



# Substance report

June 2022



**Per- and poly-fluoroalkyl substances (PFAS)**



science and policy  
for a healthy future



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The EEA has since updated this document to reflect the work developed before the conclusion of HBM4EU, with the support of the CGL and other colleagues.

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## Glossary

PFASs compounds		
Abbreviated name	Full name	CAS No
PFOA	Perfluorooctanoic acid	335-67-1
PFOS	Perfluorooctane sulphonate	1763-23-1
PFNA	Perfluoro-n-nonanoic acid	375-95-1
PFDA	Perfluoro-n-decanoic acid	335-76-2
PFU(n)DA	Perfluoro-n-undecanoic acid	2058-94-8
PFDoDA	Perfluorodeconoic Acid	307-55-1
PFTTrDA	Perfluoro-n-tridecanoic acid	72629-94-8
PFTeDA	Perfluoro-n-tetradecanoic acid	376-06-7
PFHxS	Perfluoro-1-hexanesulfonate	355-46-4
FOSA	Perfluorooctylsulfonamide	754-91-6
PFOSA	Perfluorooctanesulfonic acid amide	754-91-6
HFPO-DA	Hexafluoropropylene oxide-dimer acid	
GenX	Hexafluoropropylene oxide-dimer acid	
8:2 FTOH	8:2 fluorotelomer alcohol	678-39-7
Abbreviations		
AOP	Adverse Outcome Pathway	
CAS	Chemical Abstracts Service	
C&L	Classification & Labelling	
CLP	Classification, Labelling, Packaging	
DNEL	Derived No-Effect Level (DNEL)	
ECHA	European Chemicals Agency	
EDC	Endocrine Disrupting Chemical	
EFSA	European Food Safety Authority	
HBM	Human Biomonitoring	
HBM-GV	Human Biomonitoring Guidance Values	
IARC	International Agency for Research on Cancer	
KEMI	Swedish Chemicals Agency	
OECD	Organisation for Economic Co-operation and Development	
REACH	Registration, Evaluation, Authorisation and Restriction of Chemicals	
SVHC	Substance of Very High Concern	
TDI	Tolerable Daily Intake	
TWI	Tolerable Weekly Intake	
C&L Classification Names		
Carc	Carcinogenicity	
Acute Tox	Acute Toxicity	
Repr	Reproductive Toxicity	

PFASs compounds	
Muta	Mutagenicity
STOT RE	Specific target organ toxicity - repeated exposure
STOT SE	Specific target organ toxicity- single exposure
Eye Dam/ Irrit.	Eye Damage / Eye Irritation
Resp Sens.	Respiratory Sensitivity
Skin Corr / Irrit.	Skin Corrosion/Irritation
Skin Sens.	Skin Sensitivity
Properties of concern	
R	Toxic to Reproduction
ED	Endocrine Disrupting
SS	Skin sensitising
PBT	Persistent, Bioaccumulative and Toxic
vPvB	Very Persistent, Very Bioaccumulative

## 1 Key Messages

- Per- and polyfluoroalkyl substances (PFASs) are a large group of man-made chemicals that have been extensively used in a wide number of different industrial and consumer applications due to their useful physicochemical properties (e.g., durability, water- and oil-repellence, and high chemical and thermal stability).
- PFASs are persistent in the environment and tend to bioaccumulate in food chains. Many PFASs are also shown to be toxic to human health. Several have been linked to a number of negative health effects including increased cholesterol levels, reduced infant birth weights, effects on the immune system, increased risk for cancer, and thyroid hormone disruption.
- For the general population, the main route of exposure to PFASs will be through the consumption of drinking water or through food. Humans can also be exposed through consumer uses (e.g., textile products) and in occupational settings (e.g., in PFASs production or industrial use).
- HBM4EU Aligned Studies<sup>1</sup> (2014-2021) have generated baseline levels of internal exposure to 12 PFASs for European teenagers (1957 samples; age: 12-18 years).
- 14.26 % of the European teenagers tested exceed the internal serum level of 6.9 µg/L PFASs, EFSA's<sup>2</sup> guideline value for a tolerable weekly intake of 4.4 ng/kg. The maximum exceedance from individual studies was 23.8 %. Highest median values are observed in studies conducted in Northern and Western Europe.
- PFASs data from 17 HBM-studies can already be consulted in the online European HBM dashboard. Current exposure exceeds the EFSA Guidance values for PFASs in some parts of the EU population.
- PFASs concentrations are in general higher in men with a trend on participants with higher educational level having higher exposure levels. In some studies, higher levels of PFASs were observed with increasing age.

<sup>1</sup> The HBM4EU Aligned Studies are a survey aimed at collecting HBM samples and data as harmonised as possible from (national) studies to derive current internal exposure data representative for the European population/citizens across a geographic spread

- From the HBM4EU data collections, a decreasing trend for PFOA and PFOS concentrations can be derived, while this is not the case for other PFASs

## 2 Introduction

HBM4EU is a project funded under Horizon 2020, running from 2017 to June 2022. It generates knowledge to inform the safe management of chemicals and protection of human health in Europe. HBM4EU uses human biomonitoring (HBM) to monitor the actual human exposure to chemicals and resulting health impacts and to improve chemical risk assessment. HBM4EU compares data from across Europe, which allows an understanding of regional differences and helps to identify vulnerable groups, in order to inform targeted measures to reduce exposure. The results of the HBM4EU project are aimed at supporting policy development, by providing a key evidence base in the understanding of exposure and impacts to toxic chemicals.

If you would like to read more about the project itself, please visit the HBM4EU [website](#).

### How to use this document

This document provides a summary of the known and suspected adverse human health effects of per- and polyfluoroalkyl substances (PFASs) and describes the main exposure pathways for humans. It also indicates where HBM could be of value in the development of EU policy, along with the remaining challenges in determining PFASs exposure for humans. This brief is intended to inform policy makers and other interested stakeholders on the value of HBM to establish the EU population's exposure to PFASs compounds.

This policy brief is based largely on the [scoping document](#) and [research brief](#) for PFASs, as well as the accompanying reports on [legislative mapping](#), and the summary of HBM4EU results in relation to the [key policy questions](#). Where necessary, additional information from ECHA documents including the Classification and Labelling (C&L) Inventory, and legislative text for relevant EU policy areas, have also been used for this brief.

### Overview of PFASs

Per- and polyfluoroalkyl substances (PFASs) are a family of man-made chemicals that have been extensively used in a wide number of different industrial and consumer applications since the 1950s due to their unique physical and chemical properties (Glüge et al., 2020).

PFASs consist of a fully (per-) or partly (poly-) fluorinated carbon chain connected to different 'functional groups' (see OECD, 2013). The length of the chain (i.e. the number of carbon atoms), the number of fluorine atoms, and the type of functional groups will vary widely between different compounds. This possible set of variations means that the PFASs family are made up of a huge number of different individual chemical structures. The OECD (OECD, 2018) estimate approximately 4,700 known individual chemical substances, while others estimate that closer to around 6,000 substances belong to the PFASs family. Recently a classification tree to browse per- and polyfluoroalkyl substances (PFAS) and other fluorinated compounds has been created in PubChem. Searching for PFAS using the OECD PFAS definition of saturated CF<sub>2</sub> (Report ENV/CBC/MONO(2021)25 from 9 July 2021) results in more than 6 million entries<sup>2</sup>.

Based on the length of the fluorinated carbon chain, PFASs can be identified as either 'short-chain' or 'long-chain' (OECD, 2013). This chain length can result in different physical and chemical properties that influence how the substance behaves in the environment and the effect on human

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<sup>2</sup> <https://pubchem.ncbi.nlm.nih.gov/classification/#hid=120>

health. A general classification of different PFASs based on a commonly agreed terminology has been provided by OECD [here](#). For further information on the definition, identity and categorisation of PFASs, see OECD (2018) [here](#).

Uses of PFASs include textiles; leather; cosmetic products; food contact materials; paper and board; firefighting foams; household articles and consumer mixtures; construction products; lubricants and greases; industrial chemicals used in chrome plating; semiconductors; mixtures for treatment of skis; medical devices and apparel; applications within oil; gas and mining industry and refrigeration and cooling applications; transportation (automotive, aviation etc.) and photographic surface layers.

Some of the unique physicochemical properties of PFASs that have made them so useful and popular in these applications (such as extremely low surface tension and/or durable water- and oil-repellence, and high chemical and thermal stability) are also associated with potentially negative impacts on the environment and human health.

PFASs are extremely persistent chemicals that accumulate in food chains, humans, and the environment. Their resistance to degradation, and high mobility in the environment mean that PFASs are now found everywhere, including in remote environments such as the Arctic (OECD, 2013; European Commission, 2019) and have been observed to contaminate water and soil in most EU countries and it is extremely difficult and costly to clean up such contamination.

Within the past decade, several 'long-chain' PFASs (e.g., PFOS, PFOA) have been recognised as extremely persistent, bioaccumulative and toxic and are now restricted or banned under EU legislation (see Section 5). More recently, there have been mounting concerns and evidence that 'shorter chain' PFASs (e.g., HFPO-DA and PFBS) are persistent and mobile in the environment, potentially leading to ground water contamination in future. This is a serious concern, particularly where manufacturers and industry may have switched from longer chain to shorter chain PFASs following the previous regulatory actions.

According to KEMI (2017) there are 2,817 PFASs commercially in use. Adequate data is only available for about 15 % of these substances, whereas for 40 % data are missing entirely (KEMI, 2017). Many fluorinated substances enter the EU through the import of articles (e.g., textiles) and for the most part these are not monitored (KEMI, 2015) providing an indirect exposure source. The lack of data concerns identification, use, and exposure beside from toxicity and ecotoxicity. However, more and more chemicals with PFAS structure are becoming known. Recently a classification tree to browse per- and polyfluoroalkyl substances (PFAS) and other fluorinated compounds has been created in PubChem. Searching for PFAS using the OECD PFAS definition of saturated CF<sub>2</sub> ([Report ENV/CBC/MONO\(2021\)25 from 9 July 2021](#)) results in more than 6 million entries<sup>3</sup>.

PFASs have been studied in detail by a number of authoritative bodies. For example, the Organisation for Economic Co-operation and Development (OECD) has developed a comprehensive [database](#) of possible of PFASs, including precursors. As discussed in TURI (2020), PFASs have been recommended for listing under the Toxic or Hazardous Substances (TURA) List by the TURA Science Advisory Board (SAB). It has been noted by TURI (2020) that because there are several thousand known PFASs chemical, the SAB has determined that it is not practical to review each chemical individually, and, although many of these chemicals are being discharged into the environment, many of them have not been studied with regard to health or environmental effects.

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<sup>3</sup> <https://pubchem.ncbi.nlm.nih.gov/classification/#hid=120>

The scoping document covers a large number of individual PFASs compounds – for purposes of this policy brief, the discussion, for example on specific health effects, exposure, and policy aspects, is limited mostly to the substances designated as Category A (18 individual substances), see Section 7.

### 3 Human exposure to PFAS

PFASs are synthetic, man-made substances so are not naturally released to the environment but are released through human activities and uses. The total number of sites potentially emitting PFASs in Europe is estimated to be in the order of 100 000 (Nordic Council of Ministers, 2019). PFASs are ubiquitous in the environment and organisms across Europe (Valsecchi et al., 2013), and have been detected in air, soil, plants, biota and humans (Houde et al., 2006 – see, [OECD, 2015](#)).

As discussed in Section 2, PFASs are highly persistent in the environment. It has been shown that PFASs accumulate in the environment and have been found to contaminate surface, ground- and drinking water (Eschauzier et al., 2012; Sun et al., 2016; Gebbink et al., 2017), and accumulate in plants (Ghisi et al., 2019). Whereas most data are available for the small group of long-chain PFASs, there is also considerable exposure to other fluorinated alternatives (such as Gen-X), for example in drinking water (Gebbink et al., 2017).

Humans can be exposed to PFASs directly (e.g., via diet, drinking water, use of consumer products) and indirectly through transformation of ‘precursor’ substances e.g., polyfluoroalkyl phosphate esters (PAPs), fluorotelomer alcohols (FTOHs), fluorotelomer iodides (FTIs) and fluorotelomer acrylate (FTA) in the environment. The precursor contribution to PFASs daily exposures can contribute from <10% to >50% (Gebbink et al., 2015).

An overview of the main sources of exposure (environmental, occupational, consumer) and exposure pathways (oral, inhalation, dermal) is provided in **Error! Reference source not found.** Additional information on these sources and pathways is provided below in Section 3.1 and Appendix 1.

#### Key exposure sources – General population

PFASs appear to be ubiquitous in human blood based on the widespread detection of these substances in human serum samples (see ATSDR, 2018, EFSA, 2020). The key exposure routes for PFASs to humans have been outlined by the [EEA](#) and include the following aspects (see Appendix 1):

- For the general population, the main PFASs sources of exposure are drinking water (e.g., WHO, 2017; Banzhaf et al, 2017), the diet, consumer products and dust (EFSA, 2018).
- For populations that have elevated levels of PFASs in water supplies, the primary route of exposure is expected to be ingestion of contaminated drinking water (ATSDR (2018).
- In food, fish species at the top of the food chain and shellfish are significant sources of PFASs exposure (EFSA, 2020).
- Livestock and poultry raised on contaminated land can accumulate PFASs in their meat, milk, and eggs (Ingelido et al., 2018; Numata et al., 2014).
- Other key sources of dietary exposure for PFASs include fruit, and vegetables, as well as drinking water (EFSA, 2020).
- The EFSA opinion on PFASs (EFSA, 2018, 2020) concluded that, based on the estimated exposure, that parts of the European population exceed the derived tolerable daily intake, which is of concern.

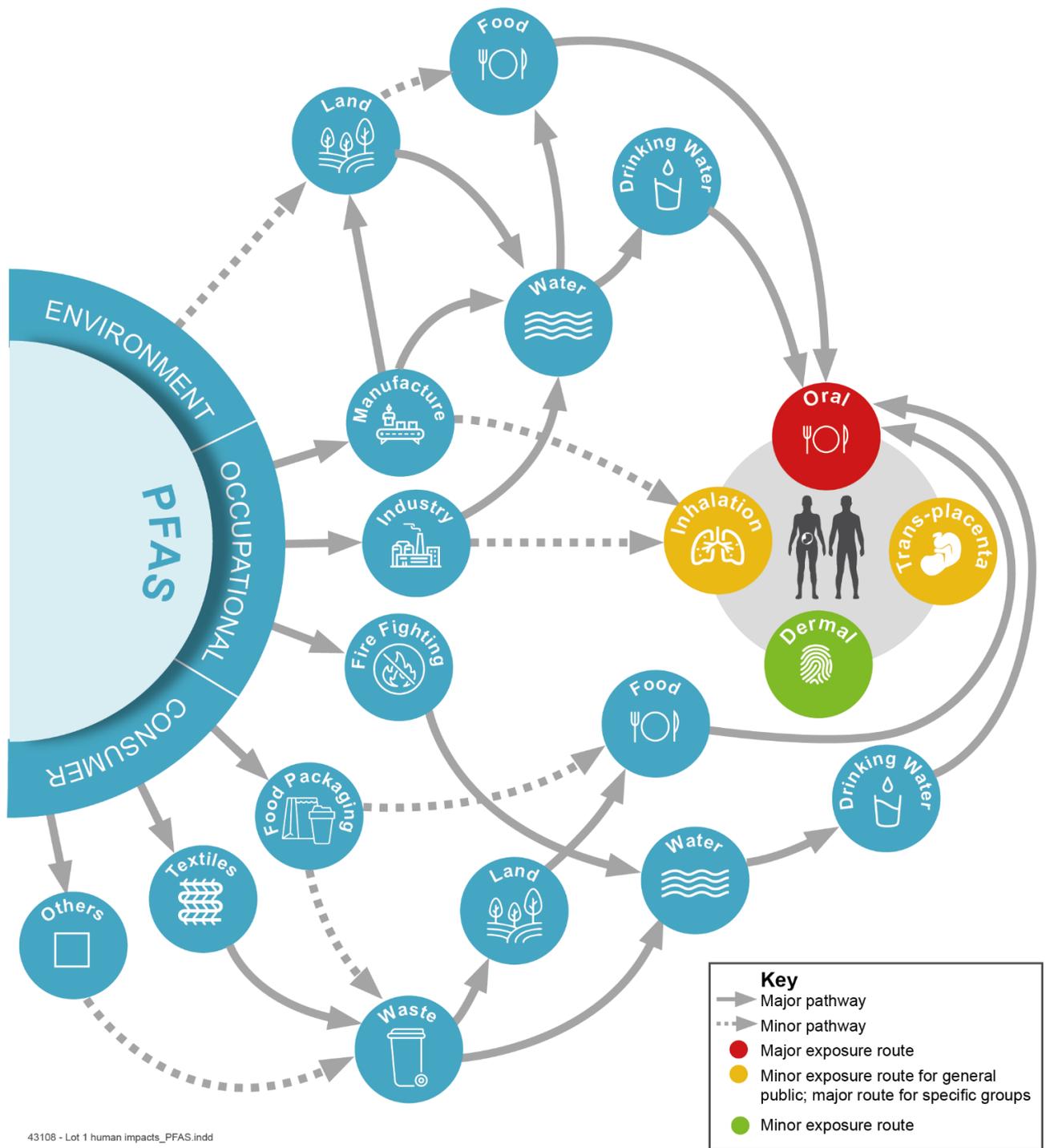
- Toddlers and other children are noted to have approximately two-fold higher mean intake (from food) than adults (EFSA, 2020).
- PFASs are present in various consumer products e.g. in textiles (e.g., waterproof clothing), but also in paper, paints, and inks, polishing and cleaning products, food packaging, cosmetic products, and drugs and medical devices. (Danish EPA, 2018; Schultes et al., 2018; Nordic Council of Ministers, 2019)
- Emissions to the environment occur via industrial wastewater releases, as well as emissions to air from industrial production sites followed by deposition onto soil and water bodies. Industrial and urban wastewater treatment plants are also a significant source of PFASs, via air, water, and sludge (Hamid, et al., 2016; Eriksson et al., 2017)
- Reuse of contaminated sewage sludge as fertilisers has led to PFASs pollution of soil (Ghisi et al., 2019) and water in Austria, Germany, Switzerland, and the US (Nordic Council of Ministers, 2019).
- The maternal transfer of PFASs to offspring can occur, both prenatally (in utero) and postnatally (via breastfeeding) (ATSDR, 2018; EFSA, 2020).

### **Key exposure sources – Occupational**

Individuals involved in activities with prolonged use of PFAS-containing products, such as the application of protective coatings for fabrics and carpet and the use of paper coatings or ski wax, may have higher levels of exposure to PFASs compounds than the general population (ATSDR, 2018). Production and use of PFASs have been the main sources of PFASs exposure over time (Wang et al., 2014a, 2014b; Hu et al., 2016) for instance from fluoropolymer production installations and from the use of PFAS-containing firefighting foams. Areas around industrial production, manufacturing and application sites have been found to be particularly contaminated by PFASs, and therefore represents a concern for environmental exposure and indirect exposure to the general population.

### **Vulnerable populations**

It is expected that the highest exposures to PFASs can occur for PFASs production and manufacturing workers, as well as communities located near fluorochemical facilities, and individuals with prolonged use PFAS-containing products (ATSDR, 2018). These populations may have higher exposure to PFASs than the general population based on elevated concentrations of these substances measured in air, soil, sediment, surface water, groundwater, and vegetation surrounding these facilities.



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**Figure 3.1 Overview of exposure route and pathways for PFAS**  
 Dermal edermal exposure is currently investigated by EFSA

## 4 Health impacts of PFAS

### Overview of key health impacts of PFAS

Some PFASs have been documented as bioaccumulative and/or toxic substances. For example, PFOA, HFPO-DA, PFBS, PFNA, PFDA, PFTeDA, PFTrDA, PFU(n)DA and PFHxS, are included in the substances of very high concern (SVHC) List under the REACH Regulation due to their very persistent and very bioaccumulative (vPvB) or persistent, bioaccumulative and toxic (PBT) properties, or properties of equivalent concern to those of vPvB/PBT and carcinogenic, mutagenic, or reprotoxic (CMR) substances. In line with this, some PFASs are classified in the EU as toxic to reproduction, the liver, and as suspected carcinogens (see below).

An overview of current EU (ECHA C&L Inventory) and/or IARC classification of PFASs is provided in Table 4.1 below (see Glossary for full list of terms/classifications). It should be noted that, because of the large number of PFASs compounds covered in the HBM4EU project (there are 80+ substances named in the [scoping document](#)) this overview is limited to a number of specific PFASs listed in Category A (see Table 7.1, Section 7.1).

Table 4.1 Overview of CLP classifications for PFAS

Substance	Properties of concern						Category according to CLP criteria							ECHA info card			
	Toxic to Reproduction (R)	Endocrine Disrupting (ED)	Skin sensitising (SS)	Persistent, Bioaccumulative and Toxic (PBT)	Very Persistent / Very Bioaccumulative (vPvB)	Persistent Organic Pollutant (POP)	Carcinogenicity	Acute Toxicity	Reproductive Toxicity	Mutagenicity	Specific target organ tox (repeated exposure)	Specific target organ tox (single exposure)	Eye Damage/ Eye Irritation		Respiratory Sensitivity	Skin Corrosion/ Irritation	Skin Sensitivity
PFOA							2*	4*	1B*		1*		1*				<a href="#">link</a>
PFOS							2*	4*	1B*		1*		1*				<a href="#">link</a>
PFNA							2*	4*	1B*		1*		1*				<a href="#">link</a>
PFDA							2*		2*								<a href="#">link</a>
PFU(n)DA								4					2		2		<a href="#">link</a>
PFDODA												3	2		2		<a href="#">link</a>
PFTrDA																	<a href="#">link</a>
PFTeDA															1B		<a href="#">link</a>
PFHxS								4							1B		<a href="#">link</a>
FOSA, PFOSA								3					3	2	2		<a href="#">link</a>

\* Harmonised classification under the CLP Regulation. (Other classifications are those notified to the CLP inventory but without harmonised EU classification.). Blank cells denote a lack of classification.

**Key:**



The human health effects of PFASs have been evaluated in a large number of epidemiology and animal studies (see, for example, in-depth discussion in EFSA, 2020 and ATSDR, 2018). An overview of the evidence from epidemiological and animal research into the health impacts of PFASs on humans (as presented in ATSDR, 2018) is provided in Table 4.2 below.

**Table 4.2 Overview of health effects evidence on PFASs (from ATSDR, 2018 and EFSA, 2020)**

Health impact	Human studies (epidemiology)	Animal studies
<b>Liver / cholesterol effects</b>	<ul style="list-style-type: none"> <li>PFOA, PFOS, and PFHxS linked with liver damage</li> <li>Clear evidence for an association between exposure to PFOA, PFOS, PFNA, and increases in cholesterol levels.</li> </ul>	<ul style="list-style-type: none"> <li>Studies indicate the liver is a sensitive target of PFOA, PFOS, PFHxS, PFNA, PFDeA, PFUA, PFBA, PFBS, PFDoA, and PFHpA toxicity (e.g., linked with increases in liver weight).</li> </ul>
<b>Cardiovascular effects</b>	<ul style="list-style-type: none"> <li>Evidence for an association between serum PFOA and PFOS and pregnancy-induced hypertension and/or pre-eclampsia.</li> <li>Epidemiological studies provide insufficient evidence to conclude on associations between exposure to PFASs and increased risk of cardiovascular disease.</li> </ul>	<ul style="list-style-type: none"> <li>No evidence presented in ATSDR, 2018 report</li> </ul>
<b>Endocrine effects</b>	<ul style="list-style-type: none"> <li>Suggestive evidence of a link between serum PFOA and PFOS and an increased risk of thyroid disease.</li> </ul>	<ul style="list-style-type: none"> <li>Many PFASs decreased the levels of thyroid hormones.</li> </ul>
<b>Immune effects</b>	<ul style="list-style-type: none"> <li>PFOS and PFOA are associated with reduced antibody response to vaccination, observed in several studies.</li> <li>Evidence is suggestive of a link between serum PFOA, PFOS, PFNA, PFHxS, and PFDeA levels and decreased antibody responses to vaccines.</li> <li>Increased propensity for infections in children exposed in the womb up to age 4 and the frequency of use antibiotics until adolescent age.</li> <li>Inconsistent associations with asthma and allergy</li> </ul>	<ul style="list-style-type: none"> <li>Evidence suggests that immune endpoints are sensitive targets of PFOA and PFOS toxicity. The most commonly reported effect was an impaired response to antigens.</li> <li>PFOS and PFOA have been shown to cause a reduced response to vaccination (T-cell dependent antibody response) and PFOS also caused a reduced resistance to infection. It is concluded that the immune system is a prime target of PFASs.</li> </ul>

<b>Reproductive effects</b>	<ul style="list-style-type: none"> <li>• A suggestive link between serum PFOA and PFOS levels and an increased risk of lower fertility.</li> <li>• There may well be a causal association between PFOS and PFOA and birth weight</li> <li>• No associations between other PFASs and fertility and reproductive outcomes in both males and females</li> </ul>	<ul style="list-style-type: none"> <li>• In general, studies of PFOA and PFOS have not found alterations in fertility.</li> <li>• Effects on male reproductive parameters have been reported for PFNA and PFDA.</li> </ul>
<b>Developmental effects</b>	<ul style="list-style-type: none"> <li>• Link between serum PFOA and PFOS and small decreases in birth weight.</li> </ul>	<ul style="list-style-type: none"> <li>• Indicated that developmental endpoints are targets of PFOA, PFOS, PFHxS, PFNA, PFDeA, PFUA, and PFBA toxicity. The developmental effects include decreases in body weight and decreases in survival rate.</li> </ul>
<b>Neurological effects</b>	<ul style="list-style-type: none"> <li>• Epidemiological studies provide no evidence for associations between exposure to PFASs and neurodevelopmental outcomes, growth in infancy or childhood, neurobehavioural, neuropsychiatric, cognitive outcomes.</li> </ul>	<ul style="list-style-type: none"> <li>• Studies with PFDA, PFHxS and PFDoDA indicate developmental neurotoxic effects</li> </ul>
<b>Carcinogenicity</b>	<ul style="list-style-type: none"> <li>• Epidemiological studies provide insufficient support for carcinogenicity of PFOS and PFOA in humans.</li> </ul>	<ul style="list-style-type: none"> <li>• Limited number of studies are available that indicate that PFOS and PFOA are tumour promoters in rodent livers.</li> </ul>

For some health effects, the level of evidence from human and animal studies is sufficient to establish a link to PFASs exposure. For example, [The Agency for Toxic Substances and Disease Registry \(ATSDR\)](#) has indicated that research involving humans suggests that high levels of certain PFASs are linked with health impacts including:

- Increased cholesterol levels
- Decreased vaccine response in children
- Changes in liver enzymes
- Increased risk of high blood pressure or pre-eclampsia in pregnant women
- Small decreases in infant birth weights
- Increased risk of kidney or testicular cancer

In animal studies, there is no evidence of cardiovascular effects, according to ATSDR.

However, for a number of other health effects, the evidence is currently lacking. EFSA (2020) has concluded that epidemiological studies currently provide insufficient evidence to conclude on associations between exposure to PFASs and increased risk of cardiovascular disease, neurodevelopment outcomes, growth in infancy or childhood, neurobehavioural, neuropsychiatric, cognitive outcomes, thyroid function, changes in kidney function and carcinogenicity in humans.

Based on consideration of the harmonised classification of PFASs compounds (see Table 4.1) and the evidence from human and animal studies, as discussed by EFSA (2020) and ATSDR (2018) (see Table 4.2 and discussion above), the health effects associated with exposure to PFASs are summarised in Figure 4.1. Further explanation on the categorisation of substances for different health effects is provided in Appendix 1.<sup>4</sup>

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<sup>4</sup> An explanation of the categorisation of the strength of evidence for the health effects presented in Figure 4.1 is provided in Appendix 2.

**Figure 4.1 Overview of health effects associated with PFAS**

Target organ of the body	Effects	Relevant Substances	 Adults (men)	 Adults (women)	 Infants / Foetuses
Organ specific/ whole body  	Cancer (stomach, liver, kidneys)	PFOS, PFOA, PFNA, PFDA			
		Other PFAS			
Liver  	Liver damage	PFOA, PFOS, and PFHxS			
		Other PFAS (e.g. PFNA, PFDeA, PFUA, PFBA, PFBS, PFDoA, and PFHpA)			
Immune system  	Reduced immune response to vaccination (e.g. toward rubella vaccine, tetanus and diphtheria); reduced resistance to infection.	PFOS, PFOA			
		Other PFAS			
Endocrine system  	Decreased thyroid hormone levels; effects on thyroid metabolism and lipid metabolism	PFOS, PFOA			
		Other PFAS			
Reproductive effects  	Increased risk of miscarriage, reduced birth weight, increased weight in adult life, and reduced fertility	PFOA, PFOS, PFNA and PFDA			
		Other PFAS			
Blood  	Increased cholesterol levels (hypercholesteremia)	PFOS, PFOA, PFNA			
		Other PFAS			
Brain/neurological system  	Developmental neurotoxic effects	All PFAS			

**Key:**  Strong evidence  Suspected  Evidence lacking  Not applicable

## 5 EU Policies on PFASs

At EU level, PFASs are regulated by a number of pieces of legislation and cross-regulation activities. These cover i) implementation of international conventions, actions and agreements, and wider chemicals legislation; ii) consumer products; iii) occupational exposure, and iv) the environment (e.g., emissions to air and water). An overview of these regulatory measures at EU level are provided in **Error! Reference source not found.** below.<sup>5</sup>

A number of other PFASs are on the REACH Candidate List of SVHCs. REACH Annex XVII restricts the use of C9-C14 PFCAs, including their salts and any combinations in the market starting from February 25, 2023. The limit value is:

- C9-C14 PFCAs and their salts < 0.0000025%
- C9-C14 PFCA-related substances < 0.000026%.

In June 2019 and January 2020, two PFASs groups (HFPO-DA and PFBS) were identified as SVHCs, based on their persistence, mobility, and toxicity, which were considered to pose a threat to human health and wildlife when exposed through the environment (including through drinking water).<sup>6</sup>

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<sup>5</sup> RPA (2020) Legislative Mapping Per/poly Fluorinated Compounds

<sup>6</sup> <https://echa.europa.eu/hot-topics/perfluoroalkyl-chemicals-pfas>

**Table 5.1 Overview of EU policy measures relating to PFAS**

<b>Chemicals</b>	<ul style="list-style-type: none"> <li>The <a href="#">EU Chemical's Strategy</a> includes specific <a href="#">Elements for an EU-strategy for PFASs</a> which sets out specific actions to minimise environmental and human exposure to PFASs, at all stages of their life cycle.</li> <li>The <b>Stockholm Convention</b> on persistent organic pollutants – PFOS and PFOA are listed under Annex A (full ban) so are restricted globally. PFHxS has also been approved for listing under Annex A</li> <li><b>POPs Regulation (2019/1021/EU)</b> - implements the Stockholm Convention on POPs and bans/restricts the manufacturing, marketing and use of POPs in the EU.</li> <li><b>REACH</b> - Several (7) (PFASs PFOA, HFPO-DA, PFBS, PFNA, PFDA, PFTeDA, PFTrDA, PFU(n)DA and PFHxS) are subject to restrictions and/or authorisations under REACH; several (7) individual PFASs are listed as SVHCs with others also being considered.</li> <li>A <a href="#">broad restriction</a> on the use of all PFASs in a wider range of applications is currently being prepared</li> <li>Restrictions on the use of all PFASs in specific applications (e.g. <a href="#">fire-fighting foams</a> and <a href="#">textiles</a>) are in preparation.</li> <li><b>CLP</b> – Several PFASs (e.g., PFOA, PFDA, PFNA) are subject to EU harmonized classification and labelling under Regulation (see Section 3).</li> <li><b>Human biomonitoring (HBM) values</b> – set by the <a href="#">HBM German Commission</a>. This includes the HBM-I-value* in blood plasma for PFOA of 2 µg/L, and for PFOS of 5µg/L for the general population, with a HBM-II-value** in blood plasma for PFOA of 10 µg/L for the general population and 5 µg/L in women of child-bearing age; and for PFOS of 20 µg/L for the general population and 10 µg/L in women of child-bearing age. These values are not in a chemicals' legislations, nor are they regulatory binding.</li> </ul>	<p><b>Food Safety</b></p> <p><i>Food Contact Materials Regulations:</i> Regulation (EC) No 1935/2004 on materials and articles intended to come into contact with food; and Commission Regulation (EU) No 10/2011 on plastic materials and articles intended to come into contact with food – PFOA is no longer permitted for use in food contact materials.</p> <p>EFSA has set a new <a href="#">safety threshold</a> for the sum of four PFAS– a group tolerable weekly intake (TWI) of 4.4 nanograms per kilogram of body weight. These PFASs are PFOA, PFOS, PFNA, PFHxS.</p> <p><b>Cosmetics</b></p> <p><b>Regulation (EC) No 1223/2009 on cosmetic products</b> (Cosmetic Products Regulation) - several PFASs (incl. PFOS, PFOPA and PTFE) are not permitted for use in cosmetic products.</p>	<b>Consumer</b>
<b>Environmental</b>	<p><i>Water</i></p> <ul style="list-style-type: none"> <li><b>The Water Framework Directive</b> - PFOS and PFOA are listed as priority hazardous substances and have set EQS values.</li> <li>Directive 98/83/EC on the quality of water intended for human consumption (<b>The Drinking Water Directive</b>) with &gt;70 individual PFASs species covered by the DWD.</li> </ul> <p><i>Waste</i></p> <ul style="list-style-type: none"> <li><b>The Waste Framework Directive</b> - Several PFASs (including PFHxS, PFBA , PFPeA ,PFHxA, PFHpA, PFBS) meet the criteria properties of waste which render it hazardous under Annex III of the Waste Framework Directive.</li> </ul>	<ul style="list-style-type: none"> <li><b>Council Directive 98/24/EC</b> on the protection of the health and safety of workers from the risks related to chemical agents at work (the Chemical Agents Directive – CAD) - 10 individual PFASs are covered by the CAD</li> </ul>	<b>Occupational</b>

\* The HBM-I-value represents the concentration of a substance in human biological material below which – according to the knowledge and judgement of the HBM Commission – there is no risk for adverse health effects and, consequently, no need for action.

\*\* The HBM-II-value represents the concentration of a substance in a human biological material above which there is an increased risk for adverse health effects and, consequently, an acute need for exposure reduction measures and the provision of biomedical advice.

## 6 Policy questions for PFAS

### 6.1 Introduction

Embedded in the substance prioritisation, stakeholders were asked to identify policy related questions that HBM4EU should address in order to contribute to the strengthening of policy ambitions on PFASs. A number of policy-related questions that relate to the commitments under this frame have been developed. Further background detail on PFASs and how the policy questions were selected is available in the [scoping document](#) and the [report on stakeholder consultation and mapping of needs](#).

### 6.2 What is the current exposure of the EU population to PFASs, and do they exceed Guidance values (reference and HBM values), where available?

We collected information on the human PFASs exposure levels in Europe sampled between 2005 and 2015. These aggregated data are now available at the European HBM dashboard. P50 values for PFOA range from 0.76 to 4.8 µg/L, PFNA levels from 0.28 to 0.86 µg/L and PFHxS from 0.18 to 1.61 µg/L. PFOS remains the dominant congener; P50 values range from 1.67 µg/L to 8.06 µg/L. These levels in newborns, children & teenagers combined, and adults support the concentration levels reported in the EFSA opinion on the risks to human health related to the presence of perfluoroalkyl substances in food (2020).

The individual data collections prepared and made available within HBM4EU also contained aggregated data stratified by age, sex and educational level. From these stratifications it can be seen that the PFASs concentrations are in general higher in men compared to women for the teenager and adult studies. This could probably be explained by the elimination of PFASs through menstruation, and for mothers also through delivery and lactation. Also, there seems to be a trend that participants with higher educational level have higher exposure levels compared with low to medium educational level. This trend was however less obvious and could not be observed in all studies and for all compounds. In some studies, higher levels of PFASs were observed with increasing age, indicating possible cumulative exposure over time. This was however not observed for all studies and not for all compounds.

The HBM4EU Aligned Studies have generated baseline levels of internal PFASs concentrations in serum/plasma samples of European 12-18 years of age from 9 sampling sites for the period 2014-2021. PFOS has the highest median values and P95 values observed, followed by PFOA, PFHxS and PFNA. In general, PFOS is the main determinant of the sum of 4 PFASs, the contribution of PFNA and PFHxS is lower **teenagers**. Risk for adverse health effects cannot be excluded: the indicator developed under HBM4EU shows that current combined exposure to PFOS, PFOA, PFNA and PFHxS exceeds the health-based guidance value based on the EFSA guideline value for a tolerable weekly intake of 4.4 ng/kg (2020) of 6.9 µg/L for  $\Sigma(\text{PFOA} + \text{PFNA} + \text{PFHxS} + \text{PFOS})$  in a fraction of the participants. Exceedances in the different studies and locations range from 1.3%-23.8% with an overall exceedance of 14.3 % and an extent of exceedance (P95/6.9 µg/L) varying from 0.74 - 1.78. The studies conducted in Western and Northern Europe had the most teenagers exceeding the guidance value.

P50 and P95 concentrations of sum of PFOS + PFHxS + PFOA + PFNA in serum are in the range of 2.55-5.15 µg/L and 5.09-12.31 µg/L across studies in teenagers. The share with exposure levels exceeding the serum level of 6.9 µg/L corresponding to the EFSA TWI ranges from 1.34-23.78 %.

All teenagers had detectable levels of PFOS in their blood samples. P50 and P95 of serum PFOS concentrations are in the range of 1.34-2.79 µg/L and 3.06-8.23 µg/L across studies. The share of individuals with exposure levels exceeding the HBM-I value of 5 µg/L ranges from 1-18 %.

P50 and P95 of serum PFOA concentrations are in the range of 0.66-1.47 µg/L and 1.03-3.12 µg/L across studies. The share of individuals with exposure levels exceeding the HBM-I value of 2 µg/L ranges from 0-20.28%.

P50 serum PFPeA concentrations are in the range of 0.02-0.10 µg/L across studies, with 5 studies with P50 < detection limit (range: 0.01-0.2 µg/L). P95 serum PFPeA concentrations are in the range of 0.17-0.19 µg/L across studies, with 5 studies with P95 < detection limit (range: 0.01-0.2 µg/L). Health-based guidance values are not available for PFPeA.

P50 serum PFHxA concentrations are in the range of 0.07-0.14 µg/L across studies, with 6 studies with P50 < detection limit (range: 0.01-0.25 µg/L). P95 serum PFHxA concentrations are in the range of 0.07-0.21 µg/L across studies, with 5 studies with P95 < detection limit (range: 0.05-0.25 µg/L). Health-based guidance values are not available for PFHxA.

P50 serum PFHpA concentrations are in the range of 0.03-0.07 µg/L across studies, with 6 studies with P50 < detection limit (range: 0.01-0.25 µg/L). P95 serum PFHpA concentrations are in the range of 0.09-0.16 µg/L across studies, with 5 studies with P95 < detection limit (range: 0.01-0.25 µg/L). Health-based guidance values are not available for PFHpA.

P50 and P95 of serum PFNA concentrations are in the range of 0.17-0.54 µg/L, with 1 study with P50 < detection limit of 0.5 µg/L, and 0.46-1.38 µg/L across studies. Health-based guidance values are not available for PFNA.

P50 and P95 of serum PFDA concentrations are in the range of 0.05-0.22 µg/L, with 3 studies with P50 < detection limit (range: 0.2-0.25 µg/L) and 0.17-0.51 µg/L across studies. Health-based guidance values are not available for PFDA.

P50 serum PFUnDA concentrations are in the range of 0.04-0.11 µg/L across studies, with 5 studies with P50 < detection limit (range: 0.004-0.25 µg/L). P95 serum PFUnDA concentrations are in the range of 0.08-0.35 µg/L across studies, with 1 study with P95 < detection limit of 0.25 µg/L. Health-based guidance values are not available for PFUnDA.

P50 serum PFDoDA concentrations of all studies are < detection limit (range: 0.005-0.25 µg/L) across studies. P95 serum PFDoDA concentrations are in the range of 0.025-0.030 µg/L across studies, with 7 studies with P95 < detection limit (range: 0.01-0.25 µg/L). Health-based guidance values are not available for PFDoDA.

P50 serum PFBS concentrations of all studies are < detection limit (range: 0.01-0.25 µg/L) across studies. P95 serum PFBS concentrations are in the range of 0.20-0.37 µg/L across studies, with 7 studies with P95 < detection limit (range: 0.01-0.25 µg/L). Health-based guidance values are not available for PFBS.

P50 and P95 of serum PFHxS concentrations are in the range of 0.23-0.68 µg/L, with 1 study with P50 < detection limit of 0.34 µg/L) and 0.43-2.32 µg/L across studies. Health-based guidance values are not available for PFHxS.

P50 serum PFHpS concentrations are in the range of 0.03-0.05 µg/L across studies (with 4 studies with P50 < detection limit (range: 0.05-0.25 µg/L). P95 serum PFHpS concentrations are in the range of 0.10-0.25 µg/L across studies, with 2 studies with P95 < detection limit (range: 0.05-0.2 µg/L). Health-based guidance values are not available for PFHpS.

Three different mixture risk assessment approaches indicate that PFASs exposure exceeds guidance values, and thus may cause adverse health effects in certain segments of the European population, thereby confirming the conclusion drawn in the recent EFSA scientific opinion. It should be mentioned that this work is meant as a scientific exploration of how to use HBM in risk assessment. This exercise explores various approaches to perform mixture risk assessment for

PFASs that are based on pragmatism and available data and therefore have their underlying assumptions, limitations and uncertainties.

### **6.3 Are there differences in exposure of the EU population to regulated and non-regulated PFASs?**

In general, there are limited data on PFASs beside PFOS, PFOA, PFHxS and PFNA, further the limits of detections are not sufficient in all studies to detect low concentrations. It can be assumed – also based on the results of the PBPK model that certain PFASs accumulate slowly over time due to their long half-life. The HBM4EU Aligned Studies show that to date, PFOS and PFOA are still the substances occurring in the highest concentrations in blood in Europe. Other PFASs compounds are also detected in many human samples. Alternative PFASs compounds (substitutes of legacy PFASs) have at present still lower exposure levels compared to regulated PFASs compounds. However, due to a large proportion of non-detects for alternative PFASs and a big difference in absolute values of LOQs reached across studies there is a need for lowering the LOQs for these compounds. Further, it has to be taken into consideration that these substances accumulate in the body over several years.

### **6.4 Has restriction of PFOS according to the POP Regulation led to a reduction in exposure, especially for children?**

The existing data collected in HBM4EU demonstrate a decrease of PFASs within the observed period. Different datasets were considered investigating the burden to the four PFASs (PFOS, PFOA, PFNA and PFHxS) in adult human serum. Although only few datasets were available and there was uncertainty related to robustness, for PFOA and PFOS – which are the most regulated PFASs – a decrease was shown between 2008 and 2015. This is particularly clear in the case of time series measurements, such as those carried out within the framework of the German Environmental Specimen Bank. When comparing the plasma samples of young adults from the German Specimen Bank in the period from 2007 to 2019, a clear decrease can be seen. While the maximum P95 (P50) value for the sum of the 4 PFASs was 28.87 (13,82) µg/l in 2007, in 2019 it is only 8.28 (4,59) µg/l.

Further, available HBM data in adult females aged 20-39 years from Belgium show a reduction of PFOS by half from 2008 to 2014, while data from Denmark show only a minimal decrease from 2010 to 2012.

Although in some individual EU countries (Austria, Germany, Belgium) decreasing time trends of PFOA and PFOS have been described, new data collected in the HBM4EU Aligned Studies still show that a fraction of the teenagers in Europe exceed guidance values for PFOA & PFOS and that substitute PFASs are detected. The HBM4EU Aligned Studies data will form baseline European exposure levels for PFASs in teenagers, allowing follow up studies to monitor increased or decreased usage.

### **6.5 Is exposure driven by diet, consumer exposure, occupation or environmental contamination?**

Regarding exposure determinants in the HBM4EU data collections, besides cohort, sex and education, diet was an important determinant of PFASs. Higher serum levels of PFNA and PFOS were associated with higher consumption of fish and seafood (increase in serum levels by 20 and 21 %, respectively) and higher consumption of eggs (increase in serum levels by 14 and 11 %, respectively). Additionally, higher exposure to PFOS was linked to higher consumption of offal (increase in exposure by 14 %) and consumption of local food (increase in exposure by 40 %). For other food items (meat, fast food, drinking water, milk), no or weak associations with individual PFASs were found.

Modelling external and internal PFASs exposure showed that food intake was the most important contributive route to the exposure of PFOS and PFOA, with percentages of 97% and 98% of the total intake, respectively. These estimations were made based on a study from Catalonia (D12.1). This is in line with the assessment of dietary exposure to PFOS and PFOA, PFNA and PFHxS by EFSA (EFSA 2018, 2020). Despite exposure in hot spot regions or specific occupational settings, diet is the main source of PFASs exposure.

Preliminary data analysis of the study to investigate PFASs exposure in chromate plating facilities indicates a statistically significant difference in PFASs plasma concentrations of chrome platers (n=52) when compared to controls (n=57) for some PFASs compounds (namely PFOA, PFHxS, L-PFHpS and L-PFOS). Median concentrations of chrome platers were at maximum about two times higher (PFHxS and L-PFHpS) than respective median concentrations of controls. P95 levels of chrome platers were at maximum about 20 times higher (L-PFHpS and L-PFOS) when compared to controls.

## **6.6 Which areas and environmental media in Europe are contaminated with PFASs?**

PFASs accumulate in the environment and have been found to contaminate surface-, ground- and drinking water and accumulate in plants. PFASs production sites, fire training areas, airports and waste disposal facilities as well as sewage treatment plants can lead to contamination of the environment, which in turn leads to exposure of people living in these areas. Currently, there are several hotspots known in different countries (e.g., Germany, Sweden, Italy, Spain, The Netherlands, Belgium, Denmark and Austria). It can be assumed that hot spots exist in most European countries.

Therefore, the need for joint action to address PFAS pollution in hotspots in the EU has been recognised and initial activities to this end have been launched within HBM4EU. An inventory of human biomonitoring studies in PFAS hotspots in Europe was created: overview the exposure levels, type and source of the contamination, exposure determinants, health research and policy impact of PFAS contamination at various hotspots across Europe.

Further, a workshop on PFAS hotspots was organized (2 May 2022). During this half-day workshop, the knowledge and challenges related to PFAS hotspots across Europe were presented and discussed with European, regional and local risk managers of contaminated sites, environmental health care workers, scientists, and regulatory authorities involved in chemical risk assessment. A guidance document on identification and monitoring, human biomonitoring, and risk communication in PFAS hotspots has been drafted. The guidance aims to be useful for policy makers and scientists confronted with new PFAS hotspots.

## **6.7 How can this feed into an assessment of the TDI for PFOS and PFOA set by EFSA?**

Workshop on the first HBM4EU research results on PFASs (April 2021) had the goal to discuss the results with selected scientists, risk assessors and risk managers to define main messages for policy making. It is clearly supported that there is an (additional) need for policy action, and the importance of dialogue between experts, risk managers and assessors was demonstrated. Concrete opportunities for the use of HBM4EU results include:

- Most recent and quality controlled HBM data are important to support the PFASs group restriction and to demonstrate that there is a need for action.
- Results on e.g. health effects, MOA, mixture risk assessment can support the qualitative risk assessment.
- HBM results can support the discussion started in the EU related to set maximum levels in food.

- At national level HBM4EU results can raise awareness among the public and among the policy makers, also since many important competences to reduce exposure are still situated at the national/local level.

A follow up workshop has been organised in March 2022; the results will feed into regulatory processes, such as the broad restriction which is planned under REACH.

From the examinations of the birth cohorts several associations have been reported: (i) increased propensity for infections in the children up to age 4 and the frequency of antibiotics use until adolescent age, (ii) poorer cardiovascular risk profile based on higher cholesterol and lipid profile, higher fasting blood glucose, BMI and blood pressure (iii) higher body weight, BMI-score and waist circumference at age 9, among boys. Further, PFASs mixture was associated with an increase in triglyceride and insulin levels and decrease in HDL cholesterol. And a modest interaction with endogenous hormones has been found. Prenatal PFASs exposure was also associated with reproductive disorders.

HBM4EU Aligned Studies results can become an important baseline to follow up effectiveness of policy measures. HBM based indicators have been developed within HBM4EU to track progress in future and can be included in state-of-the-environment reporting at EU- and national level. Continued investment in monitoring, ideally with a time interval of 2 to 3 years between data points, would be needed for this purpose.

## **6.8 What is the impact of a pending 2016 ECHA decision to restrict the manufacturing, marketing and use of PFOA under REACH?**

The data collections in HBM4EU indicate the decrease of PFOA within the observed period. This is particularly clear in the case of time series measurements, such as those carried out within the framework of the German Environmental Specimen Bank (ESB). Comparing the plasma samples of young adults from ESB in the period from 2007 to 2019 shows a decrease by half. However, although in some individual EU countries, decreasing time trends of PFOA have been described, the HBM4EU aligned studies still show that PFOS and PFOA are still the substances occurring in the highest concentrations in blood in Europe and a fraction of the teenagers in Europe exceed guidance values for PFOA. Other PFASs compounds have also been detected in many human samples. Alternative PFASs compounds have lower exposure levels compared to regulated PFASs compounds. However, due to a large proportion of non-detects for alternative PFASs and a big difference in absolute values of LOQs reached across studies there is a need for lowering the LOQ. It is of utmost importance to avoid regrettable substitutions.

## **6.9 Is it important to eliminate legacy PFASs from material cycles (i.e. waste electronic equipment) when implementing a circular economy in order to protect human health?**

This policy questions goes beyond the research focus of HBM4EU. However, due to the long half-life in humans, the exceedances of tolerable weekly intakes and internal benchmark dose levels of substances which are already restricted such as PFOS and PFOA it seems indicated to eliminate PFASs from material cycles when implementing a circular economy in order to protect human health.

## **6.10 Can differences in PFASs profiles be observed in different population groups and time periods?**

The individual existing data collections prepared and made available within HBM4EU contain aggregated data stratified by age, sex and educational level. From these stratifications it can be seen that the PFASs concentrations are in general higher in men compared to women for the teenager and adult studies. Also, there seems a trend that participants with higher educational level have higher exposure levels compared with low to medium educational level. This trend was

however less obvious and could not be observed in all studies and for all compounds. In some studies, higher levels of PFASs were observed with increasing age, indicating possible cumulative exposure over time. This was however not observed for all studies and for all compounds.

Regarding exposure determinants in the HBM4EU data collections, besides cohort, sex and education, diet was an important determinant of PFASs. Higher serum levels of PFNA and PFOS were associated with higher consumption of fish and seafood (increase in serum levels by 20 and 21 %, respectively) and higher consumption of eggs (increase in serum levels by 14 and 11 %, respectively). Additionally, higher exposure to PFOS was linked to higher consumption of offal (increase in exposure by 14 %) and consumption of local food (increase in exposure by 40 %). For other food items (meat, fast food, drinking water, milk), no or weak associations with individual PFASs were found.

Geographical differences in internal exposure can be observed in the HBM4EU Aligned Studies for PFOS, PFOA, PFNA, PFHxS and their sum. Highest median values are observed in studies conducted in Northern and Western Europe.

So far, time trend data are available for the sum  $\Sigma(\text{PFOA} + \text{PFNA} + \text{PFHxS} + \text{PFOS})$  only for Germany. When comparing the PFASs levels in plasma samples of young adults from the German Environmental Specimen Bank in the period from 2007 to 2019, a clear decrease can be seen. While the maximum P95 (P50) value for the sum of the 4 PFASs was 28.87 (13,82)  $\mu\text{g/L}$  in 2007, in 2019 it is only 8.28 (4,59)  $\mu\text{g/L}$ . Data from two mother- child studies in Vienna/Austria also showed a decline in the P50 values for the sum of the 4 PFASs from 4.3  $\mu\text{g/l}$  in 2010 to 2.2  $\mu\text{g/L}$  in 2019.

## **6.11 What are the PFASs levels and health effects in vulnerable population groups?**

AD5.5. HBM-based indicators were developed and compared with HBM-GVGenPop for several substances including PFASs, considering data of the sum of 4 PFASs (PFOS, PFOA, PFNA, PFHxS).

As described in the answer to the above Policy Questions PFASs levels in European teenagers exceed health-based guidance values, this has been visualised by the various indicators.

Based on the review in D11.4 the following associations between health outcomes and PFASs exposure were identified: cardiovascular diseases (CVD overall, hypertension, ischemic heart disease, stroke), Metabolic syndrome (metabolic syndrome overall, obesity, glucose metabolism, diabetes, lipid metabolism), reproductive health (AGD, semen quality, reproductive hormones, fertility & fecundity), fetal development, kidney function, liver function, osteoporosis, hematological system (hemoglobin levels), asthma, COPD, skin irritation and inflammation, ADHD, kidney/liver/pancreatic cancer.

In a collaboration between WP11 and WP14, an overview of the effects of endocrine disruptors, including PFASs on metabolic syndrome, was published. Some evidence of the association of PFASs, including substitutes of legacy PFASs with obesity, diabetes, and non-alcoholic fatty liver disease has been observed.

The association between pregnancy exposure to PFASs and later offspring immune function has been investigated in epidemiological data from cohort studies performed within the HBM4EU consortium. The results provided clear indications that higher maternal exposures to PFASs during pregnancy is associated with increased propensity for infections in the children up to age 4 and the frequency of antibiotics use until adolescent age. Inverse associations of maternal PFASs exposure with offspring growth trajectories and sleeping habits have been observed in mother – child cohorts. Also, associations of PFASs with poorer cardiovascular risk profile including higher total cholesterol and lipid profile, higher fasting blood glucose, BMI and blood pressure has been

observed. In another cohort prenatal exposure to PFASs was associated with higher weight, BMIz-score and waist circumference at age 9, among boys. PFASs mixture was associated with an increase in triglyceride and insulin levels and decrease in HDL cholesterol. Further, three studies provide some evidence suggesting that exposure to PFASs as observed in background exposed may modestly interact with endogenous hormones. Prenatal PFASs exposure could be associated with reproductive disorders such as preeclampsia and pregnancy hypertension, delay of menarche and abnormal menstruation/length, reduction of birth weight, length, and change in gestational length, decreases in semen quality and sperm count. One study showed correlations with the anogenital distance in girls and a risk of cerebral palsy in boys. The preliminary findings on in utero exposures and offspring cognitive development highlight that more work is needed in sufficiently powered studies using robust outcomes assessment.

Data on health impacts of different PFASs are available for a comparatively small number of PFASs, of which especially PFOS and PFOA are well researched. There is a need for human-relevant hazard and HBM data, and there are also gaps for the majority of the PFASs currently used related to uses, exposure patterns and toxicity.

## **6.12 How can mixture effects of environmental and human PFASs mixtures present to date be estimated?**

Various approaches were available and further developed to estimate the risk resulting from exposure to mixtures of PFASs. In that respect, our aim was to refine the mixture risk assessment of PFASs taking three approaches for comparison, the Relative Potency Factor (RPF) approach, the Hazard Index (HI) approach, and the sum value approach of the European Food Safety Agency (EFSA). The latter approach uses the EFSA tolerable weekly intake (TWI) value for the sum of PFOA, PFNA, PFHxS, and PFOS. The HI and RPF approach were adapted specifically with the purpose of being able to use human biomonitoring data as primary input to the risk assessment. All approaches indicate that PFASs exposure may cause adverse health effects in certain segments of the European population, thereby confirming the conclusion drawn in the recent EFSA scientific opinion (2020) on PFASs in food, supporting the need for risk management measures and thereby the need for an EU-wide restriction on PFASs.

The mixture risk assessments illustrate that HBM provides a highly valuable contribution to the assessment of the cumulative risk resulting from exposure to PFASs. It is inherent to HBM to reflect the aggregated exposure from different exposure routes and exposure sources and therefore reflects exposure from all potential sources.

It should be mentioned that this current work is a scientific exploration of how to use HBM in risk assessment. This exercise presents several approaches to perform mixture risk assessment for PFASs that are based on pragmatism, and each approach has its strengths, assumptions, uncertainties and flaws. Further refining and extending quantitative mixture risk assessment for 'forever chemicals' is recommended and possible. To support the science-based grouping of PFASs, a better understanding of the modes of action of different PFASs is needed. Further studying relative potencies of PFASs for other effects, sex, life-stages, and species, and in particular for critical effects such as immune effects would be of added value. We also recommend considering mixture exposure at the individual level rather than using summary statistics for the summation of exposure to multiple compounds. Furthermore, it is preferable to include exposure and effect data in the risk assessment at the individual level.

Positive associations with PFASs exposure and ASD/ADHD and behaviour in children were identified in several studies, however, data is inconsistent. Data on negative effects on developmental milestones are also inconsistent. In several case-studies associations with e.g. breast cancer, prostate cancer and colorectal cancer were found, but more prospective studies are needed. Related to endocrine disorders, lower testosterone and oestradiol levels as well as thyroid

hormone disruptive potential were identified. There are also indications of immunosuppressive effects in several studies, as well as positive associations with asthma and allergy. Further, prenatal PFASs exposure is associated with higher fat percentage in children leading to an increased risk of overweight/obesity, and there are associations to glucose homeostasis, dyslipidemia, high cholesterol, metabolic syndrome and risk of diabetes. Related to reproductive disorders, positive associations with preeclampsia and pregnancy hypertension were identified. Further, prenatal PFASs exposure may result in a delay of menarche and may cause abnormal menstruation/length. Prenatal exposure can also reduce birth weight, length, APGAR score and change gestational length. Decreases in semen quality and sperm count were also found. Additionally, in one study correlations with anogenital distance in girls and a risk of cerebral palsy in boys have been shown.

Based on the literature research and inventory of available effect biomarkers in WP 14 associations of PFASs exposure with certain health outcomes were identified. Positive associations with ASD/ADHD and behaviour in children were identified in several studies, however, data is inconsistent. Data on negative effects on developmental milestones are also inconsistent. In several case-studies associations with e.g. breast cancer, prostate cancer and colorectal cancer were found, but more prospective studies are needed. Related to endocrine disorders, lower testosterone and oestradiol levels as well as thyroid hormone disruptive potential were identified. There are also indications of immunosuppressive effects in several studies, as well as associations with asthma and allergy. Further, prenatal PFASs exposure is associated with higher fat percentage in children leading to an increased risk of overweight/obesity, and there are associations to glucose homeostasis, dyslipidemia, high cholesterol, metabolic syndrome and risk of diabetes. Related to reproductive disorders, positive associations with preeclampsia and pregnancy hypertension were identified. Further, prenatal PFASs exposure may result in a delay of menarche and may cause abnormal menstruation/length. Prenatal exposure can also reduce birth weight, length, APGAR score and change gestational length. Decreases in semen quality and sperm count were also found. Additionally, in one study correlations with anogenital distance in girls and a risk of cerebral palsy in boys have been shown.

AD14.5: In 702 Danish females it was demonstrated that PFASs induced xenoestrogenic transactivity is significantly inverse related to birth weight, length and head circumference. Further, combined mixture of PFASs from placenta homogenate sample provided by the Spanish INMA-Granada cohort was isolated and xenoestrogenic transactivity was measured. Similar as for the serum samples, 52% of the placenta extracts significantly induced xenoestrogenic transactivity, and 68% further enhanced the transactivity of the natural E2 receptor ligand. In addition, a literature review on PFASs exposure and thyroid homeostasis in epidemiological studies was conducted.

Maternal thyroid hormones are essential for fetal brain development and PFAA are suggested to interfere with these hormones in 2nd or 3rd trimesters. In addition, PFASs exposure may cause hypothyroid homeostasis.

In the Scoping Review of Moore et al, 2022 the association between attention deficit hyperactivity disorder (ADHD), a chronic neurodevelopment disorder and exposure to PFASs has been investigated. The evidence for an association is small but warrants further research.

The association between HBM4EU priority substances and asthma has been investigated in a scoping review from Mattila et al., 2021. The evidence for PFASs was inconclusive.

Ottenbros et al. described the use of network technics for graphical presentation of human biomonitoring data of mixtures (multiple exposure biomarkers) which facilitates the detection of exposure patterns and allows for the systematic comparison of observed exposure patterns between datasets and strata within datasets.

### **6.13 How can PFASs substances of relevance for human exposure and health be identified having in mind that more than 3000 substances are on the market?**

PFASs are included in the 3rd list of prioritised substances as outcome of the 3rd prioritization progress with the aim to produce a shortlist of priority substances intended to feed in to PARC. It is recommended to study (mixtures of) legacy and substitution PFASs as well as groups of PFASs and total organofluorine content in humans in combination with information on exposure sources and routes and with (early) biomarkers of effect and with follow up of health outcomes in longitudinal studies. We stress the need for improving the analytical methods and standards for biomonitoring and for bringing down the LOD and LOQ. We also recommend performing studies that observe paired blood-urine samples to have a better insight into exposure to short-chain PFASs.

### **6.14 How can identification and assessment (including data on (potential) adverse effects on human health and the environment) of alternatives currently hampered by CBI (Confidential Business Information) be facilitated?**

This policy question goes beyond the scope of HBM4EU.

### **6.15 How much are HBM values dependent on host characteristics and does this have implications for identifying vulnerable groups?**

The individual data collections prepared and made available within HBM4EU contained aggregated data stratified by age, sex and educational level. From these stratifications it can be seen that the PFASs concentrations are in general higher in men compared to women for the teenager and adult studies.

Also, there seems a trend that participants with higher educational level have higher exposure levels compared with low to medium educational level. This trend was however less obvious and could not be observed in all studies and for all compounds. In some studies, higher levels of PFASs were observed with increasing age, indicating possible cumulative exposure over time. This was however not observed for all studies and for all compounds.

AD12.3: A new PBPK model for PFOA and PFOS was developed based on a previously reported model within WP 12. (D12.1).

Several discussion points have been identified, e.g. highlighting the importance to obtain partitioning data from humans and of PFASs levels in human tissues in order to refine the model.

The exposure reconstruction via PBPK modelling offers unique opportunities related to HBM data interpretation. For these reconstructions, a minimum of information related to the toxicokinetic behaviour of a substance (and its metabolites) is required, which allows a translation of measured biomarker levels at a given time point with long-term daily intake patterns.

Dynamic age and gender specific PBPK models and the paediatrics case will allow to contributions to the identification of vulnerable groups.

## **7 HBM4EU outputs to date**

### **7.1 Categorisation**

Based on the large number of available PFASs substances on the market and the knowledge gaps on identity, toxicity and uses (of the alternatives), the listing of chemicals in categories A-E is an attempt to categorize possibly relevant substances that contribute to the overall PFASs burden in

humans. Several substances are listed in category A due to their restriction as PFOS and PFOA related substances, although limited or no HBM data are available. Efforts should be made to improve the methods to detect the broader spectrum of Category A substances. However, the priority for future HBM research should cover alternatives with high production volume, wide dispersive use and identified or suspected hazardous properties which qualify for SVHC identification.

The PFASs compounds within the scope of the HBM4EU project (see Section 1 and Glossary) have been categorised in the [scoping document](#) depending on availability of toxicology and human biomarker data (see Table 7.1):

Table 7.1 HBM4EU categorisation for PFAS

Category		Priority substance(s)
A	HBM data are sufficient to provide an overall picture of exposure levels across Europe, and interpretation of biomonitoring results in terms of health risks is possible.	PFOA, PFOS, PFNA, PFDA, PFU(n)DA, PFDODA, PFTrDA, PFTeDA, PFHxS, FOSA, PFOSA, n-MeFOSA, N-Et-FOSAA, Et-PFOSA-AcOH, Et-FOSAA, N-EtFOSA, SULFLURAMID, N-EtFOSE, N-MeFOSE, 8:2 diPAP, 6:2/8:2 diPAP, 8:2 monoPAP
B	HBM data exists, but not sufficiently to have a clear picture across Europe.	PFBA, PFPeA, PFHxA, PFHpA, PFBS, PFHpS, PFDS, N-Me-PFOSA-AcOH, Me-FOSAA, 6:2 FTSA, H4PFOS, THPFOS, 8:2 FTSA, PFODA, PfHxDA
C	HBM data scarcely or doesn't exist. Efforts to develop an analytical method to obtain relevant HBM results need to be done	PFBA, PFPeA, PFHxA, PFHpA, PFBS, PFHpS, PFDS, N-Me-PFOSA-AcOH, Me-FOSAA, 6:2 FTSA, H4PFOS, THPFOS, 8:2 FTSA, PFODA, PfHxDA.
D	A toxicological concern exists but HBM data are not available.	HFPO, PFCHS, 6:2/8:2 diPAP, 8:2 monoPAP, PFOPA, Perfluorinated Siloxane, FL16.119
E	Not yet identified as of toxicological concern and for which no HBM data are available	6:2 FTCA, 8:2 FTCA, 10:2 FTCA, PFECA, FBSA, MeFBSE, 6:2 PAP, 6:2 diPAP, PFHxPA, PFDPA, C8/C10 PFPiA, Denum SH, Krytox, Fomblin Z-DIAC, C3; C15-C20 PFCA, C3, C15-C20 PFSA, TFEE-5, polymers: PTFE, PVDF, PVF, TFE, HFP

## 7.2 Key outputs

### Current exposure of the EU population

The European HBM dashboard provides summary statistics for HBM data from different European countries, with 19 aggregated datasets including PFASs exposure data. In IPCHEM metadata for 33 datasets with PFASs data are available. Some harmonised datasets (however, a limited number) are currently available. The available datasets include aggregated data on exposure biomarker levels for FOSA, N-EtFOSA, N-MeFOSA, PFBA, PFBS, PFDA, PFDODA, PFDS, PFHpA, PFHxA, PFHxS, PFNA, PFOA, PFOS, PFPeA, PFTeDA, PFTrDA and/or PFUnDA, for the matrices serum, cord serum and/or breast milk. The datasets belong to studies from Belgium, Czech Republic, Slovakia and Denmark and were collected within 2000-2017. Available stratifications include information on sex, age groups (including infants <1 yr), ISCED, current non-smokers, and current non-smokers vs smokers. From the existing European data collections studies including analyses of PFOS, PFOA, PFNA and PFHxS dated between 2008 and 2015, were selected. For PFHxS concentrations in urine samples were reported in 3 studies involving toddlers, 1 study involving children and 5 studies involving adults and aggregated data for PFOA and PFOS were reported in 4 studies involving toddlers, 1 study involving children, 1 study involving teenagers and 6 studies involving adults. All studies included at least 116 participants. Despite variations in design, populations, analytical methods, and geographic location, the median

concentrations in the different European studies are rather similar, with ratios between the highest and lowest median concentration always being less than 10. Based on existing HBM data, D10.4 presents the first annual list of European reference values (ERV).

For the investigation of PFASs exposure, samples were collected between 2014-2021 across 9 sampling sites in Europe (Norway, Sweden, Greece, Slovenia, Spain, Slovakia, France, Belgium, Germany) in the HBM4EU Aligned Studies in teenagers of 12-19 years old: 477 samples from the North, 445 from the South, 292 from the East and 743 from the West. Thus, a total of 1957 samples were available. In these samples 8 to 12 PFASs were measured including PFPeA, PFHxA, PFHpA, PFOA, PFNA, PFDA, PFUnDA, PFDODA, PFBS, PFHxS, PFHpS, and PFOS (sum of all isomers). Not all biomarkers were analyzed in all contributing studies, therefore number of sampling sites and data points can vary per biomarker.

In addition, within WP8.5 (Targeted occupational studies with EU added values and coordination of activities) occupational PFASs exposure is investigated together with chromate exposure. Therefore, plasma samples of workers are analysed from five studies including a total of 155 samples. Results will be published later in 2022.

In order to answer questions on exposure levels and sources and further support current and future HBM studies, WP7 has produced a variety of materials to provide the groundwork for a harmonised approach to study planning and conduct in Europe. These materials are:

- data platform with information on existing, ongoing and planned general and occupational HBM studies in the HBM4EU consortium
- manuals/guidelines for study planning and conduct
- Standard Operating Procedures for qualified recruitment of participants, fieldwork, sampling and exchange of samples · different kinds of questionnaires for various age groups and substance (groups)
- influence of thawing and freezing procedures on the integrity of biobanked samples in the literature and supplied a respective concept for a quality study
- templates for the communication with participants
- A R-script was developed to calculate the aggregated data in a standardized and comparable way

In WP 8.4, Targeted fieldwork in combining HES and HBM surveys influencing and interfering factors for sampling and storage were identified, whereas for each substance group recommendations were made to avoid sample contamination or inappropriate storage conditions that may influence sample quality and hence the outcome of the analysis. For PFASs, specific recommendations are given, e.g. avoiding Teflon and other fluoropolymers as well as glass in the sampling material, and information on shipment and biobanking. In an additional deliverable (AD7.2), a literature search and a concept for a sample quality study on impact of thawing and freezing on integrity of human samples was conducted.

For the Quality Assurance/Quality Control Scheme in the HBM4EU project (ICI/EQUAS) (WP9.4), the rounds 1-3 of proficiency testing for the determination of PFASs in serum including PFPeA, PFHxA, PFHpA, PFOA, PFNA, PFDA, PFUnDA, PFDODA, PFBS, PFHxS, PFHpS, PFOS (sum of all isomers) were concluded. The number of qualified laboratories for PFASs analysis after the 3rd round comprises 21.

Impact indicators have been established and used to answer the policy questions.

Considerations for the future: There is a need for extending analytical methods to include more substances (PFAS) in the existing methods, such as short-chain PFASs (PFBA, PFOeA, trifluoroacetic acid (TFA)) as well as intermediates from of fluorotelomer precursor PFASs (FTCAs, FTUnCAs) which may have a different toxicity compared to PFAAs. In addition, the implementation

of analytical methods for the analysis of “total” PFASs is needed (this would be also future policy relevant for PFASs restriction and the EU PFASs strategy).

### **Differences in exposure to regulated and non-regulated PFAS**

As described in the answer to the policy question 1 various activities and efforts have been made to assess the exposure of the European population to various PFASs. HBM4EU existing and new HBM data collections and work on indicators (generating indicators incorporating HBM-GV) have been performed in order to assess the exposure of the European population to PFASs.

### **Restriction of PFOS according to the POP Regulation**

Concerning time trends observed in Human Biomonitoring studies the EFSA opinion on PFOS and PFOA in food states that generally, after the year 2000, the concentrations in serum/plasma of PFOS, PFOA and in some studies PFHxS have decreased, while the concentrations of PFNA, PFDA and PFUnDA have increased. No clear trends have been reported for other PFASs (EFSA, 2020).

Within the HBM4EU Aligned Studies, PFASs have been studied in teenagers. Thus, no new data on exposure of children will become available. Time trends have also been explored within WP 10 using existing data, though limitations due to different study populations and geographical areas need to be considered.

Within WP5 HBM indicators have been developed. Indicators are illustrators of data that bring information in an easy and accessible way to a broader audience be it scientists, policy makers or the general population. AD5.5 mainly used PFASs data from DEMOCOPHES and the first HBM4EU data collections to illustrate PFASs exposure and effects. PFASs data from adult women in Denmark and Belgium were available and have been used.

### **Exposure sources**

When identifying exposure levels and sources as well as groups at risk, WP7 questionnaires are of great value. They can be used to set up new studies and allow the harmonized collection of data on a participant’s individual characteristics and their potential exposure pathways from different sources (sociodemographic characteristics, residential environment/home exposures, dietary habits, lifestyle, occupational exposure and health status) with PFASs. For PFASs, questionnaires for adults, adolescents and children are available.

Within the HBM4EU Aligned Studies, PFASs exposure has been studied in teenagers. In addition, a targeted occupational study on hexavalent chromium (Cr(VI)) is conducted. In addition to chromium analysis, plasma samples of workers from five studies including a total of 155 samples are analysed for PFASs, as these substances are used in chromium plating. The study also includes the analyses of several effect biomarkers which are made mainly with the participants own funding. Results will be available in June 2022.

Physiology-based toxicokinetic (PBTK) modelling has been used to link external exposure to internal dose in humans (e.g. concentration in urine) by describing the process of absorption, distribution, metabolism and excretion (ADME) that a substance undergoes in living organisms. Within HBM4EU a review of published models for PFASs was performed. Based on the PBPK models available, an improvement of the model was conducted for PFOS and PFOA and validated with human experimental data. Currently, the model is available for oral and inhalation exposure and will be extended for dermal exposure for the named two PFASs. An age-dependent PBTK

model involving physiological and biological changes encompassing the full course of a human lifetime (children, adult and elderly) has been developed. This PBTK model considers age-dependent changes of system parameters like tissue volume, tissue blood flow, renal elimination and plasma protein binding for estimating age-related risk from chemical exposure. These models are being adopted for running different case scenarios including sensitive age groups, which can be applied as a tool for policymaking. Comprehensive work on external and internal exposure modelling has been performed, which is published in deliverables related to “from HBM to Exposure” within work package 12 (see respective Deliverables: D12.1-12.5, AD 12.9, AD 12.12 and AD12.13). Ongoing work of exposure assessment of the European population to PFASs will allow to develop an extended pan-European exposure modelling.

Additionally, as stated in the answer to policy question 1, different work has been undertaken for the identification of the exposure of the EU population to PFASs. As described above exposure of European teenagers will be investigated, this will be accompanied by the assessment of determinants of exposure.

### **Environmental contamination**

A substance-group-specific statistical analysis plan has been developed for PFASs. Variables for assessing environmental contamination have been identified: place of birth, place of residence (near a fluorochemical industrial facility, near civilian airports, military bases, wastewater treatment facilities, or firefighting training facilities, near agricultural areas characterised by the use of soil conditioners), years of residence, consumption of tap water, use/consumption of groundwater or surface water, locally produced food, own grown vegetables, own raised livestock, fish and seafood from a local body of water.

Primary focus of HBM4EU is exploring the background exposure of the general population and no specific studies in known hotspot areas were performed. HBM4EU study materials can however be used in national studies performed to investigate certain contamination cases, and several partners in HBM4EU are involved in studies investigating exposure in contaminated regions; the results of these studies will be used to answer the respective HBM4EU policy questions.

A working group has been formed in HBM4EU which is currently developing a guidance document on how to deal with Human Biomonitoring, health risk assessment and risk communication in (PFASs) hot spots. .(

Activity report WP5: Setting up a network of experts for developing a guidance document on how to deal with human biomonitoring in PFAS hotspots.

### **EFSA assessment of the TDI for PFOS and PFOA**

Suitable PFASs studies for the examination of exposure-health relationships were identified within WP13.3, including studies in pregnant women, children and adolescents, adults and the elderly. Additionally, individual data from several birth cohorts were examined for associations of PFASs exposure and low birth weight, and birth weight as continuous outcome. Also, associations between PFASs and thyroid function in mothers and their newborn have been investigated.

WP10/WP14 are currently studying associations between PFASs exposure, epigenetic and clinical biomarkers of effect, and asthma, allergy, metabolic disorders and sexual maturation in teenagers.

Results of the HBM4EU Aligned Studies feed into the policy processes identified, including the broad restriction of PFASs under REACH but also the chemical strategy for sustainability (toxic free environment).

HBM studies are very useful to inform policy makers and citizens and can be a vehicle for (pro)active communication on environmental health risks. An additional deliverable has been prepared within WP5 (Translation of results into policy: 'Timelines of Opportunity',) in which a strategy has been proposed to systematically map both the 'policy timeline(s)' and 'HBM4EU timeline(s)', in order to identify potential windows of opportunity for policy uptake. The strategy takes into account different types of HBM4EU output as well as different types of policy processes that might benefit from HBM data. Furthermore, HBM4EU has contributed to the public consultation of the recent EFSA opinion on PFASs, 2020 by submitting feedback and comments. Experts of the CONTAM panel presented and discussed the essential parts of the opinion with the HBM4EU PFASs community via webex on April 7.

Two science to policy workshops have been organised with members of the EU-Commission, ECHA and EFSA. HBM studies have contributed to putting PFASs high on the political agenda and HBM4EU results can further support ongoing EU processes.

### **Impact of the pending 2016 ECHA decision to restrict PFOA under REACH?**

Human biomonitoring data are available in many countries but are heterogeneous with respect to age groups and substances measured. The reported median-values and 95th percentiles of the individual existing studies on PFASs were averaged (by taking the median) over the different studies of newborns, children & teenagers combined, and adults. These levels support the concentration levels reported in the EFSA opinion. Data on PFASs exposure were summarized in the Deliverable D10.4 and D10.6. Metadata and aggregated data from HBM studies having measured PFASs were integrated in IPCHEM. The European HBM dashboard visualises the aggregated HBM data; accessible via the HBM4EU website.

### **Different population groups and time periods**

Efforts to assess PFASs exposure within HBM4EU are described above. Different research protocols have been developed to further analyse the PFASs data, including European exposure levels, exposure distributions, geographical comparisons, exposure determinants, exposure-effect associations. More results are expected to be published in the course of 2022.

To study differences in PFASs profiles in different time periods, analysis of time trend studies is needed, which are currently not available at European level. The HBM4EU Aligned Studies data will form baseline European exposure levels for PFASs in teenagers, allowing follow up studies to monitor increased or decreased usage.

Expanding the analytical range of PFASs compounds (including total extractable fluorine content to determine the amount of currently unidentified PFAS) was not possible under HBM4EU. This should be taken up in follow up initiatives such as the Partnership for the Assessment of Risks from Chemicals (PARC).

### **Health effects in vulnerable population groups**

The recent draft scientific opinion (EFSA 2020) points out that toddlers and other children had approximately two-fold higher mean intake than older age groups (adolescents, adults, elderly, very elderly). The CONTAM Panel concluded further, that parts of the European population exceed the tolerable weekly intake (TWI), which is of concern. Therefore, it can be assumed that toddlers and children are a vulnerable population group and are exposed to PFASs levels, which are cause of concern.

As described above, PFASs exposure has been examined in European teenagers in the HBM4EU Aligned Studies. Within WP 14.2, biomarkers of effect according to their utility in human studies were selected. Nuclear receptor activation, kisspeptin, thyroid and sex hormones and gonadotropins, glucose markers, serum lipids, adipokines, and oxidative stress are currently being measured in part of the HBM4EU Aligned Studies and associations with PFASs exposure and asthma, allergy, BMI, waist circumference, body fat mass, glucose intolerance, diabetes, hypertension, pubertal development, and sexual maturation (as far as data are available) are studied.

Within WP 13.1 reviews were conducted to investigate main adverse effects of PFASs. The association between exposure to PFASs and cholesterol/lipid metabolism and inflammatory responses with links to cardiovascular disease has been investigated and published in Critical Reviews in Toxicology (Fragki et al., 2020). In addition, associations between PFASs exposures and hypertension, overweight/obesity and insulin resistance have been investigated using the AOP help finder tool (Kaiser et al. in preparation). Epidemiological studies published between 2007 and 2021 were reviewed for adverse birth outcomes associated with prenatal exposure to PFASs (Gundacker et al. in preparation). In addition, the key characteristics of PFAS-induced immunomodulation and immunotoxicity were summarised and discussed (Ehrlich et al. in preparation).

In D 13.4 report on AOPs of priority substances first results addressing these endpoints were compiled. In D13.5. gaps for the establishment of AOPs were identified, and the need for required studies described.

Suitable studies for the examination of exposure-health relationships of PFASs were identified within WP13.2. Health effects in humans based on birth and adult cohorts (D13.3) including studies in pregnant women, children and adolescents, adults and the elderly. Additionally, individual data from several birth cohorts were examined regarding associations of PFASs exposure and low birth weight, and birth weight as continuous outcome. A similar investigation has been conducted for the assessment of associations between PFASs and thyroid function in mothers and their newborn.

The additional deliverable AD13.3, provides a concise summary on current progress, achievements as well as future plans in Task 13.2 cohorts within HBM4EU time frame, and also beyond. Within the Deliverable D11.4 (combination of health examination surveys with HBM: review on potential health effects) opportunities and obstacles of combining HBM and health studies have been summarised. Further, an extended review of the potential health effects of the HBM4EU priority substances has been performed.

## **Mixtures**

As described above (Policy Question 10) suitable PFASs studies for the examination of exposure-health relationships were identified within WP13.2 (D13.3.) including studies in pregnant women, children and adolescents, adults and the elderly. Additionally, individual data from several birth cohorts have been merged for the examination of associations of PFASs exposure and low birth weight, and birth weight as continuous outcome. A similar investigation has been conducted for the assessment of associations between PFASs and thyroid function in mothers and their newborn. For more detailed information see also Policy Questions No. 1 and No. 10.

Within WP 14 effect biomarkers for PFASs were successfully established.

A methodology was developed to extract and fractionate the real mixture of PFASs from human serum samples. Placental extracts (alpha fractions), containing mixtures of persistent and lipophilic chemicals, showed significant anti-androgenic activity. The hormonal profile from placental tissue was quantified, as well as some epigenetic markers such as Histone H2AX phosphorylation

(Gamma-H2AX), trimethylation of histone 3 at lysine (H3K4me) and DNA methylation of BDNF, in addition to untargeted metabolomic analysis. Finally, 8OHdG levels were assessed in urine samples coupled to the placentas from the same women. This work has shown that chemical mixtures isolated from human samples can be assessed, and its biological activity quantified using different biomarkers cell-based tools. Placenta tissue could be used as a relevant biological matrix to assess both exposure and effect biomarkers. The placenta can also be used to explore the implementation of novel effect biomarkers in human biomonitoring programs, due to the volume and availability of this biological sample.

In addition, there were activities related to the assessment of the biological effects of PFASs mixtures using in vitro biomarkers of combined activity, related to the generation of new knowledge on AOPs by in vitro research for the mechanism for PFASs on liver-cholesterol and lipid metabolism and intracellular levels of PFASs, and related to the identification of exposure-health associations from a cohort study (human milk biobanking).

AD 14.6: The pilot study with INMA-Granada samples has demonstrated the technical feasibility to implement novel biomarkers of brain function such as BDNF, at different levels of biological organisation, in HBM4EU aligned studies. Moreover, the use of cell-based bioassays may help to better understand how specific chemical mixtures contained in human samples may exert combined hormonal activities. Finally, the development of chemical's mixture isolation, such as PFASs, opens the possibility of assessing specific chemical families isolated from human samples, with potential implications for risk assessment and policy making.

Within WP 5.3, three different mixture risk assessment strategies were compared that all used HBM data as primary input (D5.8). This preliminary mixture assessment has been updated in D5.11 with the data from the aligned studies in teenagers. The first approach is the EFSA sum value approach, based on the recently derived EFSA TWI. The internal guidance value of 6.9 ng/mL will be compared to the sum of exposure to the 'EFSA-4' (PFOA, PFNA, PFOS, PFHxS). Additionally, two other approaches are introduced, that both are able to incorporate more than 4 PFASs in the risk assessment as cumulative exposure. These are the hazard index approach and the relative potency factor approach. All three approaches illustrate a cumulative risk in certain parts of the participating study cohorts and thereby confirm EFSA's earlier conclusions (EFSA 2020). Moreover, these exemplary mixture risk assessments show various possibilities by which HBM data may inform policy.

## Data gaps

For the majority of the several thousand currently used PFASs considerable data gaps exist related to current uses, exposure patterns and toxicity. Besides regulatory action which is called for by member states and the European Commission also research is needed. HBM4EU PFASs experts have submitted a statement to the public consultation on the draft scientific opinion on the risks to human health related to the presence of perfluoroalkyl substances in food, which states among others:

- There is a need for Human Biomonitoring data for PFASs other than those addressed in the risk assessment (specifically those which are used/formed in high volumes as a result of substituting legacy PFAS).
- There is a need to measure the total organofluorine content in humans in order to assess the magnitude of the so far unknown or not yet assessable contribution of PFASs in humans. Further, non-target analytical methods could be used to identify new relevant substances.
- More longitudinal epidemiological PFASs studies are needed. Research on immunotoxicity, endocrine disruption and birth outcomes is required. Research on other

toxicological endpoints is also needed including effects on the lungs/respiratory system from prenatal exposure, and cancers such as breast cancer in adults.

- Research on adverse outcome pathways is needed.
- To support the science-based grouping of PFASs, a better understanding of the modes of action of different PFASs is needed.
- Further studying relative potencies of PFASs for mixture risk assessment would be of added value.

## Exposure models

A new PBPK model for PFOA and PFOS was developed based on a previously reported model within WP 12. (D12.1). For validation purposes, data on PFOA and PFOS in human tissues from people living in the area of study (Tarragona County) were used.

In AD12.3 exposure estimates were derived based on a consumption study and they were used as input for the PBPK model to estimate the concentration of 11 PFASs in human tissues. PKs were estimated by using data on PFASs concentrations in plasma and autopsy tissue samples.

For model parameterisation, data on PFASs concentrations in blood and human tissues from three studies were used.

In general, the assessment of the models showed that there is a lack in data supporting integrated exposure, and another significant problem is the data quality. Further, there is a need for the detailed description of exposure mechanisms that are not straightforward such as inhalation or food ingestion.

AD12.5: The PBPK model was used for exposure reconstruction. Data used was from the Tarragona cohort study, and only PFOS and PFOA were considered. Exposure estimates: Daily intakes were estimated with a constant oral exposure scenario.

Ongoing work in WP12 concerns a dynamic age and gender specific PBPK model, and paediatrics case study.

Excretion values are depending on several factors e.g.:

- 1) genetic factors (for example SNPs in transporter protein genes),
- 2) sex - females of reproductive age excrete more due to menstruation and reproduction
- 3) kidney health, gut health, gut microbiome - changes in excretion or reabsorption by either route leads to changes in serum levels, and so induces an association (+ve or -ve) between HBM and various health conditions correlated with renal or gut health. Reduced eGFR/kidney function is associated with reduced excretion higher serum PFAS

Knowledge of which factors lead to increased vulnerability is still incomplete. There is still a need for research in this area.

## 8 Key data gaps

Based on the [policy questions](#) ongoing work to address the knowledge gaps is summarised in the table below. If you would like to read more about the work packages (WP), please visit the HBM4EU [website](#).

**Table 8.1 Summary of knowledge gaps**

#	Theme	Knowledge gaps and activities needed	Relevant HBM4EU WPs <sup>7</sup>
1	<b>Exposure and health effects</b>	Proceed with collecting, combining, harmonising, and comparing existing exposure data on PFASs.	WP 10
		A detailed data gap analysis should be performed, taking the respective human health related endpoints into consideration to address the question if health-based guidelines are met or not, as well as a consideration of mixture effects.	WP 5, 8, 9,10,15
2	<b>Vulnerable groups</b>	To identify differences in the exposure levels of unregulated and regulated PFASs between countries and time periods, and to identify the main reasons for differences in exposure.	WP 10, 12
		Current exposure levels in vulnerable populations (e.g., children and those affected by health effects linked to the potential PFASs exposure) to be investigated, preferable with methods, which allow identifying Cat A, B and C substances as well as the total PFASs burden.	WP 8, 9, 13
		Study how PFASs affect AOPs that lead to critical endpoints in humans such as effects on liver and thyroid, developmental toxicity, immunotoxicity and non-carcinogenic toxicogenicity.	WP 13
3	<b>Impact of regulatory controls</b>	New targeted studies identifying a multitude of PFASs in human blood and urine including newly developed methods, and validation and harmonisation of those methods in order to integrate them in planned and ongoing.	WP 8, 9,14
		Exposure of children to PFASs should be investigated, complemented by measurement of transaminases, cholesterol, immune parameters, and thyroid hormones.	WP 8,9,10,14
		The identification, assessment, and monitoring of alternatives.	WP 4,5,10
4	<b>Informing policy</b>	The detailed EFSA assessment shall be used within HBM4EU for defining data gaps and refining research questions. Combining several comparable studies will allow for more robust assessment of health outcomes in terms or broader exposure range and examination of rare health outcomes.	WP 13
5	<b>Circular economy</b>	Research on the life cycle of products may identify potential exposure routes (e.g., studies near landfills could clarify if PFASs exposure occurs).	WP 7,8,9

<sup>7</sup> HBM4EU WP description: WP1: Programme management and coordination; WP2: Knowledge hub; WP4: Prioritisation and input to the annual work plan; WP5: Translation of results into policy; WP6: Sustainability and capacity building; WP7: Survey design and fieldwork preparation; WP8: Targeted field work surveys and alignment at EU level; WP9: Laboratory analysis and quality assurance; WP10: Data management and analysis; WP11: Linking HBM, health surveys and registers; WP12: From HBM to exposure; WP13: Establishing exposure health relationships; WP14: Effect biomarkers; WP15: Mixtures, HBM and human health risks; WP16: Emerging chemicals

#	Theme	Knowledge gaps and activities needed	Relevant HBM4EU WPs <sup>7</sup>
6	<b>Practical challenges</b>	Identification of compounds to be prioritized for further information on exposure and/or toxicity to be measured in HBM studies	WP 4,5
		Identify lead chemicals in mixtures of PFAS	WP 14, 15
		Develop mixture risk assessment methodologies for PFAS	WP5
		Design new studies that measure these exposure biomarkers	WP 8
		Fuller characterization of role of gender, existing disease use of medicines and other causes affecting measured HBM in serum	WP 8, 10

## 9 Future recommendations

The results depicting widespread exposure and exceeding of health based guidance values in European teenagers as well as the various associations with adverse outcomes detected in cohort studies call for further regulatory action and will be fed into the regulatory/policy framework.

On the other hand recommendations can be also made to be investigated within the European Partnership for Risk Assessment PARC.

The following thematic areas, research questions and data gaps have been identified:

Linking human body burdens to dietary exposure and locally produced food: develop more targeted questionnaires to identify and quantify exposure sources in more detail.

Human biomonitoring and health effects in PFASs hotspots.

Human biomonitoring in order to evaluate the effectiveness of the PFASs restriction at EU level, follow up on indicators.

Development and harmonization of new methods, EOF, TOP assay, multi methods, bioassays in combination with target analytics for human, food and environmental monitoring.

Assess types of PFASs emitted, quantities and sources.

Detection of PFASs in biota and in alternative human samples and tissues: urine, faeces, fatty tissues, organs (liver, brain, lung, spleen, lymph nodes).

Further clarification of the relationship between external and internal exposure levels.

Investigate possibilities to enhance PFASs elimination from the body; determine and investigate half life's in humans.

Develop recommendations for local food consumption and soil sanitation in contaminated areas.

Research on health effects such as immunotoxicity, endocrine disruption, birth outcomes, cancer, thyroid disease, alteration of thyroid hormone levels.

Toxicity, Mode of Action, Relative Potencies – for mixture risk assessment, mechanistic pathways, AOP development.

Combining NAM - in vitro and in silico methods - with human epidemiology and human biomonitoring.

## 10 References

HBM4EU, 2019, Scoping documents for PFASs, part of the D4.9 scoping document set.

HBM4EU, 2019, prioritised substance group: per-/polyfluoroalkyl substances (PFASs) policy-related questions.

HBM4EU, 2020, Legislative mapping per/poly fluorinated compounds, summary document prepared by RPA on behalf of the European Environment Agency.

Agency for Toxic Substances and Disease Registry (2018). Toxicological Profile for Perfluoroalkyls. Available at : <https://www.atsdr.cdc.gov/ToxProfiles/tp.asp?id=1117&tid=237>.

Banzhaf, S., Filipovic, M., Lewis, J., Sparrenbom, C.J. and Barthel, R. (2017). A review of contamination of surface-, ground-, and drinking water in Sweden by perfluoroalkyl and polyfluoroalkyl substances (PFAS). *Ambio*, 46 (3), p.p. 335-346.

Barry, V., Winqvist, A., Steenland, K. (2013). Perfluorooctanoic acid (PFOA) exposures and incident cancers among adults living near a chemical plant. *Environ. Health Persp.* 121(11-12), p.p.1313-1318.

Danish EPA (2018). Risk assessment of fluorinated substances in cosmetic products, Survey of chemical substances in consumer products No 169, Available at: <https://mst.dk/service/publikationer/publikationsarkiv/2018/nov/risk-assessment-of-fluorinated-substances-in-cosmetic-products/>

EFSA (2018). Risk to human health related to the presence of perfluorooctane sulfonic acid and perfluorooctanoic acid in food. Available at: <https://www.efsa.europa.eu/en/efsajournal/pub/5194>

EFSA (2020). Risk to human health related to the presence of perfluoroalkyl substances in food, Available at: <https://efsa.onlinelibrary.wiley.com/doi/epdf/10.2903/j.efsa.2020.6223>

Eriksson, U., et al. (2017). Contribution of precursor compounds to the release of per- and polyfluoroalkyl substances (PFAS) from wastewater treatment plants (WWTPs), *Journal of Environmental Sciences (China)*, 61, p.p. 80-90.

Eschauzier, C., et al. (2012). Impact of Treatment Processes on the Removal of Perfluoroalkyl Acids from the Drinking Water Production Chain, *Environ. Sci. Technol.* 46 (3), p.p. 1708-1715.

European Commission (2019). Elements for an EU-strategy for PFASs. Available at: <https://www.regjeringen.no/contentassets/1439a5cc9e82467385ea9f090f3c7bd7/fluor---eu-strategy-for-PFAS---december-19.pdf>.

Gebbink, W. A., et al. (2015). Estimating human exposure to PFOS isomers and PFCA homologues: The relative importance of direct and indirect (precursor) exposure', *Environment International*, 74, p.p. 160-169.

Gebbink, W. A., et al. (2017). Presence of Emerging Per- and Polyfluoroalkyl Substances (PFAS) in River and Drinking Water near a Fluorochemical Production Plant in the Netherlands, *Environmental Science & Technology*, 51(19), p.p. 11057-11065.

Ghisi, R. et al. (2019). Accumulation of perfluorinated alkyl substances (PFAS) in agricultural plants: A review, *Environmental Research*, 169, p.p. 326-341.

Glüge, J., Scheringer, M., Cousins, I. T., DeWitt, J.C., Goldenman, G., Herzke, D., Lohmann, R., Ng, C.A., Trier, X., Wangj, Z. (2020). An overview of the uses of per- and polyfluoroalkyl substances (PFAS). *Environ. Sci.: Processes Impacts*, 22, p.p. 2345-2373.

Hamid, H. and Li, L. (2016). Role of wastewater treatment plant (WWTP) in environmental cycling of poly- and perfluoroalkyl (PFAS) compounds, *Ecocycles* 2(2), p.p. 43-53.

- Houde, M., et al. (2006). Biological Monitoring of Polyfluoroalkyl Substances: A Review, *Environmental Science & Technology*, 40(11), p.p. 3463-3473.
- Hu, X. C., et al. (2016). Detection of Poly- and Perfluoroalkyl Substances (PFAS) in U.S. Drinking Water Linked to Industrial Sites, Military Fire Training Areas, and Wastewater Treatment Plants, *Environmental Science & Technology Letters*. 3 (10), p.p. 344-350.
- Ingelido, A. M., et al. (2018). Biomonitoring of perfluorinated compounds in adults exposed to contaminated drinking water in the Veneto Region, Italy, *Environment International*, 110, p.p. 149-159.
- KEMI (2015). Occurrence and use of highly fluorinated substances and alternatives. Swedish Chemicals Agency, Stockholm, Sweden. Available at: <http://www.kemi.se/en/global/rapporter/2015/report-7-15-occurrence-and-use-of-highly-fluorinated-substances-and-alternatives.pdf>
- KEMI (2017). A Nordic workshop on joint strategies for per- and polyfluorinated substances (PFAS), background document, presentations. May 4-5th, Stockholm, Sweden. Available at : <http://norden.diva-portal.org/smash/get/diva2:1272489/FULLTEXT02.pdf>
- Nordic Council of Ministers (2019). The cost of inaction - A socioeconomic analysis of environmental and health impacts linked to exposure to PFASs, Tema Nord No 516.
- National Toxicology Programme (2016). Immunotoxicity associated with exposure to perfluorooctanoic acid or perfluorooctane sulfonate. NTP Monograph. National Institute of Environmental Health Sciences. US Department of health and human services.
- Numata, J., et al. (2014). Toxicokinetics of Seven Perfluoroalkyl Sulfonic and Carboxylic Acids in Pigs Fed a Contaminated Diet, *Journal of Agricultural and Food Chemistry* 62(28), p.p. 6861-6870.
- OECD (2013). OECD/UNEP Global PFC Group, Synthesis paper on per- and polyfluorinated chemicals (PFCs), Environment, Health and Safety, Environment Directorate, OECD.
- OECD (2015). Risk Reduction Approaches for PFASs – A Cross Country Analysis, OECD Environment, Health and Safety Publications Series on Risk Management No. 29, Available at: [https://www.oecd.org/chemicalsafety/risk-management/Risk\\_Reduction\\_Approaches%20for%20PFASs.pdf](https://www.oecd.org/chemicalsafety/risk-management/Risk_Reduction_Approaches%20for%20PFASs.pdf)
- OECD (2018). Toward a new comprehensive global database of per- and Polyfluoroalkyl substances (PFASs): Summary report on updating the OECD 2007 list of per- and polyfluoroalkyl substances (PFASs) Series on risk management, No. 39.
- Rappazzo, K. M., Coffman, E., & Hines, E. P. (2017). Exposure to Perfluorinated Alkyl Substances and Health Outcomes in Children: A Systematic Review of the Epidemiologic Literature. *International journal of environmental research and public health*, 14(7), p.p. 691.
- Schultes, L., et al. (2018). Per- and polyfluoroalkyl substances and fluorine mass balance in cosmetic products from the Swedish market: implications for environmental emissions and human exposure, *Environmental Science: Processes & Impacts*, 20(12), p.p. 1680-1690.
- Sun, M., et al. (2016). Legacy and Emerging Perfluoroalkyl Substances Are Important Drinking Water Contaminants in the Cape Fear River Watershed of North Carolina, *Environmental Science & Technology Letters*, 3(12), p.p. 415-419.
- Toxics Use Reduction Institute (2020). Per- and Poly-fluoroalkyl Substances (PFAS): Policy Analysis, Draft, October 2020. [https://www.turi.org/content/download/13403/205105/file/Draft%20PFAS%20Policy%20Analysis%20October%2026%20\(002\).pdf](https://www.turi.org/content/download/13403/205105/file/Draft%20PFAS%20Policy%20Analysis%20October%2026%20(002).pdf)
- Valsecchi, S., et al. (2013). Determination of perfluorinated compounds in aquatic organisms: a review, *Analytical and Bioanalytical Chemistry*, 405(1), p.p. 143-157.

Wang, Z., et al. (2014a). 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, p.p. 62-75.

Wang, Z., et al. (2014b). Global emission inventories for C4-C14 perfluoroalkyl carboxylic acid (PFCA) homologues from 1951 to 2030, Part II: The remaining pieces of the puzzle, *Environment International*, 69, p.p. 166-176.

WHO (2017). Keeping our water clean: the case of water contamination in the Veneto Region, Italy.

# Appendix 1: Additional information on sources of information on exposure.

Source of exposure	Reference
<b>Environmental</b>	
<ul style="list-style-type: none"> <li>Emissions to the environment occur via industrial waste water releases, as well as emissions to air from industrial production sites followed by deposition onto soil and water bodies. Industrial and urban waste water treatment plants are also a significant source of PFASs, via air, water and sludge.</li> <li>The production and use of PFASs in products has resulted in the contamination of drinking water supplies in several European countries.</li> <li>The recycling of PFASs containing materials such as food contact materials and the formation of volatile fluorinated gases during waste incineration (are other possible sources of PFASs pollution.</li> <li>Reuse of contaminated sewage sludge as fertilisers has led to PFASs pollution of soil and water.</li> </ul>	<p>Scoping document</p> <p>EEA report:  <a href="https://www.eea.europa.eu/publications/emerging-chemical-risks-in-europe/emerging-chemical-risks-in-europe">https://www.eea.europa.eu/publications/emerging-chemical-risks-in-europe/emerging-chemical-risks-in-europe</a></p> <p>Banzhaf et al. (2017)</p> <p>Danish EPA (2019)</p> <p>EFSA (2018)</p> <p>Ghisi et al. (2019)</p> <p>WHO (2017)</p>
<b>Occupational</b>	
<ul style="list-style-type: none"> <li>Production and use of PFASs have been the main sources of PFASs contamination over time, for instance from fluoropolymer production installations and from the use of PFAS-containing firefighting foams.</li> <li>Other sources include PFASs produced and applied to textiles and paper and painting/printing facilities.</li> </ul>	<p>Scoping document</p> <p>EEA report:  <a href="https://www.eea.europa.eu/publications/emerging-chemical-risks-in-europe/emerging-chemical-risks-in-europe">https://www.eea.europa.eu/publications/emerging-chemical-risks-in-europe/emerging-chemical-risks-in-europe</a></p> <p>Wang et al., 2014a, 2014b; Hu et al., 2016 (in EEA report) Danish EPA, 2014</p>
<b>Consumer</b>	
<ul style="list-style-type: none"> <li>PFASs in consumer products, such as textiles, furniture, polishing and cleaning agents and creams, may contaminate dust and air, while food contact materials can contaminate food. Drugs and medical devices may be other sources.</li> <li>Direct exposure may also come via skin creams and cosmetics (Danish EPA, 2018; Schultes et al., 2018) or via air from sprays and dust from PFAS-coated textiles.</li> </ul>	<p>Scoping document.</p> <p>EEA report:  <a href="https://www.eea.europa.eu/publications/emerging-chemical-risks-in-europe/emerging-chemical-risks-in-europe">https://www.eea.europa.eu/publications/emerging-chemical-risks-in-europe/emerging-chemical-risks-in-europe</a></p> <p>Nordic Council of Ministers, 2019.</p> <p>Danish EPA, 2018.</p>

Route of exposure	Reference
<p><b>Oral</b></p> <p>For the general population, PFASs sources include drinking water, food, consumer products and dust.</p> <p>In food, fish species at the top of the food chain and shellfish are significant sources of PFASs exposure. Livestock raised on contaminated land can accumulate PFASs in their meat, milk and eggs.</p>	<p>Scoping document EFSA, 2020 Ingelido et al., 2018 Numata et al. 2014</p>
<p><b>Dermal</b></p> <p>Direct exposure may also come via skin creams and cosmetics.</p> <p>There is little knowledge on uptake via skin and the lungs, which can be severely affected by PFASs.</p>	<p>Scoping document Nørgaard et al., 2010 Sørli et al., 2020 Danish EPA, 2018</p>
<p><b>Inhalation</b></p> <p>Direct exposure may air from sprays and dust from PFAS-coated textiles.</p> <p>There is little knowledge on uptake via skin and the lungs, which can be severely affected by PFASs.</p>	<p>Scoping document : Nørgaard et al., 2010; Sørli et al., 2020.</p>
<p><b>Trans-placenta</b></p> <p>PFASs are transferred in the womb from mother to child.</p>	<p>EEA report: <a href="https://www.eea.europa.eu/publications/emerging-chemical-risks-in-europe/emerging-chemical-risks-in-europe">https://www.eea.europa.eu/publications/emerging-chemical-risks-in-europe/emerging-chemical-risks-in-europe</a></p>

## Appendix 2: Additional information on sources of information on health effects.

Human health effect	Category	Justification for category	References
Increased risk for cancer; Development of tumors in one or several organs (e.g., testicles, liver, kidney, bladder)	Suspected (PFOA, PFOS, PFNA, PFDA) Evidence lacking (other PFAS)	Based on harmonised listing of Carc. 2 and evidence presented in the scoping document, OECD (2013) report and EFSA (2020) report.	ECHA (see Table 4.1) Grandjean and Clapp (2015) – in scoping document EFSA (2020) OECD (2013)
Liver damage	Strong (PFOA, PFOS, PFHxS) Suspected (other PFASs e.g. PFNA, PFDeA, PFUA, PFBA, PFBS, PFDoA, and PFHpA)	Based on harmonised listing of STOT RE 1 and evidence presented in the scoping document, , EFSA (2020) report, and ATSDR (2018) report	ECHA (see Table 4.1) EFSA (2020) ATSDR (2018)
Reduced immune response to vaccination (e.g., toward rubella vaccine) ; reduced resistance to infection.	Suspected (PFOA, PFOS, PFNA, PFHxS, and PFDeA) Evidence lacking (other PFAS)	Based on evidence presented in the scoping document. Further evidence in OECD (2013) report and EFSA (2020) report.	Grandjean and Budtz-Jørgensen (2013) Rapazzo et al. (2017) NTP (2016) EFSA (2020) OECD (2013)
Endocrine disrupting effects - Decreased thyroid hormone levels; effects on thyroid metabolism and lipid metabolism	Suspected (PFOA, PFOS) Evidence lacking (other PFAS)	Based on evidence presented in the scoping document, OECD (2013) report and EFSA (2020) report. For example: for PFOS and PFOA adverse effects on thyroid metabolism and lipid metabolism have been reported in a multitude of epidemiological studies suggesting endocrine disrupting potential (Barry et al., 2013).	Barry et al. (2013) Grandjean and Clapp (2015) OECD (2013) EFSA (2020)
Reproductive effects - Increased risk of miscarriage, reduced birth weight, increased weight in adult life, and reduced fertility	Strong (PFOA, PFOS, PFNA, PFDA) Evidence lacking (other PFAS)	Based on harmonised listing of Repr. 1B and evidence presented in the scoping document, and EFSA (2020) report.	Halldorsson et al., 2012; Jensen et al., 2015; Joensen et al., 2013; Timmermann et al., 2014 – in scoping document EFSA (2020)

Human health effect	Category	Justification for category	References
Increased cholesterol levels (hypercholesteremia)	Suspected (PFOA, PFOS, PFNA) Evidence lacking (other PFAS)	Based on evidence presented in the EFSA (2018,2020) reports.	EFSA (2018, 2020)
Developmental neurotoxic effects	Evidence lacking (other PFAS)	Based on evidence presented in the EFSA (2018,2020) reports.	EFSA (2018, 2020)

For the categorisation of the strength of evidence for human health effects, the following criteria has been used:

- **Strong** – where the health effect is confirmed by either a harmonised classification indicating that there is a known effect (e.g. 1A or 1B for CMRs) (see Table 4.1), or where there is no applicable C&L classification, a statement in the Scoping Document that concludes there is strong evidence (or where a significant body of evidence is presented in the scoping document).
- **Suspected** – where there is either (a) a harmonised classification indicating that there is a suspected effect (e.g. category 2 CMRs or similar); (b) notified classification for that effect, or (c) where there is no applicable C&L classification, a statement in the Scoping Document (or other references presented in the Table above) that there is a suspected health impact.
- **Evidence lacking** – where a health effect is noted in the Scoping Document (or other evidence sources referenced in the Table above), but it is stated that evidence is currently lacking or there are uncertainties or inconsistencies in the available evidence.
- **Not applicable** – where a health effect does not apply to a specific group/gender