National Institute for Public Health
and the Environment
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RIVM letter report 2020-0025
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Colophon

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Synopsis

Persistent organic pollutants in human milk in the Netherlands

Humans are exposed to so-called POPs (Persistent Organic Pollutants) via food and environment during their whole life-span. POPs are man-made substances that break down slowly, accumulate in the human body and are toxic. POPs accumulate in blood and fatty tissues and thus, are also found in human milk.

According to RIVM, the concentrations of POPs in human milk generally are low. There is no risk for babies when exposed to these substances via human milk. The results therefore do not give reason to stop breastfeeding.

RIVM reports these results, based on a survey on the presence of POPs in human milk. These results also show that concentrations of POPs in human milk have decreased during the last decades. This is because of international agreements, like the POP Stockholm treaty from 2001, which ban or restrict the use of POPs. Because they break down so slowly, POPs are still present in the environment. On average it takes five to ten years for the concentration in human milk to be reduced by half.

POPs are emitted via industry, during combustion processes, but can also be found in plant protection products (pesticides). Examples of POPs are dioxins, PCBs and plant protection products like DDT and heptachlor.

Since 2009, also PFOS was added to the POP list because of the undesirable properties of the substance. It is unknown whether the amount of PFOS in human milk also decreases. The substance has been measured for the first time in human milk in the Netherlands in 2014.

For their research, RIVM collected human milk samples in 2014, that were analysed between 2014 and 2016 by the World Health Organisation (WHO). The WHO has monitored POPs in human milk since 1969. Because of these measurements, developments in the concentration of POPs in human milk can be compared between countries and monitored during time. Concentrations of POPs in human milk of Dutch women are comparable to other Western-European countries.

Keywords: persistent organic pollutants, POP, WHO, human milk, PFOS, exposure, monitoring, risk assessment

Publiekssamenvatting

POP's in moedermelk van Nederlandse vrouwen

Mensen staan hun leven lang via voedsel en het milieu bloot aan kleine hoeveelheden van zogeheten POP's (Persistent Organic Pollutants/persistente organische verontreinigingen). Dit zijn door de mens gemaakte stoffen die heel langzaam afbreken, ophopen in het lichaam en giftig zijn. POP's hopen op in bloed en vetweefsel en komen zo ook in moedermelk terecht.

Volgens het RIVM gaat het in Nederland over het algemeen om lage concentraties POP's in moedermelk. Er zijn geen risico's voor baby's als ze via moedermelk aan deze stoffen blootstaan. Er is daarom geen aanleiding te stoppen met borstvoeding.

Ook blijkt dat de concentraties van POP's in moedermelk de afgelopen decennia lager zijn geworden. Dat komt door internationale afspraken, vooral het POP-verdrag van Stockholm uit 2001, om het gebruik van deze stoffen te verbieden of alleen onder strenge voorwaarden toe te staan. Doordat de stoffen zo langzaam afbreken, komen ze nog steeds voor in het milieu. De concentratie POP's in moedermelk is gemiddeld genomen pas na 5 tot 10 jaar gehalveerd.

POP's kunnen onder andere vrijkomen in de industrie, bij verbrandingen of kunnen in gewasbeschermingsmiddelen zitten. Voorbeelden van POP's zijn dioxines, PCB's en gewasbeschermingsmiddelen als DDT en heptachloor.

Sinds 2009 staat ook PFOS op de POP-lijst vanwege de ongewenste eigenschappen van deze stof. Het is niet bekend of de hoeveelheid PFOS in moedermelk afneemt. Deze stof is namelijk in 2014 voor het eerst in het meetprogramma opgenomen.

Het RIVM heeft voor dit onderzoek in 2014 monsters moedermelk verzameld, die tussen 2014 en 2016 door de Wereldgezondheidsorganisatie (WHO) zijn geanalyseerd. De WHO meet sinds 1969 wereldwijd POP's in moedermelk. Hierdoor kunnen ontwikkelingen in de concentratie van POP's in moedermelk tussen landen en door de jaren heen worden vergeleken. De concentraties van POP's in moedermelk van Nederlandse vrouwen zijn vergelijkbaar met die in andere Westerse landen.

Het RIVM beveelt aan om de concentraties van POP's in moedermelk de komende jaren te blijven meten. Dan kan ook de concentratie van stoffen die later aan de lijst POP's zijn toegevoegd, zoals PFOS en verwante verbindingen, in de gaten worden gehouden.

Kernwoorden: POP, persistente organische verontreinigingen, WHO, moedermelk, PFOS, blootstelling, monitoring, risicobeoordeling

Contents

Summary

The text in this summary is a collation of the summarizing texts in the blocks at the beginning of each section.

Persistent organic pollutants (POPs) are man-made substances that are toxic, degrade very slowly and accumulate in the environment and organisms. They easily distribute through the environment and may be found in remote areas like the polar region. POPs are emitted via industry, during combustion processes, but can also be found in plant protection products (pesticides). Examples of these substances are PCBs, dioxins, DDT and PFOS. International regulations, like the Stockholm Convention, restrict or ban the use and emissions of these POPs.

Human exposure to POPs is continuous, to small amounts via the environment and, ultimately, food. In mammals POPs tend to have strong bioaccumulating properties in lipid tissues, like breast tissue. Part of the POPs present in the human body thus ends up in human milk. The concentration in human milk reflects the amount of POPs accumulated over time in the mother's body (maternal body burden). The concentration in milk furthermore can be used to assess health risks due to the intake of POPs for babies and infants.

In order to evaluate the effectiveness of measures in the Stockholm convention, the United Nations (UNEP) and World Health Organisation (WHO) have regularly collected POP monitoring data in human milk. This enables the analysis of global trends in time of POP concentrations in human milk. The Netherlands participated in this survey in 2014 and several times before.

Material and methods

Logistics and collection of human milk samples in 2014

In 2014, human milk samples were collected from mothers that had given birth for the first time. These mothers were approached via Maternity Home Help Centres. Mothers were born and raised in the Netherlands. Milk was collected between six and ten days after childbirth. Of the 61 samples received, 50 were randomly selected and pooled into one sample for analysis. Chemical analysis was performed in the period 2014-2016 by WHO reference laboratories abroad, using validated techniques. For each substance, different detection limits were available. The UNEP/WHO study was aimed at obtaining concentrations of POPs in human milk per country. This then allows a comparison of the results of different countries and world regions. Because of this, a pooled sample per country was sufficient.

The participants in the 2014 study were representatives for all Dutch mothers. Samples were received from areas throughout the Netherlands, both in urban and rural environments. Also the age and Body Mass Index (BMI) of the mothers was representative. Characteristics of the study population in other countries were comparable.

Exposure assessment
Potential risks to new-borns and infants due to exposure to POPs via human milk were assessed by RIVM. As a first step, the daily exposure is calculated. To do so, intake amounts of POPs were calculated (expressed in nanograms per kilogram body weight of the child per day) using measured concentrations in human milk. This was performed for new-borns (with a body weight of 3 kg) and for one-year old infants (with a bodyweight of 10 kg). For both scenarios, a milk consumption of 800 mL per day was assumed.

Risk assessment
To obtain a first indication of risks, exposures were compared to a tolerable daily exposure level for humans, the so-called Health Based Guidance Value (HBGV). This value is considered to protect the whole population, including children and elderly, against health effects due to repeated exposure. For the POPs in this study, HBGVs used were published by the European Food Safety Authority (EFSA). For PFOS, the value published earlier by RIVM was used. In the first step these HBGVs were compared with the daily exposure resulting from breastfeeding. For dioxins, BDE-153 and PFOS, a more elaborated risk assessment had to be performed. In this second step, the modelled body burden was compared to the values underpinning the HBGV.

Trends during time and comparison with other countries
RIVM combined data from the current study with data from previous studies from 1969 to 2003. Although methods differed slightly over the years, these data can be used to assess trends in time for a number of POPs. For POPs with levels below detection limits, this was not possible. Besides this, no trends in time could be established for the PFAS substances (PFOS, PFOA), since they were only analysed in 2014. Another Dutch study, published in scientific literature in 2017 (by Čechová et al., 2017) could not be used for this analysis since their methods differed too much. Trends in time were also compared to similar data from other countries.

Results and Discussion

POP concentrations in Dutch human milk in 2014
The results of the pooled sample reflect the arithmetic mean value of the 50 individual samples. This means that part of the individual samples had a concentration above the reported pooled value, and the other part below. Chemical analysis showed that a number of POPs was present in Dutch human milk in 2014 (Table 1). This concerns substances from the 'Dirty Dozen' chemicals (the first POP list), but also substances that were more recently put on the POP list, like PFOS and PBDEs. Some additional substances that are formally not POPs were also analysed, like a number of PFAS substances. Most PFAS substances were not present in concentrations above detection limits.

Table 1. POP concentrations in a Dutch pooled human milk sample in 2014. Substances with concentrations below detection limits are not shown in the table. Bold exposure levels exceed the HBGV. For the group of dioxins/furans/dl-PCBs, see the main text.

POP	**Concentration (ng/g milk lipid; for PFOS and PFHxS in ng/L)**	**Exposure (ng/kg body weight/day)**		**HBGV (ng/kg body weight/day)**	**Source for HBGV**
		Newborns (3 kg)	**Infants (10 kg)**		
PFOS	45	**12**	3.6	6.25	Zeilmaker et al. (2016, 2018)
PFHxS[a]	11	2.9	0.9	20.8	Zeilmaker et al. (2016, 2018)
Chlordane (group) Oxy-chlordane Trans-nonachlor[d]	2.5 2.2	19.2	5.8	500	EFSA (2007a)
Dieldrin	2.1	16.8	5.0	100	EFSA (2005a)
DDT group p,p'-DDE p,p'-DDT	94.9 82.3 3.1	759	228	10,000	EFSA (2006a)
Heptachlor	2.2	17.6	5.3	100	EFSA (2007b)
Hexachlorobenzene	9.5	76	23	170	EFSA (2006b)
Toxaphene Parlar 26 Parlar 50 Parlar 62	2.4 0.6 0.9 0.8	19.2	5.8	Not available	
β-Hexachlorocyclohexane Beta-HCH	6.9 6.9	55.2	16.6	5,000[c]	EFSA (2005b)
PBDEs					
BDE-47	0.492	3.9	1.2	68.8	EFSA (2011a)
BDE-99	0.132	1.1	0.32	1.7	EFSA (2011a)
BDE-153	0.741	**5.9**	1.8	3.8	EFSA (2011a)
Remaining PBDEs[b]	0.227	1.8	0.5	Not available	
HBCDD Alpha-HBCDD	0.6 0.6	4.8	1.4	375	EFSA (2011b)

[a] Not listed as a POP in the Stockholm Convention, but also analysed in the 2014 survey

[b] BDEs -17, -28, -66, -100, -154

Risk assessment step 1: comparison of exposure with HBGV values

In the first step of the risk assessment, a worst case scenario is used to determine for which substances a risk can be excluded. For substances for which this is not the case, a more elaborated risk assessment is performed in step 2. When the intake of POPs via human milk was compared to the HBGV, for most POPs no risks are identified (Table 1). Only for BDE-153, PFOS, and the combined group dioxins/furans/dl-PCBs a more elaborated risk assessment was needed in step 2.

Risk assessment step 2: elaborated risk assessment

Following international risk assessment protocols, in a second step a more elaborate risk assessment was performed for BDE-153, PFOS, and the combined group dioxins/furans/dl-PCBs. The HBGVs for these substances, which were also used in step 1, were based on the bioaccumulating properties of these substances during the life-span of humans. In this step 2 exposure scenario, the bioaccumulation in the body of the neonate during breastfeeding and during the rest of its life was modelled. The modelled accumulation was then compared to bioaccumulated amounts that reflect the effect values underpinning the HBGV. The model simulations showed that dioxins, BDE-153 and PFOS in Dutch human milk do not pose a health risk for new-borns and infants. In the case of PFOS, the risk assessment may have to be updated when EFSA publishes revised HBGV values mid 2020.

Uncertainties in the risk assessment

As this assessment is based on a pooled milk sample, for part of the individual samples the POP levels may have been higher. This means that for some individual children, tolerable levels for the dioxin group may have been exceeded in 2014. It is important, however, to point here also at other uncertainties surrounding the present risk assessment. These uncertainties may lead to both under- or overestimation of the risk. On the one hand, worst-case assumptions were applied that may overestimate the risk. This concerned the duration of the period in which children exclusively received human milk, and the amount of milk consumed per day. Furthermore, samples were taken immediately after birth while it is known that concentrations decrease during the breastfeeding period. On the other hand, risk may have been underestimated because for PFOS and BDE-153 no other exposure routes were taken into account (e.g., other food sources, house dust). Furthermore, possible mixture toxicity was also not included in the risk assessment, except for the dioxin group.

Comparison with other countries and trends in time

Trends in time in Dutch human milk could be assessed for the dioxins, PCBs, p,p'-DDE (metabolite of DDT), HCB, β-HCH and PBDE. For all other substances, not enough data above detection limits were available. For PFAS-substances like PFOS and PFOA, no trends could be established since these substances were only analysed in the last monitoring campaign in 2014.

The results (see Figure 1 as an example) clearly show that POP concentrations in human milk decrease in time. The concentrations of these substances are reduced by 50% every 5 to 10 years.

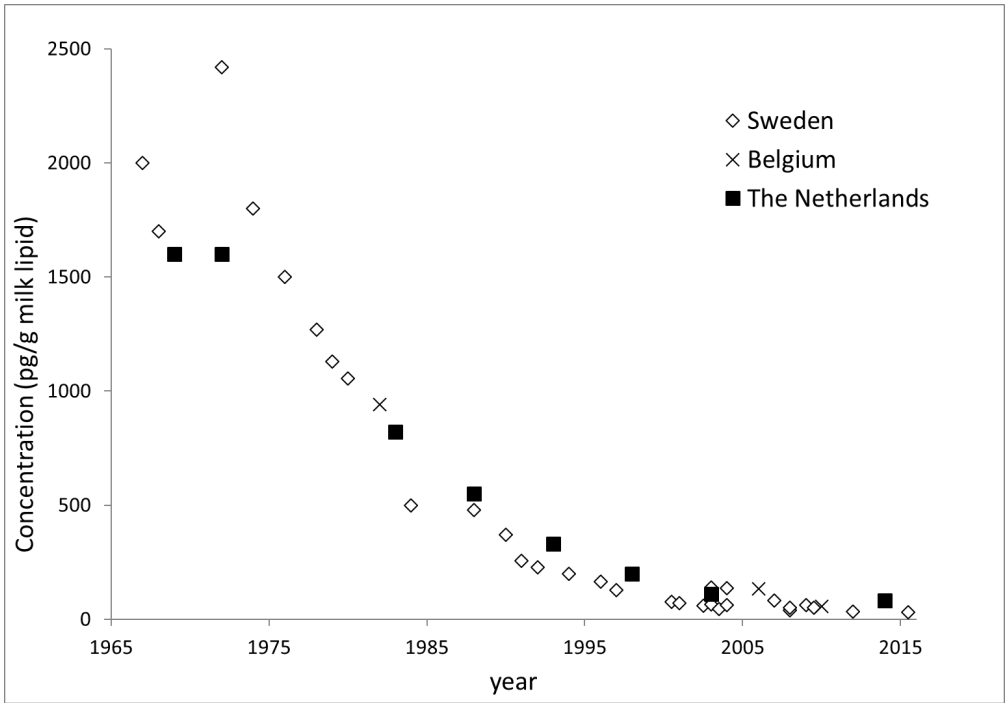


Figure 1. Trend in time for p,p'-DDE in Sweden, Belgium and The Netherlands. Comparable trends are observed for the other substances.

Considering the substances that were detected, the Netherlands showed an intermediate position amongst the neighbouring countries (Figure 1). Some variability in POP patterns is observed, which may be explained by different national regulations, production, use and consumption patterns.

Conclusions

The concentration of POPs in human milk is a good proxy for the occurrence of these substances in our food and the environment. Measurements from 2014 show that 14 of the 23 POPs are present in human milk, but in very low concentrations.

For almost all substances a risk for children that receive breastfeeding can be excluded already in the first, worst-case, step of the assessment. Exceptions were BDE-153, PFOS, and the combined group dioxins/furans/dl-PCBs. Thus, an elaborated, more precise, risk assessment was performed for these substances. Calculated concentrations in the body of the neonate or infant during breastfeeding and during the rest of his life show that POPs in Dutch human milk do not pose a health risk to new-borns and infants. This is based on the pooled sample. The results therefore do not give reason to stop breastfeeding.

The results show that POP concentrations in human milk decrease by 50% every 5-10 years. This shows that exposure of breast-fed babies and infants to these substances is substantially decreased after the substances are incorporated as POPs in the Stockholm convention. Although the decrease is noted, it takes decades before the concentrations of some of these substances fall below the detection

limits in human milk. For PFOS, no trends could be established since these substances were only analysed in the last monitoring campaign. The POP concentrations, including PFOS, in the Netherlands were comparable to those in neighbouring countries.

Recommendations
Various Dutch and international studies have indicated that beneficial long term health effects of breastfeeding outweigh the negative effects of POPs in human milk. The information in the present study is no reason to draw other conclusions.

For PFOS, the risk assessment was based on a HBGV that was derived by RIVM. After RIVM published this value, EFSA has published a provisional, more critical HBGV. However, in 2018 RIVM expressed concerns regarding the method used by EFSA to derive a provisional HBGV value. The latter value is currently under revision. The current risk assessment may have to be revised when EFSA has published their revised HBGV value (mid 2020).

RIVM recommends to continue the monitoring on a regular basis, especially for POPs that have been added to the POP-list more recently. This way, it can be monitored whether their concentrations also decrease because of measures taken by the Stockholm Convention.

The current measurements were based on a pooled sample of 50 individual human milk samples. When more information at a national scale is necessary, for instance on variability between samples, it is recommended to analyse separate samples in future monitoring campaigns.

Samenvatting

De tekst in deze samenvatting komt overeen met de (Engelstalige) samenvattende teksten in de kaders aan het begin van ieder onderdeel van het rapport.

Persistente organische verontreinigingen (POPs; Persistent Organic Pollutants) zijn door de mens gemaakte giftige stoffen die heel langzaam afbreken en in het milieu en organismen opstapelen. Ze kunnen zich gemakkelijk verspreiden waardoor ze worden gevonden in gebieden ver weg van de plek waar ze zijn gebruikt of gemaakt, zoals de poolregio. POP's komen onder andere vrij bij toepassingen in de industrie en bij verbranding, maar ze zaten ook in gewasbeschermingsmiddelen. Voorbeelden van deze stoffen zijn PCB's, dioxines, DDT en PFOS. Internationale wet- en regelgeving, zoals het VN Verdrag van Stockholm uit 2001, zorgt ervoor dat het gebruik en de uitstoot van deze stoffen wordt gereguleerd of verboden.

Mensen staan continu, maar in kleine hoeveelheden, bloot aan POP's. Dat gebeurt via het (leef)milieu en uiteindelijk ook via voedsel. In mensen en andere zoogdieren stapelen POP's zich sterk op in vetweefsel, zoals borstweefsel. Een deel van de in het lichaam aanwezige POP's komt zo in moedermelk terecht. Daarom geeft de moedermelk de hoeveelheid POP's weer die de moeder gedurende haar leven heeft opgenomen (body burden). De concentratie in moedermelk kan ook worden gebruikt om gezondheidsrisico's voor zuigelingen in te schatten van POP's die ze via moedermelk binnenkrijgen.

Om de effectiviteit van de maatregelen van het Verdrag van Stockholm te meten, meten de VN (UNEP) en de Wereldgezondheidsorganisatie (WHO) sinds 1969 geregeld de hoeveelheid POP's in moedermelk. Daardoor kan wereldwijd worden gevolgd hoe de concentraties van deze stoffen in moedermelk zich ontwikkelen. Nederland heeft in 2014 met dit onderzoek meegedaan, net als een aantal eerdere keren.

Materiaal en methoden
Logistiek en verzameling
In 2014 heeft het RIVM monsters van moedermelk verzameld van moeders die voor de eerste keer zijn bevallen. Deze moeders zijn via kraamcentra benaderd. De moeders waren geboren en opgegroeid in Nederland. De melk is 6 tot 10 dagen na de geboorte verzameld. Van de 61 ontvangen monsters zijn er *at random* 50 bij elkaar gevoegd zodat ze als één monster zijn geanalyseerd. Referentielaboratoria van de WHO in het buitenland hebben deze chemische analyse tussen 2014 en 2016 uitgevoerd, met gevalideerde technieken. Voor elke stof gelden andere detectielimieten. Het doel van het UNEP/WHO-onderzoek is om een beeld van de concentraties POP's in moedermelk per land te krijgen. Hiermee kunnen de resultaten van verschillende landen en regio's in de wereld met elkaar worden vergeleken.

De deelnemers aan het onderzoek van 2014 waren representatief voor alle Nederlandse moeders. Monsters zijn ontvangen uit verschillende

gebieden in Nederland, zowel uit stedelijke als plattelandsgebieden. Ook de leeftijd en de Body Mass Index (BMI) van de moeders was gemiddeld. De moeders van de studies uit de andere landen hadden vergelijkbare eigenschappen.

Beoordeling van de blootstelling

Het RIVM heeft de potentiële risico's voor zuigelingen door blootstelling aan POP's via moedermelk beoordeeld. Als eerste stap is daarvoor de dagelijkse blootstelling berekend. Hiervoor zijn ingenomen hoeveelheden POP's berekend (uitgedrukt als nanogram per kilogram lichaamsgewicht van het kind per dag) met behulp van de gemeten concentraties in moedermelk. Dat is gedaan voor pasgeborenen (met een lichaamsgewicht van 3 kilogram) en zuigelingen van 1 jaar (met een lichaamsgewicht van 10 kilogram). Bij beide scenario's is ervan uitgegaan dat zij 800 milliliter melk per dag drinken.

Risicobeoordeling

Om een eerste indicatie van risico's te krijgen, is de berekende blootstelling vergeleken met het aanvaardbaar niveau voor de dagelijkse blootstelling van mensen, de zogenoemde Health Based Guidance Value (HBGV). Deze waarde beschermt de hele bevolking, inclusief kinderen en ouderen, tegen gezondheidseffecten wanneer zij geregeld aan deze stoffen blootstaan. Voor deze studie zijn HBGV's van EFSA gebruikt om de risico's van POP's te beoordelen. Voor PFOS is de waarde van het RIVM gebruikt. In de eerste stap zijn deze HBGV's vergeleken met de dagelijkse blootstelling via borstvoeding. Voor dioxines, BDE-153 en PFOS was een uitgebreidere risicobeoordeling nodig. In deze tweede stap is de berekende concentratie in het lichaam vergeleken met de waarden waarop de HBGV is gebaseerd.

Tijdreeksen en vergelijking met andere landen

Het RIVM heeft de data van deze studie vergeleken met data van eerdere WHO-studies van 1969 tot 2003. Hoewel de gebruikte analytische methoden licht van elkaar afweken, konden de data worden gebruikt om de ontwikkelingen van een aantal POP's door de tijd te analyseren. Dit kon alleen voor POP's die in moedermelk in aantoonbare concentraties aanwezig waren. Voor PFAS-stoffen (onder meer PFOS en PFOA) konden geen tijdreeksen worden geanalyseerd, aangezien deze stoffen alleen in 2014 zijn geanalyseerd. Een Nederlandse wetenschappelijk studie uit 2017 (Čechová et al. 2017) kon voor deze analyse niet worden gebruikt omdat hun methode te veel afweek van de huidige methode. Tijdreeksen zijn ook vergeleken met andere Europese landen.

Resultaten en discussie

POP concentraties in Nederlandse moedermelk in 2014

De uitkomst van de 50 samengevoegde monsters geeft de 'rekenkundig gemiddelde waarde' aan van alle afzonderlijke monsters. Dat betekent dat van een deel van de afzonderlijke monsters de concentratie hoger ligt dan de geometrische gemiddelde waarden en van het andere deel de concentratie lager ligt. De analyses toonden aan dat een aantal POP's in Nederlandse moedermelk zitten (Tabel 1). Dit zijn stoffen van de 'Dirty Dozen'-lijst (de eerste POP-lijst), maar ook stoffen die pas later aan de lijst zijn toegevoegd, zoals PFOS en PBDE's. Sommige stoffen die

formeel niet op de POP-lijst staan, zoals een aantal PFAS-stoffen, zijn ook geanalyseerd. De concentraties van de meeste van deze stoffen kwamen echter niet boven de detectielimiet.

Tabel 2. POP-concentraties in een Nederlands samengevoegd monster van moedermelk uit 2014. Stoffen met concentraties beneden de detectielimiet zijn niet in de tabel opgenomen. Dit betreft α- Hexachloorcyclohexaan, γ-Hexachloorcyclohexaan (lindaan), Chlordecone, Hexabroombiphenyl, Pentachloorbenzeen en endosulfan, SCCPs, en de PFAS-stoffen PFPeA, PFHxA, PFHpA, PFOA, PFNA, PFDA, PFUnDA, PFBuS, en PFDS. Vetgedrukte waardes overschrijden de HBGV, en voor deze stoffen kan een risico niet worden uitgesloten in deze eerste stap van de risicobeoordeling. Voor de dioxine-groep, zie de hoofdtekst.

POP	**Concentratie (ng/g melkvet; voor PFOS en PFHxS in ng/L)**	**Blootstelling (ng/kg lichaamsgewicht/dag)**		**HBGV (ng/kg lich.gewicht /dag)**	**Bron voor HBGV**
		Pasgeborenen (3 kg)	**1-jaar oud (10 kg)**		
PFOS	45	**12**	3,6	6,25	Zeilmaker et al. (2016, 2018)
PFHxS[a]	11	2,9	0,9	20,8	Zeilmaker et al. (2016, 2018)
Chlordaan (groep) Oxy-chlordaan Trans-nonachloor[d]	2,4 2,4 2,2	19,2	5,8	500	EFSA (2007a)
Dieldrin	2,1	16,8	5,0	100	EFSA (2005a)
DDT groep p,p'-DDE p,p'-DDT	94,9 82,3 3,1	759	228	10.000	EFSA (2006a)
Heptachloor	2,2	17,6	5,3	100	EFSA (2007b)
Hexachlorobenzeen	9.5	76	23	170	EFSA (2006b)
Toxapheen Parlar 26 Parlar 50 Parlar 62	2,4 0,6 0,9 0,8	19,2	5,8	Niet beschikbaar	
β-Hexachlorocyclohexaan Beta-HCH	6,9 6,9	55,2	16,6	5.000[c]	EFSA (2005b)
PBDEs					
BDE-47	0,492	3,9	1,2	68,8	EFSA (2011a)

POP	Concentratie (ng/g melkvet; voor PFOS en PFHxS in ng/L)	Blootstelling (ng/kg lichaamsgewicht/dag)		HBGV (ng/kg lich.gewicht /dag)	Bron voor HBGV
		Pasgeborenen (3 kg)	1-jaar oud (10 kg)		
BDE-99	0,132	1,1	0,32	1,7	EFSA (2011a)
BDE-153	0,741	**5,9**	1,8	3,8	EFSA (2011a)
Overige PBDEs[b]	0,227	1,8	0,5	Niet beschikbaar	
HBCDD Alpha-HBCDD	0,6 0,6	4,8	1,4	375	EFSA (2011b)

[a] Niet op de POP-lijst van het Verdrag van Stockholm, maar wel geanalyseerd in 2014.
[b] BDEs -17, -28, -66, -100, -154

Risicobeoordeling stap 1: vergelijking blootstelling met HBGV-waarden
In de eerste stap van de risicobeoordeling wordt vanuit een worst-case scenario bepaald voor welke stoffen een risico kan worden uitgesloten. Voor stoffen waarvoor dit niet geldt, is een uitgebreidere risicobeoordeling in stap 2 nodig. Wanneer de hoeveelheid die pasgeborenen en 1-jarigen via moedermelk binnenkrijgen, in stap 1 wordt vergeleken met de HBGV, dan blijkt er voor de meeste POP's geen risico te zijn (Tabel 2). Alleen voor BDE-153, PFOS, en de gecombineerde groep dioxines/furanen/dl-PCBs was een uitgebreidere risicobeoordeling nodig in stap 2.

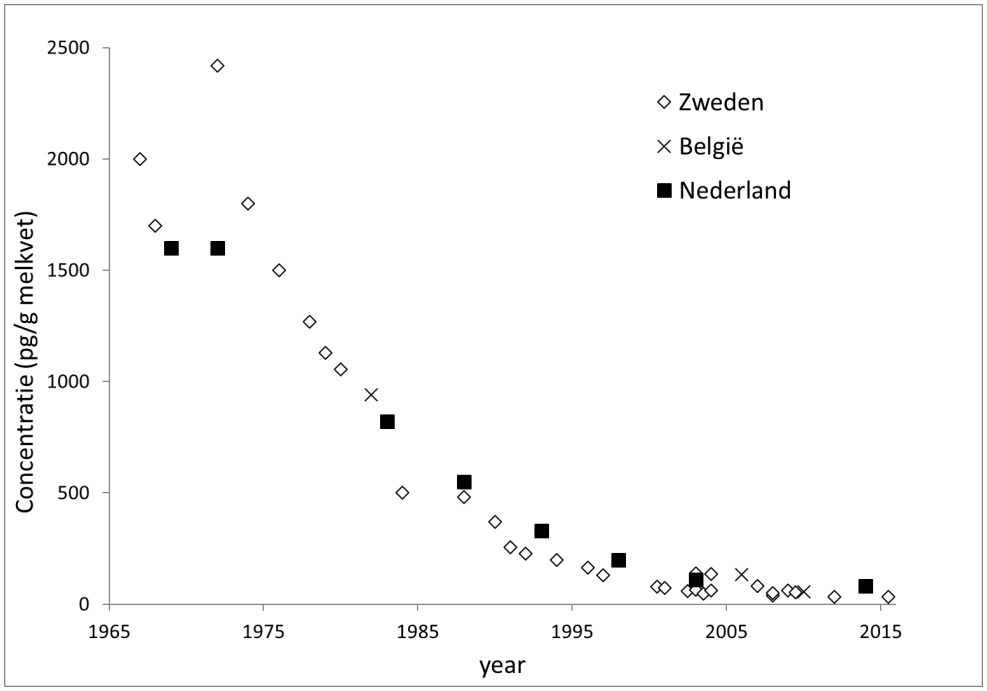
Risicobeoordeling stap 2: uitgebreide risicobeoordeling
Overeenkomstig met internationale protocollen voor risicobeoordelingen is voor BDE-153, PFOS, en de gecombineerde groep dioxines/furanen/dl-PCBs als tweede stap een uitgebreide risicobeoordeling uitgevoerd. De HBGVs, die ook in stap 1 zijn gebruikt voor deze stoffen gaan ervan uit dat POP's zich gedurende het leven van de mens ophopen in het lichaam. Dit heet bioaccumulatie. Daarom is in stap 2 gemodelleerd hoe groot deze accumulatie in het lichaam van de zuigeling in die periode en gedurende de rest van zijn leven is. Deze hoeveelheid in het lichaam is vervolgens vergeleken met het niveau dat verwacht wordt na blootstelling aan de HBGV. De modelresultaten laten zien dat dioxines, BDE-153 als PFOS in Nederlandse moedermelk geen gezondheidsrisico vormen voor zuigelingen. Voor PFOS kan het nodig zijn de beoordeling aan te passen als EFSA de herziene HBGV-waarden publiceert. Deze worden halverwege 2020 verwacht.

Onzekerheden in de risicobeoordeling
Omdat deze beoordeling is gebaseerd op een samengesteld monster, is het mogelijk dat het risico voor een deel van de individuele monsters hoger ligt. Dit betekent dat voor sommige individuele kinderen, in 2014 het acceptabele niveau van dioxines in moedermelk kan zijn overschreden. Het is belangrijk om te wijzen op onzekerheden. Deze kunnen leiden tot een overschatting van het risico, maar ook tot een onderschatting hiervan. Aan de ene kant is met worst-case aannames gewerkt die het berekende risico hoger kunnen laten uitvallen. Dat

betrof de duur van de periode waarin kinderen alleen borstvoeding kregen en de hoeveelheid melk die zij per dag dronken. Daarnaast zijn de monsters vlak na de geboorte genomen terwijl de concentraties POP's steeds lager worden in de periode dat de moeder borstvoeding geeft. Aan de andere kant kunnen de risico's ook zijn onderschat. Zo zijn voor PFOS en BDE-153 geen andere blootstellingsroutes meegenomen (zoals andere voedselbronnen en huisstof).
Verder is mogelijke mengseltoxiciteit ook buiten beschouwing gelaten, behalve voor de dioxines.

Vergelijking met andere landen en tijdreeksen
Tijdreeksen in Nederlandse moedermelk konden worden geanalyseerd voor dioxines, PCBs, p,p'-DDE (een afbraakstof van DDT), HCB, β-HCH en PBDE. Voor alle andere stoffen waren niet genoeg data boven de detectielimiet beschikbaar. Voor PFOS kon geen tijdreeks worden gemaakt omdat deze stof alleen in 2014 is gemeten.

De resultaten (zie Figuur 2 als voorbeeld) laten duidelijk zijn dat POP-concentraties in moedermelk gedurende de tijd afnemen. De concentratie van deze stoffen neemt iedere 5 tot 10 jaar met 50 procent af.



Figuur 2. Tijdreeks voor p,p'-DDE in moedermelk in Zweden, België en Nederland. Vergelijkbare ontwikkelingen zijn gezien bij de andere stoffen.

De concentratie van de aangetroffen stoffen in Nederland was vergelijkbaar met die in andere landen (zie Figuur 2). De waargenomen variaties kunnen worden verklaard door verschillen in nationale wet- en regelgeving, productie, gebruik en eetgewoonten.

Conclusies
De aanwezigheid van POP's in moedermelk geeft de mate aan waarin mensen aan deze stoffen blootstaan via voedsel en de leefomgeving.

Metingen uit 2014 laten zien dat 14 van de 23 POP's in moedermelk zitten, in zeer lage concentraties.

Van bijna alle stoffen kan een risico voor zuigelingen al in de eerste worst-case stap van de risicobeoordeling worden uitgesloten. Een uitzondering zijn BDE-153, PFOS en de gecombineerde groep dioxines/furanen/dl-PCBs. Voor deze stoffen is daarom een nauwkeurigere, uitgebreidere risicobeoordeling uitgevoerd. De berekende concentratie van deze stoffen in het lichaam van de zuigeling in die periode en gedurende de rest van zijn leven laat zien dat de POPs in Nederlandse borstvoeding geen gezondheidsrisico vormen voor zuigelingen. Dit is gebaseerd op de metingen in het mengmonster. De gemeten waardes geven geen aanleiding te stoppen met het geven van borstvoeding.

Uit resultaten blijkt duidelijk dat POP-concentraties in moedermelk elke 5 tot 10 jaar met 50 procent afnemen. Hieruit blijkt dat de blootstelling van zuigelingen aan deze stoffen flink is afgenomen nadat de stoffen zijn gereguleerd via het Verdrag van Stockholm. Maar doordat de stoffen langzaam afbreken in het milieu, zitten ze, lang nadat maatregelen zijn genomen, toch nog in moedermelk.

Voor PFOS kon geen tijdreeks worden gemaakt omdat deze stof alleen in 2014 is gemeten. De concentraties van POP's, inclusief PFOS, in Nederlandse moedermelk waren vergelijkbaar met de concentraties in ons omringende landen.

Aanbevelingen
Verschillende Nederlandse en internationale studies hebben laten zien dat de langdurige voordelen voor de gezondheid van borstvoeding opwegen tegen de mogelijke negatieve effecten vanwege de aanwezigheid van POP's. De resultaten van de huidige studie geven geen aanwijzingen om deze conclusies te veranderen. Moeders wordt aangeraden om de richtlijnen van de kraamcentra te volgen.

Voor PFOS was de risicobeoordeling gebaseerd op de HBGV die het RIVM heeft bepaald. Sindsdien heeft de EFSA een voorlopige, strengere waarde gepubliceerd. In 2018 heeft het RIVM bezwaren geuit tegen de methode die is gebruikt om deze waarde voor PFOS af te leiden. Momenteel wordt EFSA's HBGV herzien. De risicobeoordeling moet wellicht worden aangepast na publicatie van EFSA's herziene HBGV-waarde (halverwege 2020).

Het RIVM beveelt aan om de hoeveelheid POP's in moedermelk regelmatig te monitoren. Dit is vooral van belang voor de POPs die onlangs op de POP-lijst zijn gezet. Hiervan kan dan worden gevolgd of de concentraties ook afnemen door maatregelen onder het Verdrag van Stockholm.

De huidige metingen zijn gebaseerd op een samengevoegd monster van 50 individuele monsters. Als meer informatie op nationaal niveau gewenst is, bijvoorbeeld over de variatie tussen de monsters, dan kan het nodig zijn om in de toekomst ook de POPs in individuele monsters te meten.

1 Introduction

Persistent organic pollutants (POPs) are man-made substances that are toxic, degrade very slowly and accumulate in the environment and organisms. They easily distribute through the environment and may be found in remote areas like the polar region. POPs are emitted via industry, during combustion processes, but can also be found in plant protection products (pesticides). Examples of these substances are PCBs, dioxins, DDT and PFOS. International regulations, like the Stockholm Convention, restrict or ban the use and emissions of these POPs.

Human exposure to POPs is continuous, to small amounts via the environment and, ultimately, food. In mammals POPs tend to have strong bioaccumulating properties in lipid tissues, like breast tissue. Part of the POPs present in the human body thus ends up in human milk. The concentration in human milk reflects the amount of POPs accumulated over time in the mother's body (maternal body burden). The concentration in milk furthermore can be used to assess health risks due to the intake of POPs for babies and infants.

In order to evaluate the effectiveness of measures in the Stockholm convention, the United Nations (UNEP) and World Health Organisation (WHO) have regularly collected POP monitoring data in human milk. This enables the analysis of global trends in time of POP concentrations in human milk. The Netherlands participated in this survey in 2014 and several times before.

Persistent organic pollutants (POPs) are man-made toxic and persistent substances that have the potential for long-range transport. This means they do not degrade very fast and thus accumulate in the environment, and that they may be found in remote areas like the polar region. Therefore, the effects of POPs on the environment and human health are complex. Once it has been determined that a substance meets the POP criteria, its use will be banned or restricted. The use and emissions of many POPs has already been restricted or banned since the 1970s via various national and international policies (Bruinen de Bruin and Janssen, 2012). More recently, POPs have been regulated on a global scale by the Stockholm Convention through prohibiting their production and use (UNEP, 2016a).

When the Convention came into force in 2004, twelve persistent organic pollutants, the so-called Dirty Dozen[1], were included. Since then, additional substances have been added to the Convention[2]. Thus, most

[1] The original "Dirty Dozen" under the Stockholm Convention were:
Pesticides: aldrin, chlordane, DDT, dieldrin, endrin, heptachlor, mirex, toxaphene, hexachlorobenzene
Industrial chemicals: hexachlorobenzene, polychlorinated biphenyls (PCBs);
Industrial by-products: hexachlorobenzene; polychlorinated dibenzo-p-dioxins and polychlorinated dibenzofurans (PCDD/PCDF), and polychlorinated biphenyls (PCBs).

[2] α-, β- and γ-hexachlorocyclohexane (= lindane), hexabromobiphenyl, pentachlorobenzene, chlordecone, endosulfan, hexabromocyclododecane (HBCDD), bromodiphenylethers (PBDEs, commercial mixtures of penta- and octa-bromodiphenylethers; in the measurments present as tetrabromodiphenyl ether and

POPs are organic substances which were widely used as pesticides or industrial chemicals or which were formed unintentionally as a by-product, and all of them have been detected in environmental compartments all over the world. Although regulation through the Stockholm Convention prevents a further increase of environmental concentrations, POPs already distributed in the environment may still pose a risk to humans and environment through several exposure routes. All POPs are listed on the website of the Stockholm Convention[3].

Human milk is considered to be an appropriate and non-invasive matrix to get an impression of the exposure of the mother, i.e. the adult general population, as well as the infants. Human milk has a high lipid content and this provides an ideal partitioning matrix for POPs, which in majority are lipophilic. However, also in case of amphoteric substances like perfluor carboxylic acid or sulfonic acids human milk provides a suitable biomonitoring matrix. Because of this, the POP concentration in human milk reflects the POP amount in the maternal body ("body burden") due to previous (long-term) exposure, the latter reflecting environmental concentrations. Additionally, using these concentrations the potential risks of POPs in human milk may be assessed.

The World Health Organisation (WHO) has collected national POPs monitoring data every four to five years since 1976. Since 1987, the WHO has also coordinated monitoring the presence of a number of POPs in human milk. Since 2007 this work is carried out in close cooperation with the United Nations Environment Programme (UNEP) (WHO, 2007). UNEP decided to monitor the effectiveness of measures in the Stockholm Convention by sampling human milk in different regions of the world on a regular basis.

In the Netherlands, human milk samples were collected for national projects on POPs in 1969 and 1972/73, and for the 1st, 2nd and 3rd WHO coordinated human milk programme in 1987-1989, 1992-1993 and 2000-2003 (UNEP, 2009). The present study reports on the results of the Dutch milk samples for the 6th WHO/UNEP global POP survey (2013-2014) and the corresponding risk assessment. The 2014 monitoring results were further analysed with respect to previous Dutch data as published in Tuinstra, 1971; Greve, 1974; Greve, 1985; Liem et al., 1989; Greve and van Zoonen, 1990; Albers et al., 1993; Cuijpers et al., 1997; Zeilmaker et al., 2002; Zeilmaker, 2004. This allowed for the establishment of trends in time of POPs in human milk in the Netherlands. Finally, the trends were compared with the ones in comparable countries (Belgium, Germany, Sweden).

Although only PFOS is listed in the Convention as a POP, a number of PFAS-substances (PFPeA, PFHxA, PFHpA, PFOA, PFNA, PFDA, PFUnDA, PFBuS, PFDS) were also measured in the milk samples. PFOA has been added to the Convention in 2019.

pentabromodiphenyl ether and hexabromodiphenyl ether and heptabromodiphenyl ether respectively), perfluorooctane sulfonic acid (PFOS) were added up to 2014 when sampling took place. Since then hexachlorobutadiene, short-chain chlorinated paraffins, polychlorinated naphthalenes, pentachlorophenol, and decaBDE have been added. Just before 2020 PFOA and dicofol have been added to the Convention.

[3] http://www.pops.int/TheConvention/ThePOPs/AllPOPs/tabid/2509/Default.aspx.

2 Material and methods

2.1 Collection and analysis of human milk samples in 2014

In 2014, human milk samples were collected from mothers that had given birth for the first time. These mothers were approached via Maternity Home Help Centres. Mothers were born and raised in the Netherlands. Milk was collected between six and ten days after childbirth. Of the 61 samples received, 50 were randomly selected and pooled into one sample for analysis. Chemical analysis was performed in the period 2014-2016 by WHO reference laboratories abroad, using validated techniques. For each substance, different detection limits were available. The UNEP/WHO study was aimed at obtaining concentrations of POPs in human milk per country. This then allows a comparison of the results of different countries and world regions. Because of this, a pooled sample per country was sufficient.

The participants in the 2014 study were representatives for all Dutch mothers. Samples were received from areas throughout the Netherlands, both in urban and rural environments. Also the age and Body Mass Index (BMI) of the mothers was representative. Characteristics of the study population in other countries were comparable.

2.1.1 Logistics and collection of human milk samples

Collection, storage and transport of the samples were carried out under the flag of the WHO/UNEP mother milk census, which also organised the chemical analysis of the various substances. Coordination, logistics and collection of individual samples were organised by the National Institute of Public Health and the Environment (RIVM, Bilthoven, the Netherlands). Collection of the milk samples mainly followed the WHO guidelines (WHO, 2007). In line with the earlier Dutch sampling strategy, collection deviated from the WHO guidelines on the following three points:

a. Because of the age distribution of Dutch primipari mothers (giving birth for the first time) no upper age limit was set for collecting the human milk.
b. Milk was collected between six and ten days after childbirth, instead of between three and eight weeks.
c. To guarantee that the samples reflected historic Dutch exposure circumstances, mothers should have been born and raised in the Netherlands.

In the period January – April 2014 about 40-50 Maternity Home Help Centres (Dutch: Kraamcentra) throughout the Netherlands were asked whether they were willing to select a number of mothers fulfilling the above described criteria. Based on the previous 2003 POP survey in the Netherlands a response of at least 90% of the Maternity Home Help Centres was expected The mothers were approached through these centres to ask whether they were willing to cooperate. Samples were collected in the period of 1 to 30 May 2014. The WHO guidelines request a total of 50 samples of individual mothers of at least 50 mL milk each.

The sampling strategy aimed to collect, nationwide, 100 individual milk samples of 100 mL each, anticipating a less than 100% response.

Mothers received a package through the Maternity Centres containing information on the survey, sample vials, a breast pump and instructions on milk collection, storage and port-free shipping. All glassware for the collection of the individual samples and the preparation of the pooled sample was provided by the WHO reference laboratory, the Chemisches und Veterinäruntersuchungsamt Freiburg (CuVF), Freiburg, Germany.

Milk was collected by the individual mothers and sent to RIKILT (Wageningen University). After sampling, the sample was stored in the refrigerator at about 4 °C for a maximum of 72 hours. To prevent bacterial deterioration, the human milk was preserved by addition of potassium dichromate. Each individual sample and the corresponding questionnaire was labelled with a unique identification code.

Of the approached 40 – 50 centres, eleven participated in the collection of milk samples, to actually result in sixty-two samples from nine centres to be received, of which one was of a second born child This sample was discarded. Of the remaining 61 samples, 50 samples were randomly selected for further analysis. Of these samples, 25 mL was taken and pooled, resulting in a pooled sample of 50 x 25 mL = 1250 mL. The pooled sample was prepared at the end of June 2014. The centres were located in various parts of the Netherlands (e.g. Limburg, Zeeland, Drenthe, Zuid-Holland, Noord-Holland, Flevoland, Friesland).

As the 2014 POP concentration was determined in a pooled milk sample its value reflects the (arithmetic) mean of the concentration in 50 samples, with unknown variability in the Dutch population being unknown, i.e. half of the samples will have had concentrations below this value, and the other half above.

2.1.2 Study participants

Nine of the Maternity Home Help Centres that were approached reacted positively. These centres were located throughout the Netherlands, namely in Amsterdam, Gulpen, Dronten, Eindhoven, Woerden, Assen, Groningen, Venlo and Joure. Overall, the women participating in the milk surveys were representative for Dutch mothers having their first child (~30 years old, see Table 3).

Table 3. Demographic data from the Dutch women of whom human milk was used in the different human milk surveys.

year	**# of participants**	**age (years)**	**BMI (kg/m^2)**	**pregnancies**	**collection period**	**reference**
1969	50	18-32	not reported	not reported	Day 6	Tuinstra, 1971
1972	76	not reported	not reported	not reported	Week 1-7	Greve, 1974
1983	275	27.2±3.5	not reported	1^{st}-5^{th}	Day 8-12	Greve et al., 1985, Greve 1985
1988	329	30.0±3.8	24±3	1^{st}-5^{th}	Day 6-10	Albers et al., 1993
1993	103	29.3	22.2	1^{st}	Day 6-10	Cuijpers et al., 1997
1998	102	28.3±3.1	25.1±3.0	1^{st}	Day 6-10	Zeilmaker et al., 2002
2003	100	29	25.9	1^{st}	Day 6-10	Zeilmaker, 2004
2014	50	30.9 ± 4.0	23.5 ± 3.7	1^{st}	Day 6-10	Current report

The Body Mass Index (BMI) of the women participating in the different studies has been comparable between the different studies and therefore the POP concentrations in the different studies can be compared directly.

The mean age of women to have their first child in Belgium, Germany and Sweden is comparable to the age in the Netherlands (OECD, 2019). Thus, results from these countries may be compared to the Netherlands. The average age in the Netherlands of women to have their first child has been increasing since the 1960's (CBS, 2019). The average age was 24 years in 1970 and has increased to 29.4 in 2014. This influences the amount of time the women have been exposed to POPs and thus may be reflected in the POP concentrations in human milk.

2.1.3 Chemical analysis

The pooled sample was analysed for POPs and Short-Chain Chlorinated Paraffins (SCCPs) by the European Union Reference Laboratory for halogenated persistent organic pollutants in feed and food located at the Chemisches und Veterinäruntersuchungsamt (CuVF) Freiburg, Germany and for PFAS-substances by Örebro University (Örebro, Sweden). Those institutes are the WHO reference laboratories for POP analysis. The methods applied for the analysis of dioxins/furans, PCBs and organochlorine pesticides are described in Van Leeuwen and Malisch (2002), WHO (2007) and UNEP (2013). The method used for the PFAS-substances has been validated and described earlier by Kärrman et al. (2007)(see Annex 1). The complete chemical analysis was finalised in 2016. The detection limits of all measurements in 2014 are provided in Annexes 1 and 2.

2.2 Exposure assessment

Potential risks to new-borns and infants due to exposure to POPs via human milk were assessed by RIVM. As a first step, the daily exposure is calculated. To do so, intake amounts of POPs were calculated (expressed in nanograms per kilogram body weight of the child per day) using measured concentrations in human milk. This was performed for new-borns (with a body weight of 3 kg) and for one-year old infants (with a bodyweight of 10 kg). For both scenarios, a milk consumption of 800 mL per day was assumed.

2.2.1 Calculation of amount consumed

To determine the exposure of breast-fed new-borns and infants to POPs, measured milk concentrations were used to calculate intake amounts (in µg per kg body weight per day or ng per kg body weight per day).

The intake amount was calculated as the product of the measured POP concentration in the pooled sample (in amount per g milk lipid), a daily milk lipid consumption of 24 g (equivalent to 800 mL human milk [De Winter et al., 2003; EFSA, 2011a]), containing 3 % milk fat (measured value), divided by a body weight of 3 kg at birth, or 10 kg at 1 year of age, (growth curves 0-1 year old boys and girls were obtained from TNO, 2019), see also Annex 4).

2.2.2 *TEF-approach for dioxins: calculation of TEQs*

Dioxins are a group of substances. The risk assessment for dioxins and dioxin-like substances followed a different approach than for the other substances. The dioxin exposure was calculated for the mixture of PCDDs (dioxins), PCDFs (furans) and dl-PCBs (dioxin-like PCB congeners). Of this mixture, 2378-TCDD (a PCDD) is the most toxic substance. Thus, this substance was assigned a toxic potency score of 1. Accordingly, other components of the mixture were assigned a score relative to 1, depending on their toxicity. This is the Toxic Equivalency Factor (TEF; the TEF of 2378-TCDD thus is 1). The exposure to the total mixture of dioxins/furans/dl-PCBs then was reported as the *total*, i.e. PCDD/PCDF/dl-PCB, Toxicity Equivalence (TEQ). This is the weighed sum of the concentrations of all individual PCDD/PCDF/dl-PCB substances in the mixture with their corresponding TEF.

Measurements were transformed to WHO-TEQ_{2005}-values using the calculation method provided in Van den Berg et al. (2006). For TEQ values developed earlier, and known as WHO-TEQ_{1998}, original measurement data were converted into WHO-TEQ_{2005} values.

When comparing trends in time, sometimes TEQs are derived for a specified subset of substances. However, in the context of risk assessment only the total TEQ is of relevance. The reason for this is that the total TEQ reflects the exposure expressed in terms of TCDD equivalents, in pg TEQ/kg body weight /day and therefore may directly be compared to the Health Based Guidance Value (HBGV) (see section 2.3.2) which is determined for 2378-TCDD.

2.2.3 *Kinetic modelling of body burdens*

In general, for the oral route of exposure, chemical risk assessment consists of comparing the exposure to the HBGV, i.e. the life-long, daily, exposure level without toxic risk. When the exposure remains below the HBGV, toxic risk is negligible. However, when exposure exceeds the HBGV a risk cannot be excluded and the risk assessment proceeds to a more elaborated assessment. When appropriate, for this elaborated assessment bioaccumulation is modelled using a kinetic model. This then feeds into the risk assessment (see section 2.3).

In concordance with international risk assessment procedures a more elaborated risk assessment based on bioaccumulation was performed for BDE-153, PFOS, and the combined group PCDD/PCDF/dl-PCBs. This so-called body burden approach is the default risk assessment procedure for the dioxin TEQ, PBDEs and perfluoro substances like PFOA and PFOS (WHO (2000), ATSDR (2015), EFSA (2011; 2018), US EPA (2016a,b), SCF (2000,2001), US EPA (2012)).

Relevant HBGVs for PBDEs, the perfluoro substances PFOA and PFOS and the dioxin 2378-TCDD all have been derived with the aid of a one-compartmental modeling approach (EFSA, 2011a; ATSDR, 2015; Zeilmaker et al*.,* 2016; EFSA, 2018a). In this report, one-compartment models were applied to calculate the body burden over time (for technical details, see Annex 4). In the model approach a body weight of 6.5 kg was taken as the average body with during the first year of life.

As, internationally, for BDE-153 the risk assessment is based on the accumulation in the body, i.e. the whole-body concentration, the risk assessment of this substance was based on homogenous distribution over the body. This modelling needs two model parameters: the fraction of the administered chemical which is absorbed from the gastro-intestinal tract and the rate constant for the removal of the chemical from the body. This approach was applied for BDE-153 using parameters of EFSA (2011). For the simulation of the whole-body kinetics of BDE-153, the daily intake was calculated based on the measured BDE-153 concentration of 0.74 ng/g milk lipid (see section 3.1). This resulted in a daily BDE-153 intake of 2.7 ng/kg body weight/day (for details, see Annex 4).

Internationally the risk assessment of PFOS and dioxins is based on the concentration of these substances in the blood. For this reason the risk assessment of these substance was based on non-homogenous distribution over the body. In this case, whole body kinetics are modelled using one additional model parameter, i.e. the so-called Volume of Distribution (V_d). This is used to relate the whole body concentration to the serum concentration (PFOS) or the serum lipid concentration (dioxins). For the simulation of the PFOS kinetics, model parameters from Olsen et al. (2007), Zeilmaker et al. (2016) and ATSDR (2015) were used. For PFOS the daily intake was calculated using the measured PFOS concentration of 45 ng/L (section 3.1). This resulted in a daily PFOS intake of 5.5 ng/kg body weight/day (for details, see Annex 4).

Furthermore, for dioxins, the risk assessment is based on the concentration in the body lipid or serum lipid (Van der Molen et al., 1996; Kreuzer et al., 1997; US EPA, 2012; Béchaux et al. 2014; EFSA, 2018a). Modelling for the risk assessment of the combined group of dioxins/furans/dl-PCBs focused on 2378-TCDD, using the measured data for the whole group. The toxicokinetic modeling for this substance may be performed for serum lipid of neonates (Kreuzer et al., 1997) or adults (Carrier et al., 1995a,b; Van der Molen et al. 1996; 2000). In the case of adults, the effect of the age-dependent increase in amount of body lipid on the concentration in this body lipids is then taken into account, the so-called "apparent elimination by dilution" effect (Carrier et al., 1995a,b; Van der Molen et al., 1998, 2000). However, when modelling the kinetics in neonates neither Kreuzer et al., (1997) nor EFSA (2018, using a modified Carrier model) incorporated this "apparent elimination by dilution", thus overestimating the 2378-TCDD concentration in body lipid and the corresponding toxic risk for neonates and probably also for infants. In addition, Kreuzer et al. (1997) and Van der Molen et al. (1998, 2000) illustrated the importance of faecal lipid excretion as route of elimination. For the risk assessment in this report, the modelling approaches of Carrier, Kreuzer and Van der Molen were combined (see Annex 4). Human kinetics of 2378-TCDD were thus described with a (deterministic) one-compartmental model incorporating elimination in the liver, elimination through faeces and the "apparent elimination by dilution" effect. For the simulation of the kinetics of the dioxin/furans/dl-PCB group as PCDD/PCDF/dl-PCB TEQ kinetics (section 2.2.2), the daily intake was calculated using the measured PCDD/PCDF/dl-PCB TEQ concentration of 7.2 ng TEQ/g milk lipid (section 3.1). This resulted in a

daily PCDD/PCDF/dl-PCB TEQ intake of 26.6 pg TEQ/kg body weight/day for an average child (for details, see Annex 4).

2.3 Risk Assessment

To obtain a first indication of risks, exposure concentrations were compared to a tolerable daily exposure level for humans, the so-called Health Based Guidance Value (HBGV). This value is considered to protect the whole population, including children and elderly, against health effects due to repeated exposure. For the POPs in this study, HBGVs used were published by the European Food Safety Authority (EFSA). For PFOS, the value published earlier by RIVM was used. In the first step these HBGVs were compared with the daily exposure resulting from breastfeeding. For dioxins, BDE-153 and PFOS, a more elaborated risk assessment had to be performed. In this second step, the modelled body burden was compared to the values underpinning the HBGV.

2.3.1 General principles

To assess risks of new-borns and infants due to exposure to individual POPs, the exposure via human milk (as amount consumed per kg body weight per day) was compared to an exposure level which is considered to be a tolerable level. For humans this is usually the HBGV.

The current report focusses on exposure via human milk only. Of course, after the breastfeeding period, exposure will also take place via food and the environment (e.g. house dust), and for PFOS also via drinking water. The scope of the current assessment is on breast-fed children only and not on the mothers nursing these children. Because children are at the most sensitive life-stage, the risk assessment for children should also protect the mothers.

Except for dioxins (see section 2.3.3), a tolerable human exposure level reflecting the exposure via breastfeeding is not available. Therefore, for all POPs other than dioxins the exposure via breastfeeding was compared to the HBGV. The HBGV protects against toxic effects that may be caused by acute, sub-acute exposure (during a relatively short period of time) or semi-chronic or chronic exposure (during long periods of time or even life-long). So, in case where a relative short exposure like the one resulting from breastfeeding is below the HBGV, an appreciable risk can be excluded for any type of toxicity. However, in case the exposure exceeds the HBGV an appreciable risk cannot be excluded and a more elaborated risk assessment is needed.

Basically, the risk assessment thus consists of two steps:

1. A risk assessment performed based on a direct comparison of the exposure with the HBGV (Step1). When the exposure exceeds the HBGV a more elaborated risk assessment was performed (Step 2, see below).
2. In case POPs bioaccumulate, the derivation of the HBGV explicitly takes this process into account (“body burden approach”). For such chemicals kinetic modelling can be applied to determine the extent and duration by which a temporary exposure exceeding the HBGV affects long-term bioaccumulation and, hence, toxic risk (as revealed by exposure at the level of the HBGV). For this

reason a body burden risk assessment approach was performed for POPs which temporarily exceeded the HBGV during a one-year breast feeding period, i.e. the dioxin TEQ, the PBDE BDE-153 and the perfluoro substance PFOS. Currently this body burden approach is the default risk assessment procedure for substances which show large inter-species differences in bioaccumulation, like the dioxin TEQ, PBDEs and perfluoro substances like PFOA and PFOS (WHO (2000), ATSDR (2015), EFSA (2011; 2018), US EPA (2016a,b), SCF (2000,2001), US EPA (2012)). See section 2.2.3 for an explanation of the modelling approach used.

2.3.2 HBGV values

For the comparison of the exposure with the Health Based Guidance Value (HBGV), HBGVs as published by EFSA (see Table 4 in section 3.1) were used for all substances except PFOS.

For PFOS the HBGV derived by RIVM (Zeilmaker et al., 2018) is used. In 2016, RIVM derived a HBGV for PFOA (12.5 ng/kg body weight/day) based on hepatic toxicity, i.e. increased liver weight, in rodents (Zeilmaker et al., 2016). The HBGV was derived using an interspecies extrapolation to make the rodent value applicable to humans. Besides this, RIVM has derived "Relative Potency Factors" (RPFs) for several PFASs, including PFOS. These RPFs scale the potency of individual PFAS congeners relative to PFOA, with PFOA having an RPF of 1 (comparable to the TEF approach as explained in section 2.2.2, but in this case only based on liver toxicity). Using this method, the RPF of PFOS was 2, indicating a two-fold higher potency of PFOS relative to PFOA to induce liver toxicity (Zeilmaker et al., 2018). This RPF can then be used to derive a HBGV for PFOS, based on the HBGV for PFOA of 12.5 ng/kg body weight/day). The HBGV for PFOS, reflecting the two-fold higher potency, is 12.5/2 = 6.25 ng/kg body weight/day. Similarly, PFHxS showed an RPF of 0.6, corresponding with a HBGV of 12.5/0.6 = 20.8 ng/kg body weight/day.

Since RIVM derived the HBGV for PFOA in 2016 (Zeilmaker et al., 2016) and PFOS in 2018 (Zeilmaker et al., 2018), the ATSDR (Agency for Toxic Substances and Disease Registry) and EFSA (European Food Safety Authority) have derived substantial lower HBGVs for PFOA and PFOS (ATSDR, 2018; EFSA, 2018b).

EFSA has derived a provisional HBGV of 0.9 ng/kg body weight/day for PFOA and 1.9 ng/kg body weight/day for PFOS, using epidemiological data, i.e. increased serum cholesterol in adults, (EFSA, 2018b). RIVM and some other scientific institutes in Europe questioned the scientific basis of the health-based guidance value published by EFSA (EFSA, 2018c). Given the discussions and the provisional nature of the EFSA conclusions, RIVM at the moment does not use the EFSA HBGVs for PFOA and PFOS for the risk assessment. Currently EFSA conducts a risk assessment for exposure to other perfluorinated substances, including a possible re-evaluation of the HBGV of PFOA and PFOS. The EFSA opinion is expected to be available mid-2020.

ATSDR derived Minimum Risk Levels (MRLs) for "intermediate exposure" (15–364 days) of 3 ng PFOA/kg body weight/day and 2 ng PFOS/kg body weight based on developmental toxicity induced in a two-generation experiment with rats (ATSDR, 2018).

Compared with the HBGV used in this risk assessment both ATSDR and EFSA have thus derived substantially lower safe exposure levels for PFOA and PFOS. This reflects the current scientific uncertainty in the HBGVs for PFOA and PFOS. If new information on HBGVs becomes available RIVM will evaluate the need to update its PFOA and PFOS risk assessments. This also applies to the current assessment of health risks for neonates and infants due to uptake of PFOA and PFOS via human milk.

2.3.3 Specific approach for dioxins

For dioxins, the TEF approach was used. Besides this, dioxins are the only (group of) POPs for which an HBGV is available that was derived taking exposure via breastfeeding into account. This HBGV (0.25 pg PCDD/PCDF/dl-PCB TEQ/kg body weight/day) was derived under condition that new-borns exclusively received breastfeeding from birth up to the age of one year (EFSA, 2018a).

EFSA based the HBGV on a serum level of 7 pg PCDD/PCDF/g serum lipid to be reached at the age of 9 years, i.e. the age at which disturbed spermatogenesis at adult age is induced. Dl-PCBs were excluded from the analysis, which may lead to an underestimation of the HBGV. The serum concentration was modelled from birth up to 9 years, leading to this value of 7 pg/g serum lipid. The HBGV resulted from this modelling exercise and is the value that exactly causes this exposure level. EFSA's model assumed a daily TEQ intake during 1 year of breastfeeding of 165 pg TEQ (based on 800 mL milk/day, 3.5% milk fat, 5.9 pg TEQ/g milk lipid). After breastfeeding, in order to compensate for the relatively high dietary intake of infants compared to adults, up to the age of nine years, infants received a dietary exposure equal to two times the HBGV.

Thus, the EFSA approach implies that the value of 165 pg TEQ per day is acceptable exposure level for human milk. To relate this to body weight, for a neonate this amounts to 165/3 = 55 pg TEQ/kg body weight/day, while for an infant of one year this amounts to 165/10 = 17 TEQ/kg body weight/day. An appreciable risk of the exposure to dioxins in human milk thus is negligible when this exposure is lower than the 55 pg TEQ/kg body weight at birth and 17 pg TEQ/kg body weight at one year of age.

2.4 Trends during time and comparison with other countries

RIVM combined data from the current study with data from previous studies from 1969 to 2003. Although methods differed slightly over the years, these data can be used to assess trends in time for a number of POPs. For POPs with levels below detection limits, this was not possible. Besides this, no trends in time could be established for the PFAS substances (PFOS, PFOA), since they were only analysed in 2014. Another Dutch study, published in scientific literature in 2017 (by Čechová et al., 2017) could not be used for this analysis since their methods differed too much. Trends in time were also compared to similar data from other countries.

2.4.1 Data availability

To assess the trend in time, data from all Dutch studies carried out previously were combined with the new data. In the national studies from 1969 to 1988, no restriction regarding the number of pregnancies were set (see Table 3), but both mother and child should be apparently healthy, including a normal pregnancy. The study population in 1993, 1998 and 2000/2003 included only primipari women (pregnant of their 1st child) living in the Netherlands for >5 years. In 2014, only primipari women born and raised in the Netherlands were included. No age limit was set in any of the Dutch milk surveys. The samples in 1993, 1998 and 2000/2003 were analysed individually.

Data provided for POPs in human milk in the Netherlands in Čechová et al. (2017) were not used in this analysis, since these samples were taken in various years and could not be assigned to a specific year for trend analysis. Besides this, they consisted of samples from women that did not give birth for the first time, which makes it difficult to compare them to the other studies after 1993.

Women were requested to collect between 50 and 100 mL of human milk in the different surveys. During the surveys in 1969, 1972 and 1983, human milk was collected at ~Day 6, Week 1-7, and Day 8-12. Since 1987, human milk was collected during Day 6 to Day 10 after delivery. In the survey of 1972, women were requested to collect one sample of 50-100 mL per week from week 1 to 7, in order to determine the trend during lactation. As this did not reveal significant differences in the POP concentration between the various weekly samples, the average of all weekly samples was considered representative for the POP concentration in human milk in 1972 (Greve, 1974).

In the case of trend in time analyses, not only the total TEQ (dioxins+furans+dl-PCBs) was used, but also its individual components, all reflected in sub-TEQs. Using these sub-TEQ only serves a practical purpose, i.e. to assess whether the trend in time of dioxins (PCDDs), furans (PCDFs) or dioxin like-PCB (dl-PCB) sub-TEQs deviates from that of the total TEQ.

2.4.2 Chemical analysis

The milk samples of the Dutch national surveys were analysed by different national analytical laboratories. During the years, different

methods were applied. Data on the methodology are summarised in Annex 3.

Different analytical techniques have been used to determine the POP concentrations in human milk in the period from 1969 to 2014. In the national surveys, when analytical techniques were used for the first time, samples of human milk from previous studies were also re-analysed in order to determine if the new technique would lead to differences in POP concentrations. This was not the case.

2.4.3 Calculation of trends in time

Data which can be used to establish trends in time were only available for p,p'-DDE, hexachlorobenzene (HCB), β-HCH, dioxins and furans (PCDD/PCDF), dioxin-like-PCBs (dl-PCBs) and indicator PCBs (I-PCBs). Median concentrations of these data were used when available. As the decrease in concentrations is not linear but exponential, data were log-transformed. To estimate half-lives for each substance, the log-transformed data were fitted linearly in Excel using the least squares optimisation method.

Measurements were transformed to WHO-TEQ-values for dioxins and furans (PCDD/PCDF), dioxin-like-PCBs (dl-PCBs) and total TEQ using the calculation method provided in Van den Berg et al (2006), as described in section 2.2.2. For TEQ values developed earlier, originally known as WHO-TEQ_{1998}, original data were used to calculate WHO-TEQ_{2005} values.

2.4.4 Comparison with other countries

The concentration of several POPs in Dutch human milk for which time-trends were available were compared with those in human milk from Belgium, Germany and Sweden, where available. For Sweden comparable long-term data are available. Most recent data for these countries were available for the period 2009 to 2012. For Belgium data from Heyndrickx & Maes (1969), Atuma & Vaz (1986), Colles et al (2008) and Croes et al (2012) were used for DDE, HCB and β-HCH. For Germany data from BFR (2000) were used for HCB and β-HCH and from Vieth et al (2011) for PCDD/DF, dl-PCBs and total TEQ. As explained in section 2.4.3, data by Vieth et al (2011) expressed in TEQ_{1998} were transformed to TEQ_{2005} values for comparison. The transfer factors used were 0.88, 0.66 and 0.74 respectively and were estimated using German human milk data from 2000 and 2005 from the German dioxin database (2018) and from Raab et al (2008) respectively. The Swedish data for DDE, HCB and β-HCH were retrieved from Noren and Meironyte (2000), Lignell et al (2005), Lignell et al (2009b), Lignell et al (2014), Lignell et al (2016) and Gyllanhammar et al 2017. The Swedish data for PCDD/DF, dl-PCBs and total TEQ were retrieved from Fång et al., 2013. As for the Netherlands only one PFOS sample was available (the 2014 measurement), no trend in time could be established for this substance.

3 Results and discussion

3.1 POP concentrations in Dutch human milk in 2014

The results of the pooled sample reflect the arithmetic mean value of the 50 individual samples. This means that part of the individual samples had a concentration above the reported pooled value, and the other part below. Chemical analysis showed that a number of POPs was present in Dutch human milk in 2014. This concerns substances from the 'Dirty Dozen' chemicals (the first POP list), but also substances that were more recently put on the POP list, like PFOS and PBDEs. Some additional substances that are formally not POPs were also analysed, like a number of PFAS substances. Most PFAS substances were not present in concentrations above detection limits.

Of the 'Dirty Dozen' chemicals (see Chapter 1), only aldrin, endrin and mirex were not detected in the pooled human milk sample in 2014, the other nine substances were detected (see Table 4). Of the new POPs, β-hexachlorocyclohexane, tetra-, penta-, hexa- and hepta BDEs, trans-nonachlor, PFOS and HBCDD were detected, while α-hexachlorocyclohexane, γ-hexachlorocyclohexane (lindane), chlordecone, hexabromobiphenyl, pentachlorobenzene and endosulfan were not detected. Furthermore, some substances that were not included in the Stockholm POP convention were measured as well. Of these, SCCPs and the PFAS substances PFPeA, PFHxA, PFHpA, PFOA, PFNA, PFDA, PFUnDA, PFBuS, and PFDS were not detected, whereas PFHxS was detectable.

Results for the substances that were reported in the study by Čechová et al. (2017), namely pp-DDE, pp-DDT, HCB and indicator PCBs, were considerably lower than reported here. Concentrations for the first three were 35-50% lower, for indicator-PCBs only 3%. The fact that 60% of the mothers which donated milk in the Čechová et al. study delivered their second or third baby may have been the cause of the lower concentrations. The locations of collection in the Čechová et al. study (Zwolle, Purmerend and Den Helder) do not provide an obvious reason for the difference in concentration compared to the locations in this study. The measurements of γ-HCH and dieldrin could not be compared as the results in the current study were below the detection limit; the concentration of mirex in Čechová et al. (2017) was considerably higher than the detection limit in our study.

Table 4. POP concentrations in a Dutch pooled human milk sample in 2014. Substances with concentrations below detection limits are not shown in the table. Exposure levels were calculated as explained in section 2.2.1. Bold exposure levels exceed the HBGV. For the group of dioxins/furans/dl-PCBs, see the main text.

POP	**Concentration (ng/g milk lipid; for PFOS and PFHxS in ng/L)**	**Exposure (ng/kg body weight/day)**		**HBGV (ng/kg body weight/day)**	**Source for HBGV**
		New-borns (3 kg)	**Infants (10 kg)**		
PFOS	45	**12**	3.6	6.25	Zeilmaker et al. (2016, 2018)
PFHxS[a]	11	2.9	0.9	20.8	Zeilmaker et al. (2016, 2018)
Chlordane (group) Oxy-chlordane Trans-nonachlor[d]	2.4 2.4 2.2	19.2	5.8	500	EFSA (2007a)
Dieldrin	2.1	16.8	5.0	100	EFSA (2005a)
DDT group p,p'-DDE p,p'-DDT	94.9 82.3 3.1	759	228	10,000	EFSA (2006a)
Heptachlor	2.2	17.6	5.3	100	EFSA (2007b)
Hexachlorobenzene	9.5	76	23	170	EFSA (2006b)
Toxaphene Parlar 26 Parlar 50 Parlar 62	2.4 0.6 0.9 0.8	19.2	5.8	Not available	
β-Hexachlorocyclohexane Beta-HCH	6.9 6.9	55.2	16.6	5000	EFSA (2005b)
PBDEs					
BDE-47	0.492	3.9	1.2	68.8	EFSA (2011a)
BDE-99	0.132	1.1	0.32	1.7	EFSA (2011a)
BDE-153	0.741	**5.9**	1.8	3.8	EFSA (2011a)
Remaining PBDEs[b]	0.227	1.8	0.5	Not available	
HBCDD Alpha-HBCDD	0.6 0.6	4.8	1.4	375	EFSA (2011b)

[a] Not listed as a POP in the Stockholm Convention, but also analysed in the 2014 survey
[b] BDEs -17, -28, -66, -100, -154

Regarding the occurrence of dioxins, furans and dl-PCBs, 7 PCDD congeners, 10 PCDF congeners and 8 mono-ortho and 4 non-ortho dl-PCB congeners were detected (for occurrence data of individual congeners, see Annex 2). These data corresponded to a total TEQ of 7.2 pg TEQ/g milk lipid and sub-TEQs of 4.5 pg PCDD/PCDF TEQ/g milk lipid and 2.7 pg dl-PCB TEQ/g milk lipid. For more explanation on the TEQ method, see section 2.2.2.

3.2 Risk assessment

3.2.1 Risk Assessment Step 1: comparison of exposure with HBGV values

In the first step of the risk assessment, a worst case scenario is used to determine for which substances a risk can be excluded. For substances for which this is not the case, a more elaborated risk assessment is performed in step 2. When the intake of POPs via human milk was compared to the HBGV, for most POPs no risks are identified (Table 1). Only for BDE-153, PFOS, and the combined group dioxins/furans/dl-PCBs a more elaborated risk assessment was needed in step 2.

Table 4 shows the calculated daily exposure and the effect levels (HBGV) for the POPs present in human milk above detection limits. The exposure levels (in ng/kg body weight per day) for neonates are higher than those for infants, because the former are based on a body weight of 3 kg and the exposure levels of infants are based on a body weight of 10 kg. The apparent decrease in the exposure levels is thus based on intake levels that stay the same (800 mL milk with constant concentrations and fat content) and body weights that increase.

In the case of BDE-153 and PFOS the exposure to these substances via human milk showed a temporary exceedance of the HBGV in neonates, but not in one-year olds. For dioxins, a slightly different approach was followed because an HBGV was available which also takes exposure via breastfeeding into account. The measured value of 7.2 pg TEQ/g milk lipid corresponds to an exposure level of 58 TEQ/ kg body weight/day at birth and 17 TEQ/kg body weight/day at one year of age. These exposures equal the dioxin breastfeeding exposure EFSA used when deriving the HBGV for dioxins. Whether this exposure leads to bioaccumulation indicative for a toxic risk, i.e. leads to exceedance of EFSA's toxicity benchmark (7 pg TEQ/g serum lipid at age of 9 years) is addressed in step 2 of the risk assessment.

The exposure assessment is based on several assumptions which influence the outcomes of the risk assessment. First, it is based on the POP concentration from a pooled sample. This means that the exposure of part of the breast-fed children may have been higher. On the other hand, the exposure assessment is based on exclusive breastfeeding during the first year, no other food sources and a daily human milk intake of 800 mL. In the Netherlands the median milk intake increases from 670 mL/day immediately after birth to 840 mL/day after 6 months, to decline to 400 mL/day after 12 months (De Winter et al., 2003).

Peeters et al. (2015) reported the percentage of Dutch neonates which exclusively receive breastfeeding in the period between birth and 6 months of age. At birth this percentage amounts to 80%, but this

quickly drops to 58% at the age of 1 month. In the period between 1 and 6 months a linear weekly decrease of 0.9% was observed, resulting in 39% of the neonates receiving exclusive breastfeeding for a period up to 6 months (95% CI: 33-45%). Extrapolating the linear decrease to 12 months of age, i.e. assuming exclusive breastfeeding to continue, resulted in an expected percentage of 12% (95% CI: 1-21%). From this it can be concluded that in the Netherlands few children exclusively receive breastfeeding for a period up to one year. The applied breastfeeding period of maximally one year therefore is to be considered a reasonable "worst case" duration.

Finally, the POP concentration in human milk was assumed constant throughout the breastfeeding period, whereas in reality it slightly decreases (Ulaszewska et al., 2011).

Another factor of uncertainty in the risk assessment is the choice of HBGV values. For this report, the values proposed by EFSA were used (see section 2.3.2), except for PFOS. For PFOS, RIVM derived a HBGV in 2016 but since then, EFSA and ATSDR also published new safe exposure levels. When these EFSA and ATSDR values would be used for the risk assessment instead of the value that RIVM derived, the calculated risk would be higher. However, these values are still discussed internationally and EFSA is currently working on a revision of its proposed value. Thus, these values were not used for the current risk assessment. However, the risk assessment for PFOS may have to be updated after EFSA publishes the outcome of the revision (see section 2.3.2).

Here it should also be noted that step 1 of the risk assessment provides the internationally accepted default approach for evaluating the exposure from POPs via breastfeeding. This approach deviates from the PFAS mixture approach as recently applied in Nyberg et al. (2018) and the qualitative approach in which the presence of POPs in milk is already *a priori* considered to be a concern without further risk assessment (UNEP, 2017).

3.2.2 *Risk Assessment Step 2: Simulating bioaccumulation*

Following international risk assessment protocols, in a second step a more elaborate risk assessment was performed for BDE-153, PFOS, and the combined group dioxins/furans/dl-PCBs. The HBGVs for these substances, which were also used in step 1, were based on the bioaccumulating properties of these substances during the life-span of humans. In this step 2 exposure scenario, the bioaccumulation in the body of the neonate during breastfeeding and during the rest of its life was modelled. The modelled accumulation was then compared to bioaccumulated amounts that reflect the effect values underpinning the HBGV. The model simulations showed that dioxins, BDE-153 and PFOS in Dutch human milk do not pose a health risk for new-borns and infants. In the case of PFOS, the risk assessment may have to be updated when EFSA publishes revised HBGV values mid 2020.

In step 2 of the risk assessment (see section 2.2) for BDE-153, PFOS, and the combined dioxin group (PCDDs/PCDFs/dl-PCBs) the bioaccumulation of these substances in the human body was simulated. In short, the accumulation in the human body or the serum lipids resulting from a one-year breastfeeding period was modelled (for details see section 2.2.3). This accumulation was compared with the accumulation corresponding to exposure at the level of the HBGV of these chemicals. The extent and the duration of the exceedance of HBGV by the exposure levels was calculated. For technical details see Annex 4.

For BDE-153, the HBGV refers to neurodevelopmental disturbance resulting from prenatal, *in utero*, exposure or postnatal exposure from birth into adulthood. To prevent this effect to occur, the whole-body burden of BDE-153 in infants, toddlers, young children, adolescents and pregnant women must remain below the body burden corresponding with the HBGV, which is 25 µg/kg body weight (for details, see Annex 4). Note that each of these life stages represents a sensitive window for the induction of neurodevelopmental toxicity. Figure 3 shows the simulation of the accumulation of BDE-153 in the human body resulting from the calculated one-year intake via human milk (black lines), compared to the accumulation resulting from the exposure at the level of the HBGV. As shown, at all ages the accumulation resulting from breastfeeding remained below the accumulation corresponding with the HBGV, indicating that no toxic risk results from the simulated one-year breastfeeding period.

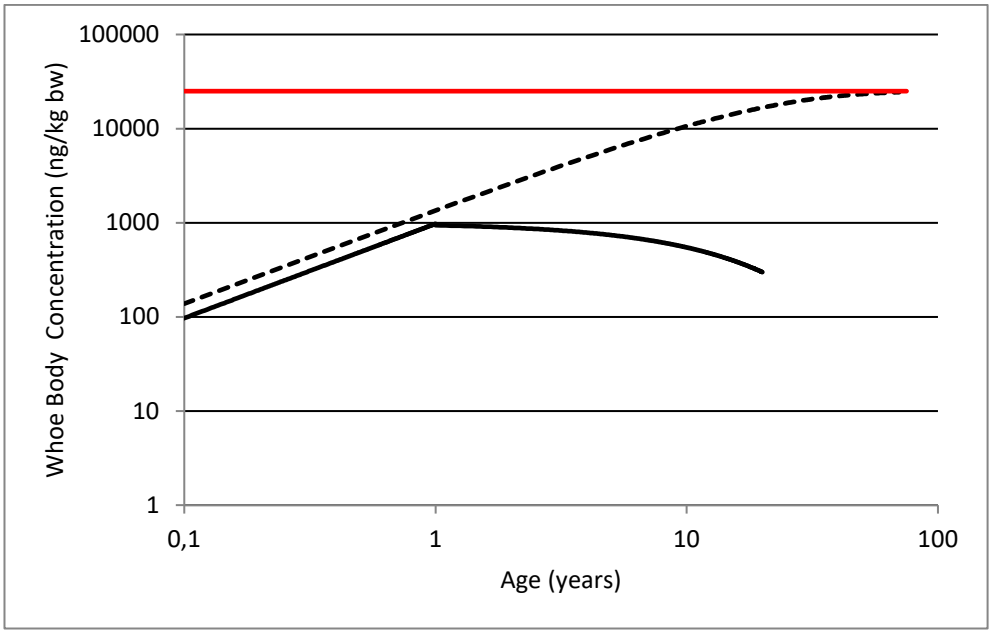


Figure 3. Simulation of the time-course of the human body burden of BDE-153 resulting from a 1-year breastfeeding period followed by a depuration period up to the age of 19 years. Solid black line: human body burden based on measured milk intake in 2014 (milk concentration: 0.74 ng/g milk lipid; median intake during the breastfeeding period: 2.7 ng BDE-153/kg body weight/day). Dashed black line: time-course of the tolerable human body burden for the most critical effect: postnatally induced neurodevelopmental toxicity, corresponding with an intake at the level of the HBGV, i.e. 3.8 ng BDE-153/kg body weight/day. For technical specification, see Annex 4). Red horizontal line: tolerable human body burden for postnatally induced neurodevelopmental toxicity (25 µg BDE-153/kg body weight).

For PFOS, RIVM's HBGV refers to liver toxicity (increased liver weight) resulting from life-long dietary daily exposure. To prevent this effect the PFOS serum concentration (after life-long exposure) must remain below the serum concentration corresponding with the HBGV, i.e. 90 µg PFOS/L serum. Figure 4 shows the model result of the accumulation of PFOS in human serum resulting from a one-year intake via human milk compared to the accumulation resulting from the exposure at the level of RIVM's HBGV. The PFOS serum concentration resulting from breastfeeding was initially close to, but not above, the accumulation corresponding with the RIVM HBGV.

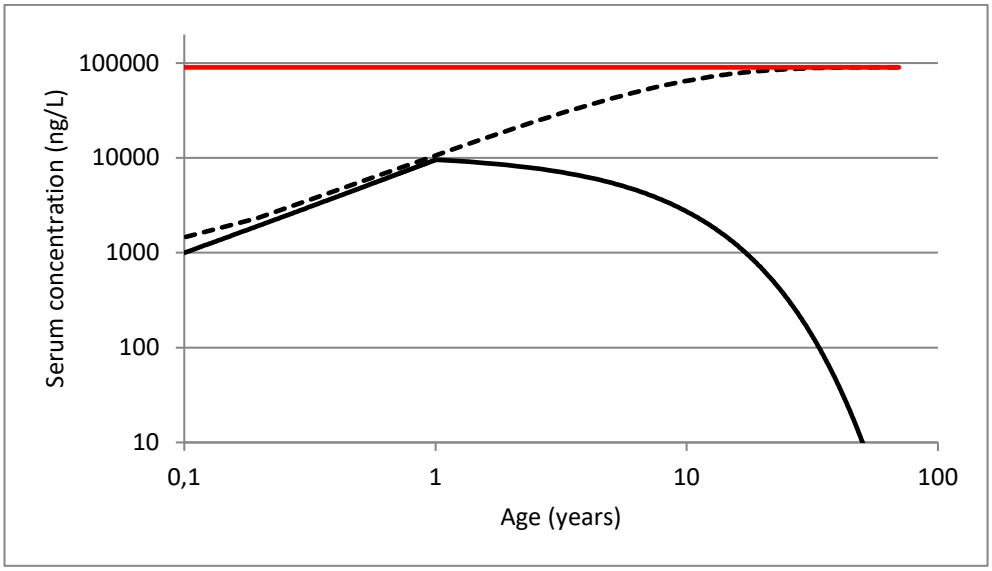


Figure 4. Simulation of the time-course of the human body burden of PFOS resulting from a 1-year breastfeeding followed by a depuration period up to the age of 19 years. Solid black line: human body burden based on measured intake in 2014 (milk concentration: 45 ng/L, average intake during the breastfeeding period: 5.5 ng/kg body weight/day). Dashed black line: time-course of the tolerable human body burden for the most critical effect: hepatotoxicity, corresponding with an intake at the level of the RIVM HBGV, i.e. 6.25 ng PFOS/kg body weight/day. For technical specification, see Annex 4). Red horizontal line: tolerable human body burden for hepatotoxicity (90 µg PFOS/L serum).

For the dioxin group (PCDD/PCDF/dl-PCB TEQ) the HBGV refers to disturbed spermatogenesis at adult age, linked to a serum level of 7 pg PCDD/PCDF TEQ/serum lipid at 9 years of age. Figure 5 shows the simulated accumulation of the PCDD/PCDF/dl-PCB TEQ in body lipid from birth to adulthood resulting from one year of breastfeeding with human milk containing 7.2 pg PCDD/PCDF/dl-PCB TEQ/g milk lipid, as measured in the 2014 pooled sample. At the end of the breastfeeding period (one year) the dioxin concentration in body lipid amounted 21 pg TEQ/g lipid. After one year, dietary exposure is added to this model, following the EFSA method. The model shows that after a child reaches one year of age, the concentration in body lipid declined to a level of 5 pg TEQ/g lipid at the age of nine years. The latter value is slightly lower than EFSA's HBGV of 7 pg TEQ/g lipid.

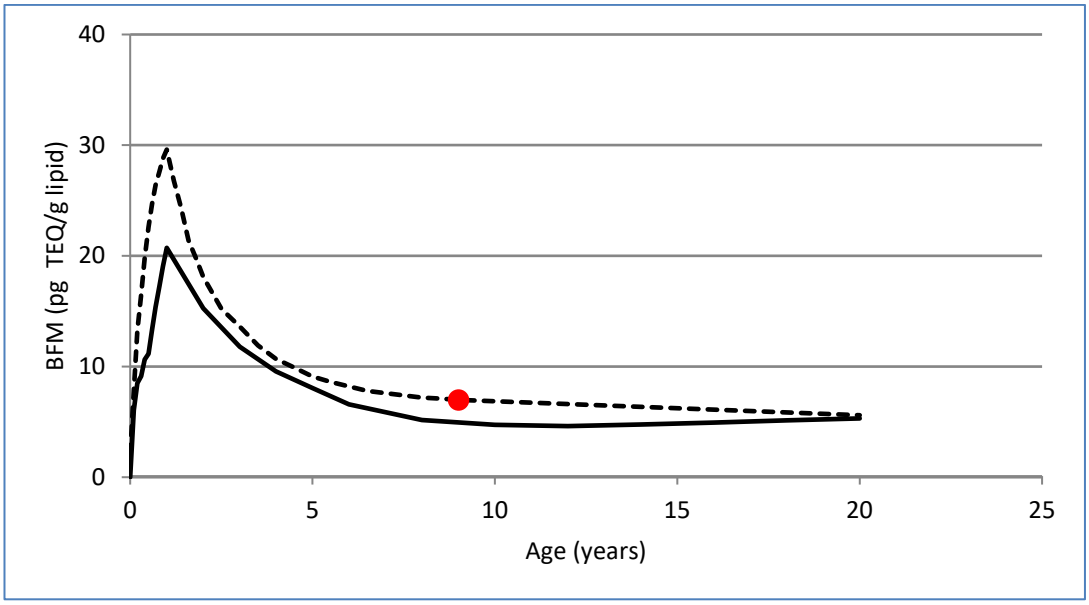


Figure 5. Simulation of the time-course of the PCDD/PCDF/dl-PCB TEQ concentration in the body fat mass (BFM; pg/g lipid) after combined exposure from human milk (first year) and the diet (from one year onwards). Solid black line: RIVM model, milk concentration: 7.2 pg TEQ/g lipid, average intake during the breastfeeding period: 26.6 pg TEQ/kg body weight/day, based on an average body weight of 6.5 kg during the first year of life). Dashed black line: tolerable body burden for the most critical effect: postnatally induced disturbed spermatogenesis (data taken from EFSA, 2018a, Figure 14). For technical specification, see Annex 4). Red dot: EFSA Benchmark for postnatally induced male reproductive toxicity, i.e. 7 pg TEQ/g serum lipid at age of 9 years).

3.3 Uncertainties in the risk assessment

As this assessment is based on a pooled milk sample, for part of the individual samples the POP levels may have been higher. This means that for some individual children, tolerable levels for the dioxin group may have been exceeded in 2014. It is important, however, to point here also at other uncertainties surrounding the present risk assessment. These uncertainties may lead to both under- or overestimation of the risk. On the one hand, worst-case assumptions were applied that may overestimate the risk. This concerned the duration of the period in which children exclusively received human milk, and the amount of milk consumed per day. Furthermore, samples were taken immediately after birth while it is known that concentrations decrease during the breastfeeding period. On the other hand, risk may have been underestimated because for PFOS and BDE-153 no other exposure routes were taken into account (e.g., other food sources, house dust). Furthermore, possible mixture toxicity was also not included in the risk assessment, except for the dioxin group.

The risk assessment is based on a pooled sample. Taking variability between individual samples into account, it may be assumed that risks for part of the samples may have been higher. However, for the current results this inter-individual variability in milk concentrations is unknown and thus, it is unknown which fraction of the children may have received milk with substance concentrations above the safe level. However,

previous RIVM monitoring has shown that this inter-individual variability of the dioxin concentration in human milk is limited (see Annex 5 and 6 and references therein). 95 Percent of all samples were within a factor of 1.7 to 2.3 from the median result, depending on the substance. Assuming the maximum factor of 2.3, this means that for PFOS and BDE-153 only for a short period of time individual samples may have exceeded the safe concentrations for life-long exposure. Note the logarithmic scale in Figures 3 and 4 when comparing the modelled concentrations with the safe concentrations for these substances. For the dioxin group, applying a factor of 2.3 would imply that in 2014 part of the individual samples exceeded tolerable levels. This means that for some individual children, risk levels may have been exceeded, by up to a factor of 1.6. This possible risk has also been identified by other authors. However, they concluded that beneficial long-term health effects of breastfeeding outweigh the negative effects of POPs in human milk (Buijssen et al., 2015; UNEP, 2013; van den Berg et al., 2017). The results from the present study give no reason to draw other conclusions.

Given a half-live of 9.5 years for the dioxin-group (PCDD/PCDF/dl-PCB) in human milk in the Netherlands (see Table 7) at present (2020) such exceedance may not be expected anymore. However, there are no recent data available to confirm this.

Further uncertainties regarding the risk assessment may work both ways. On the one hand, worst-case assumptions were applied when modelling exposure. It was assumed that children exclusively received breastfeeding during the first year, with no other food sources and a relatively high constant human milk intake (see section 3.2.1). Furthermore, it was assumed that concentrations of POPs remained constant during the one-year breastfeeding period, although it is known that during this time, concentrations decrease.

On the other hand, for PFOS and BDE-153 no other exposure routes were taken into account (e.g., other food sources, house dust). The current report focusses on exposure via human milk only. Of course, after the breastfeeding period, exposure will also take place via food and the environment (e.g. house dust), and for PFOS also via drinking water. Possible mixture toxicity was also not included in the risk assessment, except for the dioxin group.

It is difficult to quantitatively weigh the factors that may increase the potential risks versus those that may reduce them.

For PFOS it should also be mentioned that there is an ongoing discussion on the magnitude of the HBGV. At present EFSA and ATSDR recommend lower values for PFOS than those currently used in this report. In 2018 RIVM expressed concerns regarding the method of derivation of EFSA's provisional HBGV value (EFSA, 2018c). EFSA is currently re-evaluating this HBGV. Depending on the outcome of EFSA's re-evaluation in 2020, the current assessment of health risks for neonates and infants due to uptake of PFOS via human milk may have to be revised.

3.4 Comparison with other countries and trends in time

Trends in time in Dutch human milk could be assessed for the dioxins, PCBs, p,p'-DDE (metabolite of DDT), HCB, β-HCH and PBDE. For all other substances, not enough data above detection limits were available. For PFAS-substances like PFOS and PFOA, no trends could be established since these substances were only analysed in the last monitoring campaign in 2014. The results clearly show that POP concentrations in human milk decrease in time. The concentrations of these substances are reduced by 50% every 5 to 10 years. Considering the substances that were detected, the Netherlands showed an intermediate position amongst the neighbouring countries (Figure 1). Some variability in POP patterns is observed, which may be explained by different national regulations, production, use and consumption patterns.

The Netherlands participated in the first, second, third and sixth WHO/UNEP milk survey round (1987-89, 1992-94, 2000-03 and 2014 respectively). In addition, since 1969 in the Netherlands also the concentrations of substances in human milk from the DDT group (mainly *p,p'*-DDE), HCB and β-HCH was investigated. The dioxins/furans and various PCBs were added since 1983. More recently the PBDEs were added. Thus, temporal trends in Dutch human milk can be assessed for the dioxins/furans, PCBs, *p,p'*-DDE (metabolite of DDT), HCB, β-HCH and PBDE. The measured POP concentrations in Dutch human milk for these substances are shown in Table 5.

Table 5. Concentrations of several POPs in Dutch human milk. References for the subsequent years are provided in Annex 5 and Annex 6. Data from all years are reported as median values based on multiple measurements (See Annex 5/6), except for 2014 when a pooled sample was analysed.

		POP concentration							
Year	**Lipid (%)**	***p,p'*-DDE (ng/g lipid)**	**HCB (ng/g lipid)**	**B-HCH (ng/g lipid)**	**PCDD/ PCDF WHO-TEQ$_{2005}$ (pg/g lipid)**	**PCB WHO TEQ$_{2005}$ (pg/g lipid)**	**Total WHO TEQ$_{2005}$ (pg/g lipid)**	**I-PCB (ng/g lipid)**	**Σ4PBDE (ng/g lipid)**
1969	1.9	1600	+	280	-	-	-	-	-
1972	3.4	1600	860	280	-	**-**	**-**	**-**	**-**
1983	2.7	820	190	100	-	-	-	317	-
1988	2.7	550	100	80	33.6	-	-	245	1.05
1993	2.6	330	40	50	22	10.2	32.2	262.5	4.77
1998	2.7	197.8	29.2	21.4	16	7.6	23.6	176.2	2.82
2003	-	111.4	15.8	-	9.4	4.8	14.2	86	3.11
2014	3.0	82.3	9.5	6.9	4.5	2.7	7.2	40.1	1.53

I-PCB = indicator PCBs: PCB 28, PCB 52, PCB 101, PCB 138, PCB 153, PCB 180
Σ4PBDE = BDE47 + BDE99 + BDE100 + BDE153.
+ detected in human milk, but no concentration reported.
- not analysed

In Annex 7, POP concentrations in various European countries from the last 15 years are summarized. The Dutch human milk POP data fit well within the available data set for various West European (UN/WEOG)

countries. The substances below the detection limit in the Netherlands were also below the detection limit in other West European countries.

Considering the substances that were detected, the Netherlands showed an intermediate position among the neighbouring countries. The Nordic countries showed relatively low concentrations for most substances, whereas Belgium and Luxemburg showed relatively high concentrations for some. When comparing results for various countries, it should be kept in mind that a year difference in sampling already leads to a 7 to 10% decrease, assuming a half-life between 7 and 10 years.

The variability in the different POP concentrations in human milk between the different countries, although small, indicates that exposure is country specific and that concentrations in one country cannot be used to estimate the POP exposure in another country. This may have to do with production taking place in a certain country, or with specific food consumption patterns. Often these use patterns are related to cultural habits (fish consumption in Norway), regulations (*e.g.* fire safety regulations in the case of PBDEs), or different use patterns due to legislations allowing a product or a specific application in one country but not in another (e.g. Meijer et al 2008).

The PFOS concentration in Dutch human milk is comparable to that measured in the same period in Sweden, around 45-50 ng/L (Nyberg et al 2018). The Swedish data showed a considerable decrease in PFOS concentrations in Swedish human milk since 1990 after an initial increase between 1970 and 1990. This trend is also reflected in UNEP (2017) where an increase of PFOS in the 1980s and 1990s is reported, reflecting industrial production. In Western Europe, this is followed by a decline. Human milk data from other North Western European countries showed considerably higher PFOS concentrations than the current study for the Netherlands, but may have to be corrected for sampling date. Norwegian data collected between 2002 and 2006 were 126.7 ng/L, German data from 2006 were between 113 and 123 ng/L and Italian data collected in 2010 were 57 ng/L (Iszatt et al 2019; Voelkel et al 2008; Barbarossa et al 2013). The PFHxS concentration of 11 ng/L in the Netherlands is slightly higher than the concentration of 6-10 ng/L measured in Swedish human milk in the same period (Nyberg et al 2018). Other PFHxS data are limited.

Using these measured concentrations (Table 5), trends in time can be modelled, as shown in Figures 6-12, with figures in a logarithmic scale model results in Annex 8. For Sweden, Germany and Belgium the data from the references in section 2.4.4 were used. The half-lives reflect the decrease of the substance in the Dutch environment, according to the method described in section 2.4.3. The half-life is the time needed for a 50% decrease of a concentration and is different for each substance. For all substances analysed, a clear decrease is observed. Half-lives for all substances are mostly between 5 and 10 years. That means that the concentrations of these substances are reduced by 50% every 5 to 10 years.

The metabolite p,p'-DDE has a longer half-life in human milk than its mother substance p,p'-DDT and the other metabolite p,p'-TDE. This is

confirmed by known half-lives in human milk (Noren & Meironyte, 2000) and the environment (Quinsey et al 1995). That means that p,p'-DDE will persist longer than the other two substances. In our data the half-live of DDE was estimated to be 9.4 years.

The PCDD/PCDF and dl-PCB concentrations have decreased in human milk since the measurements for these POPs started (in 1988 and 1993, respectively) with half-lives of 9 and 11 years. Half-lives of indicator PCBs and total WHO TEQ_{2005} were almost identical with 10 and 9.6 years respectively. HCB is the POP with the shortest half-life (6.2 years) and dl-PCB has the longest half-life of the POPs for which trends in time are available.

PBDE concentrations have initially increased since the start of measurements but show a decrease between 2003 and 2014. The maximum PBDE concentration in human milk was most likely reached between 1993 and 2003. Around this period the production and use of commercial penta- and commercial octa-BDE in Europe was finished resulting in decreasing concentrations in the environment.

The modelled half-live of the Σ4BDEs was 12.8 years using the data from 1993 onwards. However, considering the variation in the data, the reliability of this value is limited.

For PFOS, no trend could be established since it was only analysed in the last monitoring campaign. Figure 13 shows the single measurement of PFOS in the Netherlands in 2014 compared to the measurements in Sweden, starting in the 1970's. It should be noted however, that the Swedish data for PFOS was based on only one sampling location, and that the analytical techniques used may not be according to current standards.

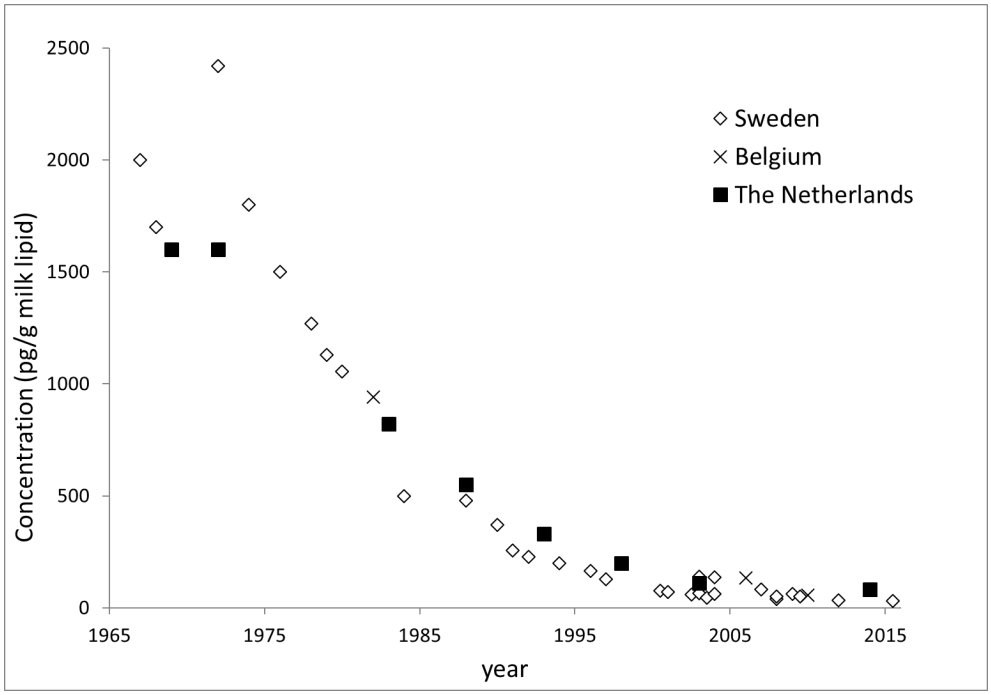


Figure 6. Trend in time for p,p'-DDE in Sweden, Belgium and The Netherlands.

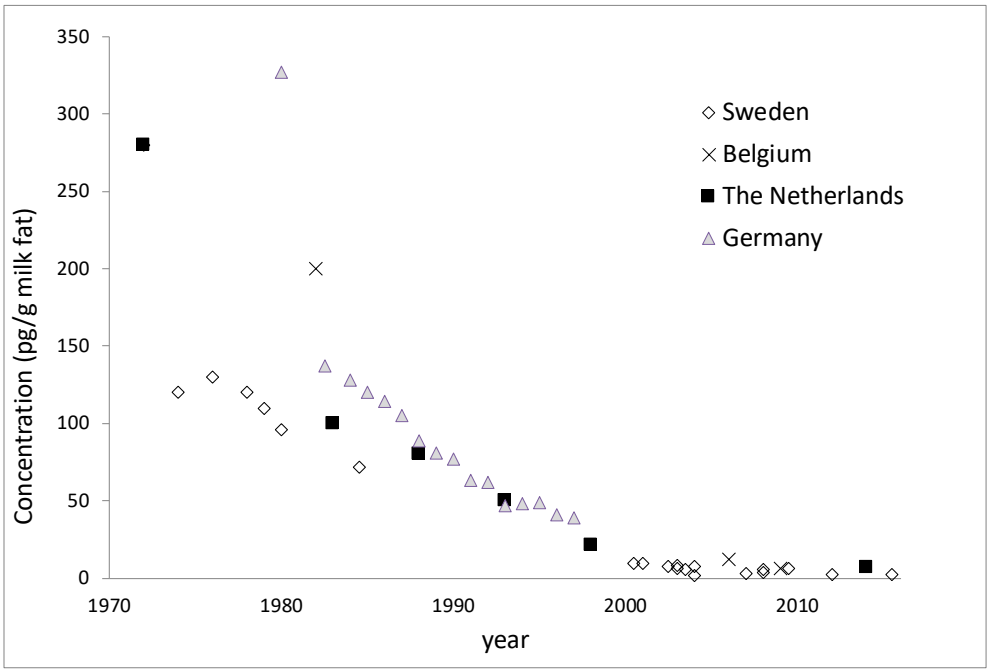


Figure 7. Trend in time for beta-hexachlorocyclohexane (β-HCH) in Sweden, Belgium, The Netherlands and Germany.

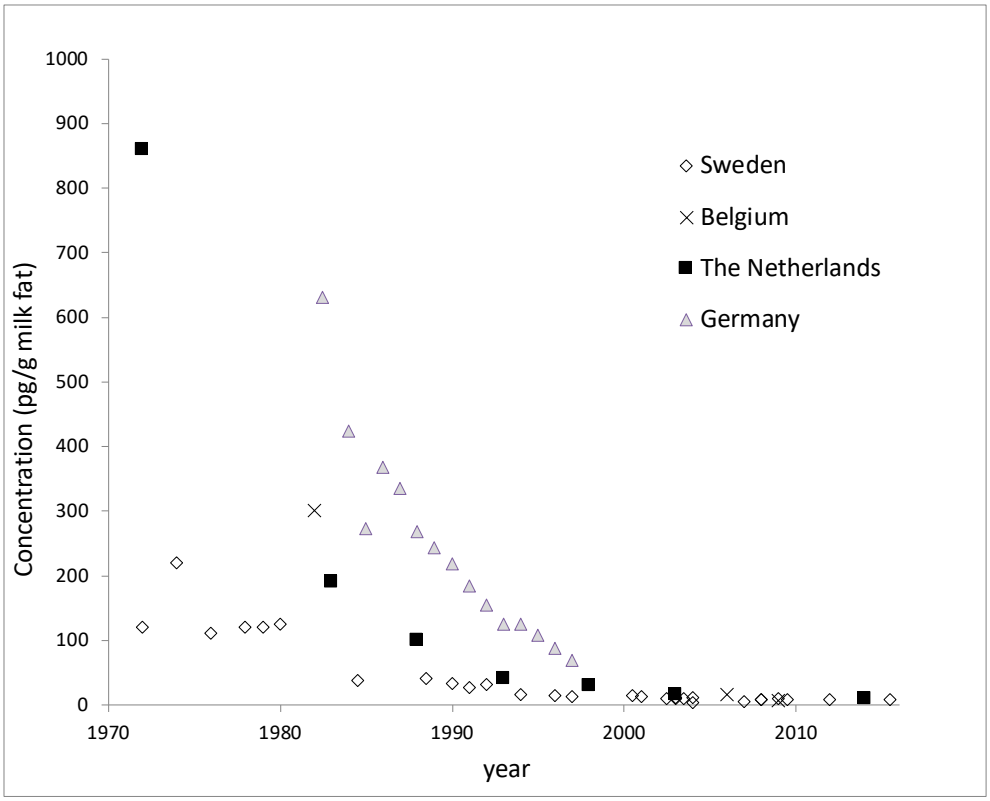


Figure 8. Trend in time for hexachlorobenzene (HCB) in Sweden, Belgium, The Netherlands and Germany.

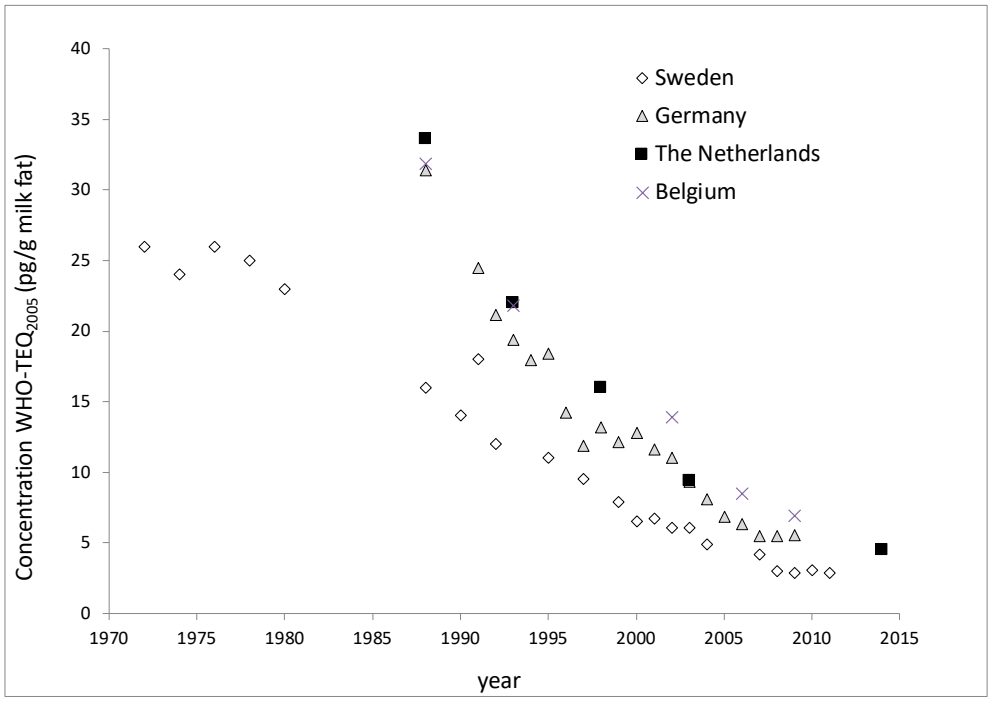


Figure 9. Trend in time for PCDD/PCDF in Sweden, Germany, The Netherlands and Belgium.

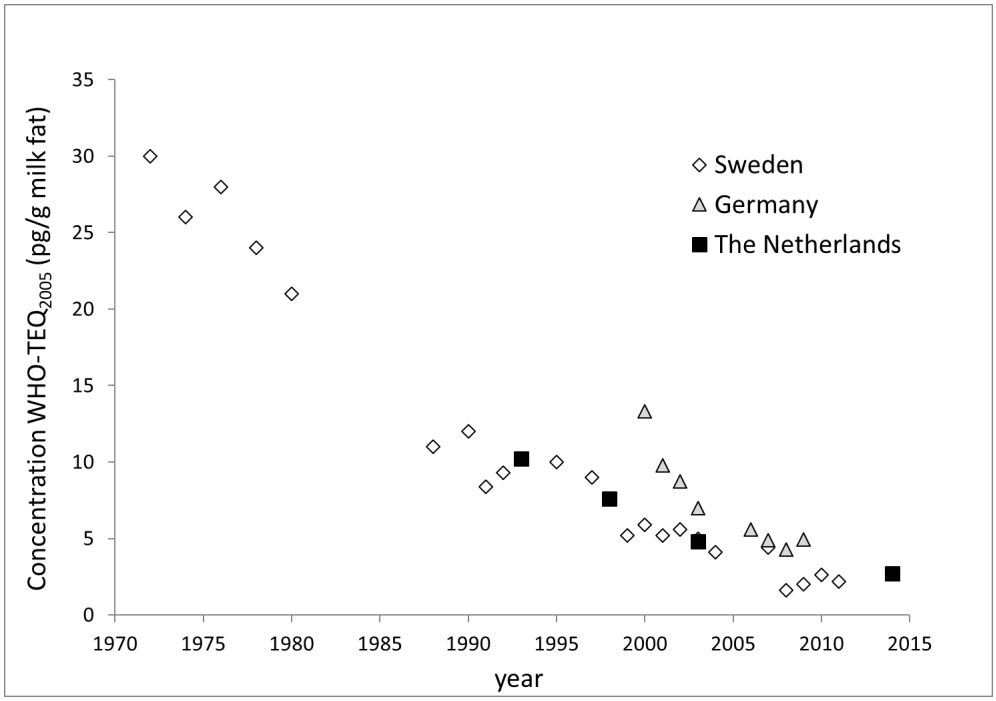


Figure 10. Trend in time for dl-PCBs in the Netherlands, Germany and Sweden.

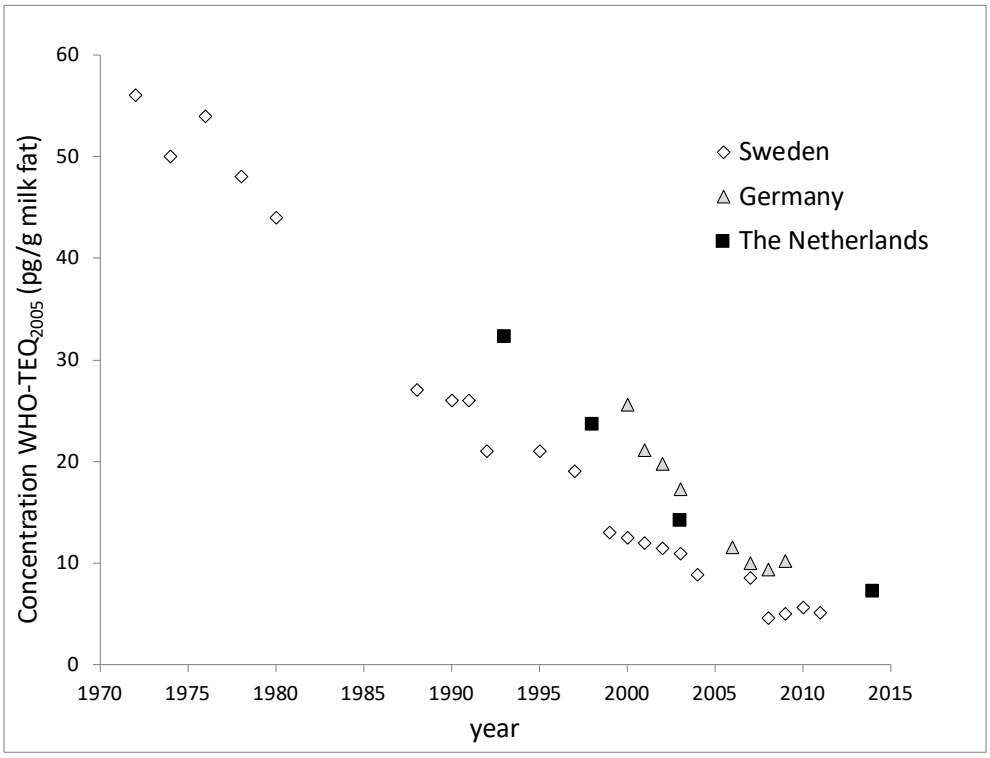


Figure 11. Trend in time for total PCDD/PCDF/dl-PCB TEQ in the Netherlands, Germany and Sweden.

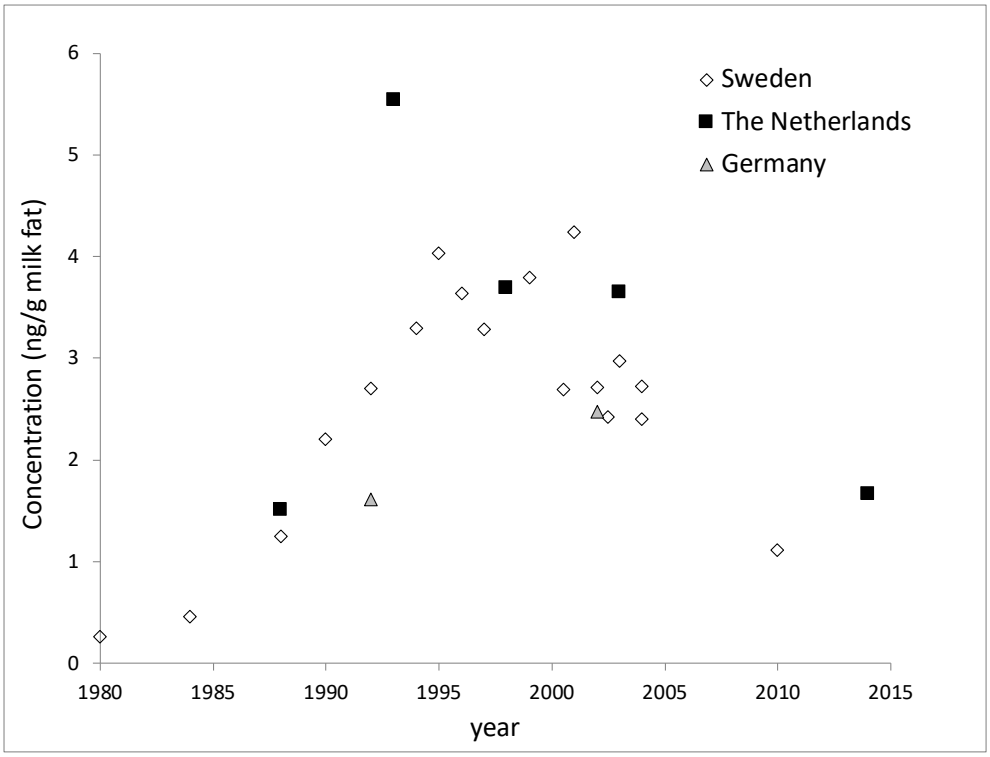


Figure 12. Trend in time for PBDE (BDE-47, BDE-99, BDE-100, and BDE-153) in the Netherlands, Germany and Sweden.

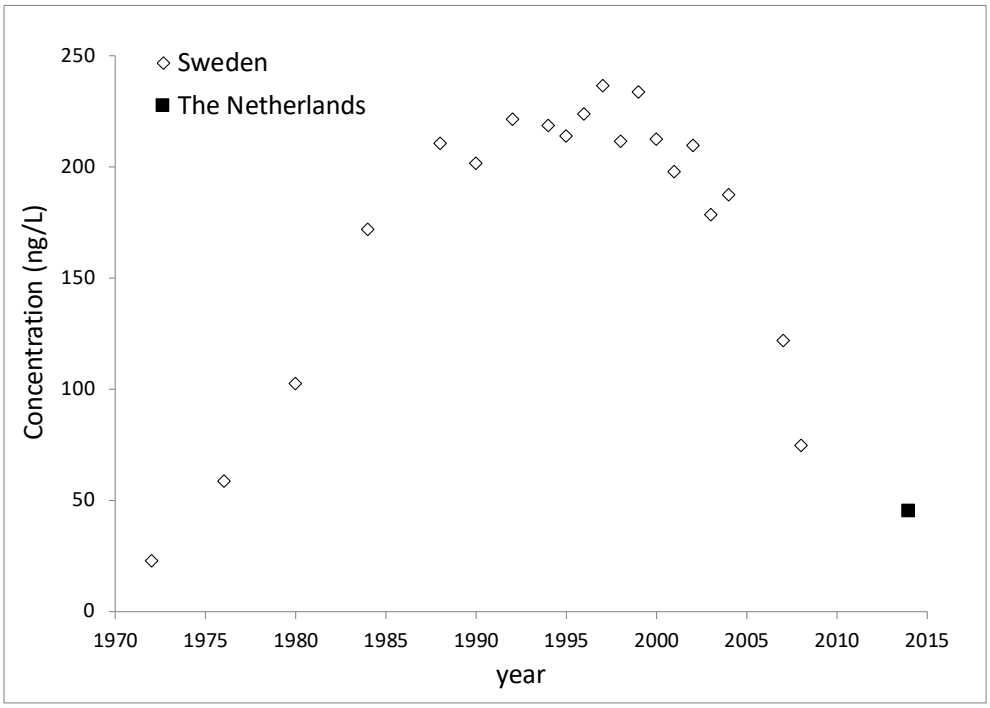


Figure 13. Trend in time for PFOS in the Netherlands and Sweden.

The decrease in time for the POPs presented in Figures 6-12 is in line with previously described data for Western Europe by the WHO in the period 1988 to 2007. The decrease in human milk concentrations reflects reduced exposure and/or reduced environmental concentrations of the POPs due to international restrictions and bans on the production and use of these POPs and efforts to reduce unintentional emission. Besides this, national measures, either because of implementation of the Stockholm Convention or because of other reasons, will also add to the decrease observed. DDT, HCH, HCB, PCBs and commercial octa-BDE and penta-BDE were prohibited or severely restricted in the Netherlands in 1973, 1991, 1973, 1985, 2002 and 2004, respectively.

In Table 7, the calculated half-lives are compared to half-lives that were calculated for other countries, using the data from the references in section 2.4.4. Half-lives were shortest in Belgium and Germany and longest for the Netherlands and Sweden. The longest and shortest half-lives were not always calculated for the same substance. DDE, HCB and β-HCH showed relatively low half-lives compared to the dioxins and furans. PCBs showed half-lives that were considerably larger.

Table 7. Half-lives (in years) for the concentration of various POPs in human milk in the Netherlands, Belgium, Sweden and Germany based on the data presented in Figures 6-12.

Substance	The Netherlands	Belgium	Germany	Sweden
DDE	9.4	7.1	-	7.0
HCB	6.2	5.1	4.8	7.9
β-HCH	7.7	5.5	6.4	6.2
PCDD/Fs	8.9	8.3	9.8	11.4
dl-PCBs	10.8	-	6.2	10.0
total TEQ	9.5	-	6.1	10.8
indicator-PCBs	10.0	8.1	11.7	13.3

- = not enough data to estimate half-lives.

In Annex 9, some additional information is provided on the TEQ_{2005} values.

Thus, trend data showed a decrease in POP concentrations in human milk for most POPs under the Stockholm Convention. Several POPs are, however, still present in measurable amounts years after they were regulated. Furthermore, the presence of some new POPs in human milk (e.g. PFOS, BDEs, HBCDD) was confirmed. Because of a lack of data, trends could not be established in all cases. It is therefore important to continue investigating the POP concentrations, especially the more recently added ones, on a regular basis in milk surveys. This way, it can be determined if measures taken by the Stockholm Convention are effective on a country-by-country basis.

4 Conclusions and recommendations

4.1 Conclusions

The concentration of POPs in human milk is a good proxy for the occurrence of these substances in our food and the environment. Measurements from 2014 show that many POPs are present in Dutch human milk, but in low concentrations compared to the concentrations measured in the 1970s and 1980s. Of the original 'Dirty Dozen', which were added to the Stockholm Convention in 2004, nine are still detectable in Dutch human milk in 2014. Of the 11 new POPs, five were detectable.

To assess the risks to exposed new-borns and infants, first HBGVs (Health Based Guidance Values) were compared to the intake of the various POPs through the consumption of human milk. For almost all substances a risk for children that receive breastfeeding can be excluded already in the first, worst-case, step of the assessment. Exceptions were BDE-153, PFOS, and the combined group dioxins/furans/dl-PCBs.

Thus, an elaborated, more precise, risk assessment was performed for these substances. Calculated concentrations in the body of the neonate or infant during breastfeeding and during the rest of his life show that POPs in Dutch human milk do not pose a health risk to new-borns and infants. This is based on the pooled sample. This means that for some individual children, tolerable levels for the dioxin group may have been exceeded in 2014. It is important, however, to point here also at other uncertainties surrounding the present risk assessment. These uncertainties may work both ways. On the one hand, worst-case assumptions were applied when modelling exposure (e.g. children exclusively received breastfeeding in relatively large amounts, concentrations of POPs that remained constant despite it being known that they decrease during the breastfeeding period). On the other hand, for PFOS and BDE-153 no other exposure routes were taken into account (e.g, other food sources, house dust). Possible mixture toxicity was also not included in the risk assessment, except for the dioxin group. It is difficult to quantitatively weigh the factors that may increase the potential risks versus those that may reduce them. For PFOS it should also be mentioned that there is an ongoing discussion on the magnitude of the HBGV. At present EFSA and ATSDR recommend lower values for PFOS than those currently used in this report.

Substances that are incorporated as POPs in the Stockholm Convention, are phased out or their use is restricted. An analysis of the trend in time of the concentrations in human milk showed a decrease by 50% every 5-10 years for most substances. This also shows that although the decrease is noted, it takes decades before the concentrations of some of these substances fall below the detection limits. For PFOS, no trend could be established since it was only analysed in the last monitoring campaign. The concentrations of POPs in the Netherlands were comparable to those in neighbouring countries.

4.2 Recommendations

Various studies have indicated that beneficial long-term health effects of breastfeeding outweigh the negative effects of POPs in human milk (Buijssen et al., 2015; UNEP, 2013; van den Berg et al., 2017). The results from the present study give no reason to draw other conclusions.

It is recommended to continue the monitoring on a regular basis, in particular for upcoming POPs, to determine if the measures taken by the Stockholm Convention are effective. Environmental data and analysis of the long-term trends in time of POP concentrations in food and food consumption may provide more insight into the reasons behind the variability of POP human milk concentrations in different European countries.

Many PFAS substances were below the detection limit with the current analytical methods. It is important to further develop the analytical methods for the PFAS substances in order to reduce the measurements below the detection limit and the uncertainty in the exposure assessment.

The current measurements were based on a pooled sample of 50 individual human milk samples, which suits the goals of the WHO and UNEP to obtain data from countries all over the world. When more information at a national scale is necessary, for instance on the variability in concentrations between the samples, it is recommended to analyse individual samples in future monitoring campaigns.

For PFOS, the risk assessment was based on a HBGV that was derived by RIVM. After RIVM published this value, EFSA has published a provisional, more critical HBGV. However, in 2018 RIVM expressed concerns regarding the method used by EFSA to derive a provisional HBGV value. The latter value is currently under revision. The current risk assessment may have to be revised when EFSA has published their revised HBGV value (mid 2020).

Finally, mixture toxicity was not considered in the present study, except for the dioxin/furan/dl-PCB group. In future studies, methods to assess mixture toxicity may be applied, if available.

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6 Abbreviations

Alpha-HBCDD	Alpha-Hexabromocyclododecane
ATSDR	Agency for Toxic Substances and Disease Registry
β-HCH	β-Hexachlorocyclohexane
γ-HCH	γ-Hexachlorocyclohexane
BDE	Bromodiphenyl ether
BMI	Body Mass Index
CVUA	Chemisches und Veterinäruntersuchungsamt
dl-PCB	Dioxin-like Poly Chlorinated Biphenyl
DDT	Dichlorodiphenyltrichloroethane
EFSA	European Food Safety Authority
HBCDD	Hexabromocyclododecane
HBGV	Health Based Guidance Value
HCB	Hexachlorobenzene
HCH	Hexachlorocyclohexane
I-PCB	Indicator Poly Chlorinated Biphenyl
PBDE	Polybromodiphenylether
PCB	Poly Chlorinated Biphenyl
PCDD	Polychlorinated dibenzodioxin
PCDF	Polychlorinated dibenzofuran
PFAS	Per- and polyfluoroalkyl substances
PFHxS	Perfluorohexane Sulfonate
PFOA	Perfluorooctanoic acid
PFOS	Perfluorooctane sulfonate
p,p'-DDE	p,p'- Dichlorodiphenyldichloroethylene
p,p'-DDT	p,p'- Dichlorodiphenyltrichloroethane
POP	Persistent Organic Pollutants
RPF	Relative Potency Factors
SCCPs	Short-Chain Chlorinated Paraffins
TCDD	Tetrachlorodibenzodioxin
TEF	Toxic Equivalency Factor
TEQ	Toxicity Equivalence
UNEP	United Nations Environment Programme
WHO	World Health Organisation

7 References

Albers JMC, Kreis IA, van Zoonen P, Liem AKD, Theelen RMC (1993). Verontreiniging van moedermelk met gechloreerde koolwaterstoffen in Nederland 1988. RIVM report 638815002. RIVM, Bilthoven, the Netherlands.

Antignac JP, Main KM, Virtanen HE, Boquien CY, Marchand P, Venisseau A, Guiffard I, Bichon E, Wohlfahrt-Veje C, Legrand A, Boscher C, Skakkebæk NE, Toppari J, Le Bizec B (2016). Country-specific chemical signatures of persistent organic pollutants (POPs) in breast milk of French, Danish and Finnish women. Environ Pollut 218: 728-738.

ATSDR, Agency for Toxic Substances and Disease Registry (2015). Draft Toxicological profile for Perfluoroalkyls.

ATSDR, Agency for Toxic Substances and Disease Registry (2018). Toxicological Profile for Perfluoroalkyls Draft for Public Comment June 2018.

Atuma SS, Vaz R (1986). A pilot study on levels of organochlorine substances in human milk in Nigeria. Intern J Anal Chem 26:187-192.

Barbarossa A, Masetti R, Gazzotti T, Zama D, Astolfi A, Veyrand B, Pession A, Pagliuca G. (2013). Perfluoroalkyl substances in human milk: A first survey in Italy. Environ Int 51: 27–30

Béchaux C, Zeilmaker M, Merlo M, Bokkers B, Crépet A (2014). An integrative risk assessment approach for persistent chemicals: A case study on dioxins, furans and dioxin-like PCBs in France. Regul Toxicol Pharmacol 70 (1): 261-269.

BFR (2000). Trends der Rückstandsgehalte in Frauenmilch der Bundesrepublik Deutschland – Aufbau der Frauenmilch- und Dioxin-Humandatenbank am BgVV1. Empfehlung der Nationalen Stillkommission vom 10. August 2000. BFR, Berlin, Germany.

Bruinen de Bruin Y, Janssen MPM (2012). PCB's in bouwmateriaal in Nederland. RIVM letter report 601356003. RIVM, Bilthoven, the Netherlands.

Buijssen M, Jajou R, van Kessel FGB, Vonk Noordegraaf-Schouten MJM, Zeilmaker MJ, Wijga AH, van Rossum CTM (2015). Health effects of breastfeeding: an update: Systematic literature review. RIVM Rapport 2015-0043. RIVM, Bilthoven, the Netherlands.

Carrier G, Brunet RC, Brodeur J (1995a). Modeling of the toxicokinetics of polychlorinated dibenzo-*p*-dioxins and dibenzofurans in mammalians, including humans. I. Nonlinear distribution of PCDD/PCDF body burden between liver and adipose tissue. Toxicol Appl Pharmacol 131: 253-266.

Carrier G, Brunet RC, Brodeur J (1995b). Modeling of the toxicokinetics of polychlorinated dibenzo-*p*-dioxins and dibenzofurans in mammalians, including humans. II. Kinetics of absorption and disposition of PCDDs/PCDFs. Toxicol Appl Pharmacol 131: 267-276.

CBS (2019). Ouderlijk gezin beïnvloedt timing eerste moederschap. https://www.cbs.nl/nl-nl/achtergrond/2014/29/ouderlijk-gezin-beinvloedt-timing-eerste-moederschap., Accessed on 20 December 2019.

Čechová E, Scheringer M, Seifertová M, Mikeš O, Kroupová K, Kuta J, Forns J, Eggesbø M, Quaak I, De Cock M, Van de Bor M, Patayováe H, Murínová ĽP, Kočan A (2017). Developmental neurotoxicants in humanmilk: Comparison of levels and intakes in three European countries. Sci Total Environ 579: 637–645.

Colles A, Koppen G, Hanot V, Nelen V, Dewolf MC, Noël E, Malisch R, Kotz A, Kypke KBiot P, Vinkx C, Schoeters G (2008). Fourth WHO-coordinated survey of human milk for persistent organic pollutants (POPs): Belgian results. Chemosphere 73: 907–914.

Croes K, Colles A, Koppen G, Govarts E, Bruckers L, Van de Mieroop E, Nelen V, Covaci A, Dirtu AC, Thomsen C, Haug LS, Becher G, Mampaey M, schoeters G, van Larebeke N, Baeyens W (2012). Persistent organic pollutants (POPs) in human milk: A biomonitoring study in rural areas of Flanders (Belgium). Chemosphere 89 (8): 988-994.

Cuijpers CEJ, Liem AKD, Albers JMC, Kreis IA, Lebret E (1997). Verontreiniging van moedermelk met gechloreerde koolwaterstoffen in Nederland, 1993. RIVM report 529102004. RIVM, Bilthoven, The Netherlands.

De Winter-Sorkina R, Bakker MI, Baumann RA, Hoogerbrugge R, Zeilmaker MJ (2003). Exposure assessment of Dutch nursing infants to brominated flame retardants. RIVM report 310305002/2003. RIVM, Bilthoven, The Netherlands.

EFSA (2005a) Opinion of the Scientific Panel on Contaminants in the food chain on a request from the Commission related to aldrin and dieldrin as undesirable substance in animal feed. The EFSA Journal, 85: 1-43.

EFSA (2005b) Opinion of the Scientific Panel on contaminants in the food chain on a request from the commission related to gamma-HCH and other hexachlorocyclohexanes as an undesirable substance in animal feed. Question No EFSA-Q-2003-067. Adopted on 4 July 2005. The EFSA Journal, 250, 1 – 39.

EFSA (2006a) Opinion of the Scientific Panel on Contaminants in the food chain on a request from the Commission related to DDT as an undesirable substance in animal feed. EFSA Journal 433, 1-69.

EFSA (2006b) Opinion of the Scientific Panel on contaminants in the food chain on a request from the commission related to hexachlorobenzene as an undesirable substance in animal feed. Question No EFSA-Q-2005-185. Adopted on 13 September 2006. The EFSA Journal, 402, 1 – 49.

EFSA (2007a) Chlordane as undesirable substance in animal feed. Scientific Panel on Contaminants in the Food Chain (Question N° EFSA-Q-2005-181). Adopted on 7 November 2007. The EFSA Journal, 582, 1-53.

EFSA (2007b) Opinion of the Scientific Panel on contaminants in the food chain on a request from the commission related to heptachlor as an undesirable substance in animal feed. Question No EFSA-Q-2005-142. Adopted on 26 April 2007. The EFSA Journal, 478, 1-48.

EFSA (2011a) Scientific Opinion on Polybrominated Diphenyl Ethers (PBDEs) in Food. EFSA Panel on Contaminants in the Food Chain (CONTAM). European Food Safety Authority (EFSA), Parma, Italy. The EFSA Journal, 9(5), 2156.

EFSA (2011b) Scientific Opinion on Hexabromocyclododecanes (HBCDDs) in Food. EFSA Panel on Contaminants in the Food Chain (CONTAM). European Food Safety Authority (EFSA), Parma, Italy. The EFSA Journal. 9(7), 2296.

EFSA (2018a) EFSA Panel on Contaminants in the Food Chain (CONTAM). Risk for animal and human health related to the presence of dioxins and dioxin-like PCBs in feed and food. EFSA Journal 16(11): 5333.

EFSA (2018b) Panel on Contaminants in the Food Chain (CONTAM), Risk to human health related to the presence of perfluorooctane sulfonic acid and perfluorooctanoic acid in food. EFSA Journal 16(12): 5194.

EFSA (2018c). Minutes of the expert meeting on perfluoorooctane sulfonic acid and perfluorooctanoic acid in food assessment. EFSA/CONTAM/3503. www.efsa.europa.eu/sites/default/files/news/efsa-contam-3503.pdf. Accessed on February 3, 2020.

Fång J, Nyberg E, Bignert A, Bergman Ä (2013). Temporal trends of polychlorinated dibenzo-p-dioxins and dibenzofurans and dioxin-like polychlorinated biphenyls in mothers' milk from Sweden, 1972–2011. Environ Int 60: 224–231.

German dioxin database (2018). http://www.dioxindb.de/index-e.html Login via https://popdioxin.uba.de/DioxinWSClient/logon.do?language=de; accessed in December 2019.

Greve PA (1974). Organochloorbestrijdingsmiddelen in moedermelk. RIV report 44/74 Tox-RoB (in Dutch).

Greve PA (1985). Organochlorine pesticides and PCBs in human tissues from Dutch citizens (1968-1983). RIVM report 638205004. RIVM, Bilthoven, the Netherlands.

Greve PA, Van Hulst SJ, Van Harten DC, Janssen GE, Heusinkveld HAG, Verschraagen C (1985). Chemische contaminanten in moedermelk – Deelrapport 3: organochloorbestrijdingsmiddelen en verwante verbindingen. RIVM report 638307003. RIVM, Bilthoven, the Netherlands.

Greve PA, van Zoonen P (1990). Organochlorine pesticides and PCBs in tissues from Dutch citizens (1968-1986). Int J Environ An Ch 38: 265-277.

Gyllenhammar I, Glynn A, Fridén U, Cantillana T, Aune M, Lignell S. (2017). Levels of persistent halogenated organic pollutants (POP) in mother's milk from first-time mothers in Uppsala, Sweden: results from year 2015 and 2016, and temporal trends for the time period 1996-2016. Naturvårdsverket, Stockholm, Sweden.

Heyndrickx A, Maes R (1969). The excretion of chlorinated hydrocarbon insecticides in human mother's milk. J Pharm Belg. 1969 24(9):459-63.

Iszatt N, Janssen S, Lenters V, Dahl C, Stigum H, Knight R, Mandal S, Peddada S, González A, Midtvedt T, Eggesbø M. (2019).Environmental toxicants in breast milk of Norwegian mothers and gut bacteria composition and metabolites in their infants at 1 month. Microbiome (2019) 7:34: 1-14.

Kärrman A, Ericson I, van Bavel B, Darnerud PO, Aune M, Glynn A, Lignell S, Lindström G (2007). Exposure of perfluorinated chemicals through lactation: levels of matched human milk and serum and temporal trend, 1996-2004, in Sweden. Environ Health Perspect 115, 226-230.

Kreuzer PE, Csanády GA, Baur C, Kessler W, Päpke O, Greim H, Filser JG (1997). 2,3,7,8-Tetrachlorodibenzo-*p*-dioxin (TCDD) and congeners in infants. A toxicokinetic model of human lifetime body burden by TCDD with special emphasis on its uptake by nutrition. Arch Toxicol 71: 383-400.

Liem AKD, de Jong APJM, Marsman JA, den Boer AC, Groenemeijer GS, de Korte GAL, Wegman RCC, Greve PA (1989). Bestrijdingsmiddelen en industriële contaminanten in moedermelk - Deelrapport 1: polygechloreerde bifenylen, dibenzo-p-dioxinen en dibenzofuranen. RIVM report 638815001. RIVM, Bilthoven, the Netherlands.

Lignell S, Aune M, Glynn A, Cantillana T, Fridén U (2014). Levels of persistent halogenated organic pollutants (POP) in mother's milk from first-time mothers in Uppsala, Sweden: results from year 2012 and temporal trends for the time period 1996-2012. Report NV-11641-11.

Livsmedelsverket, Uppsala, Sweden. http://www.diva-portal.org/smash/get/diva2:657933/FULLTEXT01.pdf.

Meijer L, Weiss J, Van Velzen M, Brouwer A, Bergman A, Sauer PJ (2008). Serum concentrations of neutral and phenolic organohalogens in pregnant women and some of their infants in The Netherlands. Environ Sci Technol 42: 3428-3433.

Noren K, Meironyte D, 2000. Certain organochlorine and organobromine contaminants in Swedish human milk in perspective of past 20-30 years. Chemosphere 40: 1111-1123.

Nyberg E, Awad R, Bignert A Ek C, Sallsten G. Benskin JP. (2018). Inter-individual, inter-city, and temporal trends of per- and polyfluoroalkyl substances in human milk from Swedish mothers between 1972 and 2016. Environ Sci: Processes Impacts 20: 1136-1147

OECD (2019). SF2.3: Age of mothers at childbirth and age-specific fertility. https://www.oecd.org/els/soc/SF_2_3_Age_mothers_childbirth.pdf. Accessed on December 16, 2019.

Olsen GW, Burris, JM, Ehresman DJ, Froehlich JW, Seacat AM, Butenhoff JL, Zobel LR (2007) Half-Life of Serum Elimination of Perfluorooctanesulfonate, Perfluorohexanesulfonate, and Perfluorooctanoate in Retired Fluorochemical Production Workers. Environ Health Perspect 115(9): 1298-1305.

Peeters D, Lanting CI, van Wouwe JP (2015). Peiling melkvoeding van zuigelingen 2015. TNO/CH 2015 R10385. TNO, Zeist, The Netherlands.

Nyberg E, Awad R, Bignert A, Ek C, Sallsten G, Benskin JP (2018). Inter-individual, inter-city, and temporal trends of per- and polyfluoroalkyl substances in human milk from Swedisch mothers between 1972 and 2016. Environ. Sci.: Process Impacts 20: 1136 – 1147.

Quinsey PM, Donohuye DC, Ahokas JT (1995). Persistence of Organochlorines in Breast Milk of Women in Victoria, Australia. Fd Chem Toxic 33(1): 49-56.

Raab U, Preiss U, Albrecht M, Shahin N, Parlar H, Fromme H (2008). Concentrations of polybrominated diphenylethers,organochlorine compounds and nitromusks in mother's milk from Germany (Bavaria). Chemosphere 72: 87–94.

Raab U, Albrecht M, Preiss U, Fromme H (2011). Levels of Organochlor Pesticides, PCB and PCDD/PCDF in Human Breast Milk - Results of the Bavarian Breast Milk Monitoring. Organohalogen Substances 73: 1614-1617.

RIVM (2003a). Rectification analysis results of flame retardants in human milk. RIVM letter report 245/03 LAC/RB, 15 April 2003.

RIVM (2003b). Analysis of brominated flame retardants in breast milk 1988, 1993 and 1998, RIVM letter report 344/03 LAC/RB, 19 June 2003.

RIVM (2004). Analysis of brominated flame retardants in breast milk 2003, RIVM letter report 01/04 LAC/BB/bb, 29 June 2004.

SCF, Scientific Committee on Food (2000). Opinion of the SCF on the Risk Assessment of Dioxins and Dioxin-like PCBs in Food. SCF/CS/CNTM/DIOXIN/8 Final.

SCF, Scientific Committee on Food (2001). Opinion of the Scientific Commission on Food on the Risk Assessment of Dioxinss and Dioxins-like PCBs in Food, Update based on new scientific information available since the adoption of the SCF opinion of the 22th November 2000. CS/CNTM/DIOXIN/20 final.

TNO, 2019. https://www.tno.nl/nl/aandachtsgebieden/gezond-leven/roadmaps/youth/groeidiagrammen-in-pdf-formaat. Accessed on December 16, 2019.

Tuinstra LGMT (1971). Organochlorine insecticide residues in human milk in the Leiden region. Netherlands Milk and Dairy Journal 25: 24-32.

Ulaszewska MM, Zuccato E, Davoli E (2011). PCDD/Fs and dioxin-like PCBs in human milk and estimation of infants' daily intake: A review. Chemosphere 83 (6): 774-782.

UNEP (2009). Global Monitoring Plan for Persistent Organic Pollutants Under the Stockholm Convention Article 16 on Effectiveness Evaluation: First regional Monitoring Report WEOG region.

UNEP (2013a). Guidance on the global monitoring plan for persistent organic pollutants. Geneva, UNEP, UNEP/POPS/COP.7/INF/39

UNEP (2015). Global Monitoring Plan for Persistent Organic Pollutants Under the Stockholm Convention Article 16 on Effectiveness Evaluation: Second regional Monitoring Report WEOG region.

UNEP (2016a). http://chm.pops.int/default.aspx Accessed January 20, 2020.

UNEP (2016b). http://www.pops-gmp.org/. Database at https://www.pops-gmp.org/index.php?pg=gmp-data-warehouse Accessed online on January 20, 2020.

UNEP (2017). Second Global Monitoring report. Geneva, UNEP/POPS/COP.8//INF/38. http://chm.pops.int/Implementation/GlobalMonitoringPlan/MonitoringReports/tabid/525/Default.aspx

US EPA (2012). EPA's Reanalysis of Key Issues Related to Dioxin Toxicity and Response to NAS Comments, Volume 1 (CAS No. 1746-01-6). EPA/600/R-10/038F.

US EPA (2016a). Health Effects Support Document for Perfluorooctane Sulfonate (PFOS). EPA 822-R-16-002.

US EPA (2016b). Health Effects Support Document for Perfluorooctanoic Acid (PFOA). EPA 822-R-16-003.

Van den Berg M, Birnbaum LS, Denison M, DeVito M, Farland W, Feeley M, Fiedler H, Hakansson H, Hanberg A, Haws L, Rose M, Safe S, Schrenk D, Tohyama C, Tritscher A, Tuomisto J, Tysklind M, Walker N, Peterson RE (2006). The 2005 World Health Organization reevaluation of human and mammalian toxic equivalency Factors for dioxins and dioxin-like substances. Toxicol Sci 93: 223–241.

Van den Berg M, Kypke K, Kotz A, Tritscher A, Yong Lee S, Magulova K, Fiedler H, Malisch R (2017). WHO/UNEP global surveys of PCDDs, PCDFs, PCBs and DDTs in human milk and benefit-risk evaluation of breastfeeding. Arch Toxicol 91(1): 83-96.

Van der Molen GW, Kooijman SALM, Slob W (1996). A generic toxicokinetic model for persistent lipophilic substances in humans: an application to TCDD. Fundam Appl Toxicol 31 (1): 83-94.

Van der Molen GW (1998) A Physiologically-Based Mathematical Model for the Long-Term Kinetics of Dioxins and Furans in Humans. Thesis, ISBN 90-9011844-6.

Molen, GW van der, Kooijman BALM, Wittsiepe J, Schrey P, Flesch-Janys D, Slob W (2000). Estimation of dioxin and furan elimination rates with a pharmacokinetic model. J Exp Anal Env Epidemiol 10: 579-585.

Van Leeuwen FXR, Malisch, R (2002). 3rd WHO-coordinated exposure study results of the third round of the WHO-coordinated exposure study on the levels of PCDDs and PCDFs in human milk. Organohalogen Substances 56: 311-316.

Vieth B, Albrecht M, Bruns-Weller E, Fromme H, Fürst P, Knoll A, Pydde E (2011). Time trends and current levels of dioxins and dioxin-like PCBs in human milk from Germany. Organohalogen Substances 73: 1559-1562.

Voelkel W, Genzel-Boroviczeny O, Demmelmair H, Gebauer C, Koletzko B, Twardella D, Raab U, Fromme H (2008). Perfluorooctane sulphonate (PFOS) and perfluorooctanoic acid (PFOA) in human breast milk: Results of a pilot study. Int J Hyg Environ Health 211: 440–446.

WHO (2000). Consultation on assessment of the health risk of dioxins; re-evaluation of the tolerable daily intake (TDI): Executive Summary. Food Addit & Contam 17 (4): 223-240.

WHO (2007). Fourth WHO-Coordinated Survey of Human Milk for Persistent Organic Pollutants in Cooperation with UNEP Guidelines for Developing a National Protocol (Revised 1 Oct 2007).

Zeilmaker MJ (2004). Analysis of dioxins and organochlore pesticides in breast milk 2003, RIVM letter report 041069/04. RIVM, Bilthoven, The Netherlands.

Zeilmaker MJ, Houweling DA, Cuijpers CEJ, Hoogerbrugge R, Baumann RA (2002). Verontreiniging van moedermelk met gechloreerde koolwaterstoffen in Nederland: niveaus in 1998 en tijd-trends. RIVM report 5291102012/2002. RIVM, Bilthoven, The Netherlands.

Zeilmaker MJ, Janssen P, Versteegh A, van Pul A, de Vries W, Bokkers B, Wuijts S, Oomen A, Herremans J. (2016). Risk assessment of the emission of PFOA: Location: Dupont/Chemours, Dordrecht, The Netherlands. RIVM Report 2016-0049. RIVM, Bilthoven, The Netherlands.

Zeilmaker MJ, Fragki S, Verbruggen EMJ, Bokkers BGH, Lijzen JPA (2018). Mixture exposure to PFAS: A Relative Potency Factor approach, RIVM report 2018-0070. RIVM, Bilthoven, The Netherlands.

8 Annexes

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Annex 1 Perfluor analytical protocol and results

ÖREBRO UNIVERSITY
SCHOOL OF SCIENCE
AND TECHNOLOGY

Örebro, 12 January 2016

Results report
PFOS human milk data

Summary:
Human milk data has been produced on perfluorooctane sulfonic acid (PFOS) which was added to the Stockholm Convention in 2009, and related perfluorinated alkyl acids.

The report includes one pooled human milk sample from the Netherlands.

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Results
Levels of perfluorooctane sulfonic acid (PFOS), perfluorooctanoic acid (PFOA) and perfluorohexanesulfonic acid (PFHxS) in ng/L milk are presented below. It should be noted that the linear PFOS isomer was measured, and that branched isomers were difficult to quantify with present method due to low PFOS levels in human milk.

MTM Code	Sample Code	L-PFOS	PFOA	PFHxS
DL14-015:12	140368983 Netherlands	45	<80	11

Other perfluorinated alkyl acids monitored were below there respective MDL as given below, in ng/L. The MDL for PFOS was 30 ng/L and 10 ng/L for PFHxS.

PFPeA	<30
PFHxA	<50
PFHpA	<20
PFNA	<25
PFDA	<50
PFUnDA	<50
PFBuS	<200
PFDS	<50

Materials and methods
The samples were extracted using weak anion exchange, solid-phase extraction (Waters Oasis® WAX)[5]. Labeled internal standards ($^{18}O_2$PFHxS, $^{13}C_4$PFOS, $^{13}C_4$PFOA) Wellington laboratories, Guelph, Canada) and 2 mL formic acid/water (1:1) were added to 1 mL milk. The solution was sonicated for 15 min and centrifuged at 10 000 x g for 30 minutes. The supernatant was extracted and the perfluorinated substances were eluted with 1 mL 2% ammonium hydroxide in methanol, after washing the sorbent with 2 mL sodium acetate buffer solution, pH 4, and 2 mL 40% methanol in water. The volume of the extracts was reduced to 30 µL by using nitrogen, and 20 µL 2 mM ammonium acetate in water was added. Performance standards, $^{13}C_8$PFOA, $^{13}C_8$PFOS, $^{13}C_3$PFHxS (Wellington laboratories, Guelph, Canada), were added to the extracts before injection. Analysis was performed using an Acquity UPLC coupled to a Xevo-TQS MS/MS (Waters Corporation, Milford, US) with an atmospheric electrospray interface operating in negative ion mode. Separation was performed on an Acquity BEH C18 2.1 x 100 mm, 1.7 µm kept at 50°C. An extra guard column (PFC isolator, Waters Corporation, Milford, US) was inserted between the pump and injector to trap contaminants originating from the LC system. Injection volume was 10 µL and the flow rate was set to 300 µL/min. A gradient program was employed delivering mobile phases consisted of 2 mM ammonium acetate in methanol, and 2 mM ammonium acetate in water.

Quality control and quality assurance
The method used has been validated and described earlier [1]. Concentration of the analytes in the samples was calculated using internal standard quantification. A minimum of five-point calibration curve was used. Two product ions were monitored for each substance and the ratio between the two product ions in the samples were

calculated and compared to an authentic standard, and did not exceed 50%. Recoveries of internal standards were monitored for each sample. The recovery of $^{13}C_4$PFOS was 75%, $^{13}C_4$PFOA 69% and $^{13}C_3$PFHxS 78% for the pooled sample. Reproducibility was assessed by including an in-house quality control sample that has been analyzed at 14 previous occasions. The comparison is shown below.

ng/L	**PFOS**	**PFOA**	**PFHxS**
n=14 (average±95%CI)	84 ± 3.3	159 ± 5.2	96 ± 2
This batch	95	169	104

One procedural blank was performed for each six milk samples. Milk levels are reported when the signal in the sample is higher than the average+ 3 standard deviations of the signal measured in procedural blanks. Further quality control includes successful participation in interlaboratory studies on PFASs in milk, one example is the 2009/2010[2] proficiency study.

References:

1. Kärrman, A., Ericson, I., van Bavel, B., Darnerud, PO, Aune, M. Lignell, S., Glynn, A., Lindström, G. 2007. *Environmental Health Perspectives* 115, 2:226-230.
2. 4th Fluoros Intercalibration 2010, coordinated by Gunilla Lindström and Stefan van Leeuwen.

Annex 2 Results of POP analyses in 2014

Chemisches und
Veterinäruntersuchungsamt Freiburg

Report of results: Basic POPs

Sample type: human breast milk
Country: Netherlands
Sample no. 140368983
Date: 18.09.14
Lipid content [%]: 3.0

		Concentration ng/g lipid weight
Aldrin		nd
Chlordane group	1)	2.4
alpha-chlordane		nd
gamma-chlordane		nd
oxy-chlordane		2.5
Trans-nonachlor		2.2
Dieldrin		2.1
DDT group	2)	94.9
o,p'-DDD		nd
p,p'-DDD		nd
o,p'-DDE		nd
p,p'-DDE		82.3
o,p'-DDT		nd
p,p'-DDT		3.1
Endrin group	3)	nd
Endrin		nd
Endrin ketone		nd
Heptachlor group	4)	2.2
Heptachlor		nd
Heptachlor-epoxide cis		2.3
Heptachlor-epoxide trans		nd
Hexachlorobenzene		9.5
Hexachlorocyclohexane (HCH) group		
alpha-HCH		nd
beta-HCH		6.9
gamma-HCH		nd
Parlar (toxaphene) group	5)	2.4
Parlar 26		0.6
Parlar 50		0.9
Parlar 62		0.8
Mirex		nd
Hexabrombiphenyl		nd
Pentachlorobenzene		nd

Chlordecone		nd
HBCD group	6)	0.6
alpha-HBCD		0.6
beta-HBCD		nd
gamma-HBCD		nd

Explanations:
nd = not detected (< 0.5 ng/g fat)
1) sum of alpha-chlordane, beta-chlordane and oxychlordane, calculated as chlordane
2) sum of o,p'-DDT, p,p'-DDT, p,p'-DDE and p,p'-DDD, calculated as DDT
3) sum of endrin and endrin ketone, calculated as endrin
4) sum of heptachlor and heptachlor-epoxid (cis/trans), calculated as heptachlor
5) sum of parlar 26, parlar 50 and parlar 62

HBCD group
nd = not detected (< 0.1 ng/g fat)
6) sum of alpha-HBCD, beta-HBCD and gamma-HBCD

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Report of results: PBDE

Sample type: human breast milk
Country: Netherlands
Sample no. 140368983
Date: 29/09/14
Lipid content [%]: 3.0

Polybrominated diphenyl ethers	**Concentration** pg/g lipid weight
BDE 17	6.9
BDE 28	29.7
BDE 47	492
BDE 66	5.9
BDE 99	132
BDE 100	163
BDE 138	< 14
BDE 153	741
BDE 154	21.3
BDE 183	< 66
Sum PBDE (lower bound)	1591

Explanations:
< [LOQ] Below limit of quantification (LOQ)

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Report of results: PCDD/Fs

Sample type: human breast milk
Country: Netherlands
Sample no. 140368983
Date: 18.09.14
Lipid content [%]: 3.0

2,3,7,8-substituted PCDF/PCDD	**Concentration** [pg/g lipid weight]	**MU**
2,3,7,8-TCDF	0.294	
1,2,3,7,8-PeCDF	0.153	
2,3,4,7,8-PeCDF	4.30	
1,2,3,4,7,8-HxCDF	1.20	
1,2,3,6,7,8-HxCDF	1.02	
2,3,4,6,7,8-HxCDF	0.477	
1,2,3,7,8,9-HxCDF	< 0.049	
1,2,3,4,6,7,8-HpCDF	1.03	
1,2,3,4,7,8,9-HpCDF	0.021	
OCDF	0.203	
2,3,7,8-TCDD	0.584	
1,2,3,7,8-PeCDD	1.61	
1,2,3,4,7,8-HxCDD	0.852	
1,2,3,6,7,8-HxCDD	4.41	
1,2,3,7,8,9-HxCDD	0.865	
1,2,3,4,6,7,8-HpCDD	4.86	
OCDD	40.0	
WHO-PCDD/F-TEQ (upperbound)	4.48	± 0.87
WHO-PCDD/F-TEQ (mediumbound)	4.48	± 0.87
WHO-PCDD/F-TEQ (lowerbound)	4.48	± 0.87

Explanations:

< [LOQ]	Below limit of quantification (LOQ)
Upperbound	Use of LOQ for the contribution of each non-quantified congener to the TEQ
Mediumbound	Use of half of LOQ for the contribution of each non-quantified congener to the TEQ
Lowerbound	Use of zero for the contribution of each non-quantified congener to the TEQ
MU	Expanded measurement uncertainty (level of confidence about 95 %)

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Chemisches und
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Report of results: PCB

Sample type: human breast milk
Country: Netherlands
Sample no. 140368983
Date: 18.09.14
Lipid content [%]: 3.0

Indicator PCB	**Concentration** [ng/g lipid weight]	**MU**
PCB 28	1.01	
PCB 52	0.227	
PCB 101	0.274	
PCB 138	10.3	
PCB 153	18.6	
PCB 180	9.69	
Summe Indikator PCB	40.1	
Mono-ortho PCB	[ng/g lipid weight]	
PCB 105	0.763	
PCB 114	0.177	
PCB 118	3.66	
PCB 123	0.040	
PCB 156	1.85	
PCB 157	0.301	
PCB 167	0.589	
PCB 189	0.192	
Non-ortho PCB	[ng/g lipid weight]	
PCB 77	0.007	
PCB 81	0.001	
PCB 126	0.021	
PCB 169	0.013	
	[pg/g lipid weight]	
WHO-mono-ortho PCB-TEQ	0.227	
WHO-non-ortho PCB-TEQ	2.45	
WHO-PCDD/F-TEQ (upperbound)	2.68	± 0.49
WHO-PCDD/F-TEQ (mediumbound)	2.68	± 0.49
WHO-PCDD/F-TEQ (lowerbound)	2.68	± 0.49

Explanations:

< [LOQ]	Below limit of quantification (LOQ)
Upperbound	Use of LOQ for the contribution of each non-quantified congener to the TEQ
Mediumbound	Use of half of LOQ for the contribution of each non-quantified congener to the TEQ

Lowerbound	Use of zero for the contribution of each non-quantified congener to the TEQ
MU	Expanded measurement uncertainty (level of confidence about 95 %)

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Report of results: WHO-TEQ

Sample type: human breast milk
Country: Netherlands
Sample no. 140368983
Date: 18.09.14
Lipid content [%]: 3.0

WHO-TEQ	**Concentration** [ng/g lipid weight]	**MU**
WHO-PCDD/F-TEQ (upperbound)	4.48	± 0.87
WHO-PCDD/F-TEQ (mediumbound)	4.48	± 0.87
WHO-PCDD/F-TEQ (lowerbound)	4.48	± 0.87
WHO-PCDD/F-TEQ (upperbound)	2.68	± 0.49
WHO-PCDD/F-TEQ (mediumbound)	2.68	± 0.49
WHO-PCDD/F-TEQ (lowerbound)	2.68	± 0.49
WHO-PCDD/F-TEQ (upperbound)	7.16	± 1.31
WHO-PCDD/F-TEQ (mediumbound)	7.16	± 1.31
WHO-PCDD/F-TEQ (lowerbound)	7.16	± 1.31

Explanations:

< [LOQ]	Below limit of quantification (LOQ)
Upperbound	Use of LOQ for the contribution of each non-quantified congener to the TEQ
Mediumbound	Use of half of LOQ for the contribution of each non-quantified congener to the TEQ
Lowerbound	Use of zero for the contribution of each non-quantified congener to the TEQ
MU	Expanded measurement uncertainty (level of confidence about 95 %)

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Report of results: endosulfan

Sample type: human breast milk
Country: Netherlands
Sample no. 140368983
Date: 18.09.14
Lipid content [%]: 3.0

		Concentration ng/g lipid weight
Endosulfan group	1)	nd
alpha-endosulfan		nd
beta-endosulfan		nd
Endosulfan sulfat		nd

Explanations:
nd = not detected (< 0.5 ng/g fat)
1) sum of alpha-endosulfan, beta-endosulfan, endosulfan sulfat

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Report of results: SCCP

Sample type: human breast milk
Country: Netherlands
Sample no. 140368983
Date: 18.09.14
Lipid content [%]: 3.0

		Concentration ng/g lipid weight
SCCP	1)	nd

Explanations:

1)nd = not detected (< 50 ng/g fat)
sum of short-chained chlorinated paraffins

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Annex 3 Analytics used for the determination of POPs in human milk in the different Dutch surveys.

year	Analytical technique	reference
1969	Column chromatography (to obtain 1 eluent) followed by GC-TECD	Tuinstra, 1971
1972	Column chromatography (to obtain different eluents containing the different POPs) followed by GC-MS	Greve, 1974
1983	Column chromatography (to obtain different eluents containing the different POPs) followed by GC-MS	Greve et al., 1985
1988	Column chromatography (to obtain different eluents containing the different POPs) followed by GC-MS	Albers et al., 1993
1993	GC-ECD for the OCPs and the mono- and di-ortho substituted PCBs GC-MS for the dioxins, furans and non-ortho substituted PCBs	Cuijpers et al., 1997
1998	GC-HRMs for PCDD/Fs and non-ortho PCBs GC-MS for OCPs, mono-ortho PCBs and indicator PCBs	Zeilmaker et al., 2002
2003	GC-HRMS for PCDD/Fs and non-ortho PCBs GC-MS for OCPs, mono-ortho PCBs and indicator PCBs	Zeilmaker et al., 2004
2014	GC-MS for PCDDs/PCDFs, coplanar PCBs and marker PCBs and other POPs by CVUA (Freiburg, Germany), Acquity UPLC coupled to a Xevo-TQS MS/MS with an atmospheric electrospray interface operating in negative ion mode for PFOS and and related perfluorinated alkyl acids by MTM Research Centre (Örebro University, Sweden)	Current study

GC = Gas-Chromatography; TECD = Tritium Electron Capture Detector; MS = Mass Spectrometry; HRMS = High Resolution Mass Spectrometry; OCP = organochlorine pesticide

Annex 4 POP modelling approach

A 4.1 One-compartment kinetic model

HBGVs for the dietary exposure of PBDEs, the perfluoro substances PFOA and PFOS and the dioxin 2378-TCDD all have been derived with the aid of a one-compartmental modeling approach (EFSA, 2011; ATSDR, 2015; Zeilmaker *et al.,* 2016; EFSA, 2018). Depending on the available toxicokinetic data a homogeneous (PBDEs) or non-inhomogeneous (PFOS; 2378-TCDD) distribution of a chemical in the body was assumed.

A 4.2 PBDEs and HBCDD

A 4.2.1 HBGV and corresponding sensitive human time window

HBGV BDE-153: 3.8 ng/kg bw/day (for specification, see Annex 4.2.3). The HBGV refers to neurodevelopmental disturbance resulting from prenatal, *in utero*, exposure or postnatal exposure from birth into adulthood. To prevent this effect to occur the BDE-153 whole body burden has to remain below the body burden corresponding with the HBGV (for specification, see Annex 4.2.3).

A 4.2.1 HBGV body burden simulation

Kinetic model: one-compartmental with homogenous body distribution (equation A4-5, see below).
Model parameters: F_{abs} =1, k_{el} = ln(2)*4530 day^{-1} (for specification, see Annex 4.2.3).

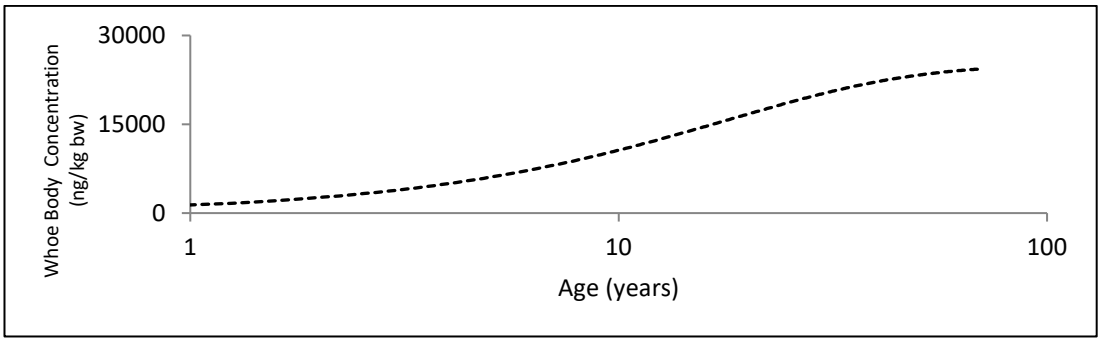


Figure A4-1 Simulation of the time-course of the human whole body concentration after lifelong, daily, intake of BDE-153 at the HBGV level of 3.8 ng (kg bw)$^{-1}$ day^{-1}, using equation A4-5. Maximal attainable whole body: 25 µg BDE-153/kg bw.

A 4.2.3 Kinetic model: specification

Defining the affinity of the blood and organ *i* by means of the "organ to blood" partition coefficient $p_i = \frac{C_i}{C_{blood}}$ it follows that:

$$A_{body} = C_{body} \cdot BW = C_{blood} \cdot (\sum_i p_i \cdot V_i + V_{Blood}) \text{ (A4-1)}$$

with:

BW	Body weight (kg)
V	blood/organ volume (L)

Homogenous distribution then is fulfilled when all organs have an "organ to blood" partition coefficient equal to one, thereby implying $C_{body} = C_{blood} = C_{organ}$.

Assuming the uptake from the gastrointestinal tract to occur via one generic absorption mechanism, instantaneous distribution over the body after uptake and elimination from the body via *i* parallel, fist-order, routes the rate of change of the *amount* a chemical in the body (A_{body}) then is:

$$\frac{d}{dt} A_{body} = F_{abs} \cdot D - \sum_i k_i \cdot A_{body} \text{ (A4-2)}$$

with:

D	daily intake (amount * day^{-1})
F_{abs}	fraction absorbed (no dimension; min: 0, max: 1)
$\sum_i k_i$	lumped whole body first order elimination rate constant (k_{el}, day^{-1})
t	exposure duration (days)

Dividing by body weight gives the corresponding rate of change of the *whole body concentration* (C_{body}), or:

$$\frac{d}{dt} C_{body} = F_{abs} \cdot D' - \sum_i k_i \cdot C_{body} \text{ (A4-3)}$$

with:

D´	daily intake (amount * (kg bw)$^{-1}$ * day^{-1})

Given a *repeated* daily oral intake D' the time-course of C_{body}, and hence C_{blood}, is given by:

$$C_{body}(t) = \frac{F_{abs} \cdot D'}{\sum_i k_i} \cdot (1 - e^{-\sum_i k_i \cdot t}) \quad \text{(A4-4)}$$

In the case of PBDEs only information on the overall whole body elimination half-life was available. Equation A4-4 therefore reduces to:

$$C_{body}(t) = \frac{F_{abs} \cdot D'}{k_{el}} \cdot (1 - e^{-k_{el} \cdot t}) \quad \text{(A4-5)}$$

For BDE-153 EFSA (2011a) provides a human elimination half-life 4530 days, corresponding with k_{el} = ln(2) *4530 day^{-1} and F_{abs} = 1 (assumption). Note that the latter is in concordance with the almost complete absorption of lipophilic POPs from breast milk (see section 4.4 below).

EFSA (2011a) also provided the following HBGVs for PBDEs, including BDE-153. In neonatal mice Post Natal Day 10 (PND 10) was found a sensitive time window for the induction of neurobehavioral toxicity (delayed habituation response, single p.o. PBDE dose) in this species. In the case of BDE-153 the toxicity data allowed a BenchMark Dose (BMD) analysis. This resulted in a $BMDL_{10}$ value of 83 µg (kg bw)$^{-1}$ for BDE-153 (effect size: 10 % decrease in the habituation response).
In the mouse PND 10 corresponds with the Brain Growth Spurt (BGS, for details see Eriksson, P., 1998, Perinatal Developmental Neurotoxicity of PCBs, Report 4897, Swedish Environmental Protection Agency, ISBN 91-620-4897-X/ISSN 0282-7298). In the mouse the BGS takes place during the first 3-4 weeks after birth, in order to peak at PND 10. However, in humans the peak of the BGS occurs in the third trimester of pregnancy, in order to gradually decrease afterwards until adulthood is reached.
Assuming 75% absorption ($F_{abs,a}$) the animal "body burden" (BB_a) on PND 10 corresponding with the $BMDL_{10}$ then is:

$$BB_a = F_{abs,a} \cdot BMDL_{10} \quad \text{(A4-6)}$$

In this way the following "body burden" was calculated: 62 µg (kg bw)$^{-1}$ (BDE-153).
Given one-compartmental "steady state" kinetic modelling the (average) life-long, daily human intake ($I_{d,h}$) leading to this "body burden" in the human body then can be calculated as:

$$BB_a = F_{abs,h} \cdot \frac{I_{d,h}}{k_{el,h}} \quad \text{(A4-7)}$$

or:

$$I_{d,h} = \frac{BB_a \cdot k_{el,h}}{F_{abs.h}} \text{ (A4-8)}$$

with $F_{abs,h}$ being the absorption fraction of BDEs from food in the human gastro-intestinal tract and $k_{el,h}$ being the PBDE specific rate constant for the removal from the body. In one-compartmental modelling $k_{el,h}$ relates to the time which is needed to remove half-the amount of a chemical from the human body, i.e. the experimentally observable half-life ($t_{1/2}$) as $k_{el,h} \cdot t_{1/2} = \ln 2$.
In this way values an $I_{d,h}$ of 9.6 ng/kg bw/day was calculated.

The calculated $I_{d,h}$ values were compared with the actual dietary long-term PBDE intake ($I_{c,h}$) by calculating the following Margin Of Exposure (MOE):

$$MOE = \frac{I_{d,h}}{I_{c,h}} \text{ (A4-9)}$$

Usually a MOE of 100, covering uncertainties and variability with respect to kinetic and dynamic differences between animal species and humans (factor 4 * 2.5 = 10) and within the human population (factor 3.2 * 3.2 = 10), is considered sufficient to conclude that there is no health concern. Since the MOE approach is based on a body burden comparison between animals and humans, the potential kinetic differences have been accounted for. Equally, by focusing on the body burden associated with a BMDL10 for neurobehavioral effects in mice induced during a relevant period for brain development, and applying this body burden to the entire life span in humans, individual difference in susceptibility has been covered. Therefore, the calculated MOE should be sufficient to cover Interspecies differences in sensitivity for the effects observed. This implies that an MOE larger than 2.5 might indicate that there is no health concern.

So, according to EFSA:

$$MOE \geq 2.5 \text{ (A4-10)}$$

or :

$$\frac{I_{d,h}}{I_{c,h}} \geq 2.5 \text{ (A4-11)}$$

Applying equation A4-11 on the calculated $I_{d,h}$ (see above) then results in a HBGV of 3.8 ng BDE-153 (kg bw)$^{-1}$ day^{-1} for BDE-153.
In a similar way HBGVs of 69 ng BDE-47 (kg bw)$^{-1}$ day^{-1}, 1.7 ng BDE-99 (kg bw)$^{-1}$ day^{-1} (EFSA, 2011a) and 375 ng HBCDD (kg bw)$^{-1}$ day^{-1} (EFSA, 2011b) were derived.

A 4.2.3 BDE-153 intake from breast milk

The daily intake was calculated on the basis of a milk intake of 800 mL day^{-1} containing 3% milk lipid (measured) and a concentration of 0.74 ng g^{-1} milk lipid (measured), an average body weight of 6.5 kg in the

period from birth to one year of age, i.e. the average of 3 kg at birth and 10 kg at one year of age, then results in a median, daily BDE-153 intake during first year of 0.74 * 0.03 * 800/6.5 = 2.73 ng (kg bw)$^{-1}$ day^{-1}.

The corresponding P95 value was estimated by multiplying this value with a factor of 2.3, i.e. the P95/P50 ratio in Dutch human milk 1998/2003 (see Annex 8), to be compared with a P95/P50 ratio of 2.0 in Belgium human milk, Croes *et al.*, Chemosphere, 2012, 89, 988 – 994). This resulted in an estimated P95 BDE-153 concentration of 1.70 ng g^{-1} milk lipid and an intake of 6.28 ng (kg bw)$^{-1}$ day^{-1}.

A 4.3 PFOS

A 4.3.1 HBGV and corresponding sensitive human time window

HBGV PFOS: 6.25 g bw day^{-1} (Zeilmaker et al.,2016; 2018).
The HBGV refers to liver toxicity (increased liver weight) resulting from life-long dietary daily exposure. To prevent this effect to occur, throughout life, the PFOS blood concentration has to remain under the serum concentration corresponding with the daily exposure to the HBGV.

A 4.3.2 HBGV serum concentration simulation

Kinetic model: one-compartmental with non-homogenous body distribution (equation A4-15, see below).
Model parameters: F_{abs} =1, k_{el} = ln(2) * 2000 day^{-1}, V_d = 0.2 L (kg bw)$^{-1}$ (for specification, see Annex 4.2.3).

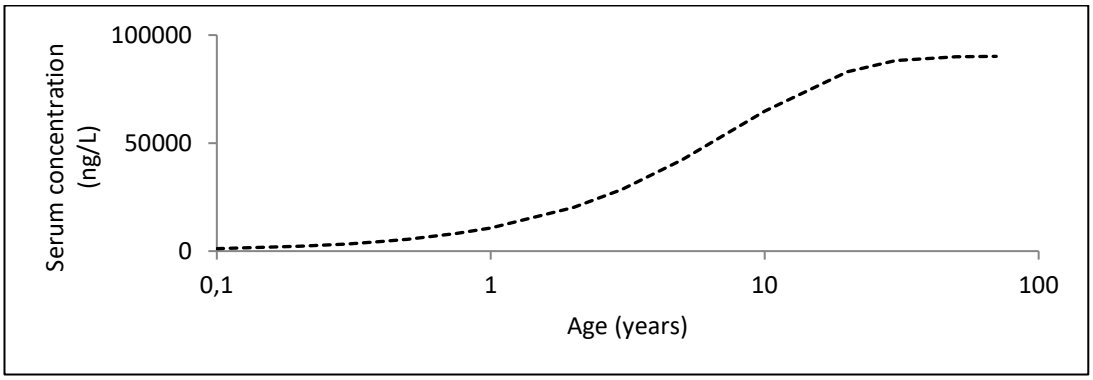


Figure A4-2 Simulation of the time-course of the human serum concentration after life-long, daily, intake of PFOS at the HBGV level of 6.25 ng (kg bw)$^{-1}$ day^{-1} using equation A4-15. Maximal attainable serum concentration: 90 µg PFOS/L serum.

A 4.3.3 Kinetic model: specification

In case of non-homogenous distribution $C_{body} \neq C_{blood} \neq C_{organ}$. Recalling $p_i = \frac{C_i}{C_{blood}}$ expressing the total amount in the body A_{body} then gives:

$$A_{Body} = C_{Body} \cdot BW = C_{Blood} \cdot (\sum_i p_i \cdot V_i + V_{blood}) \quad \text{(A4-12)}$$

Rewriting then gives:

$$C_{Body} = C_{Blood} \cdot \frac{(\sum_i p_i \cdot V_i + V_{blood})}{BW} \text{ (A4-13)}$$

Defining the Volume of Distribution V_d as $V_d = \frac{C_{body}}{C_{blood}} = \frac{(\sum_i p_i \cdot V_i + V_{blood})}{BW}$ (dimension: L (kg bw)$^{-1}$) it immediately follows that in case V_d > BW the body organs (on average) have a higher affinity for the chemical than the blood. In this case the concentration in the blood will be lower than in the organs. Such a situation is typical for lipophilic chemical such as dioxins. Reversely, V_d < BW indicates that the blood has a much higher affinity then the body organs. Hence the concentration in the blood will be higher than in body organs. This situation typically applies to chemicals which distribute in body water. However, the distribution of mobile perfluors like PFOS or PFOA also is in concordance with this principle.

Substituting $C_{body} = C_{blood} \cdot V_d$ into equation A4-4 then given the time-course for the concentration in the blood as:

$$C_{blood}(t) = \frac{F_{abs} \cdot D'}{\sum_i k_i \cdot V_d} \cdot (1 - e^{-\sum_i k_i \cdot t}) \text{ (A4-14)}$$

In the case of PFOS only information on the overall whole body elimination half-life was available. Equation A4-14 therefore reduces to:

$$C_{blood}(t) = \frac{F_{abs} \cdot D'}{k_e \cdot V_d} \cdot (1 - e^{-k_{el} \cdot t}) \text{ (A4-15)}$$

In the case of PFOS Olsen *et al.* (2007) mentions a characteristic whole body human elimination half-life of 2000 days ≈ 5.5 year to be compared with an arithmetic mean of 5.4 year (95% CI: 3.9-6.9 year) resp. geometric mean of 4.8 year (95% CI: 4.0 – 5.8 year whereas ATSDR (2015) provides a "body to serum" volume of distribution of 0.2 L/kg body weight. In concordance with ATSDR (2015) and Zeilmaker *et al.* (2016) the absorption from food was assumed complete (F_{abs} = 1).

A 4.3.4 PFOS intake from breast milk

The daily intake was calculated on the basis of a milk intake of 800 mL day^{-1} containing 3% milk lipid and a (measured) PFOS concentration of 45 ng L^{-1}, a body weight of 6.5 kg body weight in the period from birth to one year of age, i.e. the average of 3 kg at birth and 10 kg at one year of age, then results in a daily PFOS intake during first year of 45 *0.8/6.5 = 5.5 ng (kg bw)$^{-1}$ day^{-1}.

The corresponding P95 value 12.1 ng (kg bw)$^{-1}$ day^{-1} was estimated on the basis of a PFOS concentration of 99 ng L^{-1} as estimated from a

P95/P50 PFOS ratio of 2.2 in Belgium human milk (Croes *et al.*, Chemosphere, 2012, 89, 988 – 994).

Annex 4.4 Dioxin TEQ

A 4.4.1 HBGV and corresponding sensitive human time window

HBGV 2378-TCDD: 0.25 pg PCDD/PCDF-dl-PCB TEQ (kg bw)$^{-1}$ day^{-1} for dietary exposure (EFSA, 2018).
The HBGV refers to disturbed spermatogenesis at adult age which was causally linked to the PCDD/PCDF TEQ serum lipid at the age of 9 years, with serum levels exceeding 7 pg PCDD/PCDF TEQ g^{-1} serum lipid at the age of 9 years being indicative for disturbed spermatogenesis later in life.
A serum lipid level of 7 pg PCDD/PCDF TEQ g^{-1} serum lipid at the age of 9 years therefore was taken as the Point of Departure (PoD) for calculating the HBGV, *under condition* that boys have had a 1-year breastfeeding period consisting of 800 mL of breast milk containing 3.5% lipid and a (constant) concentration of 5.9 pg PCDD/PCDF/dl-PCB TEQ g^{-1} milk lipid.

In women the HBGV of 0.25 pg PCDD/PCDF-dl-PCB TEQ (kg bw)$^{-1}$ day^{-1} will lead to a body burden in women of 5.9 pg PCDD/PCDF-dl-PCB TEQ g^{-1} milk lipid at reproductive age. Feeding such breast milk to newborn boys for one year, followed by an eight year exposure period of twice the HBGV, i.e. 0.5 pg PCDD/PCDF-dl-PCB TEQ (kg bw)$^{-1}$ day^{-1}, in boys leads to a serum lipid concentration of 7 pg PCDD/PCDF/dl-PCB TEQ g^{-1} serum lipid at the age of 9 years. An exposure twice the HBGV was chosen here in order to compensate for the relative high intake of growing children in comparison with full grown adults.

Note that the epidemiological study design supplying the PoD is limited to identifying *a* narrow sensitive time window just around 9 years of age.

A 4.4.2 HBGV blood lipid simulation

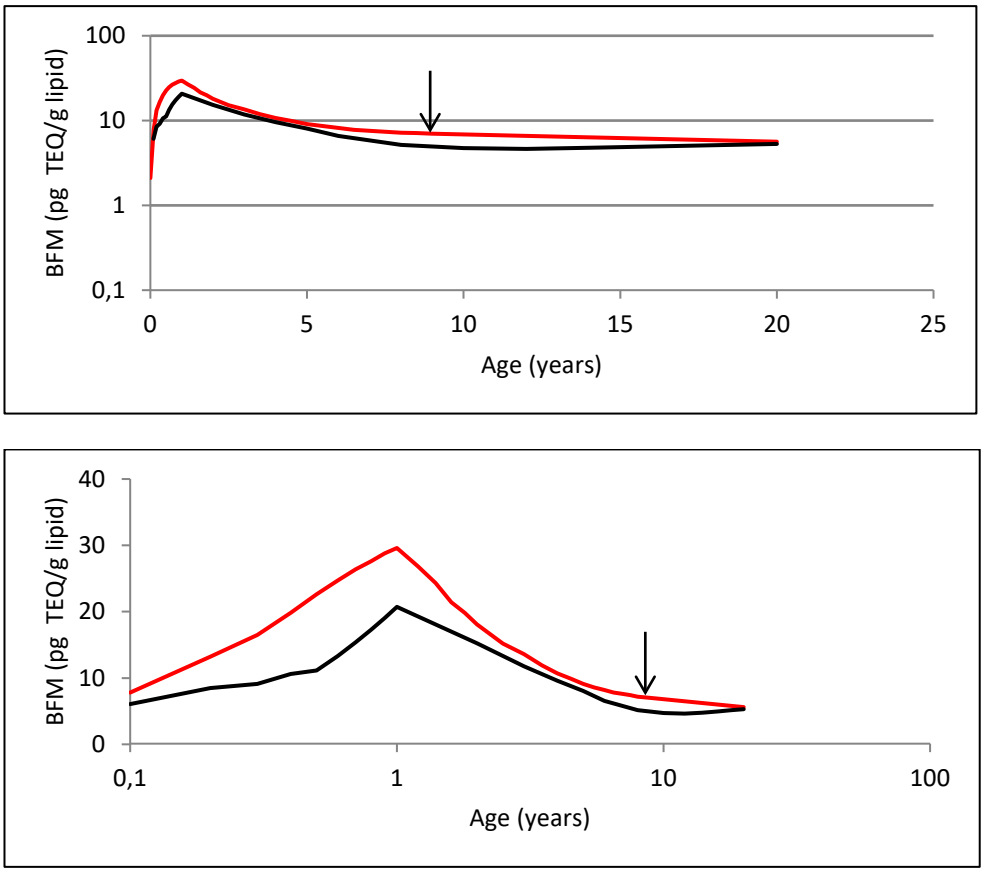


Figure A4-3 Simulation of the time-course of the human TEQ serum lipid concentration after a 1-year breast feeding period, followed by an 8-year dietary exposure at the level of two-times EFSA's HBGV (0.25 pg PCDD/PCDF/dl-PCB TEQ/kg bw/day) and an 11-year exposure period at the HBGV. A: log-linear; B: linear-log. Red line: Carrier model (EFSA, 2018; Figure 14, breast feeding: 800 mL/day, assumed 3.5% milk lipid, 5.9 pg PCDD/PCDF/dl-PCB TEQ/ milk lipid. Black line: RIVM model (this report, breast feeding: 800 mL/day, measured 3% milk lipid,7.2 pg PCDD/PCDF/dl-PCB TEQ/ milk lipid). RIVM data calculated with equation A4-18. Arrow: EFSA serum Bench Mark of 7 pg TEQ/g serum lipid at the age of 9 years indicative for disturbance of adult spermatogenesis at young adult age.

A 4.4.3 Model specification

In the case of dioxins, the concentration in the body lipid cq. serum lipid is the starting point for dioxin risk assessment (Van der Molen *et al.*, 1996; Kreuzer *et al.*, 1997; US EPA, 2012; Béchaux *et al.* 2014; EFSA, 2018). For this reason previous risk assessments of dioxins, i.e. 2378-TCDD, focused on the toxicokinetic modeling of this compound in serum lipid of neonates (Kreuzer *et al.*, 1997) or adults (Carrier *et al.*, 1995a,b; Van der Molen *et al.* 1996; 2000). Without exception this modelling described the metabolic elimination of 2378-TCDD from the body to scale in concordance to liver weight. Furthermore, in the case of adults, the age-dependent effect of the growth of body lipid on the body lipid concentration was taken into account, reflecting the so-called "apparent elimination by dilution" effect (Carrier *et al.*, 1995a,b; Van der

Molen *et al.*, 1998, 2000). However, in modelling the kinetics of neonates neither Kreuzer *et al.*, (1997) nor EFSA (2018, using a modified Carrier model) incorporated the effect of "apparent elimination by dilution" as occurring in neonates, thereby overestimating the neonatal 2378-TCDD concentration in body lipid and, hence, its corresponding toxic risk. Finally, Kreuzer *et al.* (1997) and Van der Molen (1998, 2000) illustrated the importance of fecal lipid excretion as route of elimination. In contrast to Carrier *et al.* (1995) neither the Kreuzer nor the Van der Molen modelling concept takes, next to lipid partitioning, hepatic sequestration as a dioxin distribution process into account. However, at low exposure hepatic sequestration may be neglected.

This report combines the modelling concepts of Carrier, Kreuzer and Van der Molen, as it describes the human kinetics of 2378-TCDD by means of one-compartmental kinetics incorporating hepatic elimination, fecal elimination and the "apparent elimination by dilution" effect, i.e. taking the specific neonatal growth of body lipid, referred to as "Body fat mass (BFM), into account.

Simulated BFM concentrations were compared with serum lipid concentrations found associated with the disturbance of semen quality at adult age (7 pg TEQ g^{-1} lipid at the age of 9 years, EFSA, 2018).

Absorption from breast milk
McLachlan (1993) determined the 12-day mass-balance, i.e. the difference between the total intake with breast milk and the excretion in the feces, in a 19 week old boy for 12 dibenzofurans and dibenzo-p-dioxins and 4 polychlorinated biphenyls. 2378-TCDD, penta (23478-PeCDF, 12378-PeCDD), and hexa-substituted dioxins/furans (123478-HxCDF, 123678-HxCDF, 234678-HxCDF, 123478-HxCDD, 123678-HxCDD, 123789-HxCDD) showed an absorption of 90 % or higher. The absorption of 1234678-HpCDD and 1234678-HpCDF and OCDD was found to be lower, i.e. 61/58% resp. 23%.The absorption of 2,2',4,4',5-PCB, 2,2',4,4',5,5'-PCB, 2,2',3,4,4',5'-PCB and 2,2',3,4,4',5,5'-PCB ranged from 95-96%.
Dahl *et al.* (1995) determined the 48-hour mass-balance for seven dibenzo-p-dioxins (2378-TCDD, 12378-PeCDD, 123478-HxCDD, 123678-HxCDD, 123789-HxCDD, 1234678-HpCDD, OCDD), six dibenzofurans (2378-TCDDF, 12378-PeCDF, 23478-PeCDF, 123478-HxCDF, 123678-HxCDF, 1234678-HpCDF) and three polychlorinated biphenyls (PCBs -77, 126 and 169) in four breast fed children at 1, 2, 3 and 6 months postpartum. For all tetra-, penta- and hexa-substituted dioxins/furans and biphenyls congeners the absorption was found to be over 95%. The absorption of 1234678-HpCDD/HpCDF and OCDD was found to be somewhat lower, i.e. 80/93% resp. 87%.
Abraham *et al.* (1996) determined the 5-day mass balance for four dibenzo-p-dioxins (2378-TCDD, 123678-HxCDD, 1234678-HpCDD, OCDD) and two dibenzofurans (23478-PeCDD, 12378-PeCDD) and their I-TEQ (as predessessor of the WHO_{2005} TEQ) as well as the dietary fat intake in two breast fed children at 1 and 5 months of age. The absorption of dietary fat was found to be 95% or higher. For 2378-TCDD and the I-TEQ the absorption was found to be 94% or higher resp. 91%

or higher. The absorption of 1234678-HpCDD and OCDD was found to be somewhat lower, i.e. 78% resp. 51%.
The results clearly indicated that the absorption of dioxin-like substances occurs via fat absorption (see Figure A4-4).

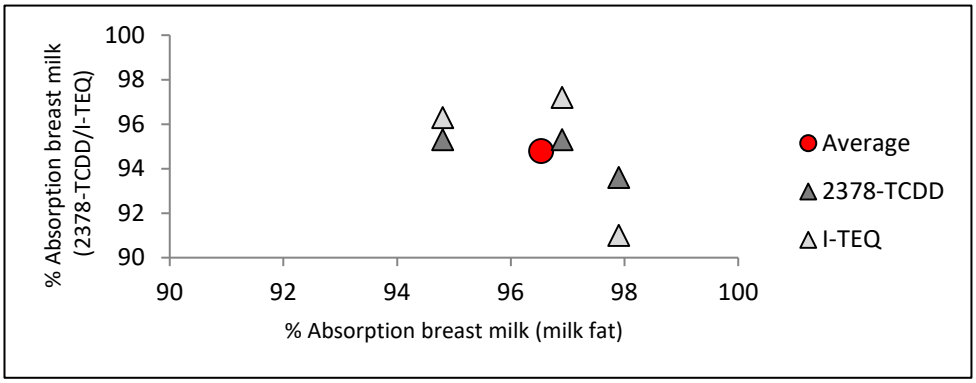


Figure A4-4 Percentage absorption of breast milk fat (x-axis) vs. % absorption of breast milk of 2378-TCDD or I-TEQ (y-axis). Grey symbols: measured values; Red symbol: average value (numerical: 97; 95). Experimental data calculated from Abraham et al. (1996).

Body Fat Mass: Age dependent half-life of 2378-TCDD
In mammals the total amount of dioxins in the body, i.e. the body burden, consists of the sum of the accumulated amounts in the adipose tissue and the liver, the latter being known as hepatic sequestration. Hepatic sequestration is an inducible process under the control of the dioxin Ah-receptor binding, followed by induction of P450 proteins (in particular P450 1A2) followed by binding of dioxins to basal and inducible P450 protein. In contrast to lipid accumulation, hepatic sequestration therefore shows clear dose-dependency. For example, in the rat inducible hepatic sequestration is observed after a single sc. ranging from 3 – 3000 ng 2378-TCDD (kg bw)$^{-1}$ (Abraham *et al.*, 1988. Similarly, for chronic dosing this range is 3.5 – 125 ng 2378-TCDD (kg bw)$^{-1}$ (Tritcher *et al.* 1992). In humans, *poisoning* cases have clearly shown hepatic sequestration to occur too. A (retrospective) kinetic analysis of 23478-PeCDF poisoning revealed significant hepatic sequestration in a patient at an (estimated) dose of 50 ng of 23478-PeCDF (kg bw)$^{-1}$ day^{-1} for an exposure period of 38 weeks (Carrier *et al.*, 1995a,b). However, this analysis also revealed that at background exposure level hepatic sequestration was negligible, i.e. the fraction of the body burden residing in the liver amounting around 1% ($f_h^{\min}$ = 0.009, Carrier *et al.*, 1995b, Figure 4), and 99% in the adipose tissue (Carrier *et al.*, 1995b, Table 4, Time weeks: 0; $C_h(0)$ = 0.35 ng kg^{-1}; $C_{at}(0)$= 4.85 ng kg^{-1}, v_h = 0.03; v_{at} = 0.20, resulting in $C_b(0) \approx$ 1.0 ng kg^{-1}). From this it can be concluded that in humans chronically exposed to dioxin in the range of pg TEQ (kg bw)$^{-1}$ day^{-1} hepatic sequestration is negligible, with dioxins virtually residing in the body's Body Fat Mass (BFM) (Van den Berg et al., 1994; Van Ede *et al.*, 2013). Note that this conclusion only holds for low background exposure and does not hold for incidental high exposure. Indeed in humans BFM (inversely) relates to elimination from the body, with the whole body half-life increasing with increasing BFM (Van der Molen *et al.,* 1996, 2000; Michalek *et al*., 1999, 2002; Kreuzer *et al.*, 1997; Milbrath *et al.*, 2009) and (hence) age in

children as well as in adults (Van der Molen *et al*., 1996, 2000; Kerger *et al.*, 2006; Milbrath *et al.*, 2009). Note that this identified the BFM as dioxin's Volume of Distribution at low background exposure.

Adopting lipid partitioning as the low dose dioxin distribution mechanism in the human body "true" elimination from the body occurs by hepatic metabolism and by excretion via fecal fat. In this context Kreuzer *et al.* (1997) provides an expression for 2378-TCDD's BFM half-life based due to these (kinetically parallel) processes, i.e.:

$$t_{1/2} = \frac{1}{\frac{1}{tm_{1/2}} + \frac{1}{tf_{1/2}}} \qquad \text{(A4-16)}$$

with:
$t_{1/2}$ BFM half-life
$tm_{1/2}$ BFM half-life due to hepatic metabolism
$tf_{1/2}$ BFM half-life due to excretion of fecal fat

However, this expression does *not* yet incorporate "apparent elimination" due to an increasing BFM, resulting in the so-called "elimination through dilution", a process applying throughout life, but in particular relevance to neonates (De Bruin *et al*., 1996; Carberry *et al.*, 2010; Wells, 2014). For in neonates the BFM increases manifold, i.e. up to 6-fold, in the first six months following birth (de Bruin *et al.*(1996). For this reason modelling an infant's BFM half-life which only depends on hepatic metabolism and fecal excretion grossly overestimates 2378-TCDD's BFM accumulation resulting from breast feeding in neonates.

For this reason Kreuzer's expression for the 2378-TCDD's BFM half-life was extended with "apparent" elimination resulting from the increase of the BFM:

$$t_{1/2} = \frac{1}{\frac{1}{tm_{1/2}} + \frac{1}{tf_{1/2}} + \frac{1}{tbfm_{1/2}}} \quad \text{(A4-17)}$$

with $tbfm_{1/2}$ the half-life due to BFM growth .
In the case of 2378-TCDD the time-course in the blood then reduces to:

$$C_{blood}(t) = \frac{F_{abs} \cdot D^{'}}{(k_1 + k_2 + k_3) \cdot V_d} \cdot (1 - e^{-(k_1+k_2+k_3)\cdot t}) \text{ (A4-18)}$$

Note that in equation A4- 18 the Volume of Distribution V_d refers to the BFM.

Contribution of metabolic clearance to BFM half-life ($tm_{1/2}$)
Kreuzer *et al.* (1997) provide the age-dependency of BMF half-life due to hepatic metabolism as:

$$tm_{1/2} = tm_{1/2,40} \cdot \frac{V(t)}{V_{40}} \cdot \left(\frac{V_{L,40}}{V_L(t)}\right)^{2/3} \text{ (A4-19)}$$

with:

$tm_{1/2,40}$	BFM half-life due to hepatic metabolism in a 40 year old adult, i.e. 10.5 year (reference value)
$V(t)$	BFM volume at age *t*
V_{40}	BFM volume at 40 years of age (reference value)
$V_{L,40}$	Liver volume at 40 years of age (reference value)
V_L(t)	Liver volume at age *t*

Assuming liver volume to be a constant fraction of body weight then gives:

$$tm_{1/2} = tm_{1/2,40} \cdot \frac{V(t)}{V_{40}} \cdot (\frac{BW_{40}}{BW(t)})^{2/3} \quad \text{(A4-20)}$$

Given the age dependent increase of the BFM (De Bruin *et al*., 1996) and body weight (see Figure A4-5) as input V_{40} was calculated at 14380 g. This then allowed for the calculation of the age-dependency of the BMF half-life due to hepatic clearance (see Figure A4-6).

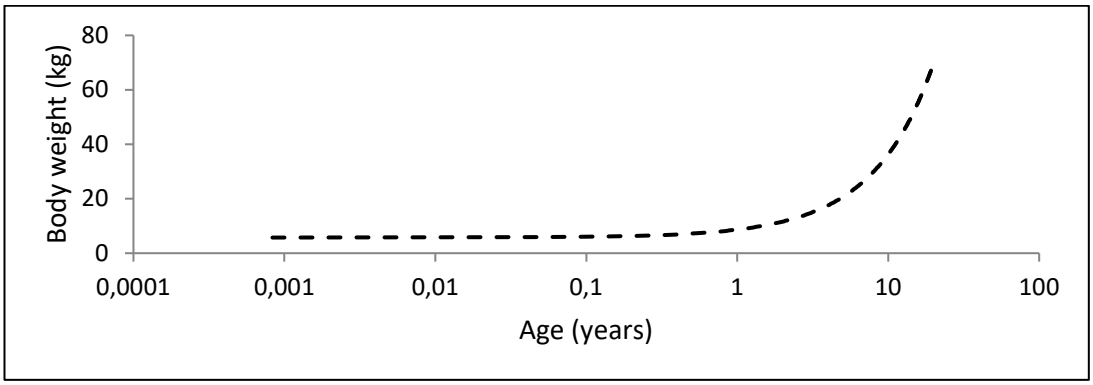


Figure A4-5 Growth curve of Dutch boys (0-20 years; source: TNO, The Netherlands). Adult body weight (20-70 years) assumed constant at 70 kg.

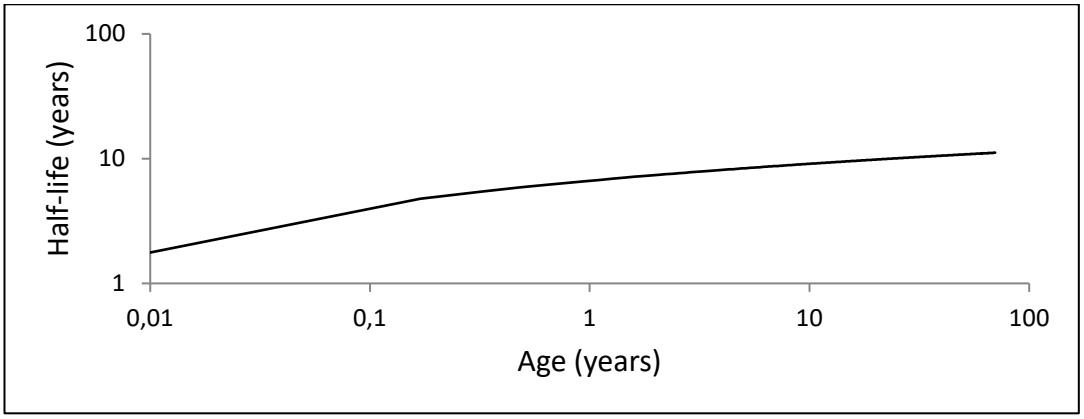


Figure A4-6 Simulation of the age dependency of the BFM half-life of 2378-TCDD resulting from hepatic clearance after Kreuzer et al. (1997).

Example calculations
V_{birth}= 350 g; V_{40} = 14380 g, BW_{40} = 70 kg, BW_{birth}= 3.5 kg, $tm_{1/2,40}$ = 10.5 years, $tm_{1/2}$ = 1.9 year
V_{70}= 17500 g; V_{40} = 14380 g, BW_{40} = 70 kg, BW_{70} = 70 kg, $tm_{1/2,40}$ = 10.5 years, $tm_{1/2}$ = 12.8 year

Contribution of fecal lipid excretion to BFM half-life ($tf_{1/2}$)
Abraham *et al.* (1996) mentions a fecal lipid excretion rate of 0.5 g lipid/day for newborns, whereas Kreuzer *et al.* (1996) mentions 3 g lipid/day for infants and 5 g lipid/day for adults.
In concordance with these references the following linear increases were used: 0.5 g lipid/day to 1.0 g lipid/day during the first year of life, 1.0 to 3.0 g lipid/day during the age period of 1-5 years, 3.0 g lipid/day to 5.0 g lipid/day during the age period of 5-20 years and 5 g lipid/day for adult age.
Given the BFM at different ages and the BFM half-life due to fecal elimination as defined in Kreuzer *et al.* (1996) this corresponds with a half-life of 1.3 years at birth, to increase linearly to 4.8 years at the age of 6 months, while remaining virtually constant at this value afterwards.

Contribution of BFM growth to BFM half-life ($tbfm_{1/2}$)
De Bruin *et al.* (1996) and Wells *et al.* (2014) provide detailed information on the age dependency of the (measured) BFM in (average) newborns, infants and young children. For example, de Bruin *et al.* and Wells *et al.* mention the BFM in newborn boys to amount around 350 g, to increase linearly to around 2250 g at the age of 6 months, to linearly increase further to 2510 g at the age of 12 months, 3280 g at the age of 5 years and 12.3 kg at the age of 20 years. From the age of 20 up to the age of 70 years the BFM was assumed to linearly increase further to 17.5 kg at the age of 70 years.

The available data allowed for the calculation of the apparent elimination half-life resulting from the growth of the BFM as a function of age. For example, estimating the BFM at birth at 350 g and 729 g at the age of 0.1 year results in an apparent "elimination" rate constant of k_{el} = $-\ln(350/729) \approx 7.35$ $year^{-1}$, corresponding with a half-life of 0.092 year over the first 0.1 years of life. Analogous the age dependency of the apparent half-life due to BFM growth was calculated (see Figure A4-7).

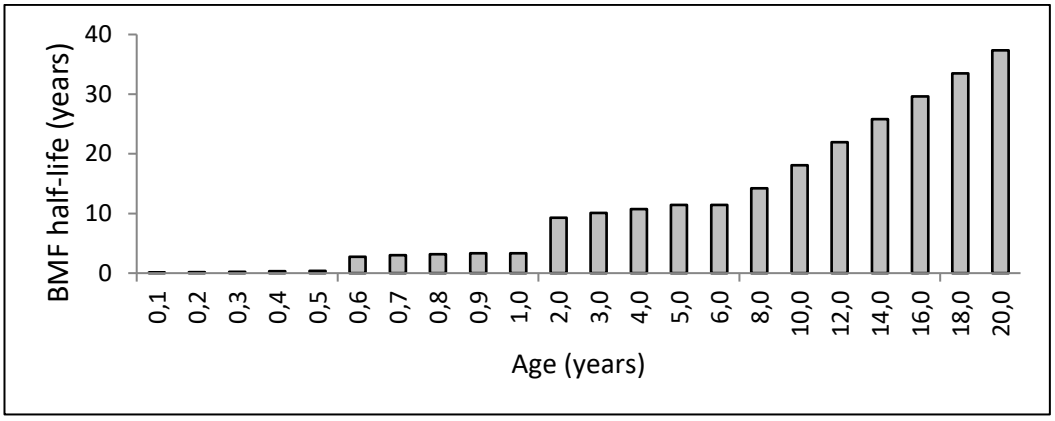


Figure A4-7 Simulation of the age dependency of the BFM half-life of 2378-TCDD resulting from its growth after De Bruin et al.(1996) and Wells et al. (2014).

BFM half-life ($t_{1/2}$)
Figure A4-8 shows the age-dependency of the BFM half-life of 2378-TCDD resulting from is hepatic clearance, fecal excretion and growth of the BFM.

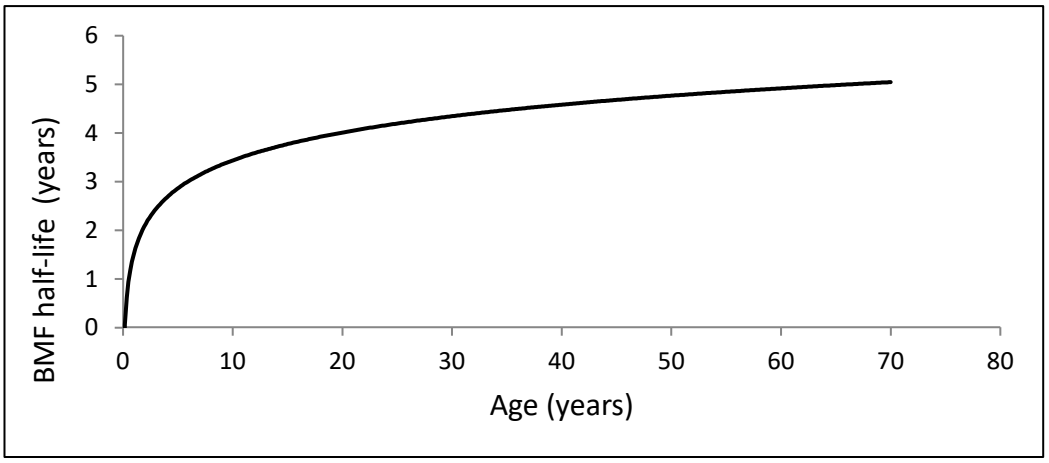


Figure A4-8 Simulation of the age dependency of the BFM half-life of 2378-TCDD resulting from the combined of hepatic clearance, fecal lipid clearance and growth of the BFM . 97). Note that the simulation holds for the average human, i.e. the average hepatic clearance, fecal clearance and BFM growth. For a 70 year old a BFM of 17.5 kg was used, corresponding with a BMI of 25% at a body weight of 70 kg.

Calculation of $t_{1/2}$: Experimental verification
Kreuzer *et al.* (1997) provide data on the levels of 2378-TCDD and I-TEQ in adipose tissue fat and liver fat in 3 still born children and 8 non-breast fed Sudden Death infant aged 0.4 – 26 weeks. Assuming the levels in the still borns as representative for the levels at birth allows for a kinetic analysis of the data (see Figure A4-9). This analysis resulted in half-lifes of 0.33 resp. 0.34 year for 2378-TCDD and I-TEQ in the BFM.

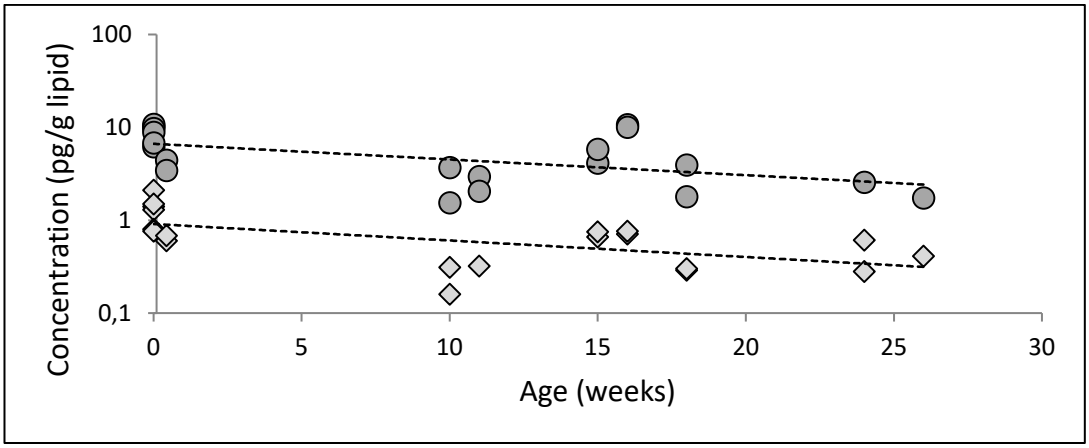


Figure A4-9 The decrease in the concentration of 2378-TCDD and I-TEQ in adipose tissue lipid and liver tissue lipid in neonates (experimental data obtained from Kreuzer et al., 1997). Light grey symbols: TCDD (half-life: 16.9 weeks) ; Dark grey symbols: I-TEQ (half-life: 17.7 weeks).

Table A4-1 presents the verification of the calculated $t_{1/2}$ on reported human half-lives of 2378-TCDD .

Table A4-12378-TCDD: Reported experimental half-life (years)

	Calculated	**Experimental (reference)**
Infant (5 months)	0.33	0.33-0.34[h] (Kreuzer *et al.*, 1997) 0.36-0.43[i] (Leung *et al.*, 2006)
Children 13.1 ± 5.5 years 19.1 ± 7.4 years 15.6 ± 6.9 years	 3.6 4.0 3.8	 1.8 + 1.1[b] (Kerger *et al.*, 2006) 2.8 ± 1.2[c] (Kerger *et al.*, 2006) 2.2 ± 1.1[d] (Kerger *et al.*, 2006)
Adults 24.6 ± 5.3 years 42.1 ± 7.4 years 68.5 ± 7.0 years	 4.1 4.4 4.9	 3.9 ± 1.4[e] (Kerger *et al.*, 2006) 6.9 (range: 4.6 – 13.9)[f] (Michalek (1999, 2002) 6.5 (P25: 5.0; P95: 8.2)[g] (Aylward *et al.*, 2013)

[b] BMI: 16.9 ± 1.7; [c]BMI: 22.9 ± 2.2; [d] BMI: 19.5 ± 2.9; [e] BMI: 28.3 ± 4.3; [f] male, 25 < BMI < 45; [g] male, BMI = 32.1 ± 4.6; [h] for analysis, see Figure A4-9; [i] Note that similar half-lives were found for other dioxin like substances: 12378-PeCDD 0.28 - 0.36 year, 23678-HeCDD 0.33 – 0.44 year, 1234678-HpCDD 0.28 – 0.36 year, OCDD 0.42 - 0.50 year, 23478-PeCDF 0.23 – 0.30 year.

Annex 4.4.4 Dioxin intake from breast milk

The daily intake was calculated on the basis of a milk intake of 800 mL day^{-1} containing a measured 3% milk lipid, and a concentration of 7.2 pg PCDD/PCDF-dl-PCB TEQ g^{-1} lipid and an average body weight of 6.5 kg body weight in the period from birth to one year of age. This then results in a daily dioxin intake during the first year of 7.2 x 0.03 x 800/6.5 = 26.6 pg PCDD/PCDF/dl-PCB TEQ (kg bw)$^{-1}$ day^{-1}.
Based on a P95/P50 ratio of 1.7 in Dutch human milk in 2003 (see Annex 9, to be compared with a P95/P50 ratio of 1.8 in Belgium human

milk, Croes *et al.*, Chemosphere, 2012, 89, 988 – 994), a P95 intake of 12.4/7.2 x 26.6 = 45.8 pg PCDD/PCDF/dl-PCB TEQ (kg bw)$^{-1}$ day^{-1} was calculated.
Children received a dietary exposure equal to 0.5 pg (kg bw)$^{-1}$ day^{-1}, i.e. two times the HBGV, from 1 year to 9 years of age, i.e. after the breastfeeding period had stopped. Thereafter the dietary exposure was reset at the level of the HBGV of 0.25 pg PCDD/PCDF-/dl-PCB TEQ (kg bw)$^{-1}$ day^{-1}. This exposure protocol is in concordance with the one used by EFSA (2018) in deriving the HBGV for dioxins.

References

Abraham K, Knoll A, Ende M, Päpke O, Helge H (1996).Intake, Fecal excretion, and body burden of Polychlorinated Dibenzo-*p*-dioxins and Dibenzofurans in breast-fed and formula fed infants. Pediatr Res, 40(5): 671 – 679.

Abraham K, Krowke R, Neubert D (1988). Pharmacokinetics and biological activity of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin. 1. Dose-dependent tissue distribution and induction of hepatic ethoxyresorufin O-deethylase in rats following a single injection. Arch Toxicol, 62: 359-368.

Aylward LL, Collins JJ, Bodner KM, Wilken M, Bodnar CM (2013). Elimination rates of dioxin congeners in former chlorophenol workers from Midland, Michigan. Env Health Perspect, 121(1): 39-45.

Carrier G, Brunet RC, Brodeur J (1995a). Modeling of the toxicokinetics of polychlorinated dibenzo-*p*-dioxins and dibenzofurans in mammalians, including humans. I. Nonlinear distribution of PCDD/PCDF body burden between liver and adipose tissue. Toxicol. Appl. Pharmacol., 131: 253-266.

Carrier G, Brunet RC, Brodeur J (1995b). Modeling of the toxicokinetics of polychlorinated dibenzo-*p*-dioxins and dibenzofurans in mammalians, including humans. II. Kinetics of absorption and disposition of PCDDs/PCDFs. Toxicol Appl Pharmacol, 131: 267-276.

Berg M van den, Jongh J de, Poiger H, Olson JR (1994). The toxicokinetics and metabolism of Polychlorinated Dibenzo-*p*-Dioxins (PCDDs) and Dibenzofurans (PCDFs) and their relevance to toxicity. Crit Rev Toxicol, 24(1):1-74.

Bruin NC de, Velthoven KAM van, Ridder M, Stijnen T, Juttman RE, Degenhart HJ, Visser HKA (1996). Standards for total body fat and fat-free mass in infants. Arch Disease in Childhood, 74: 386-399.

Carberry AE, Colditz PB, Lingwood BE (2010). Body composition from birth to 4.5 months in infants born to non-obese women. Pediatr Res, 68(1): 84-88.

Dahl P, Lindström G, Wiberg K, Rappe C (1995). Absorption of polychlorinated biphenyls, dibenzo-p-dioxins and dibenzofurans by breast fed children. Chemosphere, 30(12): 2297-2306.

Ede KI van, Aylward LL, Andersson PL, Berg M van den, Duursen MBM van (2013). Tissue distribution of dioxin like compounds: Potential impacts on systemic relative potency estimates. Toxicol Letters, 220: 294-302.

Kerger BD, Leung H-W, Scott P, Paustenbach DJ, Needham LL, Patterson DG, Gerthoux PM, Mocarelli P (2006). Age- and concentration-dependent elimination half-life of 2,3,7,8-Tetrachloro-*p*-dioxin in Seveso Children. Env Health Perspect, 114(10): 1596 – 1602.

Kreuzer PE, Csanády GA, Baur C, Kessler W, Päpke O, Greim H, Filser JG (1997). 2,3,7,8-Tetrachlorodibenzo-*p*-dioxin (TCDD) and congeners in infants. A toxicokinetic model of human lifetime body burden by TCDD with special emphasis on its uptake by nutrition. Arch Toxicol, 71: 383-400.

Leung H-W, Kerger BD, Paustenbach DJ (2006). Elimination half-lifes of selected polychlorinated dibenzodioxins and dibensofurans in breast-fed human infants. J Toxicol Env Health, Part A, 69: 437 – 443.

Schrenk D, LIpp H-P, Weimüller T, Hagenmaier H, Bock KW (1991). Assessment of biological activities of mixtures of polychlorinated dibenzo-p-dioxins: comparison between defined mixtures and their constituents. Arch Toxicol, 65: 114-118.

Thoma H (1989). Polychlordibenzo-p-dioxine und Polychlordibenzofurane in Humanproben Z Umweltchem Ökotox 1: 25-26.

Thoma H, Mücke W, Kretschmer E (1989). Concentrations of PCDD and PCDF in human fat and liver samples. Chemosphere, 18: 491-498.

Thoma H, Mücke W, Kauert G (1990). Comparison of the polychlorinated dibenzo-p-dioxin and dibenzofuran in human tissue and human liver. Chemosphere, 20: 433-442.

Tritscher AM, Goldstein JA, Portier CJ, McCoy Z, Clark GC, Lucier G (1992). Dose-response relationships for chronic exposure to 2,3,7,8-tetrachlorodibenzo-*p*-dioxin in a rat tumor promotion model: Quantification and immunolocalization of CYP1A1 and CYP1A2 in the liver. Cancer Res, 52: 3436-3442.

McLachlan MS (1993). Digestive tract absorption of polychlorinated dibenzo-*p*-dioxins, dibenzofurans, and biphenyls in a nursing infant. Toxicol Appl Pharmacol, 123: 68-72.

Michelak LE, Tripathi RC (1999). Pharmacokinetics of TCDD in veterans of operation Ranch Hand: 15-year follow up. J Toxicol Env Health, Part A, 57:369 – 378.

Michalek JE, Pirkle JL, Needham LL, Patterson DG Jr., Caudill SP, Tripathi RC, Mocerelli P (2002). Pharmacokinetics od 2,3,7,8-tetrachlorodibenzo-*p*-dioxin in Seveso adults and veterans of operation Ranch Hand. J Exp Anal Environ Epidemiol, 12: 44-53.

Milbrath MO, Wenger Y, Chang C-W, Emond C., Garabrant D, Gillespie BW, Joliet O (2009). Apparent half-lifes of dioxins, furans, and polychlorinated biphenyls as a function of age, body fat, smoking status, and breast feeding. Env Health Perspect, 117(3): 417-425.
Molen GW van der, Kooijman BALM, Slob W (1996). A generic toxicokinetic model for persistent lipophilic compounds in humans: An application to TCDD. Fundam Appl Toxicol, 31: 83-94.

Molen, GW van der, Kooijman BALM, Wittsiepe J, Schrey P, Flesch-Janys D, Slob W (2000). Estimation of dioxin and furan elimination rates with a pharmacokinetic model. J Exp Anal Env Epidemiol, 10: 579-585.

Wells JCK (2014). Towards body composition reference data for infants, children, and adolescents. Adv Nutr,5:320S-329S.

Annex 5 PBDEs (ng/g milk lipid) in Dutch human milk

Table A6-1 Monitoring 1998 (N = 103)

	BDE-17	BDE-28	BDE-47	BDE-66	BDE-85	BDE-99	BDE-100	BDE-138	BDE-153	BDE-154	BDE-183
Number of results > detection limits	10	103	103	36	13	103	103	0	103	47	100
Minimum	< 0.03	0.05	0.45	< 0.06	< 0.08	0.17	0.09	< 0.1	0.33	< 0.08	< 0.09
Maximum	0.13	0.43	6.50	0.32	0.17	2.70	1.72	< 0.1	3.88	0.26	1.90
Median	< 0.03	0.11	1.19	< 0.06	< 0.08	0.37	0.31	< 0.1	0.95	< 0.08	0.41
Average		0.13	1.53			0.53	0.37		1.03		0.45
Standard deviation		0.07	1.11			0.41	0.25		0.53		0.28
Relative standard deviation		0.51	0.73			0.78	0.69		0.52		0.62

Table A6-2 Monitoring 2003 (N = 99)

	BDE-17	BDE-28	BDE-47	BDE-66	BDE-85	BDE-99	BDE-100	BDE-138	BDE-153	BDE-154	BDE-183
Number of results > detection limits	0	96	99	7	8	99	99	1	99	22	54
Minimum	< 0.03	< 0.04	0.35	< 0.05	< 0.08	0.12	0.06	< 0.09	0.24	< 0.05	< 0.10
Maximum	< 0.05	2.01	27.46	0.27	0.44	13.70	3.46	0.13	6.56	0.50	2.07
Median	< 0.04	0.09	1.59	< 0.06	< 0.08	0.44	0.30	< 0.1	0.78	< 0.06	0.11
Average		0.14	2.23			0.76	0.44		1.00		0.22
Standard deviation		0.22	3.13			1.46	0.46		0.77		0.27
Relative standard deviation		1.56	1.41			1.92	1.05		0.77		1.23

LOQ (ng/g milk lipid): 0.03 (BDE 17), 0,02 (BDE 28), 0.03 (BDE 47), 0.06 (BDE 66), 0.05 (BDE 100), 0.05 (BDE 99), 0.08 (BDE-85), 0.05 (BDE 154), 0.04 (BDE 153), 0.1 (BDE 138), 0.09 (BDE 183).

References

RIVM (2003a). Rectification analysis results of flame retardants in human milk. RIVM letter report 245/03 LAC/RB, 15 April 2003.

RIVM (2003b) Analysis of brominated flame retardants in breast milk 1988, 1993 and 1998, RIVM letter report 344/03 LAC/RB, 19 June 2003.

RIVM (2004) Analysis of brominated flame retardants in breast milk 2003, RIVM letter report 01/04 LAC/BB/bb, 29 June 2004.

Annex 6 Dioxins/furans and PCBs in in Dutch human milk

Table A7-1 Monitoring 2003 (N=100)

Congener	**TEF**	**Mean[1] (SD)**	**Median**	**$P_{0.05}$**	**$P_{0.95}$**
Dioxins (pg/g milk lipid)					
2378-TCDD	1	1.53 (0.79)	1.46	LOD (< 0.1)	2.60
12378-PeCDD	1	3.54 (1.31)	3.38	1.85	5.54
123478-HxCDD	0.1	2.59 (1.45)	2.34	0.97	4.83
123678-HxCDD	0.1	12.12 (5.44)	10.86	4.87	22.37
123789-HxCDD	0.1	2.39 (1.13)	2.13	1.08	4.30
1234678-HpCDD	0.01	14.54 (9.65)	12.51	5.12	27.93
OCDD	0.0003	106.17 (56.00)	90.44	52.38	209.92
WHO- TEQ (dioxins)		6.96	6.53	3.61	11.63
Furans (pg/g milk lipid)					
2378-TCDF	0.1	0.34 (0.65)	0.18	LOD (< 1)	0.81
12378-PeCDF	0.03	0.24 (0.61)	0.10	LOD (< 0.1)	0.60
23478-PeCDF	0.3	7.99 (2.97)	7.83	3.89	13.14
123478-HxCDF	0.1	2.27 (0.99)	2.15	1.06	3.61
123678-HxCDF	0.1	2.06 (0.95)	1.93	0.93	3.37
123789-HxCDF	0.1	0.20 (0.55)	0.10	LOD (< 0.1)	0.46
234678-HxCDF	0.1	1.08 (0.72)	0.93	0.45	2.12
1234678-HpCDF	0.01	2.44 (1.76)	1.89	1.15	5.49
1234789-HpCDF	0.01	0.27 (0.75)	0.15	LOD (< 0.1)	0.62
OCDF	0.0003	1.05 (1.80)	0.60	LOD (< 0.3)	3.50
WHO-TEQ (furans)		3.03	2.91	1.45	5.06
WHO-TEQ (dioxins/furans)		9.99	9.44	5.06	16.69
Non-ortho PCBs (pg/g milk lipid)					
PCB 77	0.0001	7.38 (5.42)	5.21	3.29	18.85
PCB 81	0.0003	1.35 (1.13)	1.04	0.25	2.98
PCB 126	0.1	38.03 (16.54)	36.00	15.56	69.98
PCB 169	0.03	27.61 (12.71)	25.36	11.85	47.35
WHO-TEQ (non-ortho PCBs)		4.63	4.36	1.91	8.42
Mono-ortho PCBs (ng/g milk lipid)					
PCB 105	0.00003	1.50 (0.68)	1.30	0.65	2.78
PCB 114	0.00003	0.40 (0.26)	LOD (< 0.5)	LOD (< 0.5)	1.0
PCB 118	0.00003	8.02 (3.25)	7.40	4.03	14.47
PCB 123	0.00003	0.38 (0.26)	LOD (< 0.5)	LOD (< 0.5)	1.0
PCB 156	0.00003	4.32 (2.08)	4.26	1.54	7.65
PCB 157	0.00003	0.68 (0.34)	0.60	LOD (< 0.5)	1.14
PCB 167	0.00003	1.28 (0.74)	1.04	LOD (< 1.0)	2.50

Congener	TEF	Mean[1] (SD)	Median	$P_{0.05}$	$P_{0.95}$
PCB 189	0.0003	0.50 (0.31)	LOD (< 0.5)	LOD (< 0.5)	1.0
WHO-TEQ (mono-ortho PCBs)		0.51	0.50	0.28	0.95
WHO-TEQ (total)		15.13	14.3	7.25	26.06
Indicator PCBs (ng/g milk lipid)					
PCB 28		1.60 (1.24)	1.32	0.61	4.43
PCB 52		0.28 (0.13)	LOD	LOD	0.51
PCB 101		0.39 (0.26)	LOD	LOD	0.97
PCB 138		26.02 (12.45)	22.92	11.11	50.68
PCB 153		45.29 (22.45)	40.89	18.56	94.94
PCB 180		23.11 (12.09)	20.85	8.71	45.18

[1]99 individual samples, non-detects set at the LOD, i.e. < 0.1 pg/g lipid.

Reference

RIVM (2004) Analysis of dioxins and organochlore pesticides in breast milk 2003, RIVM letter report 041069/04 SIR MZ/el, 21 october 2004.

Annex 7 Data from other European countries

POP Concentrations in European human milk. Amounts in ng/g lipid, unless otherwise indicated.

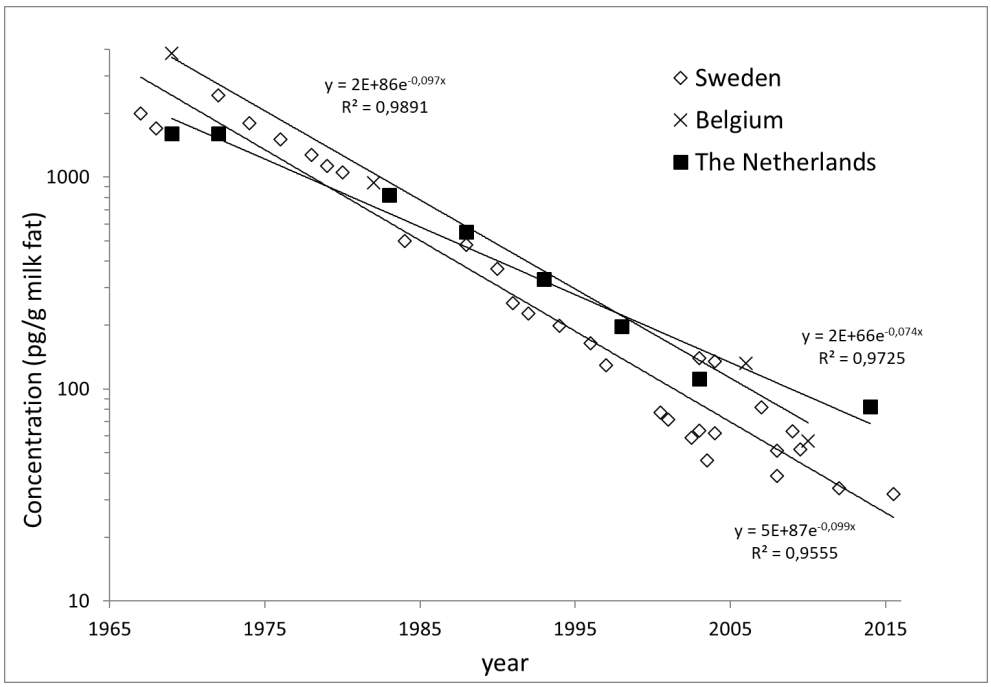
Chemical	NL[a] 2014	DE[b] 2007/08	BE[c] 2010	BE[d] 2007	LUX[f] 2006	NO[d,e] 2006/07	SW[d,e] 2006/07	FI[d] 2007	IRL[f] 2010	CH[f] 2009
Aldrin	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Chlordane group	2.4		5.6	7.8		3.6	2.2	3.8		
alphachlordane	ND		ND	ND	ND	ND	ND	ND	ND	ND
gammachlordane	ND		ND	ND	ND	ND	ND	ND	ND	ND
oxychlordane	2.5	2.0	5.6	8.0	8.06	3.7	2.3	1.0	3.02	3.75
Transnonachlor	2.2		3.3	1.7		5.7	3.0	2.8		
Dieldrin	2.1	ND	7.2	6.7	4.85	2.5	1.8	1.5	3.51	3.30
DDT group	94.9	70.0	196.0	156.3	140.27	69.6	81.9	33.1	80.12	124.62
o,p'DDD	ND		ND	ND	ND	ND	ND	ND	ND	ND
p,p' DDD	ND		ND	ND	0.97	1.0	ND	ND	ND	0.71
o,p' DDE	ND		ND	ND	ND	ND	ND	ND	ND	ND
p,p'DDE	82.3	63.0	162.0	132.3	135.68	60.0	70.2	27.8	76.83	118.73
o,p'DDT	ND		ND	ND	ND	ND	ND	ND	ND	0.73
p,p' DDT	3.1	2.0	11.0	8.8	3.62	1.6	3.6	2.1	3.29	4.45
Endosulfan group	ND									
Alpha endosulfan	ND				ND				ND	ND
Beta endosulfan	ND				ND				ND	ND
Endosulfan SO4	ND				ND				ND	ND
Endrin group	ND		ND	ND		ND	ND	ND		
Endrin	ND		ND	ND	ND	ND	ND	ND	ND	ND
Endrin ketone	ND		ND	ND		ND	ND	ND		
Heptachlor group	2.2		4.7	5.3		0.6	0.8	0.5		
Heptachlor	ND		ND	ND	ND	ND	ND	ND	ND	ND
Heptachlorepoxide cis	2.3		4.7	5.6	2.93	0.6	0.9	0.5	1.46	1.72

Chemical	NL[a] 2014	DE[b] 2007/08	BE[c] 2010	BE[d] 2007	LUX[f] 2006	NO[d,e] 2006/07	SW[d,e] 2006/07	FI[d] 2007	IRL[f] 2010	CH[f] 2009
Heptachlorepoxide trans	ND		ND	ND	ND	ND	ND	ND	ND	ND
Hexachlorobenzene	9.5	16.0	9.6	15.0	17.82	16.9	7.1	2.7	8.96	12.56
Hexachlorocyclohexane group (HCH)	6.9		8.9		17.7					
Alpha-HCH	ND	ND	ND	ND[3]	ND				ND	ND
Beta-HCH	6.9	6.0	8.9	12.0[3]	17.70				17.16	4.04
Gamma-HCH (lindane)	ND	ND	ND	0.7[3]	ND				ND	ND
Parlar (toxaphene group)	2.4		2.3	2.3		3.7	2.4	1.9		
Parlar 26	0.6		0.5	0.7	1.30	1.1	0.7	0.5	ND	0.45
Parlar 50	0.9		1.8	1.5	2.60	2.6	1.8	1.5	1.52	1.02
Parlar 62	0.8		ND	ND	ND	ND	ND	ND	ND	ND
Mirex	ND		ND	ND	ND	ND	ND	ND	ND	ND
Hexabromobiphenyl	ND								ND	ND
Pentachlorobenzene	ND								ND	ND
Chlordecone	ND									
HBCDD group	0.6		3.80	1.50	1.10					
Alpha-HBCDD	0.6		3.20	1.5	1.10					
Beta-HBCDD	ND		0.05	ND	ND					
Gamma-HBCDD	ND		0.55	ND	ND					
PCDD/PCDFs (pg WHO-TEQ/g)	4.48	4.8	8.4	10.3[3]	8.93	4.6[4]	5.0[4]		4.58	
						5.5[3]	6.0[3]			
6 Indicator PCBs	40.10		70.2	80.5	115.32				31.79	78.95
Dioxinlike PCBs (pg WHO-TEQ/g)	2.68	4.0	5.9	7.0[3]	6.75	3.2[4]	4.2[4]		2.04	4.84
						5.6[3]	6.8[2]			
PBDEs										
BDE47	0.49		0.67	0.89[3]	1.31					
BDE99	0.13		0.62	0.22[3]	0.25					

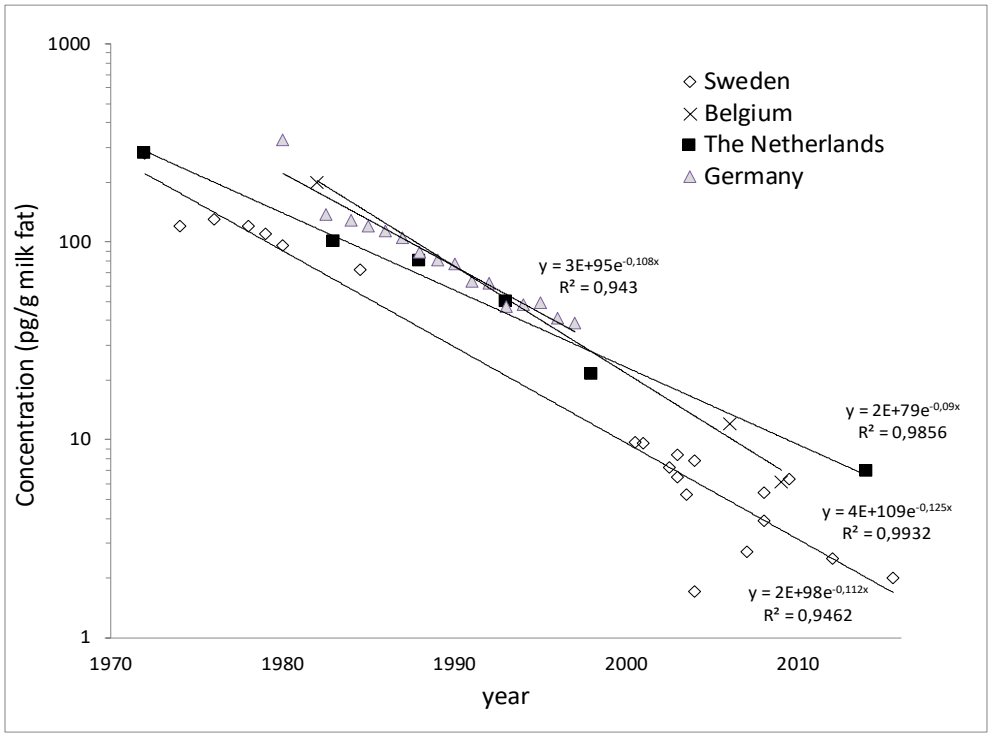
Chemical	NL[a] 2014	DE[b] 2007/08	BE[c] 2010	BE[d] 2007	LUX[f] 2006	NO[d,e] 2006/07	SW[d,e] 2006/07	FI[d] 2007	IRL[f] 2010	CH[f] 2009
BDE153	0.74		0.47	0.49[3]	0.66					
Remaining BDEs	0.27		0.27	0.35[3]	0.11					
Sum BDEs	1.63		2.03	1.95	2.33					
PFCs (ng/ml)										
LPFOS	45		130							
PFOA	<80		80							
PFHxS	11		nc							
PFPeA	<30									
PFHxA	<50									
PFHpA	<20									
PFNA	<25		nc							
PFDA	<50		ND							
PFUnDA	<50		ND							
PFBuS	<200									
PFDS	<50		ND							

Values given in ng/g lipid, TEQs given in pg/g lipid. nc = not calculated because of low levels
[a]This study
[b]Raab et al., 2011
[c]Croes et al., 2012
[d]UNEP, 2009
[e]UNEP, 2015
[f]UNEP, 2016

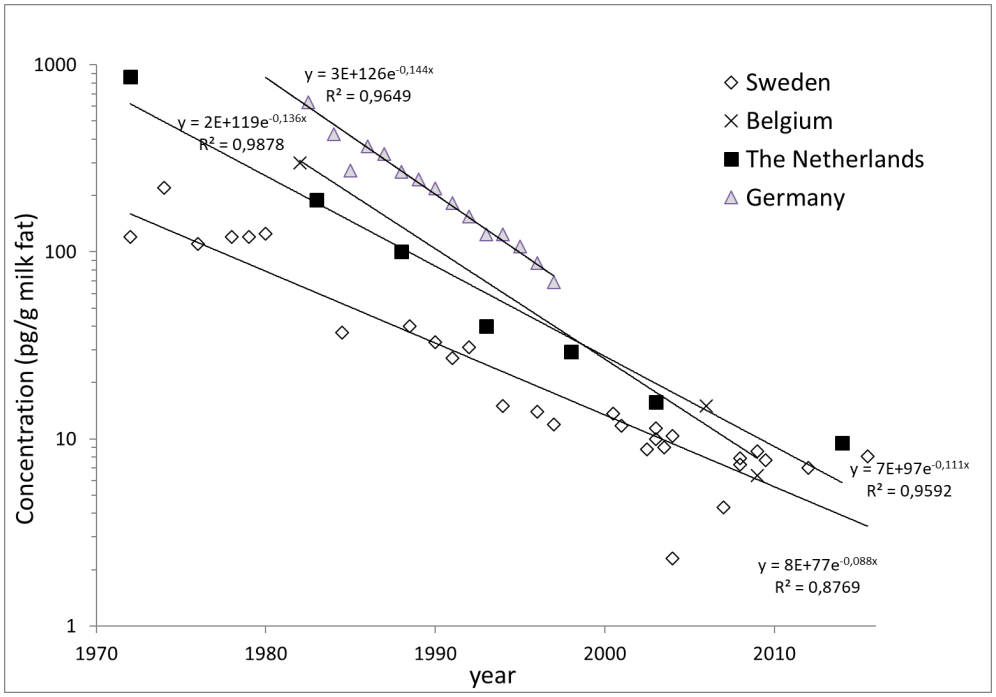
Annex 8 Trends in time of various POPs in Sweden, Belgium, Germany and The Netherlands on a logarithmic scale.



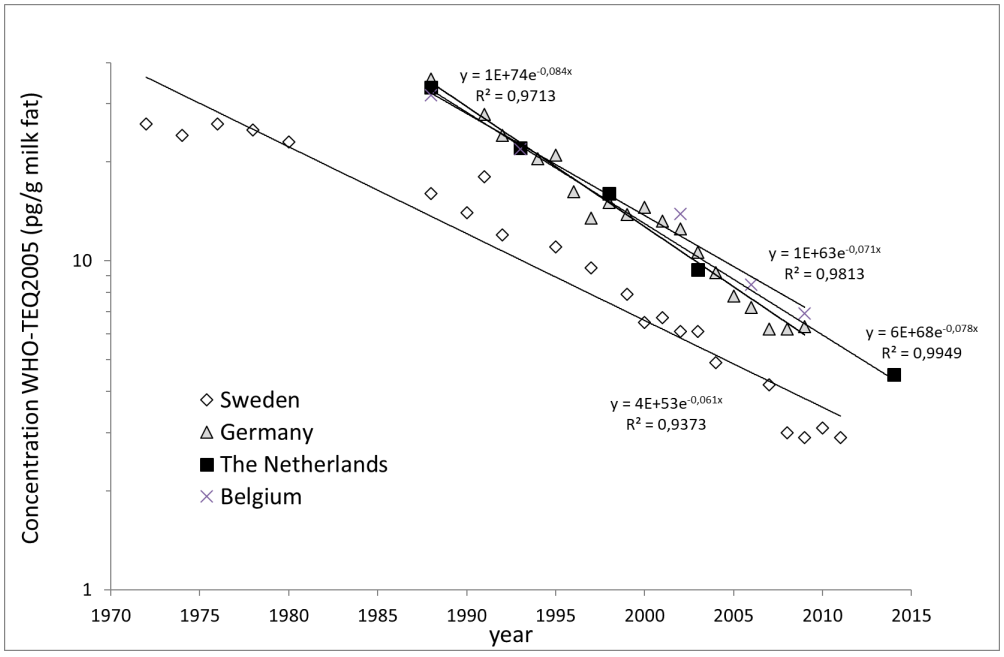
p,p'-DDE



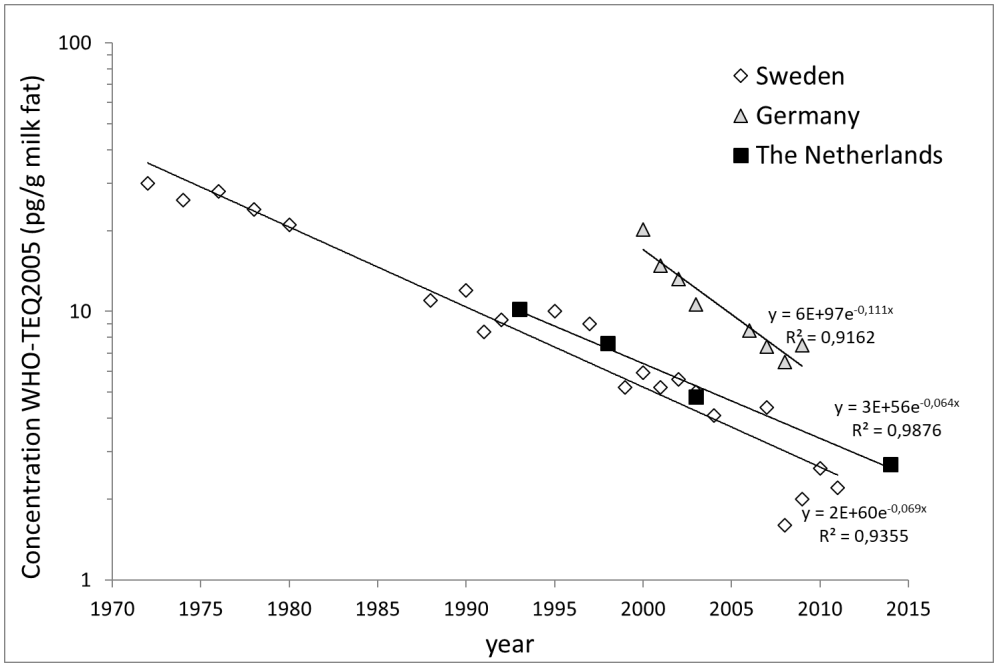
beta-hexachlorocyclohexane (β-HCH)



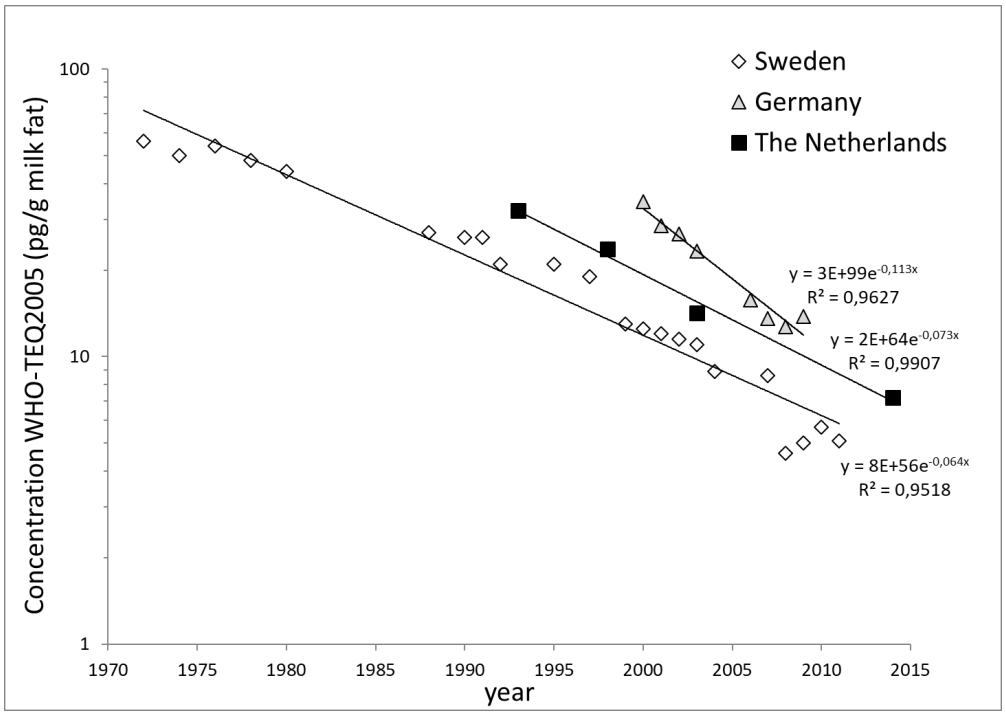
hexachlorobenzene (HCB)



PCDD/PCDF



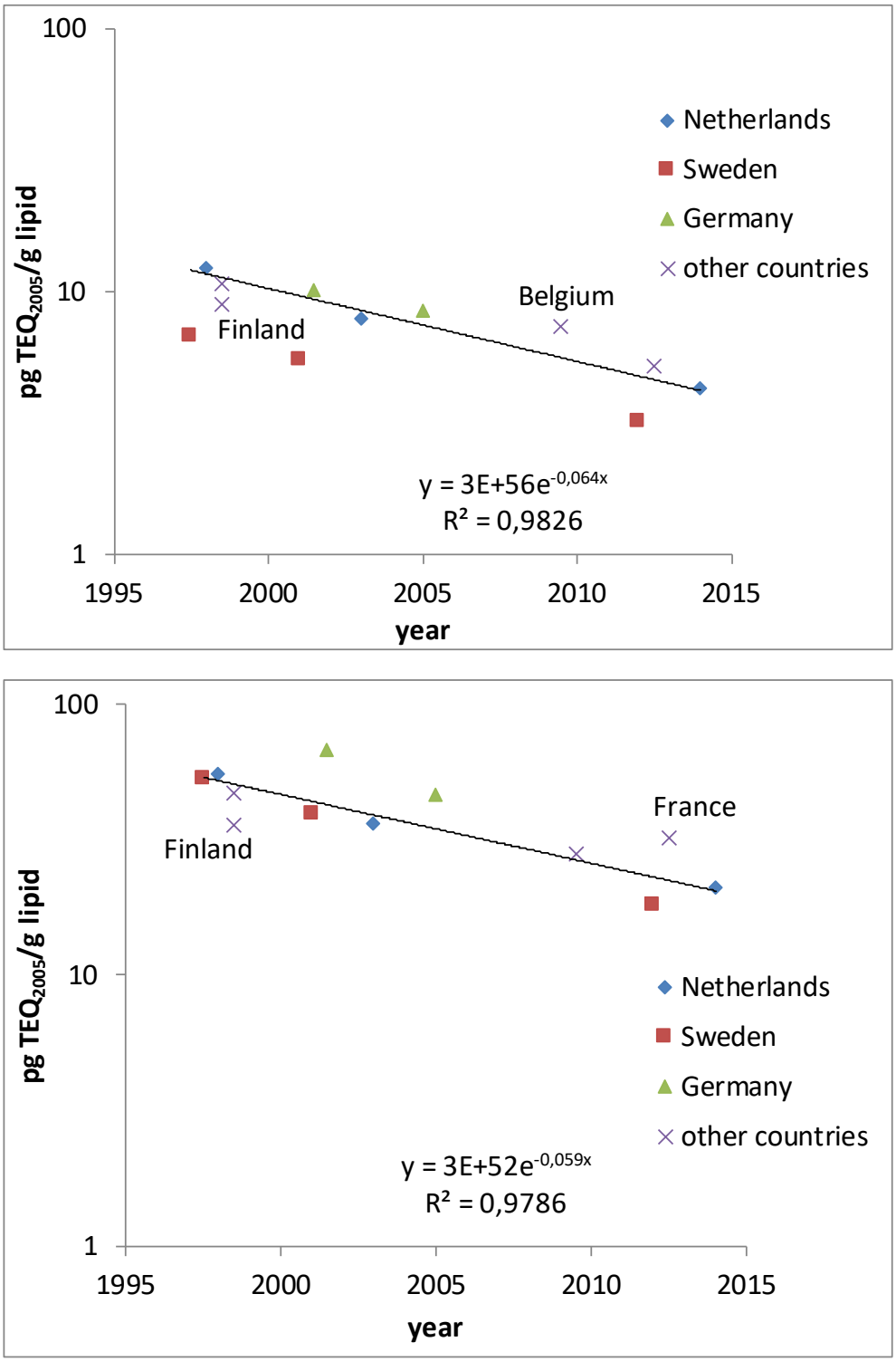
dl-PCBs



Total TEQ

Annex 9 TEQ2005

Dioxins, furans and dl-PCBs are usually combined into a TEQ-value. However, to analyse the trend in time this has limited value as the TEQ is composed of various substances and as both TEF1998 and TEF2005 may have been used (see section 2.4.4). Thus, it was decided to compare various individual congeners. Focussing on the dioxins/furans and the PCBs the data show that the dioxin/furan concentration in the Netherlands is comparable to that Germany, Belgium, France and Denmark, whereas concentration in Sweden and Finland are substantially lower (Annex 7). Concerning the PCBs (Figure 13), the concentrations in the Netherlands are intermediate. Based on limited data Fång et al (2013) conclude that the Swedish dioxin data should be at the lower end, whereas for PCBs low to medium concentrations are observed. The 1.5-2 times lower concentrations reported for France compared to Denmark and Finland (Antignac et al., 2016) can mainly be explained by the 10-year time difference in sampling. Corrected for that, France showed to have higher concentrations for PCBs compared to Denmark and Finland (annex 7).



Figures Annex 9. Concentrations in human milk of two of the most important substances determining the TEQ_{2005} values: 2,3,4,7,8-PeCDF (above) and PCB 126 (below) in ng/g lipid. The trendline is fitted to the Dutch data.

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