

Sakrapport till Naturvårdsverkets Miljöövervakning:

Tidstrend 1996-2004:

Polyklorerade dibenzo-*p*-dioxiner (PCDD) och dibenzofuraner (PCDF), polyklorerade bifenyler (PCBer), klorerade bekämpningsmedel och bromerade flamskyddsmedel i bröstmjök från förstföderskor i Uppsala.

Avtalsnummer:	215 0312, 215 0417 och 215 0415
Utförare:	Livsmedelsverket
Programområde:	Hälsorelaterad miljöövervakning
Delprogram:	Exponering via livsmedel
Undersökningar/uppdrag:	Bröstmjölksstudier
	Dioxiner, furaner, polyklorerade bifenyler, klorerade bekämpningsmedel och bromerade flamskyddsmedel i bröstmjök – tidstrend för Uppsala mellan 1996 och 2004.

SAMMANFATTNING

Under perioden 1996 till 2004 har Livsmedelsverket samlat in bröstmjölksprover från förstfödorskor i Uppsala län. Ett av syftena med studierna är att undersöka hur halterna av vissa persistenta organiska miljögifter (POP) förändras med tiden. Tidstrender för polyklorerade bifenyler (PCBer) och klorerade bekämpningsmedel mellan 1996 och 2003 har utvärderats och rapporterats tidigare (Lignell et al. 2004). I denna rapport kompletteras de tidigare utvärderade tidstrenderna med analysresultat från bröstmjölksprover som samlades in 2004. Dessutom utvärderas eventuella tidstrender för polyklorerade dibensodioxiner (PCDD), polyklorerade dibenzofuraner (PCDF), non-orto PCBer samt polybromerade difenyletrar (PBDE).

Resultaten visar att medianhalterna av PCBer (PCB 28, 105, 118, 138, 153, 156, 167, 180, 126 och 169), PCDD, PCDF och klorerade pesticider (hexaklorbensen, hexaklorcyklohexan, oxyklordan, *trans*-nonaklor, *p,p'*-DDT och *p,p'*-DDE) i bröstmjölksprover har minskat med 3-10 % per år under tidsperioden 1996 till 2004. Halterna minskade långsammast för vissa PCB kongener (PCB 28, 167 och 169) och för PCDF, medan minskningen var snabbast för DDT-föreningarna och hexaklorcyklohexan. En jämförelse av de "halveringstider" för PCBer som beräknats i vår studie med resultat från en tidigare studie i Stockholm (1972-1997) (Norén & Meironyté, 2000), visar att de uppskattade "halveringstiderna" är kortare i Uppsala än i Stockholm. En förklaring till detta är sannolikt att man i Stockholmsstudien inte tagit hänsyn till att medelåldern bland kvinnorna ökade mellan 1972 och 1997.

Inga generella trender kunde visas beträffande PBDEer i bröstmjölksprover. Halterna av BDE-47 och BDE-99 minskade, medan halterna av BDE-153 ökade under den studerade tidsperioden. Detta överensstämmer med resultat från en studie av tidstrender (1980-2004) av PBDEer i poolade bröstmjölksprover från Stockholms-området (Fängström et al. 2005).

Våra resultat visar att det är viktigt att beakta olika individuella faktorer, särskilt kvinnornas ålder, då man studerar tidstrender av POP i bröstmjölksprover. Ålder var den viktigaste förklarande variabeln för flera av de studerade föreningarna (PCB 105, PCB 118, PCB 138, PCB 153, PCB 156, PCB 167, PCB 180, PCB 169, summa PCDF, PCDD/DF TEQ, -hexaklorcyklohexan, oxyklordan, *trans*-nonaklor, *p,p'*-DDT), med förklaringsgrader på 20-46 %.

Polychlorinated dibenzo-*p*-dioxins (PCDDs) and dibenzofurans (PCDFs), polychlorinated biphenyls (PCBs), chlorinated pesticides and brominated flame retardants in breast milk from primiparae women in Uppsala County, Sweden – Levels and trends 1996-2004

INTRODUCTION

Exposure estimation is an important part of risk assessment of environmental pollutants in food. Among the Swedish human population, food is the major source of exposure to persistent organic pollutants (POPs), such as dioxins (PCDD/DFs), polychlorinated biphenyls (PCBs) and DDT-compounds. These compounds are lipophilic and accumulate in the lipid compartment of the human body. Due to the relatively high lipid content, breast milk is a good human matrix for analysis of POP levels. The POP levels in breast milk reflect the long-term exposure of the individual mother and also give information about the body burden of POPs at the time of pregnancy and nursing.

In order to estimate the body burdens of POPs among pregnant and breast feeding women, and to estimate the intake of the compounds by breast feeding infants, the Swedish National Food Administration (NFA) has made recurrent measurements of levels of POPs in human breast milk. Another aim of this project is to establish if there are temporal trends of POP levels in breast milk. Temporal trends of PCBs and chlorinated pesticides between 1996 and 2003 have been reported earlier (Lignell et al. 2004). The established time trends are now revised with data from 2004. In addition, temporal trends for dioxins, furans, non-*ortho* PCBs and brominated flame retardants are established.

MATERIAL AND METHODS

Recruitment of primiparas

Breast milk was exclusively sampled from primipara women in order to minimise variation. The recruitment in 1996-2003 (N=273) is described in Lignell et al. (2004). In addition to the mothers recruited in 1996-2003, another 32 mothers were recruited among primiparas who delivered at Uppsala University Hospital from January to December 2004. Women who delivered during the first week in every month, and on randomly selected days during this week, were asked to participate in the breast milk study. 2-3 primiparas were recruited every month. 51 women were asked to participate in the study and 32 agreed to donate breast milk. Thus, a total of 305 women were recruited from 1996 to 2004. Mothers who were born in non-Nordic countries (N=10) were excluded before the statistical analysis. After this exclusion, a total of 295 women were included in the data set.

Data on age, weight, lifestyle, medical history, dietary habits etc. of the mothers were obtained via questionnaires (Table 1).

Table 1. Characteristics of the participating mothers.

Variable	N	Mean	Median	Min	Max
Age of the mothers (yr)	295	28.7	28.7	20.9	41.4
Body Mass Index (BMI, kg/m ²)	289	22.8	22.0	16.2	37.7
Weight gain during pregnancy (% of initial wt/week)	288	0.63	0.62	0.03	1.54
Weight reduction from delivery to sampling (%) ^a	280	9.6	9.4	-1.7	21
	N	%			
Education	max 3-4 yr high school	126	43		
	1-3 yr higher education	66	23		
	>3 yr higher education	100	34		
Smoking during pregnancy ^b	Non-smoker	207	71		
	Former smoker	38	13		
	Smoker	48	16		

^aWeight reduction minus birth weight of the child in % of weight just before delivery.

^bWomen who stopped smoking before pregnancy are considered to be former smokers, and women who stopped smoking during the first or second month of pregnancy are considered to be smokers.

Breast milk sampling

The mothers sampled milk at home during the third week after delivery (approximately day 14-21 post partum). Milk was sampled during breastfeeding using a manual breast milk pump and/or a passive breast milk sampler. The women were instructed to sample milk both at the

beginning and at the end of the breastfeeding session. The goal was to sample 500 mL from each mother during 7 days of sampling. During the sampling week, the breast milk was stored in the home freezer, in acetone-washed bottles. Newly sampled milk was poured on top of the frozen milk. At the end of the sampling week, a nurse visited the mother to collect the bottles.

Analysis

The specific compounds (congeners/metabolites) that were analysed in the breast milk samples are compiled in Table 2, and the number of analysed samples is shown in Table 3.

PCBs (with a few exceptions, see below) and chlorinated pesticides were analysed at the NFA using previously described methods (Atuma and Aune 1999; Aune et al. 1999). Brominated flame retardants (polybrominated diphenyl ethers (PBDEs) and hexabromocyclododecane (HBCD)) were also analysed at the NFA using a method described in Atuma et al. (2000), with a few modifications. All samples were fortified with internal standards prior to extraction to correct for analytical losses and to ensure quality control. A number of control samples were analysed together with the samples to verify the accuracy and precision of the measurements. The laboratory is accredited for analysis of PCBs and chlorinated pesticides in human milk.

Table 2. Summary of compounds that were analysed in the breast milk samples.

Compound	Congeners, metabolites etc.
PCBs	
PCBs	28, 52, 101, 105, 114, 118, 138, 153, 156, 157, 167, 170, 180
non-ortho PCBs	77, 126, 169, 81
Chlorinated pesticides	
Hexachlorobenzene (HCB)	
Hexachlorocyclohexane	α -HCH, β -HCH γ -HCH
Chlordane	oxychlordane, trans-nonachlor
DDT	<i>p,p'</i> -DDE, <i>p,p'</i> -DDT, <i>o,p'</i> -DDT, <i>p,p'</i> -DDD, <i>o,p'</i> -DDE
Dioxins and furans	
PCDDs	7 congeners
PCDFs	10 congeners
Brominated flame retardants	
PBDE	28, 47, 66, 99, 100, 138, 153, 154, 183
HBCD	

Polychlorinated dibenzo-*p*-dioxins (PCDDs) and dibenzofurans (PCDFs) were analysed at the National Institute of Public Health and the Environment (RIVM), the Netherlands, using methods described in Glynn et al. (2001).

Non-*ortho* PCBs in samples from 1996 to 1999 were analysed either at the NFA or at RIVM. The LOQs for non-*ortho* PCBs were higher in the NFA method of analysis compared to the RIVM method. Non-*ortho* PCBs in samples collected from 2000 to 2004 were analysed at the NFA.

Calculations and statistics

Lipid adjusted breast milk POP concentrations were used in the statistical analysis since lipid-adjusted concentrations give a better estimate of the body burden than non-adjusted concentrations (see Lignell et al. 2004). In the case of concentrations below the limit of quantification (LOQ), half of this limit was taken as an estimated value in the calculations.

Breast milk levels of PCB 52, PCB 101, PCB 114, PCB 157, PCB 77, PCB 81, α -HCH, γ -HCH, *p,p'*-DDD, *o,p'*-DDE, *o,p'*-DDT, BDE-28, BDE-66, BDE-154, BDE-138 and BDE-183 were low (>50 % of the samples below LOQ), and these substances were therefore omitted from the statistical analysis. Time trends were not established for PCB 170 and HBCD since these compounds only were analysed in a few samples from the most recent years.

The distributions of the organochlorine analytical results closely followed a log-normal distribution, therefore all statistical analysis were performed on logarithmically transformed (ln) data.

Statistical analysis was performed in MINITAB[®] for Windows 12.22. Multiple linear regression was used to analyse associations between POP levels and sampling year. Independent variables (life-style factors) that have been shown to influence POP levels in breast milk (Lignell et al. 2004) were included as explanatory variables in the model. The variables considered were age of the mother (years), pre-pregnancy BMI (Body Mass Index, kg/m²) and body weight change during pregnancy as well as after delivery (Table 1). In the multiple regressions, observations with a standard residual ≥ 3 were excluded due to a large influence on the results. Observations that had a large influence on the results (as indicated by

the computer program), but for other reasons than a large standard residual, were also excluded. As a consequence of the logarithmic transformation, the influence of sampling year on POP levels is presented as percent change of the median per year, and not as change in absolute levels (Table 4). Stepwise regression was used to study how much the individual explanatory variables contributed to the variation in POP levels.

RESULTS

POP-levels in breast milk

Among the PCB congeners (excluding the non-*ortho* congeners), the di-*ortho* congener PCB 153 showed the highest median concentration (55 ng/g lipid) followed by PCB 138 (27 ng/g lipid) and PCB 180 (26 ng/g lipid) (Table 3). Among the mono-*ortho* congeners, PCB 118 showed the highest median concentration (10 ng/g lipid). PCB 28 and showed the largest variation in levels (about 60-fold). The levels of PCB 52, 101, 114 and 157 were low (>50 % of the samples below LOQ, results not shown).

Among the non-*ortho* PCBs, PCB 126 showed the highest median concentration (42 pg/g lipid), whereas the median levels of PCB 81 and PCB 77 were close to the LOQ (results not shown). Note that 47 of the 106 samples collected in 1996-1999 were analysed at RIVM. The remaining samples from 1996-1999 as well as all samples collected in 2000-2004 were analysed at the NFA. LOQs in the analyses differed between RIVM and NFA. Earlier, 26 samples (PCB 126) have been analysed at both laboratories, and a comparison of these results show that there are differences between the laboratories, but no systematic error could be detected (Wilcoxon signed rank test, $P=0.798$). Nevertheless, the fact that two different laboratories have been used adds an extra uncertainty factor to the results.

The median sumPCDD and sumPCDF levels were 107 and 13 pg/g lipid respectively. The number of samples below LOQ ranged from 0 to 4 % for the individual PCDD congeners, and from 0 to 97 % among the PCDF congeners. Congeners with >50% of the samples below LOQ were 1,2,3,4,7,8,9-HpCDF (73%) and 1,2,3,7,8,9-HxCDF (97%). Median PCDD/DF TEQ level was 8.0 pg/g lipid, and the median total TEQ level was 16 pg/g lipid.

Table 3. Concentrations of PCBs, PCDD/DFs, chlorinated pesticides and brominated flame retardants in breast milk (ng/g milk lipid, if nothing else is indicated).

Compound	N	Mean	Median	Min ^a	Max	%<LOQ ^b
PCBs						
PCB 28	295	2.8	1.9	0.5	31	11
PCB 105	295	1.3	1.1	0.4	15	32
PCB 118	295	12	10	2.9	64	0
PCB 138	295	30	27	7.8	94	0
PCB 153	295	60	55	12	186	0
PCB 156	295	4.6	3.9	1.3	24	1
PCB 167	295	1.4	1.3	0.5	5.7	21
PCB 170	92	10	9.1	2.6	26	0
PCB 180	295	28	26	5.0	84	0
mono-ortho TEQ (pg/g lipid) ^c	295	3.7	3.2	0.90	18	-
sumPCB ^d	295	142	129	35	402	-
non-ortho PCBs						
PCB 126 (pg/g lipid)	190	46	42	14	125	7
PCB 169 (pg/g lipid)	190	23	21	9.6	65	29
non-ortho TEQ (pg/g lipid) ^e	190	4.8	4.4	0.97	13	-
sumPCDD ^f	154	117	107	35	393	0-4
sumPCDF ^g	154	13	13	5.6	35	0-97
PCDD/DF TEQ (pg/g lipid)	154	8.6	8.0	3.6	23	-
total TEQ (pg/g lipid)	153	17	16	6.4	39	-
Chlorinated pesticides						
HCB	295	14	14	4.0	29	0
-HCH	295	13	11	2.7	88	0
oxychlordane	295	4.1	3.7	1.1	11	0
trans-nonachlor	295	7.2	6.5	1.7	27	0
p,p'-DDT	295	8.6	5.9	0.84	240	1
p,p'-DDE	295	111	93	20	649	0
Brominated flame retardants						
BDE-47	181	2.2	1.5	0.35	16	1
BDE-99	181	0.49	0.30	0.09	5.2	18
BDE-100	181	0.38	0.27	0.07	5.1	7
BDE-153	181	0.67	0.58	0.20	4.6	0
sumPBDE ^h	181	3.8	2.9	0.91	28	-
HBCD	69	0.47	0.32	0.14	4.4	23

^aLowest value >LOQ (limit of quantification).

^bDepending on the lipid content of the samples, the LOQ varied between 0.3 and 1.7 ng/g for PCBs, 0.1 and 4 pg/g for PCDD/DFs, 0.2 and 3.4 ng/g for pesticides and between 0.06 and 0.58 ng/g for flame retardants. LOQ in the analyses of non-ortho PCBs (PCB 126 and 169) at the NFA varied between 12 and 39 pg/g while LOQs in the analyses at RIVM were lower. Concentrations below LOQ were set to 1/2 LOQ in the calculation of means, medians, mono-ortho TEQ, sumPCB, non-ortho TEQ, sumPCDD/DF, PCDD/DF TEQ and sumPBDE.

^cIncluding CB 105, 118, 156, and 167 TEQs.

^dIncluding CB 28, 52, 101, 105, 118, 138, 153, 156, 167 and 180.

^eIncluding CB 126 and 169 TEQs.

^fSum of concentrations of 7 PCDD congeners.

^gSum of concentrations of 10 PCDF congeners.

^hincluding BDE-47, -99, -100, -153 and 154.

p,p'-DDE was the compound with the overall highest median concentration (93 ng/g lipid). The median concentrations of the other chlorinated pesticides were low (<15 ng/g lipid), with levels of *γ*-HCH, *α*-HCH, *p,p'*-DDD, *o,p'*-DDE and *o,p'*-DDT below LOQ in 84-100 % of the samples (results not shown).

Among the PBDEs, BDE-47 showed the highest median concentration (1.5 ng/g lipid). The levels of BDE-28, BDE-66, BDE-154, BDE-138 and BDE-183 were low (>50 % of the samples below LOQ, results not shown).

Temporal trends

PCBs

Multiple linear regression showed that the adjusted mean decrease in PCB levels (mono- and di-*ortho*) varied between 4.3 and 8.7 % per year depending on congener, and the calculated time to halve the concentrations in the population (the “half-time”) was 8 to 16 years (Table 4, Figure 1-2). The fastest decline was shown for PCB 118, 153 and 180, while PCB 28 and 167 declined more slowly during the time period. The regression model explained 7.2-76 % of the variation in PCB-levels, with the lowest degree of explanation for PCB 28 and the highest for PCB 180.

The adjusted mean decrease in levels of PCB 126 and PCB 169 (non-*ortho* PCBs) was 7.2 and 3.0 % per year respectively, which corresponds to “half-times” of 9 and 23 years (Table 4, Figure 2). The regression model explained 42-49 % of the variation. As mentioned before, non-*ortho* PCBs were analysed at two different laboratories, and it can not be excluded that this had an influence on the results of the regression analysis.

Generally, age had the greatest influence on the levels of PCBs in breast milk. Apart from for PCB 28, stepwise regression showed that 17-46 % of the variation in PCB levels could be explained by the variation in age of the mothers. Sampling year explained 17-24 % of the variation in levels of PCB 118, PCB 138, PCB 153, PCB 156, PCB 180 and PCB 126, while it only explained a small part (1-4 %) of the variation in levels of PCB 28, PCB 105, PCB 167 and PCB 169. BMI and weight changes during/after pregnancy only had minor influence on the PCB levels in breast milk.

Table 4. Changes in levels of POPs in breast milk from primiparae women living in Uppsala County 1996-2004. Associations between ln-transformed POP levels and year of sampling adjusted for lifestyle variables (age, BMI, weight gain during pregnancy and weight loss after delivery).

Compound	Change/yr (%) ^a		R ^{2b} (%)	"half-time" ^c	P ^d	Significant explanatory variables ^e
	Mean	sd				
PCBs						
PCB 28	-4.8	1.9	7.2	14	0.015	weight gain, weight reduction
PCB 105	-6.2	1.6	27	11	<0.0005	age, weight gain
PCB 118	-8.7	0.7	53	8	<0.0005	age, weight gain/reduction
PCB 138	-7.5	0.7	54	9	<0.0005	age, BMI, weight gain/reduction
PCB 153	-8.4	0.6	65	8	<0.0005	age, BMI, weight gain/reduction
PCB 156	-7.0	0.6	67	9	<0.0005	age, BMI, weight gain/reduction
PCB 167	-4.3	1.2	41	16	0.001	age, weight gain
PCB 180	-8.1	0.5	76	8	<0.0005	age, BMI, weight gain/reduction
mono-ortho TEQ	-6.2	0.6	63	11	<0.0005	age, BMI, weight gain/reduction
sumPCB	-7.3	0.6	66	9	<0.0005	age, BMI, weight gain/reduction
non-ortho PCBs						
PCB 126	-7.2	1.0	42	9	<0.0005	age, weight gain
PCB 169	-3.0	1.1	49	23	0.006	age, BMI, weight gain/reduction
non-ortho TEQ	-7.4	1.0	43	9	<0.0005	age, weight gain
sumPCDD ^f	-8.3	1.2	35	8	<0.0005	age, weight gain/reduction
sumPCDF ^g	-4.3	0.9	39	16	<0.0005	age, weight gain/reduction
PCDD/DF TEQ	-6.3	0.8	57	11	<0.0005	age, weight gain
Total TEQ	-6.6	0.8	61	10	<0.0005	age, BMI, weight gain/reduction
Chlorinated pesticides						
HCB	-7.8	0.5	52	9	<0.0005	age, weight gain/reduction
-HCH	-10	0.6	63	6	<0.0005	age, weight gain
oxychlorane	-6.3	0.6	58	11	<0.0005	age, BMI, weight gain/reduction
trans-nonachlor	-6.2	0.7	53	11	<0.0005	age, BMI, weight gain/reduction
p,p'-DDT	-10	1.1	32	6	<0.0005	age, weight gain
p,p'-DDE	-10	1.0	41	6	<0.0005	age, weight gain/reduction
Brominated flame retardants						
BDE-47	-3.8	1.8	1.2	18	0.039	-
BDE-99	-7.5	1.9	10	9	<0.0005	-
BDE-100	+2.0	2.1	1.3	-	0.341	-
BDE-153	+5.4	1.1	32	-	<0.0005	age, BMI, weight gain
sumPBDE	-1.5	1.6	0.4	46	0.338	weight gain

^aPercent change, decrease (-) or increase (+), of the median levels per year

^bCoefficient of determination for the whole regression model

^cThe estimated time to halve the concentrations in the population.

^dSignificance for the association between POP concentration and sampling year.

^eLifestyle variables, other than sampling year, that showed statistically significant associations to the POP levels in the multiple regression model (p<0.05).

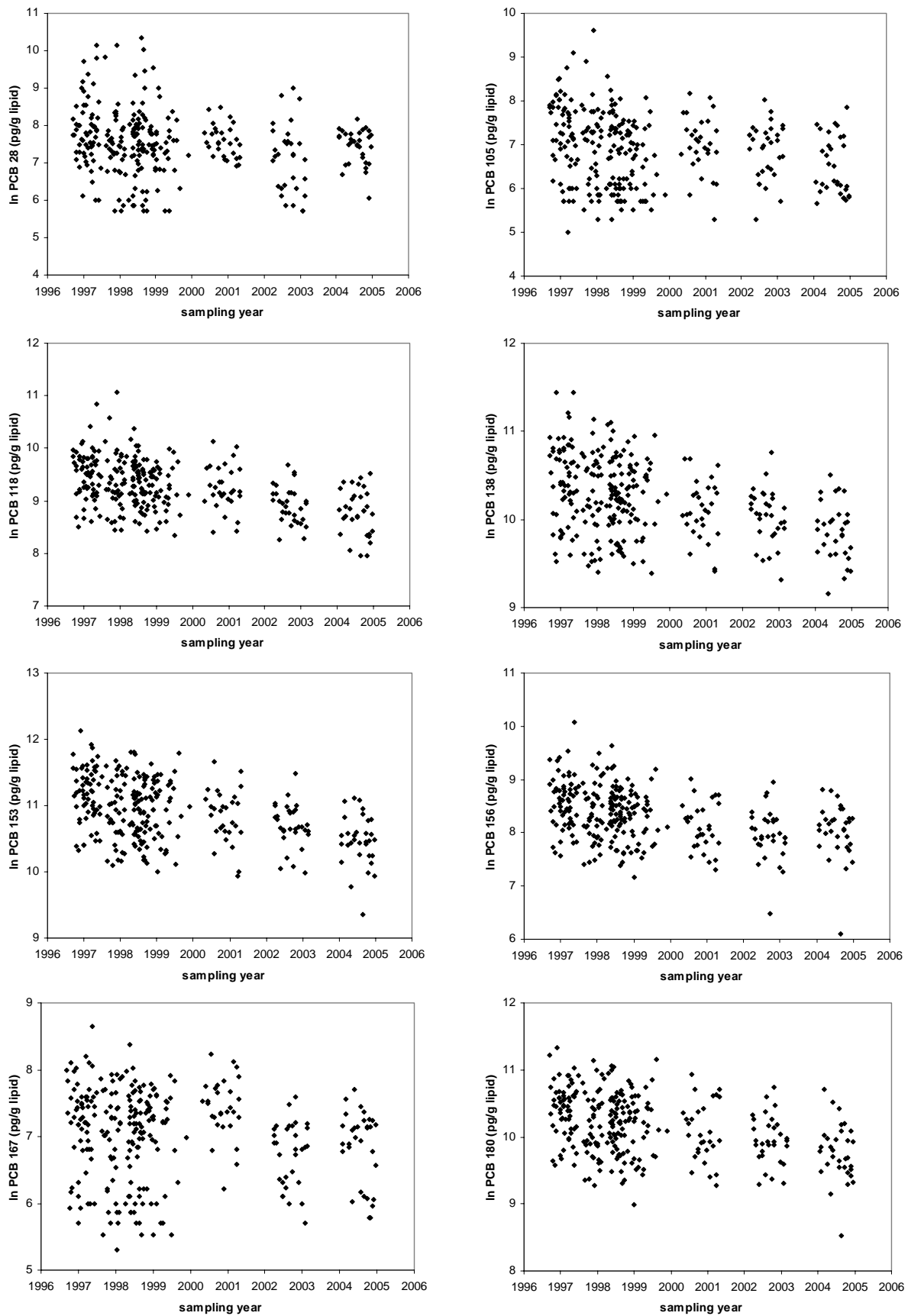


Figure 1. Temporal trends (1996-2004) of PCB concentrations (ln transformed data) in breast milk from primiparous mothers living in Uppsala County, Sweden (N=295). Note that the plots are based on raw data that have not been adjusted for life-style factors.

PCDD, PCDF and total TEQ

Multiple linear regression showed that the concentrations of sumPCDD, sumPCDF and PCDD/DF TEQ declined on average 8.3, 4.3 and 6.3 % per year, respectively, during the time period, and the “half-times” were estimated to 8, 16 and 11 years (Table 4, Figure 2). The regression model explained 35, 39 and 57 % in the levels respectively. Age explained 11-32 % and sampling year 11-22 % of the variation in levels of sumPCDD, sumPCDF and the PCDD/DF TEQ .

The adjusted mean decrease in levels of total TEQ was 6.6 % per year, which corresponds to a “half-time” of 10 years (Table 4, Figure 2). Age (31%) and sampling year (23 %) had the greatest influence on the levels of total TEQ, and the whole regression model explained 61 % of the variation.

Chlorinated pesticides

Multiple linear regression showed that the adjusted mean decrease in levels of chlorinated pesticides varied between 6.2 and 10 % per year, where *γ*-HCH, *p,p'*-DDT and *p,p'*-DDE showed the fastest decline and oxychlorane and *trans*-nonachlor the slowest (Table 4, Figure 3). The calculated “half-times” were 6-11 years. The regression model explained 32-63 % of the variation in pesticide levels, with the lowest degree of explanation for *p,p'*-DDT and *p,p'*-DDE and the highest for *γ*-HCH. Age (11-38 %) and sampling year (11-30%) had the greatest influence on the levels of pesticides in breast milk.

Brominated flame retardants

Regarding the PBDE congeners, there were no general trends during the time period (Table 4, Figure 4). Multiple linear regression showed that the levels of BDE-47 and BDE-99 decreased significantly (3.8 and 7.5 % per year respectively), while the levels of BDE-153 increased (5.4 % per year). No significant trends were shown for BDE-100 and sumPBDE. In contrast to the other POPs, there were only few significant associations between PBDE-levels and the explanatory variables included in the regression model. The regression model only explained 0.4-10 % of the variation in levels of BDE-47, BDE-99, BDE-100 and sumPBDE. However, BDE-153 deviated from the other PBDEs in this aspect, and the BDE-153 levels were significant associated to age, BMI, and weight gain during pregnancy (the regression model explained 32 % of the variation in BDE-153 levels).

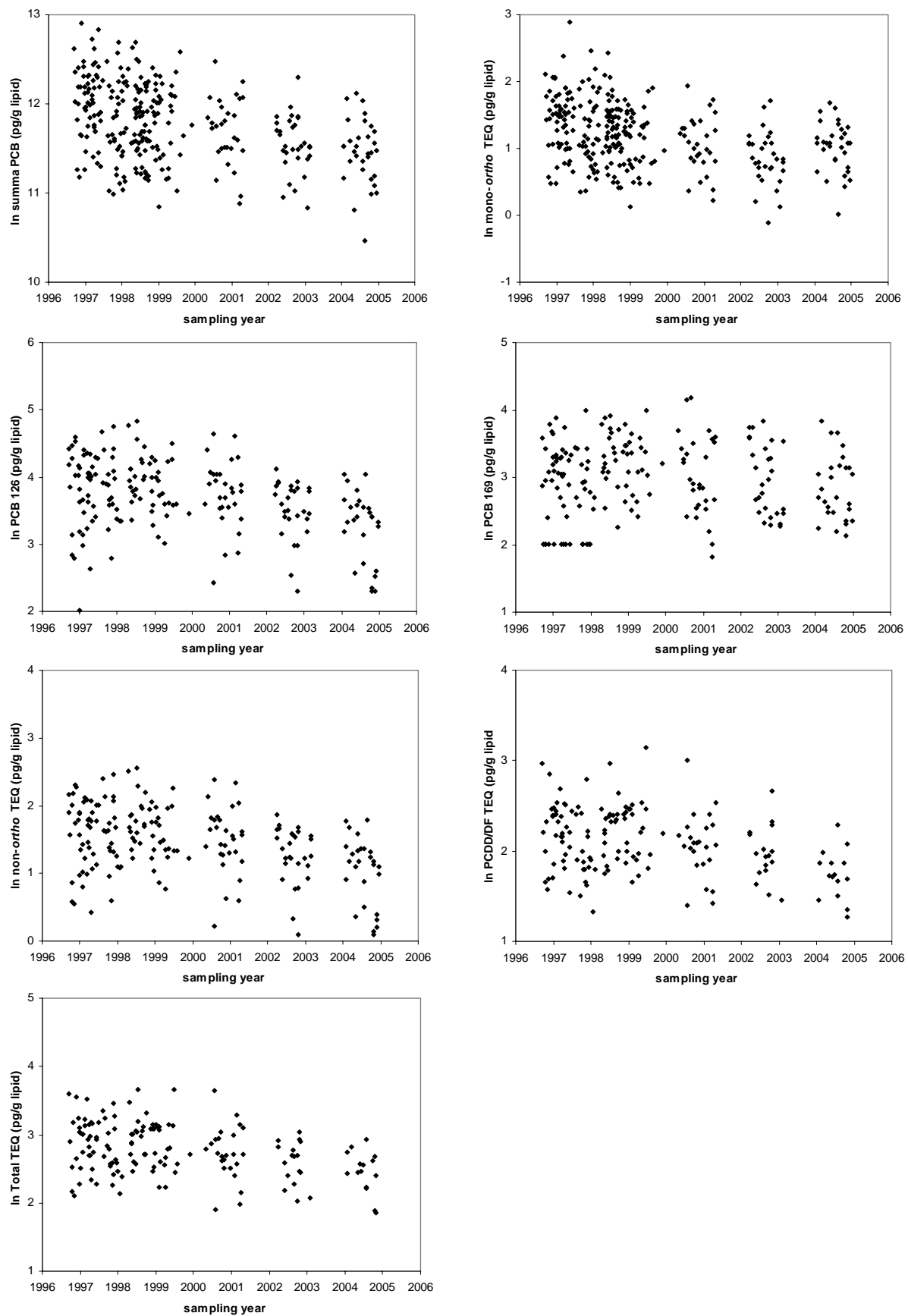


Figure 2. Temporal trends (1996-2004) of sum PCB, mono-ortho TEQ, non-ortho PCBs, non-ortho TEQ, PCDD/DF TEQ and total TEQ concentrations (ln transformed data) in breast milk from primiparous mothers living in Uppsala County, Sweden (N=153-295, see Table 3). Note that the plots are based on raw data that have not been adjusted for life-style factors.

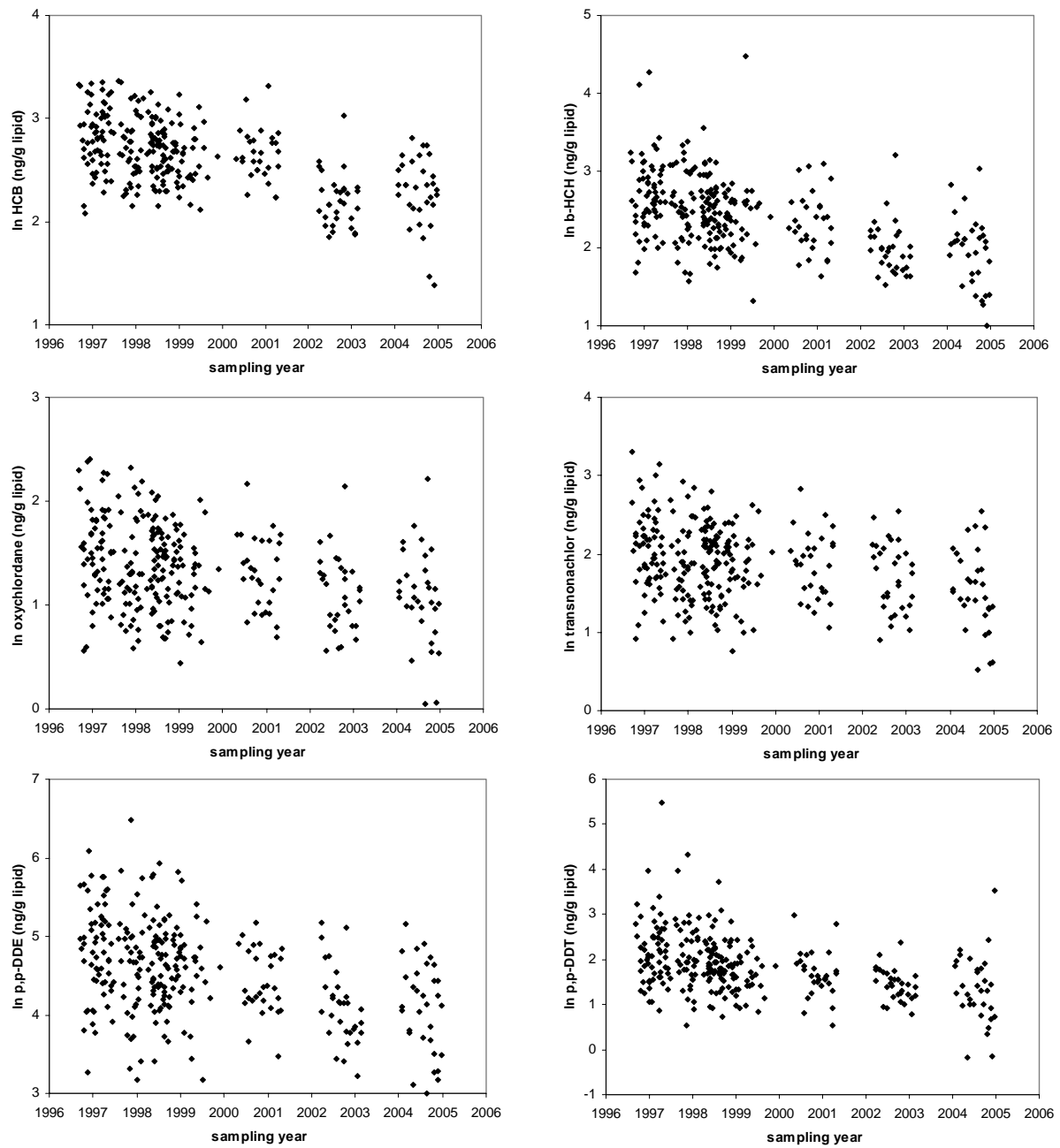


Figure 3. Temporal trends (1996-2004) of chlorinated pesticide concentrations (ln transformed data) in breast milk from primiparous mothers living in Uppsala County, Sweden (N=295). Note that the plots are based on raw data that have not been adjusted for life-style factors.

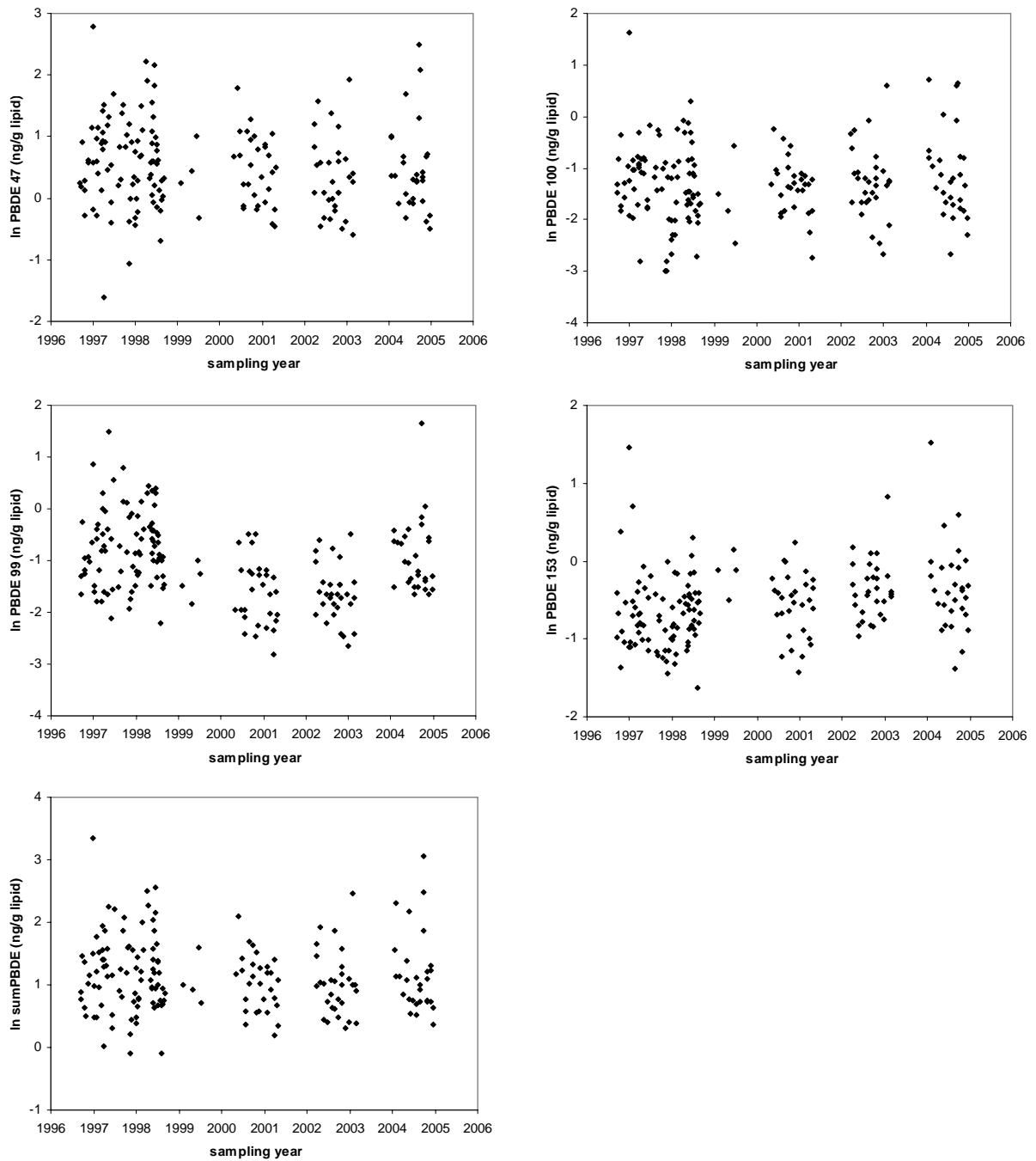


Figure 4. Temporal trends (1996-2004) of PBDE concentrations (ln transformed data) in breast milk from primiparous mothers living in Uppsala County, Sweden (N=181). Note that the plots are based on raw data that have not been adjusted for life-style factors.

DISCUSSION

Our results show that the levels of most POPs (PCBs, dioxins/furans, chlorinated pesticides) in breast milk have decreased from 1996 to 2004. This is probably a consequence of reduced levels of many POPs in the environment (and in foods) since the 1970s, i.e. a 30-year-old woman who had her first child in 1996 had been exposed to higher life-time cumulative levels of POPs before pregnancy than a 30-year-old woman who had her first child in 2004.

In the present report, the temporal trends of PCBs and chlorinated pesticides (1996-2003) that we presented earlier (Lignell et al. 2004), have been updated with data from 2004. The “half-times” for some of the compounds, especially for PCB 28, PCB 156, HCB, oxychlordane and *trans*-nonachlor, are somewhat longer in the present study compared to the earlier (Table 5). The explanation for this may be that the declines in POP levels are levelling out and/or that the calculated “half-times” are uncertain because of the relatively short time span of the study.

Table 5. Comparison of estimated “half-times” of levels of PCBs and chlorinated pesticides in breast milk from mothers living in Uppsala 1996-2003 (Lignell et al. 2004), Uppsala 1996-2004 (present report) and in Stockholm 1967-1997 (Norén & Meironyté, 2000).

Compound	Estimated “half-lives” (years)		
	Uppsala 1996-2003	Uppsala 1996-2004	Stockholm ^a
PCB 28	8	14	
PCB 105	10	11	
PCB 118	7	8	11 (1972-1997)
PCB 138	9	9	14 (1972-1997)
PCB 153	8	8	17 (1972-1997)
PCB 156	6	9	
PCB 167	15	16	
PCB 180	8	8	
sumPCDD		8	15 (1972-1997)
sumPCDF		16	11 (1972-1997)
Total TEQ		10 ^b	15 ^c (1992-1997)
HCB	7	9	6 (1974-1997)
-HCH	6	6	
oxychlordane	8	11	
<i>trans</i> -nonachlor	9	11	
<i>p,p'</i> -DDT	5	6	4 (1967-1997)
<i>p,p'</i> -DDE	5	6	6 (1972-1997)

^aTime period studied in brackets.

^bIncluding mono-*ortho* TEQ, non-*ortho* TEQ and PCDD/DF TEQ (see Table 3).

^cIncluding mono-*ortho* TEQ and PCDD/DF TEQ.

As discussed in the earlier report (Lignell et al. 2004) the estimated “half-times” of PCBs are considerably shorter in our study compared to a study of mothers from Stockholm (Norén & Meironyté, 2000) (Table 5). There are probably several reasons behind this difference. One reason could be the difference in time span of the studies. It is possible that the rate of decline was slower in the early part of the Stockholm study, when direct exposure from products containing PCB was still a reality. The most likely explanation, however, is that the Stockholm study lacked in control of life-style factors that influence breast milk levels of POPs. For instance, pooled samples, containing milk from both primiparous and multiparous women, were analysed in the Stockholm study, while we only recruited primiparous women in our study. It is well known that breast feeding is a major pathway of POP excretion in women, and consequently the POP levels are often higher in breast milk sampled after the first child is born than in breast milk sampled after subsequent deliveries (Vaz et al. 1993). Another very important factor is that the average age of the mothers donating breast milk in the Stockholm study increased between 1972-1985 (27-28 years) and 1996-1997 (30-31 years). In our study, the “half-times” were age-adjusted, thus taking care of possible differences in average age of the women between sampling years. PCB levels in breast milk increase with increasing age of the mothers. Figure 5 shows the levels of PCB 153 and PCB 180 in breast milk (from our study) divided into three different age groups.

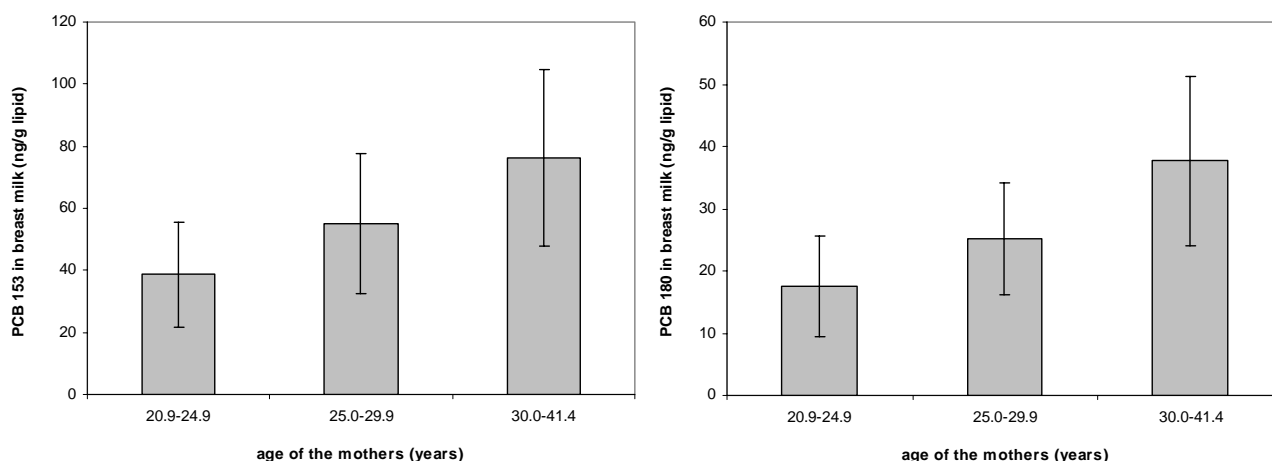


Figure 5. Mean levels (and standard deviation) of PCB 153 and PCB 180 in breast milk from primiparous mothers in Uppsala 1996-2004 (N=295) divided into three different age groups.

Regarding dioxins and furans, the “half-times” of sumPCDD and total TEQ are longer and the half-time of sumPCDF is shorter in the Stockholm study compared to our study. The reason

for the differences may be the lack in control of life-style factors in the Stockholm study that already has been discussed. We do not have any good explanation to the shorter “half-time” of sumPCDF in the Stockholm study compares to our study.

Based on the same data as we used in this report, but using data from 1996 to 2001 only, Lind et al. (2003) indicated that the levels of both BDE-47 and sumPBDE increased from 1996 to 1998, after which the levels appeared to decrease up to 2001. However, the within-year variation in individual concentrations was large, and no significant change between the median levels for 1996-1998 and those for 1998-2001 was noted. The findings by Lind et al. (2003) were similar to what has been reported by Meironyté-Guvenius (2002) in pooled samples from mothers in the Stockholm region (Meironyté et al. 1999, Meironyté-Guvenius 2002 and Norén & Meironyté 2000). Data on PBDE levels in breast milk from 2002-2004 have now been added to the data presented by Lind et al. (2003), and in a regression analysis we can show significant decreases in levels of BDE-47 and BDE-99 during the whole time period (1996-2004).

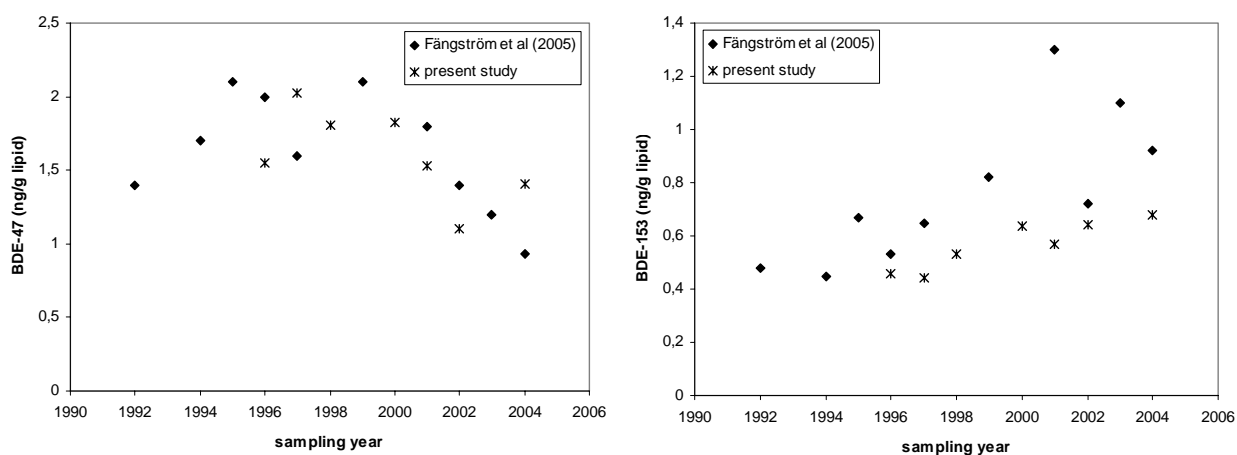


Figure 6. A comparison between data on BDE-47 and BDE-153 levels in pooled breast milk from Fångström et al. (2005) and median values of individual samples from the present study (1996-2004). The median values from 1999 and 2003 (present study) were omitted from the graph because of the small number of observations (N=4 and 5 respectively).

In a recent study by Fångström et al. (2005), PBDEs were analysed in pooled milk samples from mothers in the Stockholm region in 1980-2004 (to some extent the same pools as in the studies by Meironyté and co-workers). The results from this study indicate that that the levels of lower brominated PBDE congeners (e.g. BDE-47, BDE-99) have decreased from the middle of the 1990’s while the levels of BDE-153 have increased. This is in agreement with

the findings in our study (Figure 6), and the explanation may be a reduced use of lower brominated PBDEs and/or a higher persistence of BDE-153 compared to BDE-47. By this time, it is not possible to establish any solid temporal trend for the PBDE congeners studied in breast milk from Uppsala County. This makes it important to continue to sample breast milk for PBDE analysis in the future.

Our results show that it is important to consider life-style factors, especially age of the mothers, when temporal trends of most POPs in breast milk are studied. Age was the most important explanatory variable for several of the compounds (PCB 105, 118, 138, 153, 156, 167, 180, 169, sumPCDF, PCDD/DF TEQ, α -HCH, oxychlorane, *trans*-nonachlor, *p,p'*-DDT), and explained 20-46 % of the variation in breast milk levels of these substances. Sampling year explained >10 % of the variation in levels of all compounds except for PCB 28, PCB 105, PCB 167, PCB 169, BDE-47, BDE-99, BDE-100 and sumPBDE. Pre-pregnancy BMI and weight changes during/after pregnancy explained smaller parts of the variation in POP-levels. As mentioned earlier, the levels of many POPs in the environment have decreased since the 1970s. Consequently, the older women in our study have been exposed to higher levels than the younger women. Another explanation of the association between POP levels in breast milk and age is the bioaccumulation of these persistent substances in the body. Older primiparas have accumulated POPs in the body during a longer period than younger primiparas.

For some of the compounds (PCB 153, PCB 156, PCB 180, mono-*ortho* TEQ, sumPCB, total TEQ and α -HCH), the whole regression model explained over 60 % of the variation in breast milk levels. There are, however, still explanatory variables that have to be accounted for in order to increase the precision of estimated changes in POP levels over time. This is illustrated by the low R^2 of the regression model describing the variation in levels of PCB 28, PCB 105, *p,p'*-DDT, and PBDEs (1.2-32 %).

With the exception of BDE-153, there were particularly few significant associations between PBDE-levels and the explanatory variables in the regression model. One explanation to the absence of associations between PBDE levels and age may be that brominated flame retardants came into general use in the 1970s, when the phase out of many other POPs already had begun. In contrast to PCBs and chlorinated pesticides, the older women in our study have

consequently not been exposed to higher levels of PBDEs than the younger women. Another explanation may be that PBDEs are less persistent and do not bioaccumulate to the same extent as many other POPs. However, there is a significant association between BDE-153 levels and age, where age explains 9 % of the variation in BDE-153 levels. The reason for this can possibly be a higher persistence of BDE-153 compared to BDE-47, BDE-99 and BDE-100, and/or another usage pattern.

ACKNOWLEDGEMENT

The Swedish EPA (Environmental Protection Agency) is acknowledged for financial support. Appreciation is expressed to the participating women and to the midwives who assisted in recruitment, interviewing, and sample collection. The laboratory technicians Arpi Bergh, Ingalill Gadhasson, Martin Isaksson, Lotta Larsson and Elvy Netzel are appreciated for technical assistance.

REFERENCES

- Atuma SS, Aune M. 1999. Method for the determination of PCB congeners and chlorinated pesticides in human blood serum. *Bull Environ Contam Toxicol* 62(1), 8-15.
- Atuma S, Aune M, Darnerud PO, Cnattingius S, Wernroth ML, Wicklund Glynn A. Polybrominated diphenyl ethers (PBDEs) in human milk from Sweden. 2000. In: Lipnick RL, Jansson B, Mackay D, Petreas M, editors. *Persistent, bioaccumulative and toxic chemicals II*. Washington, DC: ACS symposium series 773, 235-242.
- Aune M, Atuma S, Darnerud PO, Wicklund-Glynn A, Cnattingius S. 1999. Analysis of organochlorine compounds in human milk. *Organohalogen Compounds* 44, 93-96.
- Fängström B, Strid A, Bergman Å. 2005. Rapport till Naturvårdsverket (Dnr 721-2653-05Mm): Temporal trends of brominated flame retardants in milk from Stockholm mothers, 1980-2004.
- Glynn A., Atuma S., Aune M., Darnerud P.O. and Cnattingius S. 2001. *Environ. Res.* 86 (3), 217-228.

- Lignell S, Darnerud PO, Aune M, Törnkvist A, Glynn A. 2004. Report to the Swedish Environmental Protection Agency: Polychlorinated biphenyls and chlorinated pesticides/metabolites in breast milk from primiparae women in Uppsala County, Sweden – levels and trends 1996-2003.
- Lind Y, Darnerud PO, Atuma S, Aune M, Becker W, Bjerselius R, Cnattingius S, Glynn A. 2003. Polybrominated diphenyl ethers in breast milk from Uppsala County, Sweden. *Environmental Research* 93, 186-194.
- Meironyté D, Norén K, Bergman Å. 1999. Analysis of polybrominated diphenyl ethers in Swedish human milk. A time-related trend study, 1972-1997. *Journal of toxicology and environmental health* 58, 329-341.
- Meironyté-Guvenius D. 2002. Organohalogen contaminants in humans with emphasis on polybrominated diphenyl ethers. Thesis, department of medical biochemistry and biophysics, Karolinska Institute, Stockholm, Sweden, ISBN 91-7349-140-3.
- Norén K, Meironyté D. 2000. Certain organochlorine and organobromine contaminants in Swedish human milk in perspective of past 20-30 years. *Chemosphere* 40, 1111-1123.
- Vaz R, Slorach SA, Hofvander Y. 1993. Organochlorine contaminants in Swedish human milk: studies conducted at the National Food Administration 1981-1990. *Food Add Contam* 10, 407-418.