Request from the Norwegian Food Safety Authority

Risk assessments of five per- and polyfluoroalkyl substances (PFAS) migrating from food contact materials (FCM) of paper and board collected on the Norwegian market

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Abbreviations and acronyms

ALT	-	alanine transferase
AST	-	aspartate aminotransferase
ATSDR	-	Agency for Toxic Substances and Disease Registry (USA)
bw	-	body weight
CAR	-	constitutive androstane receptor
CAS	-	Chemistry Abstracts Service
CI	-	confidence interval
DCFH-DA	-	2,7-dichlorofluorescein-diacetate
DTU	-	Technical University of Denmark
EFSA	-	European Food Safety Authority
EU	-	European Union
FCM	-	food contact material
FPG	-	formamidopyrimidine DNA-glycosylase
FTOH	-	fluorotelomer alcohol
GD	-	gestational day
HBGV	-	health-based guidance value
HED	-	human equivalent dose
HPLC	-	high performance liquid chromatography
IARC	-	International Agency for Research on Cancer
JECFA	-	Joint FAO/WHO Expert Committee on Food Additives
LD	-	lethal dose
LOAEL	-	lowest observed adverse effect level
LOQ	-	limit of quantification
MS	-	mass spectrometry
MTT	-	3-(4,5-dimethylthiazol-2-yl)−2,5-diphenyltetrazolium bromide
NFSA	-	Norwegian Food Safety Authority
NIPH	-	Norwegian Institute of Public Health
NOAEL	-	no observed adverse effect level
NOEL	-	no observed effect level
PAPS	-	polyfluoroalkyl phosphate esters
PFAS	-	per- and polyfluoroalkyl substances
PFBA	-	perfluorobutanoic acid
PFCA	-	perfluorocarboxylic acid
PFHxA	-	perfluorohexanoic acid
PFHxS	-	perfluorohexane sulfonic acid
PFOA	-	perfluorooctanoic acid
PFOS	-	perfluorooctane sulfonic acid
PFPeA	-	perfluoropentanoic acid
POD	-	point of departure
PPAR	-	peroxisome proliferator-activated receptor
PPD	-	postpartum day
RfD	-	reference dose
ROS	-	reactive oxygen species
RPF	-	relative potency factor
SD	-	standard deviation
TCEQ	-	Texas Commission on Environmental Quality

TDI	-	tolerable daily intake
ToR	-	terms of reference
TSH	-	thyroid-stimulating hormone
TTC	-	threshold of toxicological concern
TTE	-	transplacental transfer efficiency
TWI	-	tolerable weekly intake
T4	-	thyroxine
US EPA	-	United States Environmental Protection Agency (USA)
WHO	-	World Health Organization
6:2 FTOH	-	6:2 fluorotelomer alcohol

Norsk sammendrag

Mattilsynet har utført en studie på per- og polyfluoralkylstoffer (PFAS) som migrerer fra matkontaktmaterialer. Totalt ble 30 prøver av matkontaktmaterialer laget av papir eller papp, seks prøver fra hver av fem kategorier matkontaktmaterialer, samlet inn i Norge og analysert for ulike PFAS. Kategoriene som det ble tatt prøver av var sugerør, former for baking av muffins, pizzabokser, matkontaktmateriale av papir til å lage popkorn direkte i mikrobølgeovn hjemme, og tallerkener eller boller laget av papir eller papp som skal brukes til å varme opp mat direkte i mikrobølgeovn eller konvensjonell ovn. Testing av migrasjon til en matsimulant utført på tre separate prøver for hvert innsamlet produkt og analyser av PFAS ble utført av Danmarks Tekniske Universitet (DTU).

Mattilsynet ba Folkehelseinstituttet om å utføre risikovurderinger for voksne av de syv produktene som hadde kvantifiserbare nivåer av PFAS i migrasjonstestene. Disse risikovurderingene inkluderte fem PFAS-stoffer.

For å finne etablerte helsebaserte veiledende verdier, såkalte tålegrenser, for disse PFAS-stoffene, ble det søkt på nettsidene til et stort antall internasjonale institusjoner som utfører risikovurderinger. For å finne vitenskapelige publikasjoner som beskriver eksperimentelle studier som kan gi informasjon om trygge nivåer av spesielt de mindre studerte PFAS-stoffene ble det utført et litteratursøk i databasen OVID Medline, begrenset til publikasjoner på engelsk fra 2018 til i dag.

Ved beregning av eksponering for bruk i risikovurderinger av PFAS lekket ut fra matkontaktmaterialer antar man at 1 kg mat konsumeres daglig i løpet av livet av en person med 60 kg kroppsvekt og at denne maten er pakket inn i en kubisk beholder med 6 dm^2 overflate som frigjør stoffet.

I risikovurderingene ble det søkt etter tålegrenser for de fem PFAS-stoffene i ulike rapporter og publikasjoner for sammenligning med eksponeringsnivåene av stoffene. Som tålegrenser fastsetter Den europeiske myndighet for næringsmiddeltrygghet (EFSA) tolerabelt daglig inntak (TDI) eller tolerabelt ukentlig inntak (TWI), definert som estimater av mengden av et kjemisk stoff i luft, mat eller drikkevann som kan inntas daglig eller ukentlig hele livet uten nevneverdig helserisiko. Fordi ikke alle PFAS under vurdering i denne studien har en tålegrense etablert av EFSA, ble også slike verdier fra USA og andre land brukt i risikovurderingene.

Risikoen for skadelige helseeffekter knyttet til innholdet av PFAS-stoffer er vurdert for de 7 produktene som hadde migrasjonsnivåer av en eller flere PFAS over kvantifiseringsgrensen for analysemetoden. Disse vurderingene ble basert på sammenligninger av eksponeringsnivåer mot tilgjengelige tålegrenser. I tillegg ble metodene 'terskel for toksikologisk bekymring' (TTC) og relative potensfaktorer (RPF) brukt i risikovurderingene.

For et sugerørprodukt av papir (Referansenr. 2024/87057) var det ingen bekymring for en potensiell risiko for skadelige helseeffekter fra migrasjon av perfluoroktansyre (PFOA). For et annet sugerørprodukt (Referansenr. 2024/93980) var det ingen bekymring for en potensiell risiko for skadelige helseeffekter fra migrasjon av PFOA alene, men noe bekymring fra summen av PFOA og perfluorheksansyre (PFHxS). For et tredje sugerørprodukt av papir (Referansenr.

2024/85370) gav ikke migrasjonen av perfluorbutansyre (PFBA) bekymring for skadelige helseeffekter, mens PFOA alene, og summen av PFOA og PFHxS, gav noe bekymring for potensielle skadelige helseeffekter. For et papirtallerkenprodukt (Referansenr. 2024/092361) var det ikke risiko for skadelige helseeffekter fra verken PFBA eller perfluorpentansyre (PFPeA). For et muffinsform papirprodukt (Referansenr. 2024/94002) var det ingen bekymring for en potensiell risiko for skadelige helseeffekter fra migrasjon av verken perfluorheksansyre (PFHxA) eller 6:2 fluortelomeralkohol (6:2 FTOH). For muffinsformene (Referansenr. 2024/86295) var det ingen bekymring for en potensiell risiko for skadelige helseeffekter fra migrasjon av PFHxA, men noe bekymring angående 6:2 FTOH ved evaluering gjort med RPF. Også for et pakningsmateriale for popcorn til bruk i mikrobølgeovn (Referansenr. 2024/102124) var det en viss bekymring angående 6:2 FTOH når vurderingen ble gjort med RPF.

Konklusjonene om den potensielle helserisikoen ved inntak av disse PFAS-stoffene er mer usikre for PFPeA og 6:2 FTOH enn de er for PFOA, PFBA og PFHxA. Dette skyldes at det er mindre toksisitetsdata tilgjengelig for disse stoffene og ingen risikovurderinger basert på alle potensielle toksiske effekter er utført ennå av en anerkjent internasjonal risikovurderingsorganisasjon. Internasjonalt planlegges det for tiden videre studier av forekomst og helseeffekter av flere PFAS og vurderinger av om det kan etableres tålegrenser for disse stoffene.

Summary

The Norwegian Food Safety Authority (NFSA) performed a study on per- and polyfluoroalkyl substances (PFAS) migrating from food contact materials (FCM). In total, 30 samples of FCM made of paper or board, six product samples from each of five categories of FCM, were collected in Norway and analysed for various PFAS. The FCM categories sampled were drinking straws, forms for baking muffins, pizza boxes, paper FCM used for making popcorn directly in microwave ovens at home, and plates or bowls made of paper or board to be used for heating food directly in microwave or conventional ovens. The migration testing to a food simulant performed on three separate samples for each collected FCM item and analyses of PFAS were performed by the Technical University of Denmark (DTU).

NFSA requested the Norwegian Institute of Public Health to perform risk assessments for adults of the seven FCM that had migration levels of PFAS above the limit of quantification (LOQ) in the migration tests. The risk assessments included five PFAS.

To find established health-based guidance values (HBGV) for these PFAS, the websites of a large number of international institutions performing risk assessments were searched. To obtain primary scientific publications describing experimental studies that could give information on safe levels of especially the less studied PFAS, a literature search was performed in OVID Medline, limited to publications in English language from 2018 to present.

When calculating exposure in order to perform risk assessments of chemicals migrating from FCM, the conventional assumption used is that 1 kg of food is consumed daily over a lifetime by a person of 60 kg body weight and that this food is packaged in a cubic container of 6 dm^2 surface area releasing the substance.

In these risk assessments, HBGV for the five PFAS were searched for in various reports and publications for comparisons with the exposure levels. As HBGV, European Food Safety Authority (EFSA) establishes tolerable daily intake (TDI) or tolerable weekly intake (TWI), defined as estimates of the amount of a substance in air, food or drinking water that can be consumed daily or weekly, respectively, over a lifetime without appreciable health risk. Because not all the PFAS under evaluation in this study have a HBGV established by EFSA, also such values from USA and other countries were used.

The risk of adverse health effects related to the content of PFAS was evaluated for each of the seven products that had migration levels of one or more PFAS above the LOQ. The evaluations were based on comparisons of the exposure levels with available HBGV. In addition, the threshold of toxicological concern (TTC) and relative potency factor (RPF) approaches were used.

For a paper straw product (Reference no. 2024/87057), there was no concern for a potential risk of adverse health effects from migration of perfluorooctanoic acid (PFOA). In another straw product (Reference no. 2024/93980), there was no concern for a potential risk of adverse health effects from migration of PFOA alone, but some concern from the sum of PFOA and perfluorohexane sulfonic acid (PFHxS). For a third paper straw product (Reference no. 2024/85370), the migration of perfluorobutanoic acid (PFBA) did not cause concern for

adverse health risks, whereas PFOA alone, and the sum of PFOA and PFHxS, caused some concern for potential adverse health effects. One paper plate product (Reference no. 2024/092361) was likely to be without an appreciable risk of adverse health effects from both PFBA and perfluoropentanoic acid (PFPeA). For one paper muffin form product (Reference no. 2024/94002), there was no concern for an appreciable risk of adverse health effects from migration of either perfluorohexanoic acid (PFHxA) or 6:2 fluorotelomer alcohol (6:2 FTOH). For the muffin forms (Reference no. 2024/86295), there was no concern for a potential risk of adverse health effects from migration of PFHxA, but some concern regarding 6:2 FTOH when evaluated with RPF. Also for one microwave oven popcorn package (Reference no. 2024/102124), there was some concern regarding adverse health effects of 6:2 FTOH when evaluated with RPF.

The conclusions on the potential risk from intake of these PFAS are more uncertain for PFPeA and 6:2 FTOH, for which there are less toxicity data available and no risk assessment based on all toxicity endpoints has yet been performed by a recognized risk assessment organisation internationally, than for PFOA, PFBA and PFHxA. However, there are ongoing processes internationally planning further examination of the occurrence and health effects of additional PFAS and examination of whether a HBGV can be established for these substances.

Background

Study of per- and polyfluoroalkyl substances (PFAS) in food contact materials (FCM) in Norway

The Norwegian Food Safety Authority (NFSA) performed a study on PFAS migrating from FCM. In total, 30 samples of FCM made of paper or board, six product samples from each of five categories of FCM, were collected in Norway and analysed for various PFAS. The FCM categories sampled were drinking straws, forms for baking muffins, pizza boxes, paper FCM used for making popcorn directly in microwave ovens at home, and plates or bowls made of paper or board to be used for heating food directly in microwave or conventional ovens.

Both so-called legacy and emerging PFAS were included in this study. "Legacy" PFAS refers to long-chain PFAS, such as perfluorooctanoic acid (PFOA) and perfluorooctane sulfonate (PFOS), that have been phased out of production in numerous developed nations, while the terms "emerging" and/or "alternative" refer to short-chain PFAS and polyfluorinated compounds used as replacements for legacy PFAS (Brase et al., 2021). For information on terminology, classification and origin of the PFAS, see Buck et al. (2011).

Migration testing and analyses

The migration testing and analyses of PFAS were performed by the Technical University of Denmark (DTU) (Lerch et al., 2022; Loureiro et al., 2024). The samples were subjected to migration testing to a food simulant. Each sample was 6 cm^2 of the collected items, treated in 2 ml Eppendorf tubes containing 1.5 ml 50% ethanol (for legacy PFAS and polyfluoroalkyl phosphate esters (PAPS)) or 100% methanol (for fluorotelomer alcohols (FTOH)). The migration was done in an ultrasonic bath for 2 hours at 70°C.

The content of PFAS was analysed by a DTU FC430.3 accredited method for specific PFAS and by a high performance liquid chromatography (HPLC)-orbitrap-mass spectrometry (MS) method for suspect screening for other PFAS. The migration from samples with high PFAS content and identified by NFSA for possible follow-up actions was tested in triplicate.

The limit of quantification (LOQ) was the lowest spiking level determined when the method was validated, i.e. it is the lowest level at which a quantitative result was obtainable with a statistically significant level of analytical uncertainty (at the 95% confidence interval (CI)).

Terms of reference (ToR)

NFSA requested the Norwegian Institute of Public Health (NIPH) to perform risk assessments for adults of the seven FCM that had migration levels of PFAS above the LOQ after subtraction of the analytical uncertainty in migration tests performed on three separate samples (A, B, C) for each collected FCM item. In evaluations in which the exposure to a PFAS exceeded the health-based guidance value (HBGV), also migration levels including the analytical uncertainty should be considered. The risk assessments comprised five PFAS (Table 1).

Literature searches

The toxicity of PFOA (and PFOS) has been evaluated in a large number of studies in humans and laboratory animals, however, less toxicity data are available for other PFAS. According to the Agency for Toxic Substances and Disease Registry in USA, comparison of the toxicity of perfluoroalkyls across species is problematic due to differences in elimination half-lives, lack of adequate mechanistic data, species differences in the mechanism of toxicity for some endpoints and differences in measurement of exposure levels between epidemiological and experimental studies (ATSDR, 2021).

To look for existing risk assessments with a HBGV for these PFAS, grey literature was searched, such as the websites of the World Health Organization (WHO), Joint FAO/WHO Expert Committee on Food Additives (JECFA), Organisation for Economic Co-operation and Development (OECD), European Food Safety Authority (EFSA), Environmental Chemicals Agency (ECHA), European Medicines Agency (EMA), the European Union (EU) Commission Joint Research Centre (JRC), the Federal Institute for Risk Assessment (BfR, Germany), French National Institute for Industrial Environment and Risks (INERIS), French Agency for Food, Environmental and Occupational Health and Safety (ANSES) and the National Institute for Public Health and the Environment (RIVM, the Netherlands). In addition, various federal (the National Institutes of Health (NIH), the United States Environmental Protection Agency (US EPA) and Agency for Toxic Substances and Disease Registry (ATSDR)) and state (Texas Commission on Environmental Quality (TCEQ)) governmental institutions in USA were searched for information. The Australian Government and Food Standards Australia New Zealand (FSANZ) were also searched for HBGV for PFAS.

To obtain primary scientific publications describing experimental studies that could give information on safe levels of especially the less studied PFAS, a literature search was performed in OVID Medline. The search terms included the names, abbreviations and Chemistry Abstracts Service (CAS) no. of the five PFAS under evaluation, combined with terms for adverse health effects (health effect* OR harmful OR risk* OR adverse OR negative effect* OR toxic*) with and without another limiting term (health-based guidance value OR health based guidance value OR HBGV OR tolerabl* daily intake OR tolerabl* weekly intake OR NOAEL OR NOEL). The search was limited to publications in the English language, from 2018 to present, to obtain publications published after EFSA (2020). Relevant publications from the reference lists of the obtained reports and scientific publications were also included.

Exposure to PFAS

The plasma levels of PFAS vary substantially, due to varying exposure as well as differences in carbon chain-length, with much longer half-lives for long-chained substances, such as PFOA (C8) than for short-chained PFAS, such as perfluorobutanoic acid (PFBA) (C4),

perfluoropentanoic acid (PFPeA) (C5), perfluorohexanoic acid (PFHxA) (C6) (Xu et al., 2020; Abraham et al., 2024). 6:2 FTOH is C8, but no estimated half-life of it was found.

Table 1. Migration levels of PFAS above the LOQ minus the analytical uncertainty from three FCM samples of each product (A, B, C) (µg/kg food simulant)

FCM type	Reference no.	PFAS*	Sample A	Sample B	Sample C
Paper straw	2024/85370	PFOA	**0.059**	0.057	0.055
Paper straw	2024/85370	PFBA		0.015	
Muffin form	2024/86295	PFHxA	**0.092**	0.076	0.083
Muffin form	2024/86295	6:2 FTOH	2.970	2.079	2.283
Paper straw	2024/87057	PFOA	0.029	0.027	0.027
Straw	2024/93980	PFOA	0.028	0.028	0.030
Paper muffin form	2024/94002	PFHxA	0.055	0.050	0.051
Paper muffin form	2024/94002	6:2 FTOH	1.791	2.005	1.723
Paper plate	2024/092361	PFBA	**0.152**	0.142	0.137
Paper plate	2024/092361	PFPeA	**0.011**		
Microwave oven popcorn package	2024/102124	6:2 FTOH	7.336	**7.581**	7.296

*PFOA: perfluorooctanoic acid, CAS no. 335-67-1, PFBA: perfluorobutanoic acid, CAS no. 375-22-4, PFPeA: perfluoropentanoic acid, CAS no. 2706-90-3, PFHxA: perfluorohexanoic acid, CAS no. 307-24-4, 6:2 FTOH: 6:2 fluorotelomer alcohol, CAS no. 647-42-7. The numbers in bold are the highest levels of migration measured in this study across the seven FCM products for each of the five PFAS.

To establish regulatory migration limits for or perform risk assessments of chemicals from FCM, the conventional assumption used is that 1 kg of food is consumed daily over a lifetime by a person of 60 kg body weight (bw) and that this food is packaged in a cubic container of 6 dm^2 surface area releasing the substance (Commission Regulation (EU) No 10/2011). These assumptions are used by EFSA and other organizations. The exposures of five PFAS based on their highest level of migration from the FCM are shown in Table 2.

Table 2. Exposures to PFAS based on the highest level of migration in this study, a daily intake of 1 kg of food packaged in 6 dm^2 of the FCM and a body weight of 60 kg (for adults)

PFAS	Exposure (ng/kg bw per day)
PFOA	0.98
PFBA	2.5
PFPeA	0.18
PFHxA	1.5
6:2 FTOH	126.4

Risk assessment of PFAS

In these risk assessments, HBGV for the five PFAS have been searches for in various reports and publications for comparisons with the exposure levels. As HBGV, EFSA establishes tolerable

daily intake(TDI) or tolerable weekly intake (TWI), defined as estimates of the amount of a chemical substance in air, food or drinking water that can be taken in daily or weekly, respectively, over a lifetime without appreciable health risk. Because not all the PFAS under evaluation in this study have a HBGV established by EFSA, also such values from USA and other countries have been included.

The United States Environmental Protection Agency (US EPA), USA, defines a reference dose (RfD) as 'an estimate (with uncertainty spanning perhaps an order of magnitude) of a daily oral exposure to the human population (including sensitive subgroups) that is likely to be without an appreciable risk of deleterious effects during a lifetime (US EPA, 2022a). It can be derived from a no observed adverse effect level (NOAEL), lowest observed adverse effect level (LOAEL) or benchmark dose, with uncertainty factors generally applied to reflect limitations of the data used. Generally used in EPA's noncancer health assessments.' RfD are not enforceable standards, but they are risk assessment benchmarks. An aggregate daily exposure to a chemical at or below the RfD (expressed as 100 % or less of the RfD) is generally considered acceptable by US EPA. The individual US states can set their own RfD.

These risk assessments of PFAS do not consider exposures from other sources such as food.

Perfluorooctanoic acid (PFOA)

Health effects of PFOA

Estimated elimination half-life for PFOA in humans varies from 2.1–10.1 years (ATSDR, 2021). In a male human volunteer given a single oral PFAS mixture, Abraham et al. (2024) calculated a half-live of PFOA as point estimate (95% CI) of 2011 (1466-3206) days using isotope-labelled $^{13}C4_4$-PFOA.

Health effects are most thoroughly investigated for PFOA (and PFOS), while there is less information for other PFAS. The following information is summarized from EFSA (2018; 2020). PFOA inhibited the development of antibodies after vaccination in both laboratory animals and humans, which was considered the most sensitive effect, i.e. the effect seen at the lowest exposure in EFSA (2020). A reduced antibody formation after vaccination does not necessary indicate increased risk of illness, but a decreased resistant to infections. Some data indicated that PFOA exposure also increased the risk of infections, but these results were more limited.

Experiments on mice showed that a low PFOA exposure during the fetal stage and early after birth disrupted the development of the mammary glands. This has not been investigated for other PFAS, and it was not clear whether the same effect occurred in humans. Other developmental disorders have been investigated for several PFAS and only occur at much higher exposures.

An increase in liver weight was observed in laboratory animals at relatively low exposures to all investigated PFAS. However, because the mechanism for the effects of PFAS on the liver differs between laboratory animals and humans, this effect was not considered relevant for humans.

Liver toxicity and disturbances in fat metabolism were considered possibly relevant to humans, but were only observed at high exposure in animals. Many PFAS reduced the metabolic hormones triiodothyronine (T3) and thyroxine (T4) in laboratory animals, but did not affect the level of thyroid-stimulating hormone (TSH). The effects on the liver and on metabolic hormones occurred at much higher doses than those which affected antibody formation and mammary gland development in mice.

Many population studies have found associations between exposure to PFOA and health risk factors, such as between PFOA exposure and a small increase in cholesterol levels in adults. An association has also been shown between exposure to PFOA and a small increase in the liver enzyme alanine transferase (ALT), indicating liver toxicity. For these effects, it was not clear whether they were caused by PFOA or whether the associations were influenced by individual differences in the excretion and reabsorption of bile acids and PFOA.

It is possible that high PFOA levels in the mother during pregnancy were the cause of a somewhat lower birth weight observed in children, but this reduction was small and of unclear health significance.

A wide range of other health effects have been investigated in population studies. However, EFSA (2018; 2020) found that there was insufficient basis for drawing conclusions about causal relationships other than those effects described above.

EFSA (2020) reported a few new publications and reports that were found after the previous opinion, but these new studies did not change the conclusion on PFOA (and PFOS) made in EFSA (2018). EFSA (2020) concluded that for PFAS other than PFOA (and PFOS) the number of studies and data was limited.

Among 19 PFAS, PFOA was found to be one of those with a lower transplacental transfer efficiency (TTE) (mean ± SD, 0.82 ± 0.2) across the human placenta in a meta-analysis based upon a systematic review (Appel et al., 2022). The transmission rate of PFOA was lower when sampling occurred during pregnancy (in the 2nd-3rd trimester) than when sampling occurred at the time of delivery.

Eriksen et al. (2010) found that PFOA modestly increased the intracellular reactive oxygen species (ROS) production by 1.52-fold (95% CI, 1.37-1.67) in the 2,7-dichlorofluorescein-diacetate (DCFH-DA) assay in the human hepatocellular carcinoma cell line HepG2. However, the increase in ROS production was not concentration-dependent and PFOA did not generate DNA damage that could be detected by the alkaline Comet assay as strand breakage and alkali-labile sites or formamidopyrimidine DNA-glycosylase (FPG) sites in HepG2 cells.

PFOA did not induce mutations in *Salmonella typhimurium* strains TA98, TA100, TA1535, TA1537 and TA 1538 (Ames test) and was negative in the micronucleus test in V79 cells, in both tests with or without S9 fraction for metabolic activation (Buhrke et al., 2013). In this study, PFOA was also found to be cytotoxic in HepG2 cells, without affecting apoptosis.

PFOA did not cause significant levels of cytotoxicity and did not cause damage to human sperm DNA in the alkaline Comet assay (Emerce and Çetin, 2018).

PFOA decreased viability in the 3-(4,5-dimethylthiazol-2-yl)−2,5-diphenyltetrazolium bromide (MTT) assay and increased intracellular ROS in the DCFH-DA assay in HepG2 cells (Amstutz et al., 2022). PFOA also decreased viability in undifferentiated human THP-1 monocytes and in differentiated THP-1 macrophages in the MTT assay, and increased ROS in both of these cell types in the DCFH-DA assay (Amstutz et al., 2024).

EFSA (2018) concluded regarding genotoxicity that for PFOA (and PFOS) the available data were inconclusive. No evidence for a direct genotoxic mode of action was identified for these two substances. There was some evidence for oxidative stress induced by PFOA (and PFOS). In 2020, EFSA concluded that a few new studies did not change the conclusion made in 2018 (EFSA, 2020).

Crebelli et al. (2019) assessed genotoxicity in mice administered PFOA (0.1, 1 and 5 mg/kg bw) for five weeks through drinking water. Markers of cell toxicity, oxidative stress and DNA strand breaks were measured in the liver, the main target of toxicity of PFOA in rodents, and systemic genotoxicity was also assessed by the analysis of micronuclei in reticulocytes and spleen lymphocytes, and germ cell effects by the Comet assay on testis cells. PFOA administration at the highest dose (5 mg/kg bw) induced marked liver hypertrophy with signs of cell injury (elevated ALT and aspartate aminotransferase (AST)), with no concurrent evidence of lipid peroxidation and oxidative stress (decreased antioxidant capacity). No evidence of treatment-related genotoxicity was observed. Overall, the data indicated that severe liver toxicity induced by PFOA administration was not associated with oxidative stress.

The National Toxicology Program in USA (NTP TR 598, 2020, revised 2023) reported that PFOA did not show evidence of genotoxic activity in tests conducted by NTP. PFOA was negative in bacterial mutagenicity assays in *Salmonella typhimurium* strain TA100 and *Escherichia coli* strain WP2 uvrA pKM101, with and without 10% rat liver S9, and in strain TA98, with S9. In strain TA98 without S9, results with PFOA were equivocal. PFOA was judged to be negative in the peripheral blood micronucleus assay. There were no increases in micronucleated reticulocytes in the peripheral blood of female Sprague Dawley rats administered PFOA for 28 days via gavage. Although a positive response was indicated for male rats, the results were within the laboratory's historical control range and therefore the biological significance of the increase was questionable. No changes were noted in the percentage of reticulocytes in peripheral blood of either sex, suggesting that PFOA did not induce bone marrow toxicity.

In 2023, PFOA was classified as "carcinogenic in humans" (Group 1) by the International Agency for Research on Cancer (IARC) (Zahm et al., 2024).

HBGV for PFOA

Of the five PFAS found to migrate from FCM and evaluated in this risk assessment, only PFOA had an individual established TDI from EFSA. In 2018, a TWI value for PFOA was established by EFSA as 6 ng/kg bw per week (EFSA, 2018), corresponding to a TDI of 0.86 ng/kg bw per day. In 2020, EFSA evaluated the risk of among other PFAS the four substances PFOA, perfluorononanoic acid (PFNA), perfluorohexane sulfonic acid (PFHxS) and PFOS in food. Based on several similar effects in animals, toxicokinetics with similar accumulation and long half-lives, and observed levels in human blood, EFSA established a group TWI for the sum of these

four substances of 4.4 ng/kg bw per week (EFSA, 2020), corresponding to **0.63 ng/kg bw per day**. This value should be used for the presence of either one, two, three or four of these substances in the group, and replaced the individual TDI for PFOA from 2018. Because the data were not sufficient for derivation of potency factors, EFSA assumed by default equal, weight-based, potencies for effects of these four PFAS on the critical endpoint.

The most critical human health effect was decreased antibody response of the immune system to vaccination, which was the basis for the TWI. Because accumulation over time was considered important, a TWI instead of a TDI was established. This TWI was lower than the older TWI that EFSA set for PFOA in 2018. The reasons for the changes were new studies of health effects, new information on how the substances are distributed and excreted from the body, and that the four substances were now assessed together.

The four PFAS included in the new group TWI from 2020 make up approximately half of the amount of PFAS ingested from food (EFSA, 2020). Nevertheless, in total they make up approximately 90% of PFAS in the blood. This is because a large proportion of the other ingested PFAS come from PFBA and PFHxA (evaluated below). These are substances that are excreted relatively quickly from the body, thus, having short half-lives in humans, and less is known about their toxic properties. Therefore, these substances were not taken into account when determining the TWI for the sum of PFOA, PFNA, PFHxS and PFOS.

These four PFAS contributed approximately 46% to the sum of all PFAS in adults for which the exposure was calculated, with relative contributions of 9%, 2%, 4% and 30% for PFOA, PFNA, PFHxS and PFOS, respectively (EFSA, 2020).

For comparison (Table 3), on the US federal level, the US EPA derived an oral non-cancer RfD of 0.00002 mg/kg bw per day (**20 ng/kg bw per day**) for PFOA (US EPA, 2017). The RfD is an estimate of the daily exposure level that is likely to be without harmful effects over a lifetime, i.e. similar to a TDI. On the US state level, Texas Commission on Environmental Quality set a RfD of **12 ng/kg bw per day** for PFOA (TCEQ, 2023).

The health authorities in Australia and New Zealand established a TDI of **160 ng/kg bw per day** for PFOA based on a NOAEL for fetal toxicity in a developmental and reproductive study in mice (Australian Government, 2017).

Risk assessment of PFOA

Evaluation of exposure to PFOA alone compared with the sum TWI from EFSA (2020)

The highest migration level of PFOA reported was 0.059 µg/kg food simulant from paper straws after subtraction of the analytical uncertainty (**Reference no. 2024/85370**) (Table 1). If a person with body weight 60 kg eats 1 kg food packaged in FCM with this level of PFOA per day, the exposure is **0.98 ng/kg bw per day** or 6.88 ng/kg bw per week (Table 2), i.e. above the TWI (about 1.6 times) set for the sum of four PFAS by EFSA (2020), but not the HBGV by US EPA (2017), TCEQ (2023) or Australian Government (2017) (Table 3). However, it may not be likely that an adult person uses drinking straws every day.

For the other two (paper) straws examined in which migration of PFOA was detected above the LOQ (**Reference nos. 2024/870057** and **2024/93980**), the highest exposure to PFOA alone, **0.58 and 0.62 ng/kg bw per day**, respectively, did not exceed the TDI for PFOA (EFSA, 2020).

Table 3. Exceedance of various health-based guidance values (HBGV) for the highest migration level of PFOA from paper straws (Reference no. 2024/85370)

Institution (year)	HBGV (ng/kg bw per day)	Did the exposure to PFOA (0.98 ng/kg bw per day), exceed the HBGV?
EFSA (2020)	0.63 (for sum of 4 PFAS, including PFOA)	Yes
US EPA (2017)	20	No
TCEQ (2023)	12	No
Australian Government (2017)	160	No

For PFOA alone in all three straw products, the conclusions would be the same if the analytical uncertainty values for PFOA (Table 5, Appendix 1) were not subtracted.

Evaluation of exposure to several PFAS compared with the sum TWI from EFSA (2020)

In the paper straws with **Reference no. 2024/85370**, PFOS and PFNA were not detected in any of the three samples. PFHxS was detected in one sample (C), above LOD, but below LOQ, as 0.005 µg/kg food simulant. The highest level of PFOA in sample C was 0.067 µg/kg, and after subtraction of the analytical uncertainty of 0.012 µg/kg, the level was 0.055 µg/kg (Table 5, Appendix 1). Thus, the sum of PFHxS (0.005 µg/kg) and PFOA (0.055 µg/kg) is 0.060 µg/kg, giving an exposure of 7.0 ng/kg bw per week, exceeding the sum TWI in EFSA (2020) (which was 4.4 ng/kg bw per week) about 1.6 times. If not subtracting the analytical uncertainty for PFOA, the corresponding exposure is 8.4 µg/kg per week for sample C, thus, i.e. about the double of the sum TWI from EFSA (2020).

From the straws with **Reference no. 2024/93980**, no migration of PFOS or PFNA was detected. Migration of PFHxS was 0.014 and 0.006 µg/kg food simulant in two samples (A and C), respectively. In sample A, the highest level of PFOA was 0.034 µg/kg, and after subtraction of the analytical uncertainty (0.0061 µg/kg), the level was 0.0279 µg/kg (Table 5, Appendix 1). Thus, the sum of PFHxS (0.014 µg/kg) and PFOA (0.0279 µg/kg) is 0.042 µg/kg, giving an exposure of 4.9 ng/kg bw per week, i.e. slightly above the sum TWI from EFSA (2020). If not subtracting the analytical uncertainty for PFOA, the corresponding exposure is 5.6 µg/kg per week for sample A, i.e. slightly above the sum TWI from EFSA (2020).

In sample C, the highest level of PFOA was 0.037 µg/kg, and after subtraction of the analytical uncertainty (0.0066 µg/kg), the level was 0.0304 µg/kg (Table 5, Appendix 1). Thus, the sum of PFHxS (0.006 µg/kg) and PFOA (0.0304 µg/kg), is 0.0364 µg/kg, giving an exposure of 4.2 ng/kg bw per week, i.e. slightly below the sum TWI from EFSA (2020). If not subtracting the analytical uncertainty for PFOA, the corresponding exposure is 5.0 µg/kg per week for sample C, i.e. slightly above the sum TWI from EFSA (2020).

From the straws with **Reference no. 2024/870057**, no migration of PFOS, PFNA or PFHxS was detected, only PFOA (evaluated above).

Conclusions on PFOA

Since the highest exposures to PFOA alone in paper straws with Reference no. 2024/85370, and in two straws (Reference numbers 2024/85370 and 2024/93980) also the sum of PFOA and PFHxS, were above the TDI and sum TWI established by EFSA, respectively, there is some concern about potential risk of adverse health effects from PFOA, or PFOA and PFHxS, from these two paper straws. However, the exceedance was small in all cases. Since the highest exposure to PFOA alone in Reference no. 2024/87057 did not exceed the TDI from EFSA, these paper straws are likely to be without an appreciable risk of adverse health effects from PFOA.

Perfluorobutanoic acid (PFBA)

PFBA has an estimated elimination half-life of 72-81 hours (ATSDR, 2021). In a male human volunteer given a single oral PFAS mixture, Abraham et al. (2024) calculated a half-live of PFBA as point estimate (95% CI) of 4.18 (4.06-4.31) days using isotope-labelled $^{13}C_3$-PFBA.

PFBA contributed approximately 16% to the sum of all 17 PFAS for which the exposure was calculated in adults, however, PFBA was not included among the four PFAS for which the TWI was based on the sum of, and thus, no risk assessment was done for PFBA (EFSA, 2020).

Health effects of PFBA

PFBA ammonium (NH4+) salt was given to timed-pregnant CD-1 mice by oral gavage daily from gestational day (GD) 1 to 17 at 0, 35, 175 or 350 mg/kg bw by Das et al. (2008). PFBA did not significantly affect maternal weight gain, number of implantations, fetal viability, fetus weight or incidence of fetal malformations. Incidence of full-litter loss was significantly greater in the 350 mg/kg bw group and maternal liver weights were significantly increased at 175 and 350 mg/kg bw. PFBA exposure during pregnancy did not adversely affect neonatal survival or postnatal growth. A significant delay in eye-opening in offspring was detected in all three PFBA groups and slight delays in the onset of puberty were noted at 175 and 350 mg/kg bw. The data suggested that exposure to PFBA during pregnancy in the mouse did not produce developmental toxicity comparable to that observed with PFOA, in part, due to rapid elimination of the chemical.

Among 19 PFAS, PFBA was found to have a high TTE (mean ± SD, 2.17 ± 1) across the human placenta in a meta-analysis based upon a systematic review (Appel et al., 2022).

Butenhoff et al. (2012) performed sequential 28-day and 90-day oral toxicity studies in male and female Sprague-Dawley rats with ammonium perfluorobutyrate (NH4+PFBA) at doses up to 150 and 30 mg/kg bw per day, respectively. In the 90-day study, rats of both sex (n = 10 per sex per group) were given PFBA as NH4+PFBA in doses of 0, 1.2, 6 and 30 mg/kg bw per day by gavage. No adverse effects were observed in the females, whereas in the males increased liver weight, slight to minimal hepatocellular hypertrophy, decreased serum total cholesterol

and reduced serum T4 thyroid hormone with no change in serum TSH were observed. During recovery, liver weight, histological and cholesterol effects were resolved. The transcriptional expression of the xenosensor nuclear receptors peroxisome proliferator activated receptor (PPAR) and the constitutive androstane receptor (CAR), as well as the thyroid receptor, was increased, and expression of the cytochrome P450 (Cyp)1A1 enzyme was decreased. The NOAEL values were 6 and >150 mg/kg bw per day for male and female rats in the 28-day study, and 6 and >30 mg/kg bw per day in the 90-day study, respectively.

Weatherly et al. (2021) analysed serum chemistry, histology, immune phenotyping and gene expression to evaluate the systemic toxicity of subchronic (15-day) dermal PFBA (375 mg/kg bw) or 28-day (93.8-187.5 mg/kg bw) exposures. PFBA produced significant increases in liver and kidney weights and altered serum chemistry (all doses). Immune cell phenotyping identified significant increases in several types of immune cells. Histopathological and gene expression changes were observed in both the liver and skin. The findings indicated that PFBA induced liver toxicity and alterations of PPAR target genes, suggesting a role of a PPAR pathway.

US EPA (2022b) concluded that the available evidence indicated that developmental, thyroid and liver effects in humans were likely caused by PFBA exposure *in utero* or during adulthood. There was inadequate evidence to determine whether reproductive effects might be a potential human health hazard after PFBA exposure.

PFBA did not significantly decrease viability in the MTT assay but increased intracellular ROS in the DCFH-DA assay in HepG2 cells (Amstutz et al., 2022). PFBA also decreased viability in THP-1 monocytes and THP-1 macrophages in the MTT assay, and increased ROS in both of these cell types in the DCFH-DA assay (Amstutz et al., 2024).

PFBA did not induce mutations in *Salmonella typhimurium* strains TA98, TA100, TA1535, TA1537 and TA 1538 (Ames test) and was negative in the micronucleus test in mammalian V79 cells, in both tests with or without S9 fraction for metabolic activation (Buhrke et al., 2013).

Crebelli et al. (2019) assessed genotoxicity in mice administered PFBA (5 mg/kg bw) for five weeks through drinking water. Only mild liver hypertrophy, with no other signs of toxicity and a lower tendency for bioaccumulation compared with PFOA, was observed with PFBA. No evidence of treatment-related genotoxicity was observed.

No human or animal studies were available to give information about the potential for PFBA exposure to cause cancer. Overall, there is inadequate information to assess the carcinogenic potential of PFBA exposure (US EPA, 2022b).

HBGV for PFBA

US EPA established an oral lifetime (chronic) RfD for non-cancer effects of PFBA of 0.001 mg/kg bw per day (**1000 ng/kg bw per day**) (US EPA, 2022b), which is also reported by TCEQ (2023). The confidence in this value was considered medium and was based on hepatic and thyroid effects in rats in the 90-day study by Butenhoff et al. (2012). The NOAEL values were 6 and >30 mg/kg bw per day for male and female rats, respectively. The point of departure (POD) doses for the effects in rats used for the risk assessment were converted to human equivalent doses (HED) taking into account pharmacokinetic difference between rats and humans. The POD for humans were 1.15 and 1.27 mg/kg bw per day for increased liver hypertrophy and decreased

T4 thyroid hormone, respectively, and they used an uncertainty factor of 1000 to derive the RfD.

Risk assessment of PFBA

The highest migration of PFBA was 0.152 µg/kg food simulant from samples of paper plates (**Reference no. 2024/092361)** (Table 1). The migration from samples of paper straws (**Reference no. 2024/85370**) was 0.015 µg/kg food simulant. If a person with a body weight of 60 kg eats 1 kg food packaged in FCM with the highest level of PFBA per day (0.152 µg/kg), the exposure is **2.5 ng/kg bw per day** (Table 2), i.e. far below (about 400 times) the US EPA RfD (US EPA, 2022b).

Conclusions on PFBA

The exposure to PFBA from paper plates (Reference no. 2024/092361) or paper straws (Reference no. 2024/85370) in this study did not exceed the available RfD from US EPA, and thus, both products are likely to be without an appreciable risk of adverse health effects from PFBA.

Perfluoropentanoic acid (PFPeA)

In a male human volunteer given a single oral PFAS mixture, Abraham et al. (2024) calculated a half-live of PFPeA as point estimate (95% CI) of 0.52 (0.51-0.53) days using isotope-labelled $^{13}C_3$-PFPeA.

PFPeA was included in the sum of 17 PFAS for which the exposure was calculated in adults, however, PFPeA was not included among the four PFAS for which the TWI was based on the sum of, and thus, no risk assessment was done for PFPeA by (EFSA, 2020).

Health effects of PFPeA

Fewer relevant studies were found on PFPeA than for the other PFAS. Systemic toxicity of a daily subchronic 28-day dermal exposure to PFPeA (3.25-125 mg/kg bw) was examined in female B6C3F1 mice by Weatherly et al. (2023). PFPeA increased relative liver weight and caused hepatocellular hypertrophy. Histopathological changes were observed in both liver and skin. Gene expression changes were observed with PPAR isoforms in the liver and skin along with changes in genes involved in steatosis, fatty acid metabolism, necrosis and inflammation. These findings, along with significant detection levels in serum and urine, supported PFPeA-induced liver damage and PPARα, δ and γ receptor involvement in systemic toxicity and immunological disruption, indicating that dermal exposure to PFPeA has similar trend in liver effects compared to oral exposure and PFOA exposure.

HBGV for PFPeA

No risk assessments from EFSA or RfD value from US EPA were found for PFPeA, however, the state of Texas, USA, had a RfD for PFPeA (TCEQ, 2023). The oral lifetime (chronic) RfD for non-cancer effects of PFHxA of 0.0005 mg/kg bw per day (**500 ng/kg bw per day**) is used as a surrogate value for PFPeA, which has no useful toxicity data according to TCEQ (2023).

Risk assessment of PFPeA

The only detected migration of PFPeA above LOQ was 0.011 µg/kg food simulant from samples of paper plates (**Reference no. 2024/092361**) (Table 1). If a person with a body weight of 60 kg eats 1 kg food packaged in FCM with this level of PFHxA per day, the exposure is **0.18 ng/kg bw per day** (Table 2). The exposure to PFPeA is below (ca. 2728 times) the RfD from TCEQ (2023).

Conclusions on PFPeA

The exposure to PFPeA from paper plates (Reference no. 2024/092361) in this study did not exceed the RfD from TCEQ (2023), and thus, these paper plates are likely to be without an appreciable risk of adverse health effects from PFPeA.

Perfluorohexanoic acid (PFHxA)

PFHxA has an estimated mean half-life of 32 days (range 14-49 days) in humans according to Russell et al. (2013). However, in a male human volunteer given a single oral PFAS mixture, Abraham et al. (2024) calculated a half-live of PFHxA as point estimate (95% CI) of 1.45 (1.42-1.48) days using isotope-labelled $^{13}C_2$-PFHxA.

PFHxA does not seem to persistently bioaccumulate in the manner of many other PFAS. For example, Swedish ski wax technicians, who have high PFAS exposure, did not have significantly higher levels of PFHxA in their blood samples when compared to the general population median for their age groups, even while having concentrations of other PFAS, like PFOA, up to 44 times higher than the general population (Nilsson et al., 2010).

PFHxA contributed approximately 15% to the sum of all 17 PFAS for which the exposure was calculated in adults, however, PFHxA was not included among the 4 PFAS for which the TWI was based on the sum of, and thus, no risk assessment was done for PFHxA by (EFSA, 2020).

Health effects of PFHxA

Chengelis et al. (2009) evaluated toxic effects of PFHxA in rats administered orally by gavage at levels up to 200 mg/kg bw per day for 90 days. Lower body weight gains were noted in the 10, 50 and 200 mg/kg bw group males (without dose-response). Other changes included lower red blood cell parameters, higher reticulocyte counts and lower globulin in the 200 mg/kg bw group males and females, higher liver enzymes in males at 50 and 200 mg/kg bw, lower total protein and higher albumin/globulin ratio, and lower cholesterol and calcium in males at 200 mg/kg bw. Minimal centrilobular hepatocellular hypertrophy was present in 200 mg/kg bw group males and correlated with higher liver weights and slightly higher peroxisome β-oxidation activity at the end of the dosing period. The authors concluded that based on liver histopathology and liver weight changes, the NOAEL for oral administration was 50 and 200 mg/kg bw day for males and females, respectively.

Loveless et al. (2009) reported a 90-day subchronic toxicity study, including an one-generation reproduction part, and a developmental toxicity study in Crl:CD(SD) rats. Because the pKa of PFHxA is less than 3, it exists in the environment principally as an anion, PFHx, and therefore

they administered sodium perfluorohexanoate (NaPFHx). The doses used were 0, 20, 100 and 500 mg/kg bw per day administered by oral gavage in all three studies. The female rats in the reproductive study were dosed for approximately 70 days prior to cohabitation through gestation and lactation, for a total of ca. 126 days, and the males were dosed for ca. 110 days. In the developmental toxicity study, the female rats were dosed once daily on days 6-20 of gestation, and the offspring were sacrificed on GD 21. In the 90-day toxicity study, pathological findings (nasal lesions) were reported in male and female rats at 100 and 500 mg/kg bw per day, and adverse changes in hematology, and hepatocellular and thyroid hypertrophy, were observed at 500 mg/kg bw per day. No NaPFHx-related anatomic pathology, hepatic peroxisomal β-oxidation, neurobehavioral or clinical pathology changes were present in male or female rats administered 20 mg/kg bw per day, resulting in a NOAEL for subchronic toxicity of 20 mg/kg bw per day. For the reproductive part of the 90-day subchronic study, the parental adult rat NOAEL was 20 mg/kg bw per day, based on reduced body weight parameters, whereas the NOAEL for reproductive toxicity was 100 mg/kg bw per day, based on reduced offspring pup weights. The maternal and developmental toxicity NOAEL was 100 mg/kg bw per day, based on decreased maternal and fetal body weight at 500 mg /kg bw per day. No NaPFHx-related effects were noted on incidence of fetal, visceral or skeletal variations at any doses.

A chronic study (duration 104 weeks) was performed to evaluate the possible toxicologic and carcinogenic effects of PFHxA by daily gavage, 7 days per week, in male and female Sprague-Dawley rats (Klaunig et al., 2015). Dose levels of 0, 2.5, 15 and 100 mg/kg bw per day of PFHxA in males and 5, 30 and 200 mg/kg bw per day of PFHxA in females were used. No effects on body weight, food consumption, a functional observational battery or motor activity were observed. While no difference in survival rates in males was seen, a dose-dependent decrease in survival in PFHxA-treated females was observed. Hematology and serum chemistry were unaffected. PFHxA-related histologic changes (papillary necrosis) were noted in the kidneys of the 200 mg/kg bw per day group females. Other changes included decreases in triglyceride serum levels for males at 2.5 mg/kg bw day and decreases in serum low density lipoprotein (LDL)/very low density lipoprotein (VLDL) and increases in urinary volume at 200 mg/kg bw per day for females. Finally, there was no evidence that PFHxA was tumorigenic in male or female rats at any of the doses used.

Systemic toxicity of a daily subchronic 28-day dermal exposure to PFHxA (3.25-125 mg/kg bw) was examined in female B6C3F1 mice (5 per group) by Weatherly et al. (2023). PFHxA increased relative liver weight and caused hepatocellular hypertrophy. Histopathological changes were observed in both liver and skin. Gene expression changes were observed with PPAR isoforms in the liver and skin along with changes in genes involved in steatosis, fatty acid metabolism, necrosis and inflammation. These findings, along with significant detection levels in serum and urine, supported PFHxA-induced liver damage and PPARα, δ and γ receptor involvement in systemic toxicity and immunological disruption, indicating that dermal exposure to PFHxA has similar trend in liver effects compared to oral exposure and PFOA exposure.

The reproductive toxicity of PFHxA ammonium salt was investigated in pregnant Crl: CD1(ICR) mice (20 females/group) given once daily from GD 6-18 by Iwai and Hoberman (2014). In phase

1, the doses were 0, 100, 350 and 500 mg/kg bw per day, and in phase 2, the doses were 0, 7, 35 and 175 mg/kg bw per day. At doses of 350 and 500 mg/kg bw per day, maternal mortalities, excess salivation and changes in body weight gains occurred. The authors concluded that the maternal and reproductive NOAEL of PFHxA ammonium salt was 100 mg/kg bw per day. Pup body weights were reduced on postpartum day (PPD) 0 in all dose groups, but persisted only in the 350 and 500 mg/kg bw per day groups. Additional effects at 350 and 500 mg/kg bw per day were stillbirths, reductions in viability indices and delays in physical development. In phase 2, adverse effects occurred only in the 175 mg/kg bw per day group and consisted of increased stillborn pups, pups dying on PPD 1 and reduced pup weights on PPD 1. Thus, the NOAEL for developmental toxicity was 35 mg/kg bw per day. In an addendum publication (Iwai et al., 2019) they extended the analysis and interpretation of findings by pooling the control group information from both phases of the previous study, comparing the study findings to the incidence rates for stillbirths and postpartum viability for this species and strain of mouse observed for similar studies conducted by the same laboratory and evaluating data on the incidence and range of spontaneous eye abnormalities reported in the literature. Based on this supplemental evaluation, they claimed that the original study supported a NOAEL of 175 mg/kg bw per day for PFHxA in mice, which is a factor of 5-fold higher than previously reported. Furthermore, they noted that 175 mg/kg bw per day for maternal exposure was an unbounded NOAEL for developmental effects, i.e. the study did not establish a dose at which developmental effects may occur.

Among 19 PFAS, PFHxA was found to have a high TTE (mean ± SD, 2.71 ± 2.28) across the human placenta in a meta-analysis based upon a systematic review (Appel et al., 2022).

A critical review of data including epidemiological studies, *in vivo* and *in vitro* toxicity studies on PFHxA acute, subchronic and chronic toxicity, as well as key findings from toxicokinetic and mode of action studies was performed by Luz et al. (2019). They concluded that PFHxA was not carcinogenic, was not a selective reproductive or developmental toxicant, and did not disrupt endocrine activity. Further, they concluded that the effects caused by PFHxA exposure were largely limited to potential mild and/or reversible kidney effects that occurred at much higher doses than observed for PFOA. They calculated a chronic RfD for PFHxA of 0.25 mg/kg bw per day using benchmark dose modeling of renal papillary necrosis from a chronic rat bioassay.

Regarding health effects of PFHxA, US EPA (2023) identified endocrine, hepatic, hematopoietic and developmental toxicity of potential concerns for humans. The evidence was inadequate to determine whether PFHxA had the potential to cause adverse effects on the kidneys, reproduction, the immune system or the nervous system in humans.

PFHxA did not significantly decrease viability in the MTT assay but increased intracellular ROS in the DCFH-DA assay in HepG2 cells (Amstutz et al., 2022). PFHxA also decreased viability in THP-1 monocytes and THP-1 macrophages in the MTT assay, and increased ROS in both of these cell types in the DCFH-DA assay (Amstutz et al., 2024).

PFHxA did not significantly increase ROS production and did not generate DNA damage detected by the alkaline Comet assay as strand breakage and alkali-labile sites or FPG sites in HepG2 cells (Eriksen et al., 2010).

Regarding genotoxicity of PFHxA *in vitro*, no mutations were detected in bacterial reverse mutation (Ames) assay in *Salmonella typhimurium* strains TA98, TA100, TA1535 and TA1537, or in *Escherichia coli* strain WP2uvrA, or in chromosomal aberrations in human peripheral blood lymphocytes (Loveless et al., 2009).

PFHxA did not induce mutations in *Salmonella typhimurium* strains TA98, TA100, TA1535, TA1537 and TA 1538 (Ames test) and was negative in the micronucleus test in V79 cells, in both tests with or without S9 fraction for metabolic activation (Buhrke et al., 2013).

PFHxA did not cause significant levels of cytotoxicity and did not cause damage to human sperm DNA in the alkaline Comet assay (Emerce and Çetin, 2018).

NTP TOX 97 (2019, revised 2022) also reported that PFHxA was negative in bacterial mutagenicity tests. *In vivo*, in a 28-day study, no increase in micronucleated reticulocytes was observed in peripheral blood of male and female rats administered PFHxA (31.5-500 mg/kg bw twice daily), whereas the percentage of circulating immature erythrocytes was markedly increased, suggesting a stimulation of erythropoiesis in the bone marrow of PFHxA-treated rats (NTP TOX 97 (2019, revised 2022).

US EPA concluded that there is inadequate information to assess carcinogenic potential for PFHxA by all routes of exposure (US EPA, 2023).

HBGV for PFHxA

After considering the publications by i.a. Loveless et al. (2009), Chengelis et al. (2009) and Klaunig et al. (2015), US EPA established an oral lifetime (chronic) RfD for non-cancer effects of PFHxA of 0.0005 mg/kg bw per day (**500 ng/kg bw per day**) based on the study by Loveless et al. (2009) (US EPA, 2023). This value was based on decreased body weight at postnatal day 0 in the rat offspring. The confidence in this value was considered medium. The POD dose for this effect used for the risk assessment was converted to HED taking into account pharmacokinetic difference between rats and humans. The POD for humans was 0.048 mg/kg bw per day and an uncertainty factor of 100 was used to derive the RfD.

Risk assessment of PFHxA

The highest migration of PFHxA was 0.092 µg/kg food simulant from samples of muffins forms (**Reference no. 2024/86295**) (Table 1). If a person with a body weight of 60 kg eats 1 kg food packaged in FCM with this level of PFHxA per day, the exposure is **1.5 ng/kg bw per day** (Table 2), i.e. far below (ca. 326 times) the US EPA RfD (US EPA, 2023). PFHxA was found to migrate also from the paper muffin forms (**Reference no. 2024/94002**) and the highest value was 0.055 µg/kg food simulant in this product (Table 1). The exposure from this product is **0.9 ng/kg bw per day**, even further below the RfD.

Conclusions on PFHxA

The exposure to PFHxA from two muffin form products (Reference nos. 2024/86295 and 2024/94002) in this study did not exceed the available RfD from US EPA, and thus, both products are likely to be without an appreciable risk of adverse effects from PFHxA.

6:2 fluorotelomer alcohol (6:2 FTOH)

FTOH substances such as 6:2 FTOH can undergo environmental degradation to perfluorocarboxylic acids (PFCA), including PFOA, PFBA and PFHxA, some of which are persistent and bioaccumulative in the environment. FTOH may therefore be considered an indirect source of PFCA in the environment (EFSA, 2020).

No risk assessments from EFSA or RfD values from US EPA were found for 6:2 FTOH.

Health effects of 6:2 FTOH

6:2 FTOH was considered slightly acute toxic based on an oral lethal dose (LD)50 in rats of 1750 mg/kg bw (Serex et al., 2014). In rabbits, 6:2 FTOH was not a primary skin or eye irritant, and it did not produce a dermal sensitization response in mice (Serex et al., 2014).

According to Rice et al. (2020), some published risk assessments have assumed that perfluorohexanoic acid (PFHxA), a metabolite of 6:2 FTOH, adequately models the human health effects of 6:2 FTOH. However, they claimed based on a comparative analysis, that 6:2 FTOH was significantly more toxic than PFHxA. Therefore, the use of toxicological studies conducted with PFHxA to assess 6:2 FTOH exposure may significantly underestimate human health risk.

Toxicokinetic evaluation of 6:2 FTOH calculated times to steady state of one of its main metabolites, 5:3 fluorotelomer carboxylic acid (5:3A), in the plasma and tissues of up to a year after repeated oral exposure to rats, indicating that 5:3A may be persistent, raising concern about long-term health effects of 6:2 FTOH (Rice et al., 2024).

In a 90-day subchronic toxicity study, Crl:CD(CD) rats were exposed to 6:2 FTOH via oral gavage in doses of 0, 5, 25, 125 and 250 mg/kg bw per day (10 rats per sex per group). Deaths were observed at doses of 125 og 250 mg/kg bw per day after about three weeks of exposure and continued sporadically (Serex et al., 2014). The NOAEL in this subchronic study was 5 mg/kg bw per day, based on hematological changes and effects on the liver at higher doses (Serex et al., 2014).

To evaluate potential developmental and reproductive toxicity, 6:2 FTOH was administered by oral gavage to Sprague-Dawley rats at doses of 0, 5, 25, 125 or 250 mg/kg bw per day (O'Connor et al., 2014). In the developmental toxicity study, the pregnant dams were dosed one daily from GD 6-20. The adverse maternal toxicity observed at 250 mg/kg bw per day included reductions in body weight parameters and food consumption. Evidence of developmental toxicity was limited to increases in skeletal variations (ossification delays in the skull and rib alterations) at 250 mg/kg bw per day. There were no adverse maternal or developmental effects observed at 5, 25 or 125 mg/kg bw per day and there were no effects

on reproductive outcome or quantitative litter data at any dose level. For the one-generation reproduction toxicity study, the parental rats were dosed for at least 70 days prior to mating and throughout cohabitation (males and females), gestation and lactation period (females only), and were terminated on test day 110 (males) and PPD 22 (dams and offspring). Systemic parental and developmental toxicity were observed at 125 and 250 mg/kg bw per day. At 250 mg/kg bw per day, there was increased mortality among male and female parental rats, effects on body weight parameters, food consumption and clinical signs, and there were effects on offspring survival indices and body weights. At 125 mg/kg bw per day, there was an increase in mortality in parental males only, and parental toxicity was limited to effects on body weight gain, food consumption (lactation) and clinical signs. Uterine weights were decreased at 125 and 250 mg/kg bw per day, although no histopathological changes were observed. At 125 mg/kg bw per day, pup mortality was increased on lactation day 1 and body weights of the offspring were decreased during the second half of lactation. There was no evidence of either parental or developmental toxicity at 5 or 25 mg/kg bw per day. There were no effects on reproductive outcome at any dose level. Based on these data, 6:2 FTOH was not considered a selective reproductive or developmental toxicant at doses that induce clear maternal/parental toxicity.

In a study on CD-1 mice, 6:2 FTOH was administered by oral gavage in doses of 0, 1, 5, 25 and 100 mg/kg bw per day (Mukerji et al., 2015). The mice were euthanized after completion of the cohabitation period for parental males (on test days 107-109), on postpartum day 21 for parental females and after developmental landmark achievement for all offspring adults (on post-natal day 40-43). NOAEL for systemic toxicity was 25 mg/kg bw per day for males and 5 mg/kg bw per day for females, based on mortality, changes in body weight, hematological parameters, nutritional parameters, clinical chemistry (liver-related), liver weights and histopathology (liver, teeth, reproductive tract and mammary gland). For reproductive toxicity, the NOAEL was >100 mg/kg bw per day, i.e. no effects on reproductive outcomes were observed with any of the doses. For the offspring, the NOAEL was 25 mg/kg bw per day, with signs of delayed maturation, reduced survival and lower body weight at 100 mg/kg bw per day.

While the severity of the effects was generally greater in mice (Mukerji et al., 2015) than reported in rats (Serex et al., 2014; O'Connor et al., 2014), the overall NOAEL was identical in both species, 5 mg/kg bw per day for systemic toxicity and 25 mg/kg bw day for offspring viability/growth. Thus, 6:2 FTOH was not a selective reproductive toxicant in either species, as no effects on reproductive outcome occurred at any dose level, and any effects observed in offspring occurred at dose levels that induced mortality and severe toxicity in maternal animals.

The NOAEL for reproductive toxicity of 6:2 FTOH was >100 and >125 mg/kg bw per day in studies with mice (Mukerji et al., 2015) and rats (O'Connor et al., 2014). However, a more recent study by Xia et al. (2023) indicated that 5 mg/kg bw may be a LOAEL, not a NOAEL, for reproductive toxicity in mice in the above-mentioned studies, since they found that even the lowest concentration (5 mg/kg bw per day) of 6:2 FTOH caused slight damage to the blood-testis barrier, disorganization of testicular tissue and abnormal spermatogenesis in offspring mice.

The study by Xia et al. (2023) showed that embryonic 6:2 FTOH exposure caused reproductive toxicity by disrupting the formation of the blood-testis barrier in offspring BALB/c mice. Pregnant mice were given corn oil or 5, 25 or 125 mg/kg bw per day of 6:2 FTOH by gavage from GD 12.5-21.5. The embryonic 6:2 FTOH exposure resulted in disrupted testicular structure, low expression of tight junction protein between Sertoli cells, impaired blood-testis barrier formation and maturation, reduced sperm viability and increased malformation, and induced testicular inflammation in the offspring of mice.

6:2 FTOH did not decrease either the viability in the MTT assay or the intracellular ROS in the DCFH-DA assay in HepG2 cells (Amstutz et al., 2022). 6:2 FTOH decreased viability both in THP-1 monocytes and THP-1 macrophages in the MTT assay, but did not increase ROS in either of these cell types in the DCFH-DA assay (Amstutz et al., 2024).

6:2 FTOH was not mutagenic in the bacterial reverse mutation test or in the mouse lymphoma assay and was not clastogenic in a chromosome aberration assay in human lymphocytes (Serex et al., 2014).

HBGV for 6:2 FTOH

The LOAEL for 6:2 FTOH based on the reproductive toxicity experiment in mice was 5 mg/kg bw per day (Xia et al., 2023). Using a composite uncertainty factor (UF) of 3000 (10 for extrapolation from animals to humans, 10 for interindividual variation in human susceptibility, 10 for using a LOAEL instead of a NOAEL, 3 for subchronic rather than chronic duration of the studies) gives a tentative safe level of 6:2 FTOH in humans of **1.7 µg/kg bw per day**. However, this estimated safe level may indicate that 6:2 FTOH is not significantly more toxic than PFHxA, which had a RfD of (0.5 µg/kg bw per day) (US EPA, 2023), as claimed by Rice et al. (2020).

Risk assessment of 6:2 FTOH

The highest migration of 6:2 FTOH was 7.581 µg/kg food simulant from samples of a microwave oven popcorn package (**Reference no. 2024/102124**) (Table 1). If a person with a body weight of 60 kg eats 1 kg food packaged in FCM with this level of 6:2 FTOH per day, the exposure is **126.4 ng/kg bw per day** (Table 2), i.e. below (ca. 13 times) the tentative safe level. In two other products, muffin forms (**Reference no. 2024/86295**) and paper muffin forms (**Reference no. 2024/94002**), 6:2 FTOH was observed to migrate in lower levels than from the first product, with their highest levels of 2.970 and 2.005 µg/kg food simulant, respectively (Table 1). The exposure from these two muffin forms is **49.5 and 33.4 ng/kg bw per day**, i.e. below (ca. 34 and 51 times) the tentative safe level, respectively.

Conclusions on 6:2 FTOH

The exposure to 6:2 FTOH from the products muffin forms (Reference no. 2024/86295), paper muffin forms (Reference no. 2024/94002) and microwave oven popcorn package (Reference no. 2024/102124) in this study did not exceed the estimated tentative safe level based on experimental data from the literature, and thus, these products are likely to be without an appreciable risk of adverse effects from 6:2 FTOH.

Other approaches for risk assessment of PFAS with incomplete toxicity data

Use of the threshold of toxicological concern (TTC)

No HBGV has been established by well-known risk assessment organisations such as EFSA or US EPA for 6:2 FTOH and PFPeA. Another approach to evaluate PFAS with little available toxicity data is to use TTC. TTC is a pragmatic risk assessment tool, i.a. used for FCM, that is based on the principle of establishing a human exposure threshold value for chemicals, below which there is no appreciable risk to human health (Kroes et al., 2005; VKM, 2006). TTC can be used for substances that do not have sufficient toxicity data to perform a full risk assessment and that meet certain criteria (Kroes et al., 2005). There are several TTC values, depending on the structure and properties of the chemical under evaluation. Based on the available data, PFAS in general appear not to be genotoxic. Substances with structures that have a high potential for toxicity, such as halogen- (fluorine- in PFAS) containing substances, but are not genotoxic, called Cramer class III chemicals, have a TTC value of 90 µg/person per day or 1.5 µg/kg bw per day (**1500 ng/kg bw per day**) for a person with 60 kg body weight. Both PFPeA and 6:2 FTOH have exposure from the FCM in this study below this TTC value; the highest values being **0.18 ng/kg bw per day** and **126.4 ng/kg bw per day**, respectively (Table 2), indicating no appreciable risk for adverse health effects of these two PFAS based on TTC.

Conclusions when using TTC

Both PFPeA and 6:2 FTOH have exposures from the FCM in this study below the relevant TTC value, thus, the use of the TTC approach did not indicate an appreciable risk for adverse health effects of these two PFAS in any products.

Use of relative potency factors (RPF) versus PFOA

Another approach is to look at information about the potency of the PFAS with incomplete toxicity data sets and compare them to the potency of PFOA, for which there are established several HBGV (Table 3).

Bil et al. (2021) used RPF to compare the potency of PFAS based on increased relative liver weight in male rats exposed to PFAS for 42-90 days with PFOA. In this publication, PFOA was given a RPF of 1, PFBA of 0.05, PFHxA of 0.01, PFPeA was given a RPF of $0.01 \leq RPF \leq 0.05$ and 6:2 FTOH of 0.02. Thus, the RPF values for the lesser studied PFAS were all below the RPF for PFOA, indicating lower potency for toxic effects on the rat liver. However, these RPF do not take into account any other potential health effects of these PFAS.

Based on the newest TDI of **0.63 ng/kg bw** for PFOA (EFSA, 2020), the TDI values based on the relative potency of the other PFAS relative to PFOA would be **12.6, 63.0, 12.6-63.0 and 31.5 ng/kg bw per day** for PFBA, PFHxA, PFPeA and FTOH, respectively (Table 4). The exposure to 6:2 FTOH exceeds (4 times) the HBGV calculated based on its relative potency compared with

PFOA, whereas the exposures to PFBA, PFHxA and PFPeA do not exceed these values (Table 5. Appendix 1).

Table 4. Comparisons of exposure to PFAS with less toxicity data (Table 2) with the TDI calculated based on their potency relative to PFOA

PFAS substance	Highest exposure (ng/kg bw per day)	TDI relative to PFOA (ng/kg bw per day)	Exposure exceeds the TDI?
PFBA	2.5	12.6	No
PFHxA	1.5	63.0	No
PFPeA	0.18	12.6-63.0	No
6:2 FTOH	126.4	31.5	Yes

Conclusions when using the RPF approach

When using the RPF approach, the exposure to 6:2 FTOH in all three samples (both with the analytical uncertainty included or excluded) of the muffin forms (Reference no. 2024/86295) exceeded the TDI (up to about two times). The exposure to 6:2 FTOH in all three samples (both with the analytical uncertainty included or excluded) of the microwave oven popcorn package (Reference no. 2024/102124) exceeded the TDI (up to about five times). Thus, there is some concern about potential risk of adverse health effects from 6:2 FTOH in these two products.

For the paper muffin forms (Reference no. 2024/94002), exposure to the highest level of 6:2 FTOH in the three samples did only slightly exceed the TDI in one sample (B) with the analytical uncertainty excluded (33.4 ng/kg bw per day/31.5 ng/kg bw per day = 1.06). Thus, this product is likely to be without an appreciable risk of adverse effects from 6:2 FTOH based on RPF.

Answers to the ToR with conclusions on the individual products

The risk of adverse health effects in adults related to the migration of PFAS has been evaluated for each FCM product that had migration levels of one or more PFAS above the LOQ, taking into account the analytical uncertainty, based on comparisons of their exposure levels versus available HBGV from the literature, and using the TTC and/or the RPF approaches.

Paper straws (Reference no. 2024/85370)

The highest exposure to **PFOA** alone did slightly exceed the TDI for PFOA from EFSA (2020). The **sum of PFOA and PFHxS** also exceeded the corresponding TDI from the sum TWI from EFSA (2020). Thus, there is some concern about potential risk of adverse health effects from PFOA, and PFOA and PFHxS.

The exposure to **PFBA** from these paper straws did not exceed the available RfD from US EPA (2022b), and thus, the product is likely to be without an appreciable risk of adverse health effects from PFBA.

Conclusion: There is some concern about a potential risk of adverse health effects from PFOA, and PFOA and PFHxS, from these paper straws.

Paper straws (Reference no. 2024/87057)

The exposure to **PFOA** alone did not exceed the TDI for PFOA (EFSA, 2020). Thus, these paper straws are likely to be without an appreciable risk of adverse health effects from PFOA.

Conclusion: These paper straws are likely to be without an appreciable risk of adverse health effects from PFOA.

Straws (Reference no. 2024/93980)

The exposure to **PFOA** alone did not exceed the TDI for PFOA (EFSA, 2020). Thus, these straws are likely to be without an appreciable risk of adverse health effects from PFOA alone.

However, the exposure to the sum of **PFOA** and **PFHxS** exceeded the corresponding TDI from the sum TWI from EFSA (2020). Thus, there is some concern about potential risk of adverse health effects from PFOA and PFHxS.

Conclusion: There is some concern about a potential risk of adverse health effects from PFOA and PFHxS from these straws.

Paper plates (Reference no. 2024/092361)

The exposure to **PFBA** from paper plates did not exceed the available RfD from US EPA (2022b). Thus, these paper plates are likely to be without an appreciable risk of adverse health effects from PFBA.

The exposure to **PFPeA** from paper plates did not exceed the RfD from TCEQ (2023). Thus, these paper plates are likely to be without an appreciable risk of adverse health effects from PFPeA.

Conclusion: These paper plates are likely to be without an appreciable risk of adverse health effects from PFBA and PFPeA.

Muffin forms (Reference no. 2024/86295)

The exposure to **PFHxA** from these muffin forms did not exceed the available RfD from US EPA (2023), and thus, the product is likely to be without an appreciable risk of adverse health effects from PFHxA.

The exposure to **6:2 FTOH** from these muffin forms did not exceed the estimated tentative safe level based on experimental data from the literature (Xia et al., 2023). However, when using the RPF approach (Bil et al., 2021), the exposure to 6:2 FTOH in all three samples (both with the analytical uncertainty included or excluded) of the muffin forms exceeded the TDI (up to about two times). Thus, there is some concern about potential risk of adverse health effects from 6:2 FTOH in this product.

Conclusion: These muffin forms are likely to be without an appreciable risk of adverse health effects from PFHxA, but there is some concern for adverse health effects from 6:2 FTOH.

Paper muffin forms (Reference no. 2024/94002)

The exposure to **PFHxA** from these paper muffin forms did not exceed the available RfD from US EPA (2023). Thus, these muffin forms are likely to be without an appreciable risk of adverse health effects from PFHxA.

The exposure to **6:2 FTOH** from these paper muffin forms did not exceed the estimated tentative safe level based on experimental data from the literature (Xia et al., 2023). Thus, these paper muffin forms are likely to be without an appreciable risk of adverse health effects from 6:2 FTOH based on these data.

When using the RPF approach (Bil et al., 2021), the exposure to the highest level of 6:2 FTOH in the three samples did only slightly exceed the TDI in one sample (B) with the analytical uncertainty value excluded (33.4 ng/kg bw per day/31.5 ng/kg bw per day = 1.06). Thus, this product is likely to be without an appreciable risk of adverse effects from 6:2 FTOH based on RPF.

Conclusion: These paper muffin forms are likely to be without an appreciable risk of adverse health effects from PFHxA or 6:2 FTOH.

Microwave oven popcorn package (Reference no. 2024/102124)

The exposure to **6:2 FTOH** from the microwave oven popcorn package did not exceed the estimated tentative safe level based on experimental data from the literature (Xia et al., 2023). Thus, this popcorn package is likely to be without an appreciable risk of adverse health effects from 6:2 FTOH based on these data.

However, when using the RPF approach (Bil et al., 2021), the exposure to 6:2 FTOH in all three samples of the microwave oven popcorn package, both with the analytical uncertainty included or excluded, exceeded the TDI (up to about 5 times). Thus, there is some concern for adverse health effects from 6:2 FTOH in this microwave oven popcorn package based on the RPF approach.

Conclusion: There is some concern for adverse health effects from 6:2 FTOH in this microwave oven popcorn package.

When not stated specifically above, these conclusions are also valid if using migration levels of PFAS without subtracting the analytical uncertainty (Table 5, Appendix 1).

Uncertainty in these risk assessments

The toxicity data are more substantial for PFOA than for the other PFAS and it has been evaluated for adverse health effects to humans several times, both in Europe and in USA. The conclusions on PFOA are therefore considered more certain than for the other PFAS. PFBA and PFHxA have both recently been evaluated by US EPA. The conclusions on the potential risk from intake of PFBA and PFHxA therefore have less uncertainty than for PFPeA and 6:2 FTOH, for which there are less toxicity data available and no risk assessment based on all toxicity endpoints has yet been performed by a recognized risk assessment organisation internationally.

The RPF approach used (Bil et al., 2021) do not take into account any other potential health effects of these PFAS than effects on the liver based on experiment in rats.

Planned new risk assessments of PFAS

According to a member of the EFSA CONTAM Panel (personal communication), there are no known plans for immediate risk assessments of other PFAS in EFSA at present.

In view of the public health concerns of PFAS, the WHO initiated the development of a background document for the Guidelines for drinking-water quality on PFAS in drinking-water with a focus on PFOA and PFOS (WHO, 2023). The ongoing WHO assessment of PFAS started in 2017 and has now been expanded to consider sources of exposure beyond focusing principally on drinking-water. The review will also include further examination of the occurrence and health effects of additional substances beyond PFOA and PFOS, and a further examination of whether international HBGV can be established. The results of this WHO assessment are expected in 2026. On that basis, the European Commission will decide whether new limit values should be proposed through a targeted review of the Drinking Water Directive (DWD) (Directive (EU) 2020/2184). This updated WHO review of PFAS will also play a critical role in the planning of a future JECFA safety assessment of PFAS, which is scheduled to be undertaken in the coming years.

According to an article on fluorotelomer alcohols (FTOH) in water by an analytical company in USA (ALS, 2024), new European regulations are currently pending for 6:2 (and 8:2) FTOH, which are proposed for inclusion within a regulated sum of 24 PFAS of primary concern.

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Appendixes

Appendix 1

Table 5. Measured migration levels before and after subtraction of the analytical uncertainty for each PFAS (µg/kg food simulant)

Reference no.	PFAS	Sample A	Anal. uncert.	A – Anal. uncert.	Sample B	Anal. uncert.	B - Anal. uncert.	Sample C	Anal. uncert.	C - Anal. uncert.
2024/85370	PFOA	**0.072**	0.013	**0.059**	0.069	0.012	0.057	0.067	0.012	0.055
2024/85370	PFBA				0.048	0.034	0.015			
2024/86295	PFHxA	**0.152**	0.060	**0.092**	0.126	0.050	0.076	0.137	0.054	0.083
2024/86295	6:2 FTOH	3.610	0.640	2.970	2.529	0.450	2.079	2.773	0.490	2.283
2024/87057	PFOA	0.035	0.006	0.029	0.033	0.006	0.027	0.033	0.006	0.027
2024/93980	PFOA	0.034	0.006	0.028	0.034	0.006	0.028	0.037	0.007	0.030
2024/94002	PFHxA	0.091	0.036	0.055	0.083	0.033	0.050	0.085	0.034	0.051
2024/94002	6:2 FTOH	2.181	0.390	1.791	2.445	0.440	2.005	2.093	0.370	1.723
2024/092361	PFBA	**0.504**	0.352	**0.152**	0.472	0.330	0.142	0.454	0.317	0.137
2024/092361	PFPeA	**0.031**	0.020	**0.011**						
2024/102124	6:2 FTOH	8.946	1.610	7.336	**9.241**	1.660	**7.581**	8.896	1.600	7.296

The numbers in bold are the highest levels of migration measured in this study across the seven FCM products for each of the five PFAS.

Anal. uncert.: Analytical uncertainty.