. W., Lyu, J. L., Kwok, K. Y. and Lam, P. K. S. (2016). Perfluoroalkyl Substances (PFASs) in Marine Mammals from the South China Sea and Their Temporal Changes 2002-2014: Concern for Alternatives of PFOS? *Environmental Science & Technology* 50(13): 6728-6736.
- Lee, H., D'Eon, J. and Mabury, S. A. (2010). Biodegradation of polyfluoroalkyl phosphates as a source of perfluorinated acids to the environment. *Environ Sci Technol* 44(9): 3305-3310.

- Lee, H., De Siva, A. O. and Mabury, S. A. (2012). Dietary Bioaccumulation of Perfluorophosphonates and Perfluorophosphinates in Juvenile Rainbow Trout: Evidence of Metabolism of Perfluorophosphinates. *Environmental Science & Technology* 46(6): 3489-3497.
- Lee, H. and Mabury, S. A. (2011). A Pilot Survey of Legacy and Current Commercial Fluorinated Chemicals in Human Sera from United States Donors in 2009. *Environmental Science & Technology* 45(19): 8067-8074.
- Lee, H., Tevlin, A. G., Mabury, S. A. and Mabury, S. A. (2014). Fate of polyfluoroalkyl phosphate diesters and their metabolites in biosolids-applied soil: biodegradation and plant uptake in greenhouse and field experiments. *Environmental Science & Technology* 48(1): 340-349.
- Lehmle, H. J. (2005). Synthesis of environmentally relevant fluorinated surfactants - a review. *Chemosphere* 58(11): 1471-1496.
- Li, L., Zhai, Z. H., Liu, J. G. and Hu, J. X. (2015). Estimating industrial and domestic environmental releases of perfluorooctanoic acid and its salts in China from 2004 to 2012. *Chemosphere* 129: 100-109.
- Liu, C. and Liu, J. X. (2016). Aerobic biotransformation of polyfluoroalkyl phosphate esters (PAPs) in soil. *Environ Pollut* 212: 230-237.
- Liu, J., Lee, L. S., Nies, L. F., Nakatsu, C. H. and Turco, R. F. (2007). Biotransformation of 8 : 2 fluorotelomer alcohol in soil and by soil bacteria isolates. *Environ Sci Technol* 41(23): 8024-8030.
- Liu, J., Wang, N., Szostek, B., Buck, R. C., Panciroli, P. K., Folsom, P. W., Sulecki, L. M. and Bellin, C. A. (2010). 6-2 Fluorotelomer alcohol aerobic biodegradation in soil and mixed bacterial culture. *Chemosphere* 78(4): 437-444.
- Loi, E. I. H., Yeung, L. W. Y., Mabury, S. A. and Lam, P. K. S. (2013). Detections of Commercial Fluorosurfactants in Hong Kong Marine Environment and Human Blood: A Pilot Study. *Environmental Science & Technology* 47(9): 4677-4685.
- Lorber, M. and Egeghy, P. P. (2011). Simple Intake and Pharmacokinetic Modeling to Characterize Exposure of Americans to Perfluorooctanoic Acid, PFOA. *Environ Sci Technol* 45(19): 8006-8014.

- Ma, Z., Stramel, R. D., Yue, S., Iu, K. K., Chou, H. C. and Canfield, D. G. (2002). Color ink composition for graphic art ink jet image printers, Google Patents.
- Manzano-Salgado, C. B., Casas, M., Lopez-Espinosa, M. J., Ballester, F., Basterrechea, M., Grimalt, J. O., Jimenez, A. M., Kraus, T., Schettgen, T., Sunyer, J. and Vrijheid, M. (2015). Transfer of perfluoroalkyl substances from mother to fetus in a Spanish birth cohort. *Environmental Research* 142: 471-478.
- Martin, J. W., Ellis, D. A., Mabury, S. A., Hurley, M. D. and Wallington, T. J. (2006). Atmospheric chemistry of perfluoroalkanesulfonamides: Kinetic and product studies of the OH radical and Cl atom initiated oxidation of N-ethyl perfluorobutanesulfonamide. *Environmental Science & Technology* 40(3): 864-872.
- Martin, J. W., Mabury, S. A. and O'Brien, P. J. (2005). Metabolic products and pathways of fluorotelomer alcohols in isolated rat hepatocytes. *Chem Biol Interact* 155(3): 165-180.
- Martin, J. W., Smithwick, M. M., Braune, B. M., Hoekstra, P. F., Muir, D. C. G. and Mabury, S. A. (2004). Identification of long-chain perfluorinated acids in biota from the Canadian Arctic. *Environmental Science & Technology* 38(2): 373-380.
- McKeen, L. W. (2015). *Fluorinated Coatings and Finishes Handbook: The Definitive User's Guide*, William Andrew.
- Melzer, D., Rice, N., Depledge, M. H., Henley, W. E. and Galloway, T. S. (2010). Association between Serum Perfluorooctanoic Acid (PFOA) and Thyroid Disease in the US National Health and Nutrition Examination Survey. *Environmental Health Perspectives* 118(5): 686-692.
- Muller, C. E., Spiess, N., Gerecke, A. C., Scheringer, M. and Hungerbühler, K. (2011). Quantifying Diffuse and Point Inputs of Perfluoroalkyl Acids in a Nonindustrial River Catchment. *Environmental Science & Technology* 45(23): 9901-9909.
- Nelson, J. W., Hatch, E. E. and Webster, T. F. (2010). Exposure to polyfluoroalkyl chemicals and cholesterol, body weight, and insulin resistance in the general U.S. population. *Environ Health Perspect* 118(2): 197-202.

- Nilsson, H., Karrman, A., Rotander, A., van Bavel, B., Lindstrom, G. and Westberg, H. (2013). Biotransformation of fluorotelomer compound to perfluorocarboxylates in humans. *Environment International* 51: 8-12.
- Norway (2004). Forskrift om begrensning i bruk av helse- og miljøfarlige kjemikalier og andre produkter (produktforskriften). <https://lovdata.no/dokument/SF/forskrift/2004-06-01-922>.
- Nost, T. H., Vestergren, R., Berg, V., Nieboer, E., Odland, J. O. and Sandanger, T. M. (2014). Repeated measurements of per- and polyfluoroalkyl substances (PFASs) from 1979 to 2007 in males from Northern Norway: Assessing time trends, compound correlations and relations to age/birth cohort. *Environ Int* 67: 43-53.
- OECD (2007). Report of an OECD Workshop on Perfluorocarboxylic Acids (PFCAs) and Precursors. ENV/JM/MONO(2007)11.
- Oliaei, F., Kriens, D., Weber, R. and Watson, A. (2013). PFOS and PFC releases and associated pollution from a PFC production plant in Minnesota (USA). *Environmental Science and Pollution Research* 20(4): 1977-1992.
- Olsen, G. W., Huang, H. Y., Helzlsouer, K. J., Hansen, K. J., Butenhoff, J. L. and Mandel, J. H. (2005). Historical comparison of perfluorooctanesulfonate, perfluorooctanoate, and other fluorochemicals in human blood. *Environmental Health Perspectives* 113(5): 539-545.
- Olsen, G. W., Mair, D. C., Church, T. R., Ellefson, M. E., Reagen, W. K., Boyd, T. M., Herron, R. M., Medhdizadehkashi, Z., Nobiletti, J. B., Rios, J. A., Butenhoff, J. L. and Zobel, L. R. (2008). Decline in perfluorooctanesulfonate and other polyfluoroalkyl chemicals in American Red Cross adult blood donors, 2000-2006. *Environmental Science & Technology* 42(13): 4989-4995.
- Padilla-Sanchez, J. A. and Haug, L. S. (2016). A fast and sensitive method for the simultaneous analysis of a wide range of per- and polyfluoroalkyl substances in indoor dust using on-line solid phase extraction-ultrahigh performance liquid chromatography-time-of-flight-mass spectrometry. *J Chromatogr A* 1445: 36-45.
- Paul, A. G., Jones, K. C. and Sweetman, A. J. (2009). A first global production, emission, and environmental inventory for perfluorooctane sulfonate. *Environmental Science & Technology* 43.

- Pilot Chemical. (2016). <https://www.pilotchemical.com/products/masurf>.
- Post, G. B., Cohn, P. D. and Cooper, K. R. (2012). Perfluorooctanoic acid (PFOA), an emerging drinking water contaminant: a critical review of recent literature. *Environmental Research* 116: 93-117.
- Powley, C. R., George, S. W., Ryan, T. W. and Buck, R. C. (2005). Matrix effect-free analytical methods for determination of perfluorinated carboxylic acids in environmental matrixes. *Analytical Chemistry* 77(19): 6353-6358.
- Prevedouros, K., Cousins, I. T., Buck, R. C. and Korzeniowski, S. H. (2006). Sources, fate and transport of perfluorocarboxylates. *Environmental Science & Technology* 40(1): 32-44.
- Rand, A. A., Rooney, J. P., Butt, C. M., Meyer, J. N. and Mabury, S. A. (2014). Cellular Toxicity Associated with Exposure to Perfluorinated Carboxylates (PFCAs) and Their Metabolic Precursors. *Chemical Research in Toxicology* 27(1): 42-50.
- Rankin, K., Lee, H., Tseng, P. J. and Mabury, S. A. (2014). Investigating the biodegradability of a fluorotelomer-based acrylate polymer in a soil-plant microcosm by indirect and direct analysis. *Environmental Science & Technology* 48(21): 12783-12790.
- Rankin, K., Mabury, S. A., Jenkins, T. M. and Washington, J. W. (2016). A North American and global survey of perfluoroalkyl substances in surface soils: Distribution patterns and mode of occurrence. *Chemosphere* 161: 333-341.
- REACH (2010). COMMISSION REGULATION (EU) No 757/2010, <http://eur-lex.europa.eu/legal-content/EN/TXT/PDF/?uri=CELEX:32010R0757&from=EN>.
- Riget, F., Bossi, R., Sonne, C., Vorkamp, K. and Dietz, R. (2013). Trends of perfluorochemicals in Greenland ringed seals and polar bears: Indications of shifts to decreasing trends. *Chemosphere* 93(8): 1607-1614.
- Roos, A., Berger, U., Jarnberg, U., van Dijk, J. and Bignert, A. (2013). Increasing Concentrations of Perfluoroalkyl Acids in Scandinavian Otters (*Lutra lutra*) between 1972 and 2011: A New Threat to the Otter Population? *Environmental Science & Technology* 47(20): 11757-11765.

- Rosenmai, A. K., Nielsen, F. K., Pedersen, M., Hadrup, N., Trier, X., Christensen, J. H. and Vinggaard, A. M. (2013). Fluorochemicals used in food packaging inhibit male sex hormone synthesis. *Toxicology and Applied Pharmacology* 266(1): 132-142.
- Rotander, A., Karrman, A., van Bavel, B., Polder, A., Riget, F., Auousson, G. A., Vikingsson, G., Gabrielsen, G. W., Bloch, D. and Dam, M. (2012). Increasing levels of long-chain perfluorocarboxylic acids (PFCAs) in Arctic and North Atlantic marine mammals, 1984-2009. *Chemosphere* 86(3): 278-285.
- Routti, H., Letcher, R. J., Born, E. W., Branigan, M., Dietz, R., Evans, T. J., Fisk, A. T., Peacock, E. and Sonne, C. (2011). Spatial and temporal trends of selected trace elements in liver tissue from polar bears (*Ursus maritimus*) from Alaska, Canada and Greenland. *Journal of Environmental Monitoring* 13(8): 2260-2267.
- Rudel, H., Muller, J., Jurling, H., Bartel-Steinbach, M. and Koschorreck, J. (2011). Survey of patterns, levels, and trends of perfluorinated compounds in aquatic organisms and bird eggs from representative German ecosystems. *Environmental Science and Pollution Research* 18(9): 1457-1470.
- Salihovic, S., Karrman, A., Lindstrom, G., Lind, P. M., Lind, L. and van Bavel, B. (2013). A rapid method for the determination of perfluoroalkyl substances including structural isomers of perfluorooctane sulfonic acid in human serum using 96-well plates and column-switching ultra-high performance liquid chromatography tandem mass spectrometry. *Journal of Chromatography A* 1305: 164-170.
- Schroter-Kermani, C., Muller, J., Jurling, H., Conrad, A. and Schulte, C. (2013). Retrospective monitoring of perfluorocarboxylates and perfluorosulfonates in human plasma archived by the German Environmental Specimen Bank. *International Journal of Hygiene and Environmental Health* 216(6): 633-640.
- Schultz, M. M., Higgins, C. P., Huset, C. A., Luthy, R. G., Barofsky, D. F. and Field, J. A. (2006). Fluorochemical mass flows in a municipal wastewater treatment facility. *Environmental Science & Technology* 40(23): 7350-7357.

- Smithwick, M., Norstrom, R. J., Mabury, S. A., Solomon, K., Evans, T. J., Stirling, I., Taylor, M. K. and Muir, D. C. G. (2006). Temporal trends of perfluoroalkyl contaminants in polar bears (*Ursus maritimus*) from two locations in the North American Arctic, 1972-2002. *Environmental Science & Technology* 40(4): 1139-1143.
- Sundstrom, M., Ehresman, D. J., Bignert, A., Butenhoff, J. L., Olsen, G. W., Chang, S. C. and Bergman, A. (2011). A temporal trend study (1972-2008) of perfluorooctanesulfonate, perfluorohexanesulfonate, and perfluorooctanoate in pooled human milk samples from Stockholm, Sweden. *Environment International* 37(1): 178-183.
- Taniyasu, S., Kannan, K., Yeung, L. W. Y., Kwok, K. Y., Lam, P. K. S. and Yamashita, N. (2008). Analysis of trifluoroacetic acid and other short-chain perfluorinated acids (C2-C4) in precipitation by liquid chromatography-tandem mass spectrometry: Comparison to patterns of long-chain perfluorinated acids (C5-C18). *Analytica Chimica Acta* 619(2): 221-230.
- Taylor, K. W., Hoffman, K., Thayer, K. A. and Daniels, J. L. (2014). Polyfluoroalkyl Chemicals and Menopause among Women 20-65 Years of Age (NHANES). *Environmental Health Perspectives* 122(2): 145-150.
- Thompson, J., Eaglesham, G. and Mueller, J. (2011). Concentrations of PFOS, PFOA and other perfluorinated alkyl acids in Australian drinking water. *Chemosphere* 83(10): 1320-1325.
- Thompson, J., Lorber, M., Toms, L. M. L., Kato, K., Calafat, A. M. and Mueller, J. F. (2010). Use of simple pharmacokinetic modeling to characterize exposure of Australians to perfluorooctanoic acid and perfluorooctane sulfonic acid. *Environ Int* 36(4): 390-397.
- Toms, L. M. L., Thompson, J., Rotander, A., Hobson, P., Calafat, A. M., Kato, K., Ye, X., Broomhall, S., Harden, F. and Mueller, J. F. (2014). Decline in perfluorooctane sulfonate and perfluorooctanoate serum concentrations in an Australian population from 2002 to 2011. *Environ Int* 71: 74-80.
- Trier, X. and Numata, J. (2015). Uptake, elimination and toxicokinetic modeling of 13C4-8:2 diPAPs given with food to twelve males. Poster session presented at Fluoros workshop 2015, Colorado, United States.

- TRP (2002). Telomer Research Telomer (TRP) Update. Presented to U. S. EPA November 2002. U. S. EPA Public Docket AR226-1141.
- Ubel, F. A., Sorenson, S. D. and Roach, D. E. (1980). Health-Status of Plant Workers Exposed to Fluorochemicals - a Preliminary-Report. American Industrial Hygiene Association Journal 41(8): 584-589.
- UNEP (2009). UNEP /POPS/POPRC.1/9
[http://www.pops.int/Vdocuments/meetings/poprc/meeting_docs/en/K0582622_1-9 edited EJF.pdf](http://www.pops.int/Vdocuments/meetings/poprc/meeting_docs/en/K0582622_1-9_edited_EJF.pdf).
- Wang, N., Buck, R. C., Szostek, B., Sulecki, L. M. and Wolstenholme, B. W. (2012). 5:3 Polyfluorinated acid aerobic biotransformation in activated sludge via novel "one-carbon removal pathways". Chemosphere 87(5): 527-534.
- Wang, N., Liu, J., Buck, R. C., Korzeniowski, S. H., Wolstenholme, B. W., Folsom, P. W. and Sulecki, L. M. (2011). 6:2 fluorotelomer sulfonate aerobic biotransformation in activated sludge of waste water treatment plants. Chemosphere 82(6): 853-858.
- Wang, Z. Y., Cousins, I. T., Berger, U., Hungerbuhler, K. and Scheringer, M. (2016a). Comparative assessment of the environmental hazards of and exposure to perfluoroalkyl phosphonic and phosphinic acids (PFPA and PFPiAs): Current knowledge, gaps, challenges and research needs. Environment International 89-90: 235-247.
- Wang, P., Lu, Y. L., Wang, T. Y., Meng, J., Li, Q. F., Zhu, Z. Y., Sun, Y. J., Wang, R. S. and Giesy, J. P. (2016b). Shifts in production of perfluoroalkyl acids affect emissions and concentrations in the environment of the Xiaoqing River Basin, China. Journal of Hazardous Materials 307: 55-63.
- Wang, Y. W., Fu, J. J., Wang, T., Liang, Y., Pan, Y. Y., Cai, Y. Q. and Jiang, G. B. (2010). Distribution of Perfluorooctane Sulfonate and Other Perfluorochemicals in the Ambient Environment around a Manufacturing Facility in China. Environmental Science & Technology 44(21): 8062-8067.

- Wang, Z., Cousins, I. T., Scheringer, M., Buck, R. C. and Hungerbühler, K. (2014). Global emission inventories for C4–C14 perfluoroalkyl carboxylic acid (PFCA) homologues from 1951 to 2030, Part I: production and emissions from quantifiable sources. *Environment International* 70.
- Wang, Z. Y., Cousins, I. T., Scheringer, M. and Hungerbuhler, K. (2013). Fluorinated alternatives to long-chain perfluoroalkyl carboxylic acids (PFCAs), perfluoroalkane sulfonic acids (PFSA) and their potential precursors. *Environment International* 60: 242-248.
- Washington, J. W. and Jenkins, T. M. (2015). Abiotic Hydrolysis of Fluorotelomer-Based Polymers as a Source of Perfluorocarboxylates at the Global Scale. *Environmental Science & Technology* 49(24): 14129-14135.
- Weiner, B., Yeung, L. W. Y., Marchington, E. B., D'Agostino, L. A. and Mabury, S. A. (2013). Organic fluorine content in aqueous film forming foams (AFFFs) and biodegradation of the foam component 6 : 2 fluorotelomermercaptopalkylamido sulfonate (6 : 2 FTSAS). *Environmental Chemistry* 10(6): 486-493.
- Weiss, O., Wiesmuller, G. A., Bunte, A., Coen, T., Schmidt, C. K., Wilhelm, M. and Holzer, J. (2012). Perfluorinated compounds in the vicinity of a fire training area - Human biomonitoring among 10 persons drinking water from contaminated private wells in Cologne, Germany. *International Journal of Hygiene and Environmental Health* 215(2): 212-215.
- Vestergren, R. and Cousins, I. T. (2009). Tracking the Pathways of Human Exposure to Perfluorocarboxylates. *Environmental Science & Technology* 43(15): 5565-5575.
- Willson, M. (2010). Fluorotelomer based foams – are they safe for continued use? *Asia Pacific Fire Mag.*, 35 (2010), pp. 27–31.
- Wong, F., MacLeod, M., Mueller, J. F. and Cousins, I. T. (2014). Enhanced Elimination of Perfluorooctane Sulfonic Acid by Menstruating Women: Evidence from Population-Based Pharmacokinetic Modeling. *Environ Sci Technol* 48(15): 8807-8814.

- Xie, S. W., Wang, T. Y., Liu, S. J., Jones, K. C., Sweetman, A. J. and Lu, Y. L. (2013). Industrial source identification and emission estimation of perfluorooctane sulfonate in China. *Environment International* 52: 1-8.
- Yao, Y. M., Chang, S., Sun, H. W., Gan, Z. W., Hu, H. W., Zhao, Y. Y. and Zhang, Y. F. (2016). Neutral and ionic per- and polyfluoroalkyl substances (PFASs) in atmospheric and dry deposition samples over a source region (Tianjin, China). *Environ Pollut* 212: 449-456.
- Yeung, L. W. Y. and Mabury, S. A. (2016). Are humans exposed to increasing amounts of unidentified organofluorine? *Environmental Chemistry* 13(1): 102-110.
- Yeung, L. W. Y., Miyake, Y., Taniyasu, S., Wang, Y., Yu, H. X., So, M. K., Jiang, G. B., Wu, Y. N., Li, J. G., Giesy, J. P., Yamashita, N. and Lam, P. K. S. (2008). Perfluorinated Compounds and Total and Extractable Organic Fluorine in Human Blood Samples from China. *Environmental Science & Technology* 42(21): 8140-8145.
- Yeung, L. W. Y., De Silva, A. O., Loi, E. I. H., Marvin, C. H., Taniyasu, S., Yamashita, N., Mabury, S. A., Muir, D. C. G. and Lam, P. K. S. (2013a). Perfluoroalkyl substances and extractable organic fluorine in surface sediments and cores from Lake Ontario. *Environment International* 59: 389-397.
- Yeung, L. W. Y., Robinson, S. J., Koschorreck, J. and Mabury, S. A. (2013b). Part I. A Temporal Study of PFCAs and Their Precursors in Human Plasma from Two German Cities 1982-2009. *Environ Sci Technol* 47(8): 3865-3874.
- Yeung, L. W. Y., Robinson, S. J., Koschorreck, J. and Mabury, S. A. (2013c). Part II. A Temporal Study of PFOS and Its Precursors in Human Plasma from Two German Cities in 1982-2009. *Environmental Science & Technology* 47(8): 3875-3882.
- Yoo, H., Washington, J. W., Jenkins, T. M. and Ellington, J. J. (2011). Quantitative Determination of Perfluorochemicals and Fluorotelomer Alcohols in Plants from Biosolid-Amended Fields using LC/MS/MS and GC/MS. *Environmental Science & Technology* 45(19): 7985-7990.

- Zabaleta, I., Bizkarguenaga, E., Prieto, A., Ortiz-Zarragoitia, M., Fernandez, L. A. and Zuloaga, O. (2015). Simultaneous determination of perfluorinated compounds and their potential precursors in mussel tissue and fish muscle tissue and liver samples by liquid chromatography-electrospray-tandem mass spectrometry. *Journal of Chromatography A* 1387: 13-23.
- Zhang, L., Liu, J. G., Hu, J. X., Liu, C., Guo, W. G., Wang, Q. and Wang, H. (2012). The inventory of sources, environmental releases and risk assessment for perfluorooctane sulfonate in China. *Environ Pollut* 165: 193-198.
- Zushi, Y. and Masunaga, S. (2009). Identifying the Nonpoint Source of Perfluorinated Compounds Using a Geographic Information System Based Approach. *Environmental Toxicology and Chemistry* 28(4): 691-700.

PUBLICATIONS *in the series*
ÖREBRO STUDIES IN CHEMISTRY

1. Bäckström, Mattias, *On the Chemical State and Mobility of Lead and Other Trace Elements at the Biogeosphere/Technosphere Interface*. 2002.
2. Hagberg, Jessica, *Capillary zone electrophoresis for the analysis of low molecular weight organic acids in environmental systems*. 2002.
3. Johansson, Inger, *Characterisation of organic materials from incineration residues*. 2003.
4. Dario, Mårten, *Metal Distribution and Mobility under alkaline conditions*. 2004.
5. Karlsson, Ulrika, *Environmental levels of thallium – Influence of redox properties and anthropogenic sources*. 2006.
6. Kärrman, Anna, *Analysis and human levels of persistent perfluorinated chemicals*. 2006.
7. Karlsson Marie, *Levels of brominated flame retardants in humans and their environment: occupational and home exposure*. 2006.
8. Löthgren, Carl-Johan, *Mercury and Dioxins in a MercOx-scrubber*. 2006.
9. Jönsson, Sofie, *Microextraction for usage in environmental monitoring and modelling of nitroaromatic compounds and haloanisoles*. 2008.
10. Ericson Jogsten, Ingrid, *Assessment of human exposure to per- and polyfluorinated compounds (PFCs). Exposure through food, drinking water, house dust and indoor air*. 2011.
11. Nilsson, Helena, *Occupational exposure to fluorinated ski wax*. 2012.
12. Salihovic, Samira, *Development and Application of High-throughput Methods for Analysis of Persistent Organic Pollutants in Human Blood*. 2013.
13. Larsson, Maria, *Chemical and bioanalytical characterisation of PAH-contaminated soils. Identification, availability and mixture toxicity of AhR agonists*. 2013.

14. Kalaitzaki, Argyro, *Biocompatible microemulsions: formulation, encapsulation of bioactive compounds and their potential application*. 2014
15. Saqib, Naeem, *Distribution and chemical association of trace elements in incinerator residues and mining waste from a leaching perspective*. 2016
16. Chatzidaki, Maria, *Formulation and characterization of W/O nano-dispersions for bioactive delivery applications*. 2016
17. Geng, Dawei, *Gas Chromatography-Atmospheric Pressure Chemical Ionization-Tandem Mass Spectrometry Methods for the Determination of Environmental Contaminants*. 2016
18. Eriksson, Ulrika, *Contribution of polyfluoroalkyl phosphate esters (PAPs) and other precursor compounds to perfluoroalkyl carboxylates (PFCAs) in humans and the environment*. 2016