Report to the Swedish EPA (the Health-Related Environmental Monitoring Program)

Temporal trends of polychlorinated alkanes (PCAs) in pooled blood serum samples from first-time mothers in Uppsala, Sweden, 1997-2023

Irina Gyllenhammar, Pernilla Hedvall Kallerman
Livsmedelsverket

Thanh Wang, Idoia Beloki Ezker
Linköpings universitet

2026-04-13

NATIONELL
MILJÖÖVERVAKNING
PÅ UPPDRAG AV
NATURVÅRDSVERKET

ÄRENDENNUMMER	NV-03247-24
AVTALSNUMMER	215-24-005
PROGRAMOMRÅDE	Hälsorelaterad MÖ
DELPROGRAM	Biologiska mätdata – organiska ämnen

Temporal trends of polychlorinated alkanes (PCAs) in pooled blood serum samples from first-time mothers in Uppsala, Sweden, 1997-2023

Rapportförfattare Irina Gyllenhammar, Livsmedelsverket Pernilla Hedvall Kallerman, Livsmedelsverket Thanh Wang, Linköpings universitet Idoia Beloki Ezker, Linköpings universitet	**Utgivare** Livsmedelsverket **Postadress** Box 622, 751 26 Uppsala **Telefon** 018-175500
Rapporttitel Temporal trends of polychlorinated alkanes (PCAs) in pooled blood serum samples from first-time mothers in Uppsala, Sweden, 1997-2023	**Beställare** Naturvårdsverket 106 48 Stockholm **Finansiering** Nationell hälsorelaterad miljöövervakning
Nyckelord för plats Uppsala	
Nyckelord för ämne PCA, polyklorerade alkaner, klorparaffiner, CP, SCCP, MCCP, LCCP	
Tidpunkt för insamling av underlagsdata 1997-2023	

Sammanfattning

Sedan 1996 har Livsmedelsverket regelbundet samlat in prover från förstföderskor i Uppsala för analys av persistenta halogenerade organiska miljöföroreningar (POP). I följande rapport redovisas halterna av kort-, medium- och långkedjiga polyklorerade alkaner (PCA), även kallade klorparaffiner i poolade serumprover (n=60) insamlade under perioden 1997-2023. Blodproven togs av en barnmorska vid ett hembesök, tre veckor efter förlossningen.

Serumprover extraherades med vätske-vätskextraktion och renades därefter upp med multisilikakolonn. PCA haltbestämdes sedan med vätskekromatografi kopplad till en högupplösande masspektrometer (LC-qTOF-MS) genom analys av kloridaddukter och kvantifierades genom dekonvolutionsmetoden av homologgrupperna C10–30.

PCA påvisades i poolade serumprover från förstföderskor i Uppsala, Sverige. För de långkedjiga PCA var alla halter över metodens detektionsgräns (MDL) med en median på 160 ng/g fett. För de kort- och mellankedjiga PCA låg dock de flesta prover under MDL (63 respektive 82 %). Inga signifikanta tidstrender observerades under studieperioden, 1997–2023, vilket tyder på att det inte varit någon tydlig förändring i exponering under denna tid.

INTRODUCTION

The Swedish Food Agency has conducted recurrent sampling of breastmilk and blood from primiparous women in Uppsala since 1996, in the so-called POPUP study (Persistent Organic Pollutants in Uppsala Primiparas). The Swedish Environmental Protection Agency has funded the study since year 2000. The main aim of the study is to investigate temporal trends of exposure to persistent organic pollutants (POPs) among pregnant and nursing women.

Chlorinated paraffins are complex mixtures mainly composed of polychlorinated n-alkanes (PCAs) (Fernandes et al. 2023). They are manufactured by chlorinating paraffinic hydrocarbons of different chain lengths, producing complex mixtures comprising thousands of congeners that vary in carbon chain length and degree of chlorination. Historically, they have been grouped into short-chain (C10–C13, SCCPs), medium-chain (C14–C17, MCCPs), and long-chain (C18–C30, LCCPs) chlorinated paraffins. Nowadays, the term chlorinated paraffins is mostly used for the commercial mixtures, while PCAs is used for the more defined polychlorinated n-alkanes that are analysed (Fernandes et al. 2023). Chlorinated paraffins are widely used as flame retardants, as plasticizers in flexible polyvinyl chloride (PVC), and as lubricants in metalworking applications. Global production has increased and is estimated to exceed 1 million tonnes annually, with China as the leading producer (Gluge et al. 2016). Owing to their persistence, bioaccumulative properties, and toxicity to aquatic organisms, PCAs are of environmental and public health concern. The short-chain PCAs (SCCPs) were listed as persistent organic pollutants (POPs) under the Stockholm Convention in 2017, and medium-chain PCAs (MCCPs) were listed in 2025 (Stockholm Convention 2025).

The aim of this study was to analyse, for the first time, PCAs in serum from first-time mothers in the POPUP cohort and to evaluate temporal trends over the period 1997–2023.

MATERIALS AND METHODS

Recruitment and sampling

In the POPUP study, over 1100 first-time mothers from the general population living in Uppsala County were recruited between 1996 and 2023. The participants donated a blood sample three weeks after delivery. Blood sampling was done using 10 ml Vacutainer® or Vacuette® serum tubes and serum was stored at -20°C. The study was approved by the local ethics committee of Uppsala University, and the participating women gave informed consent prior to the inclusion in the study.

In the present study, we used pooled serum samples from the participants for analysis of PCAs. The composition of the 60 pools from 1997-2023 is given in Table 1. The total number of individual samples included in all pools was 602.

Table 1. Composition of the pooled serum samples used for analyses of PCAs.

Sampling year	**N**	**No of pools**	**N in each pool**	**Age (years)[a] mean (range)**
1997	30	3	10	28 (21-34)
1998	20	2	10	29 (23-35)
1999	10	1	10	27 (22-33)
2002	31	3	10-11	30 (24-37)
2004	32	3	10-11	29 (21-35)
2006	30	3	10	30 (19-40)
2008	30	3	10	32 (22-42)
2009	30	3	10	31 (22-40)
2011	29	3	9-10	30 (21-38)
2012	30	3	10	29 (21-38)
2013	30	3	10	29 (22-39)
2014	30	3	10	30 (20-38)
2015	30	3	10	30 (22-38)
2016	30	3	10	31 (25-37)
2017	30	3	10	29 (22-34)
2018	30	3	10	31 (24-40)
2019	30	3	10	30 (25-38)
2020	30	3	10	31 (26-43)
2021	30	3	10	32 (27-40)
2022	30	3	10	32 (26-38)
2023	30	3	10	32 (24-46)

[a]Mean age of the women donating blood during the specific sampling year.

Analysis

The chemical analyses of PCAs were performed by Linköping University. Sample preparation followed the protocol established by Yuan et al (2024) with minor modifications, and conducted in a clean room facility. Serum samples (2 mL) were spiked with 25 ng of the internal standard ($^{13}C_{12}$-1,2,5,6,9,10-hexabromocyclododecane) prior to extraction. Proteins were precipitated by adding isopropanol (iPrOH) in a 1:1 (v/v) ratio (sample:iPrOH) and vortex mixing. Subsequently, Milli-Q water was added to obtain a 1:2 (v/v) ratio of iPrOH:water, facilitating phase separation and liquid–liquid extraction (LLE). The samples were first extracted by collecting the organic phase after phase separation. The remaining aqueous phase was then subjected to sequential LLE steps to ensure exhaustive recovery of PCAs. First, a solvent mixture of n-hexane:methyl tert-butyl ether (MTBE) (9:1, v/v) containing 30% iPrOH (7:3, solvent:iPrOH) was added, mixed thoroughly, and the organic phase collected. This was followed by an additional extraction using n-hexane:MTBE (9:1, v/v), and the organic phase was again recovered. All collected organic extracts were combined in a test tube containing 1.0% (w/v) KCl solution and subjected to a washing step via LLE. After phase separation, the organic layer was collected. The aqueous phase was further extracted with n-hexane, and the organic phase was combined with the previous extracts. The pooled organic extracts were evaporated to dryness overnight under N_2 and reconstituted in1 mL hexane:dichloromethane (DCM) (1:1, v/v) before the clean-up step. For clean-up, a multilayer column packed from bottom to top with 2 g of silica (deactivated with 2.5% H_2O), 6 g of 44% sulfuric acid silica, and 4 g of anhydrous sodium sulfate was used. The column was washed with 15 mL of hexane:DCM (1:1, v/v), then the concentrated extract was loaded onto the column and eluted with 15 mL of hexane:DCM (1:1, v/v). The eluent was solvent-exchanged to acetonitrile, and 26 ng of d_{18}-1,2,5,6,9,10-hexabromocyclododecane were added as a recovery standard before instrumental analysis.

PCAs were analyzed using liquid chromatography coupled to a quadrupole time-of-flight mass spectrometer (LC-qTOF; Agilent 6550) operated in electrospray ionization negative mode (ESI−). Quantification of PCAs-$C_{10–30}$ was performed by monitoring the $[M + Cl]^-$ adduct ions, generated by addition of 0.05 mM tetramethylammonium chloride to organic and aqueous mobile phases to enhance chloride adduct formation. The contribution of each PCA homologue group was determined using the deconvolution approach originally described by Bogdal et al. (2015). For the pattern deconvolution of PCAs-$C_{10–13}$ and PCAs-$C_{14–17}$, single-chain standards (Chiron AS) were used for PCAs-C_{10} (52.5% and 58.4% Cl), PCAs-C_{11} (52.3% and 57.7% Cl), PCAs-C_{12} (53.8% and 57.3% Cl), PCAs-C_{13} (45.9% and 60.0% Cl), PCAs-C_{14} (49.2% and 58.7% Cl), PCAs-C_{15} (47.7% and 59.3% Cl), PCAs-C_{16} (51.5% and 58.4% Cl), and PCAs-C_{17} (56.3% Cl). For PCAs-$C_{18–30}$, standard mixtures (Dr. Ehrenstorfer, Augsburg, Germany) containing 36.0% and 49.0% Cl were applied, together with technical mixtures Uniclor40 (Neville Chemical Co., USA) and Paroil CW 40 (Dover Chemical Corporation, USA). Instrumental data was processed using the in-house developed workflow CPxplorer, where non-interfering m/z values were selected by applying relative abundance thresholds of 20% (relative to the base isotopic peak) in CPions (Beloki Ezker et al. 2025). For quantification in CPquant, calibration curves for individual homologue groups were accepted only when meeting a linearity criterion of $R^2 > 0.8$. Recoveries were calculated based on the ratio between the internal standard and recovery standard. Average quantified concentrations in the blanks were subtracted from the samples, and method detection limits (MDLs) were defined as three times the standard deviation of replicate blanks (Table 2). The goodness-of-pattern fit (GoPF, R^2), calculated using CPxplorer, was applied to evaluate quantification performance. The GoPF reflects the agreement between the PCA homologue distribution in the calibration standards and that observed in the sample, with a value of 1 indicating perfect concordance. The GoPF threshold was set at 0.5.

Table 2. Method detection limit (MDL) for the analytical method.

Analytes	**MDL ng/g**
PCAs-C_{10-13}	0.4
PCAs-C_{14-17}	1.4
PCAs-C_{18-30}	0.04
PCAs-C_{10-30}	1.9

Calculations and statistics

All values below the MDL were replaced with MDL/2 for statistical analysis. The results from three samples were excluded due to low recovery (12-14%). Linear regression was used to analyse associations between PCA serum concentrations and sampling year from 1997 to 2023, using Stata version 17.0. Both fresh weight (ng/g) and fat adjusted concentrations (ng/g lipid) were evaluated. All concentrations were ln-transformed before analysis.

RESULTS AND DISCUSSION

Serum concentrations of PCAs in pooled serum samples from the period 1997-2023 (n=60) are presented in Table 3. For the long-chained PCAs (C_{18-30}), all samples were above MDL and the median concentrations was 160 ng/g lipid. For the short- (C_{10-13}) and medium-chained (C_{14-17}) PCAs 63 and 82%, respectively, had serum concentrations below MDL. This observation indicates a high blank contamination of short- and medium chained PCAs, which is consistent with their ubiquitous environmental contamination. For ∑PCAs around half of the samples had detectable levels (53%) and the mean concentration was 696 ng/g lipid. In Figure 1, fat-adjusted serum levels are presented for the time period 1997-2023. The results from the linear regression analysis showed no temporal trends for PCAs in this study.

In a study of serum samples from 25 breastfeeding women in the Swedish NorthPop cohort, sampled 2020, short- and medium-chained PCAs were observed at higher levels than in the present study (median 785 and 519 ng/g lipid weight, respectively). In contrast, long-chained PCAs were lower than in the present study (median 16 ng/g lipid weight). However, detection limits differed substantially between studies, which may have influenced the comparability of the results (Yuan et al. 2024). Serum concentrations of short-chained PCAs were similar in the NorthPOP-study and in samples from adults from China sampled 2016-2018 (median 721 ng/g lipid weight), but the median concentration of medium-chained PCAs were around 4 times higher (2210 ng/g lipid weight) (Zhang et al. 2023). In a study of serum samples from 403 Belgian adults, similar results were observed for short-chained PCAs (median <0.6 ng/mL), whereas higher levels of medium-chained PCAs (median 12 ng/mL) and lower levels of long-chained PCAs (median <0.6 ng/mL) were reported compared with the present study (Cseresznye et al. 2026).

Temporal trend studies of PCAs in serum has shown increased levels of medium-chained PCAs in samples from Australian males during 2004-2015 (van Mourik et al. 2020). Higher levels were also observed for medium-chained PCAs in serum from Norwegian women in 2019 compared to 2007-2009 (Xu et al., 2022).

Dietary intake is considered the primary exposure pathway for the general population, accounting for approximately 60–88% of total PCA exposure (Yuan et al. 2022). In 2020, EFSA conducted a risk assessment of chlorinated paraffins in food and feed (EFSA 2020). In animal studies the liver, kidneys, and thyroid were identified as primary target organs for short- and medium chained PCAs, while the liver was the main target for long-chained. PCAs were analyzed in the Swedish market basket studies conducted in 2015 and 2022 (SFA 2024). The risk assessment indicated a substantial margin between estimated per capita intakes and the reference points, resulting in high Margin of Exposures (MOEs) and no identified health concerns. The estimated per capita intake of ∑PCAs was approximately tenfold higher in the 2022 market basket than in 2015. This difference may be explained by the use of different laboratories and analytical methods, as well as lower detection limits in 2015; however, an increase in PCA levels over time could not be excluded (SFA 2024). In the present study, no temporal trends were observed in serum, which could be expected if levels in food had increased between 2015 and 2022.

Table 3. Concentrations of PCA (ng/g, fresh weight) in pooled serum samples from first-time mothers from Uppsala, sampled 1997-2023.[a]

Year	Recovery (%)	Fat %	PCAs-C_{10-13}	PCAs-C_{14-17}	PCAs-C_{18-30}	PCAs-C_{10-30}
1997	41%	0.5	1.8	<MDL	0.8	2.6
1997	39%	0.5	<MDL	<MDL	0.6	<MDL
1997	44%	0.5	<MDL	<MDL	0.7	0.7
1998	38%	0.5	<MDL	<MDL	1.2	1.2
1998	41%	0.5	<MDL	<MDL	0.8	<MDL
1999	38%	0.5	<MDL	<MDL	0.5	<MDL
2002	45%	0.5	<MDL	<MDL	0.6	<MDL
2002	48%	0.6	<MDL	<MDL	0.9	<MDL
2004	50%	0.5	0.8	<MDL	0.9	1.7
2004	42%	0.5	0.8	<MDL	1.4	2.2
2004	39%	0.5	<MDL	<MDL	1.0	<MDL
2006	46%	0.5	<MDL	<MDL	0.6	<MDL
2006	49%	0.5	<MDL	<MDL	0.8	<MDL
2006	37%	0.5	<MDL	<MDL	0.7	<MDL
2008	36%	0.5	15.7	2.7	1.5	19.8
2008	37%	0.5	<MDL	<MDL	0.6	<MDL
2008	41%	0.5	<MDL	<MDL	0.5	<MDL
2009	36%	0.5	<MDL	<MDL	0.5	<MDL
2009	40%	0.5	<MDL	<MDL	0.7	<MDL
2009	56%	0.5	0.4	4.6	1.3	6.3
2011	45%	0.6	12.0	<MDL	1.0	13.0
2011	32%	0.6	0.5	2.4	1.0	3.9
2012	40%	0.5	<MDL	<MDL	0.6	<MDL
2012	54%	0.5	<MDL	2.0	1.0	3.1
2012	40%	0.5	<MDL	<MDL	0.8	0.8
2013	44%	0.5	8.6	<MDL	0.9	9.5
2013	40%	0.5	<MDL	<MDL	0.8	<MDL
2013	51%	0.5	<MDL	1.8	0.7	2.5
2014	46%	0.6	0.5	<MDL	1.0	<MDL
2014	42%	0.5	0.9	<MDL	0.9	1.8
2015	38%	0.4	14.3	<MDL	1.1	15.4
2015	59%	0.5	<MDL	<MDL	0.5	<MDL
2015	47%	0.4	<MDL	<MDL	0.8	<MDL

2016	50%	0.5	0.8	<MDL	0.7	1.5
2016	42%	0.5	<MDL	<MDL	0.6	<MDL
2016	44%	0.5	<MDL	<MDL	0.4	<MDL
2017	49%	0.5	0.6	<MDL	1.8	2.4
2017	45%	0.5	<MDL	<MDL	0.5	<MDL
2017	41%	0.6	0.5	<MDL	0.7	1.2
2018	37%	0.4	0.6	<MDL	0.8	1.4
2018	43%	0.5	<MDL	<MDL	0.5	<MDL
2018	53%	0.5	<MDL	<MDL	0.9	0.9
2019	48%	0.5	1.5	<MDL	1.1	2.6
2019	36%	0.4	6.1	<MDL	0.8	7.0
2019	57%	0.5	0.5	2.4	1.4	4.3
2020	54%	0.5	0.7	<MDL	0.8	<MDL
2020	57%	0.4	<MDL	<MDL	0.7	<MDL
2020	42%	0.5	<MDL	1.6	1.0	2.6
2021	36%	0.5	<MDL	<MDL	0.8	<MDL
2021	42%	0.5	<MDL	<MDL	0.7	<MDL
2021	51%	0.5	<MDL	<MDL	0.8	<MDL
2022	46%	0.4	<MDL	<MDL	0.8	<MDL
2022	49%	0.6	<MDL	<MDL	0.6	<MDL
2022	42%	0.5	<MDL	2.0	0.6	2.6
2023	40%	0.6	<MDL	<MDL	0.6	<MDL
2023	40%	0.5	1.9	1.6	1.3	4.8
2023	28%	0.5	0.5	1.7	2.4	4.7

[a]Three samples were excluded due to low recovery, 1 pool from 2002 (recovery 12%), 1 pool from 2011 (14%) and 1 pool from 2014 (13%).

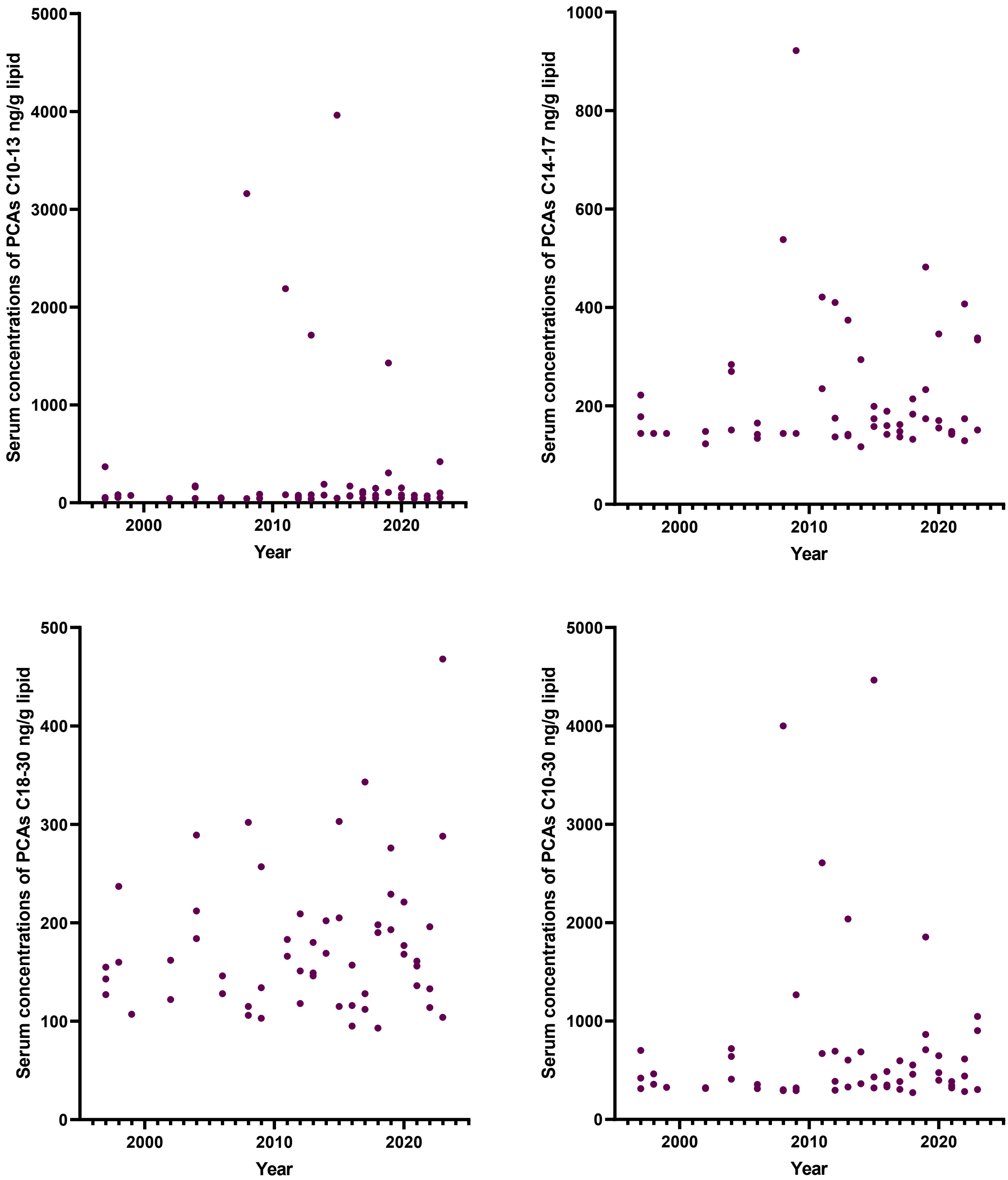

Figure 1. Concentrations of PCAs (ng/g lipid weight) in pooled serum samples from first-time mothers from Uppsala, sampled 1997-2023. All concentrations below MDL were replaced with MDL/2.

CONCLUSIONS

PCAs were detected in pooled serum samples from first-time mothers in Uppsala, Sweden; however, for short- and medium-chain PCAs most samples were below the MDL (63% and 82%, respectively). No temporal trends in concentrations were observed over the study period, 1997–2023, suggesting no clear change in population exposure during this time.

ACKNOWLEDGEMENT

The Swedish EPA (Environmental Protection Agency) is acknowledged for financial support. Appreciation is expressed to the participating women and to midwives who assisted in recruitment, interviewing, and sample collection. The laboratory staff at the Swedish Food Agency are appreciated for excellent work. We would also thank Bo Yuan at Norwegian University of Science and Technology for help in the sample preparation method.

REFERENCES

Beloki Ezker I, Yuan B, Røsrud Borgen A, Liu J, Wang Y, Thanh Wang T. 2025. Streamlining Quantification and Data Harmonization of Polychlorinated Alkanes Using a Platform-Independent Workflow. *Environ Sci Technol* (59) 22074−22084.

Bogdal C, Alsberg T, Diefenbacher PS, Macleod M, Berger U. 2015. Fast quantification of chlorinated paraffins in environmental samples by direct injection high-resolution mass spectrometry with pattern deconvolution. *Anal Chem* (87) 2852–2860.

Cseresznye A, den Ouden F, Ait Bamai Y, Engelen L, Maris E, De Paepe E, Poma G, Derrien M, Vich Vila A, Proost S, de Commer L, Verspecht C, Devolder L, Vieira-Silva S, Falony G, Hemeryck LY, Yin S, McGrath T.J, Pero-Gascon R, van Nuijs A, Covaci A. 2026. Serum PCBs, OCPs, PBDEs and PCAs: Associations with metabolic syndrome risk factors in the Flemish Gut Flora Project cohort. *Environmental Research* (296) 124068.

EFSA, Schrenk D, Bignami M, Bodin L, Chipman JK, Del Mazo J, et al. 2020. EFSA panel on contaminants in the food chain (CONTAM). Risk assessment of chlorinated paraffins in feed and food. *EFSA Journal* (18) e05991.

Fernandes AR, Krätschmer K, McGrath TJ, Yuan B, Brandsma S, Brits M, Cariou R, Letcher RJ, Mueller J, Muir D, Vetter W, Wang T, Yu G, Bergman Å. 2023. Recommended terms and abbreviations for polychlorinated alkanes (PCAs) as the predominant component of chlorinated paraffins (CPs). *TrAC Trends in Analytical Chemistry* (169) 117363.

Gluge J, Wang Z, Bogdal C, Scheringer M, Hungerbuhler K. 2016. Global production, use, and emission volumes of short-chain chlorinated paraffins - A minimum scenario. *Sci Total Environ* (573) 1132-1146.

SFA. 2024 "Swedish Market Basket Study 2022 - Per capita-based analyses of nutrients and toxic compounds in market baskets and assessment of benefit or risk." In. Uppsala, Sweden: Swedish Food Agency.

Stockholm Convention on Persistent Organic Pollutants (POPs) (2001). United Nations Environment Programme (UNEP), Geneva. Amended in 2009, 2011, 2013, 2015, 2017, 2019, 2022 and 2025. Stockholm Convention - Home page

van Mourik LM, Toms LML, He C, Banks A, Hobson P, Leonards PEG, de Boer J, Mueller JF. 2020. Evaluating age and temporal trends of chlorinated paraffins in pooled serum collected from males in Australia between 2004 and 2015. *Chemosphere* (244) 125574.

Xu S, Hansen S, Rautio A, Jarvelin MR, Abass K, Rysa J, Palaniswamy S, Huber S, Grimalt JO, Dumas P, Odland JO. 2022. Monitoring temporal trends of dioxins, organochlorine pesticides and chlorinated paraffins in pooled serum samples collected from Northern Norwegian women: The MISA cohort study. *Environ Res* (204) 111980.

Yuan B, Haug LS, Tay JH, Padilla-Sanchez JA, Papadopoulou E, de Wit CA. 2022. Dietary Intake Contributed the Most to Chlorinated Paraffin Body Burden in a Norwegian Cohort. *Environ Sci Technol* (56) 17080-17089.

Yuan B, Bignert A, Andersson PL, West CE, Domellöf M, Bergman Å. 2024. Polychlorinated alkanes in paired blood serum and breast milk in a Swedish cohort study: Matrix dependent partitioning differences compared to legacy POPs. *Environ International* (183) 108440.

Zhang J, Liao H, Chen Y, Li X, Chen R, Han S, Liu S, Yin S. 2023. Concentrations and homologue patterns of SCCPs and MCCPs in the serum of the general population of adults in Hangzhou, China. *Chemosphere* (335) 139131