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PCBs and OH-PCBs in polar bear mother–cub pairs: A comparative study based on plasma levels in 1998 and 2008

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ABSTRACT

The aim of this study was to examine the plasma concentrations and prevalence of polychlorinated biphenyls (PCBs) and hydroxylated PCB-metabolites (OH-PCBs) in polar bear (*Ursus maritimus*) mothers ($n=26$) and their 4 months old cubs-of-the-year ($n=38$) from Svalbard to gain insight into the mother–cub transfer, biotransformation and to evaluate the health risk associated with the exposure to these contaminants. As samplings were performed in 1997/1998 and 2008, we further investigated the differences in levels and pattern of PCBs between the two sampling years. The plasma concentrations of Σ_{21}PCBs (1997/1998: 5710 ± 3090 ng/g lipid weight [lw], 2008: 2560 ± 1500 ng/g lw) and Σ_6OH-PCBs (1997/1998: 228 ± 60 ng/g wet weight [ww], 2008: 80 ± 38 ng/g ww) in mothers were significantly lower in 2008 compared to in 1997/1998. In cubs, the plasma concentrations of Σ_{21}PCBs (1997/1998: 14680 ± 5350 ng/g lw, 2008: 6070 ± 2590 ng/g lw) and Σ_6OH-PCBs (1997/1998: 98 ± 23 ng/g ww, 2008: 49 ± 21 ng/g ww) were also significantly lower in 2008 than in 1997/1998. Σ_{21}PCBs in cubs was 2.7 ± 0.7 times higher than in their mothers. This is due to a significant maternal transfer of these contaminants. In contrast, Σ_6OH-PCBs in cubs were approximately 0.53 ± 0.16 times the concentration in their mothers. This indicates a lower maternal transfer of OH-PCBs compared to PCBs. The majority of the metabolite/precursor-ratios were lower in cubs compared to mothers. This may indicate that cubs have a lower endogenous capacity to biotransform PCBs to OH-PCBs than polar bear mothers. Exposure to PCBs and OH-PCBs is a potential health risk for polar bears, and the levels of PCBs and OH-PCBs in cubs from 2008 were still above levels associated with health effects in humans and wildlife.

1. Introduction

Polychlorinated biphenyls (PCBs) are still among the dominating persistent organic pollutants (POPs) in arctic mammals (Letcher et al., 2010), and the levels of PCBs in polar bears (*Ursus maritimus*) are among the highest reported in any species (e.g. Norstrom et al., 1998; Andersen et al., 2001; Dietz et al., 2004; Verreault et al., 2005a; Letcher et al., 2010; McKinney et al., 2011). In polar bears, PCBs and other POPs are suggested to cause adverse effects on the thyroid hormone system, sex steroid homeostasis, vitamin status, immune system, organ morphology and behaviour (e.g. Wiig et al., 1998; Skaare et al., 2001; Haave et al., 2003; Olsen et al., 2003; Oskam et al., 2003, 2004; Braathen et al., 2004; Lie et al., 2004, 2005; Sonne, 2010).

Adult polar bears have a well developed cytochrome P450 (CYP) enzyme system, which can metabolically biotransform PCBs to PCB-metabolites, and hydroxylated PCBs (OH-PCBs) are among the most common PCB-metabolites in polar bears (Letcher et al., 2000; Verreault et al., 2005b; Gebbink et al., 2008; Letcher et al., 2010). OH-PCBs are less hydrophobic than their parent compounds, and these metabolites bind to proteins and accumulate in blood rather than being associated with lipids as PCBs are (van den Berg, 1990; Lans et al., 1993, 1994). In adult polar bears from Svalbard (Norway), East-Greenland and Resolute Bay in the Canadian Arctic, the levels of OH-PCBs in plasma and whole blood have been reported to be even higher than the levels of PCBs in the same tissue (Sandau, 2000;Sandau et al., 2000; Sandala et al., 2004; Verreault et al., 2005b; Gebbink et al., 2008). The OH-PCBs detected in significant concentrations in polar bears and other mammals, including humans, are *meta-* or *para*-hydroxylated PCBs (Letcher et al., 2000). OH-PCBs have structural similarities to endogenous compounds, and *in vivo* and *in vitro* studies have demonstrated that OH-PCBs may disrupt the transport and

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metabolism of thyroid hormones, vitamin homeostasis, the oestrogen cycle, interrupt intercellular communication, and development of the nervous system (Brouwer and van den Berg, 1986; Rickenbacher et al., 1986; Lans et al., 1993; Brouwer et al., 1998; Schuur et al., 1998, 1999; Cheek, 1999; Kester et al., 2000; Machala et al., 2003, 2004; Meerts et al., 2004; Ptak et al., 2005; Gutleb et al., 2010).

In mammals, PCBs are transferred from mothers to their offspring through the umbilical cord and via the milk (Guvenius et al., 2003; Sørmo et al., 2003a; Greig et al., 2007; Park et al., 2008). The milk of polar bears is lipid-rich and contains high levels of hydrophobic contaminants such as PCBs (Bernhoft et al., 1997; Polischuk et al., 1995, 2002). Levels of PCBs in polar bear milk are higher than in the adult diet and the PCB levels in suckling cubs exceed the levels in their mothers (Bernhoft et al., 1997; Sandau, 2000; Polischuk et al., 2002). Contaminant levels in umbilical blood and the foetuses of polar bears are unreported. Because the polar bear foetuses are small and have minor fat reservoirs (Blix and Lentfer, 1979) for storage of hydrophobic POPs, the POPs entering their body may be more bioavailable to exert toxic effects. Hence, developing polar bears may be of particular risk from harmful effects of these pollutants (Bernhoft et al., 1997; Wiig et al., 1998; Polischuk et al., 2002). Exposure to POPs during critical developmental periods has been associated with negative health effects in wildlife, experimental animals, and humans (Colborn et al., 1993; Peterson et al., 1993; Brouwer et al., 1995, 1998; Huisman et al., 1995; Ulbrich and Stahlmann, 2004; Grandjean and Landrigan, 2006).

Because of the important role of thyroid hormones in normal growth and development of mammals, concern has been expressed about the thyroid disruptive effects of PCBs and OH-PCBs in polar bear cubs (Jenssen, 2006). Furthermore, there are indications of reproductive effects and decreased cub survival in polar bears that may be linked to high concentrations of contaminants (Derocher et al., 2003). Human infants exposed to PCBs and OH-PCBs prenatally show lower birth weight, smaller head circumference and alterations in the thyroid hormone homeostasis (Fein et al., 1984; Rylander et al., 1996; Sandau et al., 2002), and exposed children show altered neural development, cognitive, motor and learning abilities (Schantz et al., 2003; Grandjean and Landrigan, 2006; Maervoet, 2007; Park et al., 2009; Roze et al., 2009). In a recent study, OH-PCBs have been associated with alterations in the thyroid hormone homeostasis in hooded seal (*Cystophora cristata*) pups from East-Greenland (Gabrielsen et al., 2011). There are, however, few reports on PCB and OH-PCB levels in cubs-of-the-year (Sandau, 2000; Polischuk et al., 2002) and there is a lack of knowledge on mother–cub transfer of these two groups of endocrine disrupting compounds.

Starting in the 1970s, the production and use of PCBs have gradually been banned in most countries, and following the ratification of the Stockholm Convention in 2004, production, use and release of PCBs have been globally banned (www.pops.int). Thus, the levels of PCBs in adult polar bears have declined at least since the 1990s (Braune et al., 2005; Verreault et al., 2005a; McKinney et al., 2011). However, there are no reports on changes in levels of OH-PCBs in neither adult polar bears or in cubs. Information on changes in the levels of these environmental pollutants in polar bear mothers and their offspring are important for assessing the risk of exposure, and to assess to which extent international regulatory treaties and national bans of production, use and release of environmental pollutants affect levels in arctic wildlife and ecosystems.

The objective of the study was to examine the levels and prevalence of PCBs, OH-PCBs in polar bears mother and their suckling cubs to gain insight into the mother–cub transfer, biotransformation and to evaluate the health risk associated with the exposure to these contaminants. Thus, contaminant levels were analysed in plasma samples from live-caught female polar bears from Svalbard and their cubs (~4 months old, i.e. cubs-of-the-year), shortly after den emergence. As samples were obtained in 1997/1998 and 2008, we further investigated the differences in levels and patterns of PCBs and OH-PCBs in mothers and cubs from the two sampling periods.

2. Material and methods

2.1. Field sampling

In April 1997, 1998 and 2008, blood samples were collected from 3 polar bear mothers with 5 cubs, 13 mothers with 17 cubs, and 10 mothers with 16 cubs, respectively, at Svalbard, Norway. Cubs were approximately 4 months old at sampling (i.e. cubs-of-the-year), and litters comprised of either one ($n=14$) or two cubs ($n=12$). Because the quantitative rates of change over time in contaminant levels in polar bears are slow (AMAP, 2004), bears sampled in 1997 and 1998 were pooled into one group hereafter termed “1998”. Thus, the bears were divided into 4 groups based on age and sampling year: Mothers 1998, Mothers 2008, Cubs 1998, and Cubs 2008. In 1998, animals were sampled at Hopen and Edgeøya, whereas in 2008 animals were sampled at Spitsbergen and Edgeøya (Fig. 1). Latitude and longitude co-ordinates were recorded for all bears (Table 1; Fig. 1).

Capture and handling procedures followed standard protocols (Stirling et al., 1989; Derocher and Wiig, 2002) and were approved by the National Animal Research Authority (NARA), Norway. Following immobilization, and as part of the routine measurements of polar bears, a selection of morphometric variables representing the bears body size and head size were collected. Dorsal straight-line body length (SL), head length (HL) and zygomatic width (ZW) were measured in all bears, axillary girth (AG) was recorded in all mothers, and body mass (BM) was recorded by spring scale for all cubs (Derocher et al., 2005) (Table 1). Because BM was measured only for mothers from 2008, BM for all mothers was estimated based on SL and AG using a morphometric equation (Derocher and Wiig, 2002) before further recalculation into body condition index (BCI) using a BCI equation developed for polar bears (Cattet et al., 2002). The calculated BCI closely represent the polar bears true body condition. For some mothers, age was known because they had been caught previously, during their first two years of life. For the remaining mothers, age was estimated by counting annual growth layers in the cementum of an extracted vestigial premolar (Calvert and Ramsay, 1998; Christensen-Dalsgaard et al., 2010). All capture dates were transformed to capture day (1–365). Detailed information on capture day, age, and morphometric variables for the 4 groups are listed in Table 1.

Blood was collected from the femoral vein into heparinised Venoject® tubes (10 mL, Thermo Electron Corporation, Belgium) and separated into plasma and blood cells by centrifugation (3500 rpm, 10 min) within 8 h after sampling. Plasma samples were transferred to cryogenic vials and stored at −20 °C in the field and then at −70 °C in the lab freezer until analysis.

2.2. Contaminant analysis

The analyses of PCBs and OH-PCBs in the plasma samples were performed at the Laboratory of Environmental Toxicology at the Norwegian School of Veterinary Science (Oslo, Norway).

2.2.1. Extraction of PCBs and OH-PCBs

The multicomponent method used for extraction, determination of plasma lipid percentage (PL%), clean-up and analyses of the PCBs is based on the procedure originally described by Brevik (1978) with modifications described by Bernhoft et al. (1997) and Andersen et al. (2001), and extended to include OH-PCBs as described by Løken et al. (2006) and Berg et al. (2010). Briefly, the samples (~3 g of plasma) were added an internal standard (I.S.) mix consisting of PCB-29, PCB-112, PCB-207 (Ultra Scientific, RI, USA), 4′-OH-

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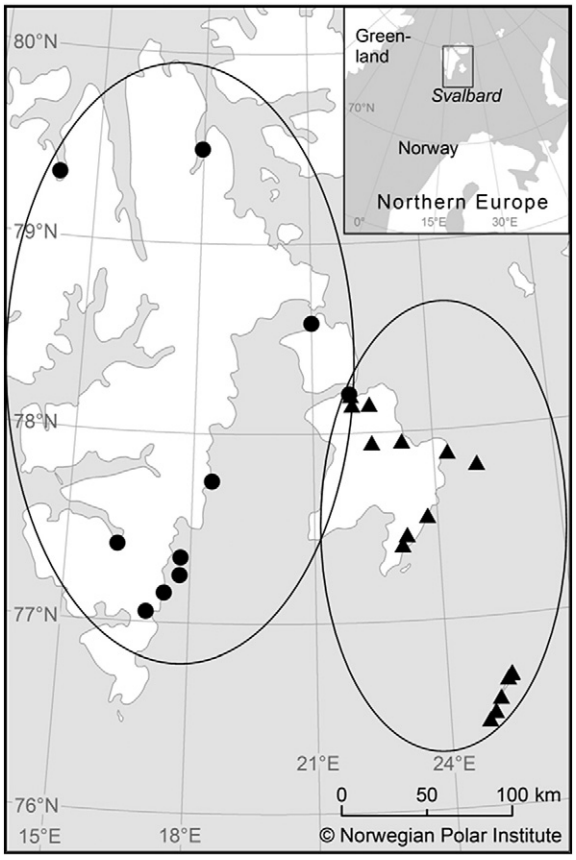

Fig. 1. Capture location of polar bear mothers with 4 months old cubs sampled in Svalbard (Norway) in 1998 ($n=16$, filled triangles) and 2008 ($n=10$, filled circles).

[$^{13}C_{12}$]CB159 and 4-OH-[$^{13}C_{12}$]CB187 (Wellington Laboratories Inc., Guelph, Ontario, Canada), 10 mL 1 M H_2SO_4 (96%) and 4 mL 2% NaCl. Lipid-extraction was performed twice with acetone:cyclohexane (2:3). PL% was determined gravimetricly using the whole extract. Sulphuric acid (H_2SO_4, 96%) was used for clean-up. The samples were then extracted twice with 1 M potassium hydroxide (KOH, Elektrokemiska Aktiebolaget, Sweden) in ethanol:water (1:1) (Ethanol, 96%, Arcus AS, Oslo, Norway and Grade 1 water). The solvent phase containing the PCBs was ready for GC-analyses after reduction to the final volume. The alkaline phase containing the phenolic analytes was treated with drops of 96% sulphuric acid (to pH ~1–2) and back-extracted three times with cyclohexane (5 mL). The OH-PCBs in the combined extracts were reduced (~1 mL) and thereafter derivatised with an acetylating reagent mixture (100 μL) of acetic acid anhydride and pyridine (1:1) to yield the acetylated analogues to the OH-PCBs. After 30 min in a heating cabinet (60 °C), the samples were cleaned with water (Grade 1, 2 mL). The organic phase containing the OH-PCBs was then reduced (~0.3 mL) and transferred to amber vials with inlets before OH-PCB analysis.

Table 1
Mean (X) ± standard deviation (SD) of capture location as latitude and longitude, capture day (1–365), age (years for mothers and months for cubs), head length (HL, mm) zygomatic width (ZW, mm), straight length (SL, cm), axillary girth (AG, cm), body mass (BM) and body condition index (BCI) of polar bear mothers and cubs sampled in Svalbard (Norway) in 1998 and 2008.

	Mothers 1998	Mothers 2008	Cubs 1998	Cubs 2008
	($n=16$)	($n=10$)	($n=16$)	($n=10$)
	X ± SD	X ± SD	X ± SD	X ± SD
Latitude (°N)	77.4 ± 0.7	78.0 ± 0.9	77.4 ± 0.7	78.0 ± 0.9
Longitude (°E)[a,b]	23.9 ± 1.3	18.0 ± 2.3	23.9 ± 1.3	18.0 ± 2.3
Capture day (1–365)[a,b]	104 ± 7	112 ± 7	104 ± 7	112 ± 7
Age (years, months)	12 ± 3	12 ± 5	~4	~4
HL (mm)[b]	347 ± 13	339 ± 9	162 ± 7	177 ± 15
ZW (mm)[b]	201 ± 10	199 ± 6	100 ± 6	107 ± 8
SL (cm)[b]	196 ± 8	192 ± 5	73 ± 6	84 ± 11
AG (cm)	114 ± 9	110 ± 5	n.m.	n.m.
BM (kg)[b,c]	185 ± 34	169 ± 17	11 ± 3	15 ± 6
BCI[d]	158 ± 13	152 ± 8	n.c.	n.c.

n.m. = not measured, n.c. = not calculated.
[a] Significant difference between mothers sampled in 1998 and 2008 ($p<0.05$).
[b] Significant difference between cubs sampled in 1998 and 2008 ($p<0.05$).
[c] Estimated body mass (BM) of mothers is based on the following equation: BM = 0.00003377 axillary girth$^{1.7515}$ straight length$^{1.3678}$ (Derocher and Wiig, 2002). BM of cubs was measured by spring scale.
[d] Body condition index (BCI) of mothers is based on the following equation: BCI = (ln body mass − 3.07 ln straight length + 10.76)/(0.17 + 0.009 ln straight length) (Cattet et al., 2002). ln = natural logarithm.

2.2.2. Quantification of PCBs and OH-PCBs

Details concerning the quantification of PCBs are described by Andersen et al. (2001). The following 28 PCB compounds were analysed in the plasma samples; PCB-28, -47, -52, -66, -74, -99, -101, -105, -114, -118, -123, -128, -137, -138, -141, -149, -151, -153, -156, -157, -167, -170, -180, -183, -187, -189, -194, and -206.

The extracts containing the acetylated analogues of the OH-PCBs in the plasma were analysed by a high-resolution gas chromatograph (Agilent 6890 Series, Agilent Technologies, Santa Clara, CA, USA) with auto sampler and split/splitless injector (Agilent 7683 Series) operated in the pulsed splitless mode. This system was connected to a quadrupole mass spectrometer (MS) (Agilent 5973 Series). The capillary column was a DB-5 MS (J&W Scientific, 60 m, 0.25 mm i.d., 0.25 μm film thickness). Hydrogen (purity: 99.999%, Yara Praxair) was used as carrier gas at a constant flow of 1 mL/min and methane (purity: 99.995, Yara Praxair) was used as reagent gas. The injected volume was 2 μL, and the injection temperature was 210 °C. Detection of the OH-PCBs and the I.S. was performed using negative chemical ionization (NCI) with the following target ions: 4′-OH-CB106 and 4′-OH-CB108 at m/z 383.8, 4-OH-CB107 at m/z 383.9, 3-OH-CB118 at m/z 309.9, 4′-OH-CB130 and 3′-OH-CB138 at m/z 345.9, 4-OH-CB146 at m/z 417.8, 4′-OH-CB159 at m/z 417.8, 4′-OH-^{13}C-CB159 at m/z 429.8, 4′-OH-CB172 and 3′-OH-CB180 at m/z 451.8, 4-OH-CB187 at m/z 451.8, and 4 –OH-^{13}C-187 at m/z 464.0. The following temperature programme was used: 90 °C (hold 1 min); 90–250 °C (35 °C/min, hold 5 min); 250–300 °C (5 °C/min, hold 5 min). The total runtime was 25.57 min. A total of 11 OH-PCBs were analysed: 4′-OH-CB106 (4-hydroxy-2′,3,3′,4′,5′-pentachlorobiphenyl), 4-OH-CB107 (4-hydroxy-2,3,3′,4′,5–pentachlorobiphenyl), 4′-OH-CB108 (4-hydroxy-2′,3,3′,4′,5-pentachlorobiphenyl), 3-OH-CB118 (3-hydroxy-2,3′,4,4′,5-pentachlorobiphenyl), 4′-OH-CB130 (4-hydroxy-2,2′,3,3′,4′,5-hexachlorobiphenyl), 3′-OH-CB138 (3-hydroxy-2,2′,3′,4,4′,5-hexachlorobiphenyl), 4-OH-CB146 (4-hydroxy-2,2′,3,4′,5,5′-hexachlorobiphenyl), 4′-OH-CB159 (4-hydroxy-2′,3,3′,4′,5,5′-hexachlorobiphenyl), 4′-OH-CB172 (4-hydroxy-2,2′,3,3′,4′,5,5′-heptachlorobiphenyl), 3′-OH-CB180 (3-hydroxy-2,2′,3′,4,4′,5,5′-heptachlorobiphenyl), and 4-OH-CB187 (4-hydroxy-2,2′,3,4′,5,5′,6-heptachlorobiphenyl). The concentrations of the 11 OH-PCBs were determined by the I.S. method using calibration curves with 6 to 10 calibration points. To prepare the calibration standards, known amounts of the authentic standards of the 11 OH-PCBs and two I.S. (Wellington Laboratories Inc., Guelph, Ontario, Canada) were derivatised by the same procedure as the plasma samples, and then mixed and diluted to desired concentrations.

2.2.3. Validation of contaminant analysis

The Laboratory of Environmental Toxicology at The Norwegian School of Veterinary Science (Oslo, Norway) is accredited for the determination of several POPs in biological material of animal origin according to the requirements of NS-EN ISO/IEC 17025 (TEST 137). Determination of OH-PCBs is not an accredited method, but is validated after the same procedure as the accredited PCB-method.

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For all the samples and for the quantification of both PCBs and OH-PCBs, standard validation procedures were used to ensure adequate quality assurance and control. Each work up series included the following samples for intra- and inter-assay validation; three blank samples (solvents) to check for interferences, a blind sample and two low-contaminated sheep blood samples spiked with a standard mix containing all analytes to determine relative recovery, and the laboratory's internal reference samples of seal blubber (PCBs) and seal blood (OH-PCBs) to determine reproducibility. For both PCBs and OH-PCBs, the accuracy, precision, linearity and sensitivity of the analyses were within the laboratory's accredited requirements. The relative recovery rates for PCBs and OH-PCBs were between 69–137%, and 69–116%, respectively. Detection limits for individual compounds were determined as three times the noise level. The detection limit for the PCBs were between 0.02 ng/g wet weight (ww) and 0.07 ng/g ww, and between 0.01 ng/g ww and 0.08 ng/g ww for the OH-PCBs.

2.3. Statistical analysis

Statistical analyses were performed using SPSS Statistical software (Version 17.0 for Windows, SPSS Inc., Chicago, IL). Shapiro–Wilk test was used to test if the data were normally distributed ($n \leq 50$). If not, variables were $\log_{10}$-transformed to normalise the data before analyses. In the text, all values are given as untransformed values and as mean ± standard deviation (SD) unless otherwise noted. The level of statistical significance was set at $p < 0.05$.

21 PCB congeners (PCB-47, -74, -99, -101, -105, -114, -118, -128, -137, -138, -153, -156, -157, -167, -170, -180, -183, -187, -189, -194, and -206) of the 28 analysed congeners, and 6 OH-PCBs (4-OH-CB107, 3′-OH-CB138, 4-OH-CB146, 4′-OH-CB159, 3′-OH-CB180, and 4-OH-CB187) of the 11 analysed OH-PCBs were detected in >60% of the individuals in the groups. Thus, Σ_{21}PCBs and Σ_6OH-PCBs represent the sum of the concentrations of the two groups of compounds detected in >60% of the polar bears. It should be noted that PCB-101 was detected in 85% and 77% of the polar bear mothers and cubs from 1998, respectively, but only in 50% and 44% of the polar bear mothers and cubs from 2008, respectively. Moreover, 4′-OH-CB172 was detected in all individuals in both years, but because this metabolite co-eluted with another hepta-chlorinated OH-PCB-isomer (Cl_7-OH-PCB), 4′-OH-CB172 was not included in Σ_6OH-PCBs. However, due to the significant concentrations of this co-eluting peak, referred to as 4′-OH-CB172/Cl_7-OH-PCB, the combined concentration of these two metabolites is presented separately. Thus, the combined concentration of these two metabolites should be considered as semi-quantitative. Furthermore, even though 4′-OH-CB130 was detected in >70% of the individuals, it co-eluted with two unknown OH-PCB-isomers. Because the concentrations of these co-eluting compounds were low (0.6 ± 0.5 ng/g ww), they are excluded from analyses. When PCBs or OH-PCBs levels were below the detection limit, random values between zero and the detection limit for the given compound were used as a replacement to avoid missing values in the statistical analyses (Verreault et al., 2005b). Because PCBs are associated with lipids, and OH-PCBs associate with proteins, PCB concentrations are expressed as ng/g lipid weight (lw), and OH-PCB concentrations as ng/g ww.

General linear models (GLM, type III sum of squares) show that litter effect (parity) explained ≥78.2% of the variation in Σ_{21}PCBs and Σ_6OH-PCBs in polar bear cubs from 1998 and 2008 when years were analysed separately ($F(1, 5) \geq 4.29$, $p \leq 0.05$). Further, litter effect explained ≥78.6% of the variation in SL, ZW, and BM of cubs from 1998 and 2008, and HL of cubs from 2008 ($F(1, 5) \geq 4.42$, $p \leq 0.001$). HL of cubs from 1998 and PL% between twins from both years were the only variables where the variance was lower within twin-pairs than between twin-pairs ($F(1, 5) \leq 2.65$, $p \geq 0.133$). This means that the variation between twin-pairs was higher than within twin-pairs for Σ_{21}PCBs, Σ_6OH-PCBs and most of the morphometric variables. Furthermore, using the mean value of the twins did not, however, allow us to use sex of cubs as a covariate on the entire data set in further analyses to explain individual variation in the response variables (morphometric variables, PL%, Σ_{21}PCBs, and Σ_6OH-PCBs), as some ($n = 7$) of the litters consisted of a male and a female cub. GLM analyses (type III sum of squares) showed that there were no significant difference in morphometric variables (SL, HL, ZW, BM), Σ_{21}PCBs and Σ_6OH-PCBs between male and female cubs in 1998 ($F(1, 20) \leq 2.16$, $p \geq 0.157$) or in 2008 ($F(1, 14) \leq 3.49$, $p \geq 0.083$) when examining all cubs for each sampling year separately. Of all the variables examined, sex influenced only on PL% in cubs from 2008 ($F(1, 5) = 5.63$, $p = 0.028$). Based on these findings, the mean values of variables for twin cubs of both different and equal sexes were used (six twin pairs from each year) in further analyses. Thus, the study was performed on 16 mother–cub pairs from 1998 and 10 mother–cub pairs from 2008.

Previous studies have shown that sampling location, time of sampling, age and condition may affect levels and toxicokinetics of PCBs in polar bears (Polischuk et al., 1995; Bernhoft et al., 1997; Letcher et al., 2000; Andersen et al., 2001; Henriksen, 2001; Polischuk et al., 2002; Olsen et al., 2003). Thus, the extent to which sampling year and other covariates (capture location [latitude and longitude], capture day, age [mothers only], BM [cubs only] and BCI [mothers only]) explained the variation in levels of Σ_{21}PCBs and Σ_6OH-PCBs in mothers and cubs, was investigated using GLM (type III sum of squares) with backward selection and sampling year as a fixed factor. In the GLM-models developed for mothers, BCI and not BM was used because BCI reflects the bears true body condition as the combined mass of fat and skeletal muscle or fat mass only relative to body size (Cattet et al., 2002) and this way is relevant for the bears fat reservoirs for storage of hydrophobic contaminants as PCBs. The BCI equation was developed based on adult bears and was unsuited for use on cubs. Homogeneity of variance was tested by Levene's test, and the assumption of homogeneity of regression slopes was tested for all models to test if the pooled slopes of regression represent the slope for both years. Because OH-PCBs are metabolites of PCBs, Σ_{21}PCBs in mothers was defined as an independent variable (in the model) when Σ_6OH-PCBs in mothers was defined as a dependent variable. Because PCBs in cubs originate from their mothers, Σ_{21}PCBs in mothers was defined as an independent variable when Σ_{21}PCBs in cubs (dependent variable) was examined. When analysing Σ_6OH-PCBs in cubs (dependent variable), Σ_{21}PCBs and Σ_6OH-PCBs in mothers, and Σ_{21}PCBs in cubs, were included as independent variables to account for all possible sources of OH-PCBs in cubs.

Correlations between the contaminant levels in mothers and their cubs were examined using Pearson's correlation test (two-tailed) or Spearman's rank correlation test (two-tailed), depending on whether the variables were normally distributed (with or without $\log_{10}$-transformation) or not, respectively. The cub–mother (CM)-ratios of contaminants were calculated to gain insight into the mother–cub transfer of PCBs and OH-PCBs. The CM-ratios for PCBs and OH-PCBs are based on lw and ww concentrations, respectively. A paired *t*-test was used to compare the CM-ratio of Σ_{21}PCBs and Σ_6OH-PCBs.

The metabolite/precursor-ratios were calculated to gain insight into the biotransformation of PCBs in mothers and cubs. Ratios are based on ww concentrations of known or suggested OH-PCB-metabolite and PCB-precursor combinations (Letcher et al., 2000; Sjödin et al., 2000; Verreault et al., 2008). Levels of contaminants and metabolite/precursor-ratios for mothers and their cubs were compared using a paired *t*-test or Wilcoxon signed rank-test (2-tailed) test depending on whether variables were normally distributed (with or without $\log_{10}$-transformation) or not, respectively. Between-year differences were analysed using Student's *t*-test or Kolmogorov–Smirnov test (K–S-test) depending on whether variables were normally distributed (with or without $\log_{10}$-transformation) or not, respectively.

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3. Results and discussion

3.1. Concentrations and prevalence of PCBs and OH-PCBs

The plasma concentration of Σ_{21}PCBs and Σ_6OH-PCBs in polar bear mothers and cubs was lower in 2008 than in 1998 ($p \leq 0.001$) (Fig. 2). In the mothers, Σ_{21}PCBs (1998: 5710 ± 3090 ng/g lw, 2008: 2560 ± 1500 ng/g lw) and Σ_6OH-PCBs (1998: 228 ± 60 ng/g ww, 2008: 80 ± 38 ng/g ww) were 55% and 65% lower in 2008 than in 1998, respectively. In cubs, Σ_{21}PCBs (1998: 14680 ± 5350 ng/g lw, 2008: 6070 ± 2590 ng/g lw) and Σ_6OH-PCBs (1998: 98 ± 23 ng/g ww, 2008: 49 ± 21 ng/g ww) were 59% and 50% lower in 2008 than in 1998, respectively. The decreased plasma levels of PCBs and OH-PCBs from 1998 to 2008 are in accordance with the temporal trend of decreasing levels of PCBs in arctic wildlife including polar bears (Henriksen, 2001; AMAP, 2004; Verreault et al., 2005a; Wolkers et al., 2008; McKinney et al., 2011). The decreasing levels of PCBs in polar bears most likely reflect the declining emissions due to national bans on production, use and release of PCBs. Thus, the establishment of international treaties (e.g. the Stockholm Convention) that aims to protect humans and wildlife against exposure to environmental contaminants has functioned as intended.

It should be noted that although sampling year was the only determinant of $\sum_{21}$PCBs in mothers and that latitude, longitude, capture day, age and BCI of mothers did not explain the plasma levels of $\sum_{21}$PCBs (Table 2), other confounding factors may still have influenced the differences in the plasma contaminant levels between the years. Previous studies have shown that dietary shifts influence on the contaminant levels and pattern in polar bears (McKinney et al., 2010). Thus, differences in the diet of the polar bears from the 2 years could have influenced on the differences in PCB concentrations reported herein. Furthermore, in 2008 the bears were captured 6° further west than in 1998 (Table 1; Fig. 1). Thus, sampling year is confounded by longitude in the present study (Table 2). Previous studies have shown that the plasma concentration of PCB-153 in adult polar bears from the Norwegian Arctic decreases by approximately 4% with each westward degree (Henriksen, 2001). Thus, spatial or between-year differences in the home-range areas of the mothers may also have influenced on the contaminant levels (Henriksen, 2001; Olsen et al., 2003), and thus the reported between-year contaminant differences.

Although the use of plasma to monitor levels of hydrophobic contaminants in seals has been questioned (Lydersen et al., 2002), several studies on polar bears have reported highly similar PCB-patterns in adipose tissue and blood, minor short-term fluctuations in the PCB-levels in plasma, and a lower relative variation in PCB concentrations in plasma compared to in adipose tissue (Henriksen, 2001; Polischuk et al., 2002; Sandala et al., 2004). Thus, in polar bears, plasma appears to be a good non-destructive matrix for monitoring changes in levels of hydrophobic contaminants.

The $\sum_6$OH-PCBs in the mothers from 1998 (228 ± 60 ng/g ww) and 2008 (80 ± 38 ng/g ww) were higher and lower, respectively, than the concentration of $\sum_{11}$OH-PCBs reported in blood of female polar bears from Svalbard sampled in 2002 (mean concentration: 173 ng/g ww) (Verreault et al., 2005b). The decreasing levels of OH-PCBs in Svalbard polar bear mothers from 1998 via 2002 to 2008 could reflect the decreasing levels of PCBs. However, as the OH-PCBs in polar bears predominantly originate from endogenous biotranformation by the CYP-enzyme system and not from consumption of contaminated prey (Letcher et al., 1996, 2000; Sandau et al., 2000; Routti et al., 2008), factors affecting the quantity of parent compounds available for biotransformation (e.g. condition, fasting and intake of food) or factors affecting biotransformation rates of PCBs (e.g. level of exposure to CYP-enzyme inducing compounds) (Safe, 1994; Letcher et al., 1996; Polischuk et al., 1995; Boon et al., 1997; Letcher et al., 2000; Polischuk et al., 2002) could have influenced the between-year differences in concentration of OH-PCBs reported herein.

Comparisons of the plasma levels of PCBs and OH-PCBs in cubs-of-the-year with previous studies on the same age-group were limited to two studies. The concentrations of $\sum_{21}$PCBs (133 ± 54 ng/g ww) and $\sum_6$OH-PCBs (98 ± 23 ng/g ww) in cubs from 1998 were somewhat lower than and comparable to, respectively, the levels of PCBs ($\sum_{24}$PCBs: ~220 ng/g ww) and OH-PCBs ($\sum_6$OH-PCBs: ~81 ng/g ww) reported in three cubs (≤ 2 years old) from Svalbard also sampled in 1998 (Sandau, 2000). Furthermore, the concentrations of $\sum_{21}$PCBs in cubs from 1998 in our study were higher than reported

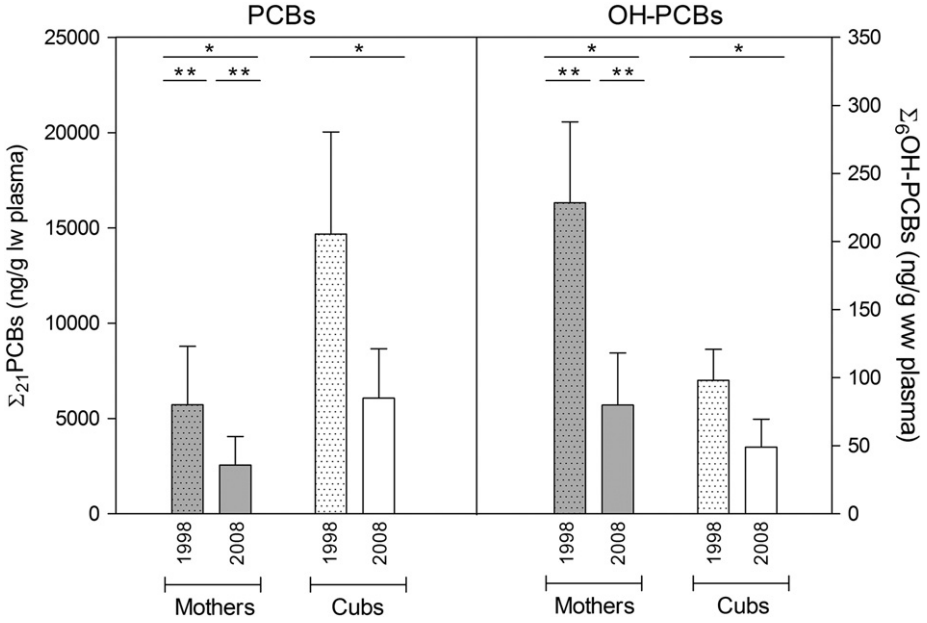

Fig. 2. Plasma concentration of $\sum_{21}$PCBs (ng/g lw) and $\sum_6$OH-PCBs (ng/g ww) in polar bear mothers in 1998 (grey bars with dots, $n = 16$), mothers in 2008 (grey bars, $n = 10$), cubs in 1998 (white bars with dots, $n = 16$), and cubs in 2008 (white bars, $n = 10$) from Svalbard presented as mean bars with standard deviation (SD) error bars. * Significant differences in concentration of $\sum_{21}$PCBs or $\sum_6$OH-PCBs between mothers sampled in 1998 and 2008, and cubs sampled in 1998 and 2008. ** Significant differences in concentration of $\sum_{21}$PCBs or $\sum_6$OH-PCBs between mothers and cubs sampled the same year.

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Table 2
Variables explaining $\sum_{21}$PCBs ($\log_{10}$ ng/g lw) and $\sum_{6}$OH-PCBs (ng/g ww) in the plasma of polar bear mothers ($n=26$) and cubs ($n=26$) sampled in Svalbard (Norway) in 1998 and 2008. Results are indicated as parameter estimates from linear models with 1998 as the reference year (β and standard error (SE) of β), F-statistics, p-values, R^2, and degrees of freedom (df) from analysis of variance. Levene's test is performed to examine equality of error variances between the groups (homogeneity). Regression analysis was done on the results from the two years together, with sampling year as a fixed factor and with backward selection.

			β	SE β	F	p	R^2 Model	df	Levene's
Mothers	$\log_{10}\Sigma_{21}$PCBs[a]	Intercept	6.36	0.07					
		Sampling year	−0.36	0.08	18.42	≤0.001	0.43	24	0.71
Mothers	Σ_6OH-PCBs	Intercept	79.90	16.64					
		Sampling year	−148.51	21.21	49.02	≤0.001	0.67	24	0.20
Cubs	$\log_{10}\Sigma_{21}$PCBs[a,b]	Intercept	1.45	0.52					
		$\log_{10}\Sigma_{21}$PCBs mothers[a]	0.84	0.08	115.02	≤0.001	0.83	24	0.42
Cubs	Σ_6OH-PCBs	Intercept	19.44	3.72					
		Σ_6OH-PCBs mothers	0.35	0.02	325.72	≤0.001	0.93	24	0.58

[a] Logarithmic values are based on pg/g lw PCB concentrations.
[b] $\log_{10}\Sigma_{21}$PCBs in cubs was explained by $\log_{10}\Sigma_{21}$PCBs mothers together with longitude ($F_{1,23}\geq 13.90$, $p\leq 0.001$) in the first model. After testing for assumption of homogeneity of regression slopes, longitude was no longer significant, and only $\log_{10}\Sigma_{21}$PCBs mothers was concluded to explain $\log_{10}\Sigma_{21}$PCBs in cubs.

in 12 cubs about 8 months old sampled in Churchill, Canada, during July to August (49 ± 29 ng/g ww) and September to November (45 ± 26 ng/g ww) between 1992 and 1995 (Polischuk et al., 2002). The difference in PCB concentrations between cubs from Svalbard and Canada most likely reflect the generally higher levels of PCBs reported in polar bears in Svalbard compared to Canada (Norstrom et al., 1998; Verreault et al., 2005a; Letcher et al., 2010; McKinney et al., 2011).

Σ_{21}PCBs consisted mainly of PCB-99, -138, -153, -170, -180 and -194 in mothers and cubs from both years (Table 3). The order of dominance in mothers from 1998 was: PCB-153 (39%) > PCB-180 (22%) > PCB-170 (11%) > PCB-99 (8%) = PCB-138 (8%) > PCB-194 (5%), in mothers from 2008: PCB-153 (38%) > PCB-180 (23%) > PCB-170 (9%) > PCB-194 (8%) > PCB-99 (7%) > PCB-138 (6%), in cubs from 1998: PCB-153 (42%) > PCB-180 (20%) > PCB-170 (10%) > PCB-99 (9%) = PCB-138 (9%) > PCB-194 (4%), and in cubs from 2008: PCB-153 (44%) > PCB-180 (18%) > PCB-99 (10%) > PCB-170 (8%) > PCB-138 (7%) > PCB-194 (5%). These congeners are the same as reported to dominate in plasma and adipose tissue in earlier polar bear studies in Alaska, Canada, Russia and Svalbard and confirm the persistence of these particular congeners (Norstrom et al., 1998; Sandau, 2000;

Table 3
Mean (X) ± standard deviation (SD) of plasma lipid percentage (PL%), plasma concentrations of PCBs (ng/g lw) and OH-PCBs (ng/g ww) in plasma samples of polar bear mothers and cubs sampled in Svalbard (Norway) 1998 and 2008.

	Mothers 1998[a] ($n=16$)		Mothers 2008[a] ($n=10$)		Cubs 1998[a] ($n=16$)		Cubs 2008[a] ($n=10$)	
	X ± SD	% detection	X ± SD	% detection	X ± SD	% detection	X ± SD	% detection
PL%[b,c,d]	1.1 ± 0.1	–	1.3 ± 0.2	–	0.9 ± 0.1	–	1.3 ± 0.1	–
PCB-47[b,c,d,e]	25 ± 11	100	10 ± 7	100	68 ± 24	100	31 ± 16	100
PCB-74[a,e]	9.0 ± 3.3	85	9.7 ± 2.5	100	7.5 ± 4.4	64	7.9 ± 2.1	100
PCB-99[b,c,d,e]	440 ± 153	100	172 ± 96	100	1340 ± 501	100	573 ± 213	100
PCB-101[b,c,f]	8.5 ± 2.3	85	5.3 ± 6.2	50	17 ± 20	77	4.1 ± 1.7	44
PCB-105[c,d,e]	11 ± 4	100	9.3 ± 2.6	100	18 ± 6	100	11 ± 5	100
PCB-114[b,c,d,e,f]	2.6 ± 0.4	100	1.8 ± 0.7	80	4.5 ± 1.0	100	2.6 ± 0.8	100
PCB-118[c,d,e]	31 ± 11	100	34 ± 16	100	61 ± 19	100	40 ± 17	100
PCB-128[b,c,d,f]	8.2 ± 2.7	100	2.3 ± 2.6	80	23 ± 10	100	4.2 ± 4.0	94
PCB-137[b,c,d,e]	43 ± 18	100	20 ± 12	100	118 ± 42	100	50 ± 19	100
PCB-138[b,c,d,e]	420 ± 139	100	171 ± 110	100	1290 ± 470	100	408 ± 193	100
PCB-153[b,c,d,e]	2200 ± 1 100	100	978 ± 611	100	6180 ± 2340	100	2690 ± 1170	100
PCB-156[b,c,d,e]	70 ± 22	100	41 ± 20	100	196 ± 52	100	121 ± 42	100
PCB-157[b,c,d,e]	59 ± 30	100	27 ± 15	100	170 ± 171	100	91 ± 40	100
PCB-167[c,e,f]	2.0 ± 0.3	85	1.9 ± 1.0	70	3.4 ± 3.4	100	1.8 ± 2.0	63
PCB-170[b,c,d,e]	663 ± 533	100	237 ± 160	100	1560 ± 789	100	519 ± 246	100
PCB-180[b,c,d,e]	1320 ± 929	100	577 ± 361	100	2890 ± 1180	100	1130 ± 522	100
PCB-183[b,c,d,e]	32 ± 15	100	19 ± 11	100	72 ± 24	100	30 ± 11	100
PCB-187[c]	6.9 ± 3.0	100	4.7 ± 3.1	100	8.5 ± 3.1	100	3.0 ± 1.5	100
PCB-189[b,c,d,e]	17 ± 13	100	7.0 ± 3.4	100	36 ± 20	100	16 ± 7	100
PCB-194[b,c,d,e]	299 ± 245	100	194 ± 122	100	530 ± 210	100	296 ± 195	100
PCB-206[c,d]	49 ± 29	100	42 ± 25	100	76 ± 23	100	46 ± 29	100
4-OH-CB107[b,c]	20 ± 11	100	7.1 ± 4.8	100	17 ± 7	100	7.7 ± 7.6	100
3′-OH-CB138[b,c]	1.9 ± 1.5	100	0.5 ± 0.2	100	1.2 ± 0.6	100	0.6 ± 0.2	100
4-OH-CB146[b,c,e]	79 ± 26	100	27 ± 13	100	37 ± 10	100	23 ± 7	100
4′-OH-CB159[e,f]	0.5 ± 0.2	100	0.6 ± 0.9	100	0.7 ± 0.3	100	0.8 ± 0.6	100
4′-OH-CB172/Cl_7-OH-PCB[b,c,e]	38 ± 11	100	12 ± 6	100	25 ± 8	100	15 ± 5	100
3′-OH-CB180[b,c]	3.3 ± 2.1	100	0.9 ± 0.2	100	1.8 ± 0.9	100	1.0 ± 0.4	100
4-OH-CB187[b,c,d,e]	125 ± 26	100	44 ± 26	100	41 ± 10	100	17 ± 8	100

[a] PCB-28, -52, -66, -123, -141, -149, -151, 4′-OH-CB106, and 4′-OH-CB108 were not detected in any individuals, while 3-OH-CB118 were detected in <40% of the individuals from 1998.
[b] Significant difference between mothers sampled in 1998 and 2008 ($p<0.05$).
[c] Significant difference between cubs sampled in 1998 and 2008 ($p<0.05$).
[d] Significant difference between mothers and cubs sampled in 1998 ($p<0.05$).
[e] Significant difference between mothers and cubs sampled in 2008 ($p<0.05$).
[f] Detected concentrations are close to the detection limit.

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Polischuk et al., 2002; Derocher et al., 2003; Dietz et al., 2004; Verreault et al., 2005a, 2005b; McKinney et al., 2011).

In our study, Σ_6OH-PCBs consisted mainly of the *para*-hydroxylated OH-PCBs; 4-OH-CB107, 4-OH-CB146 and 4-OH-CB187 in mothers and cubs for both years (Table 3). The order of dominance in mothers from 1998 was: 4-OH-CB187 (56%) > 4-OH-CB146 (34%) > 4-OH-CB107 (8%), in mothers from 2008: 4-OH-CB187 (44%) > 4-OH-CB146 (27%) > 4-OH-CB107 (7%), in cubs from 1998: 4-OH-CB187 (42%) > 4-OH-CB146 (37%) > 4-OH-CB107 (17%), and in cubs from 2008: 4-OH-CB146 (47%) > 4-OH-CB187 (34%) > 4-OH-CB107 (13%). These are the same metabolites as reported to be among the dominant metabolites in plasma in previous polar bear studies as well as in humans (Sandau, 2000; Guvenius et al., 2003; Park et al., 2008). In all bears, we also detected significant concentrations of a co-eluting peak consisting of 4′-OH-CB172 and another heptachlorinated OH-PCB-isomers (4′-OH-CB172/Cl_7-OH-PCB). Of the co-eluting isomers, Cl_7-OH-PCB was the dominating isomer. 4′-OH-CB172 has been detected in earlier polar bear studies (Sandau, 2000; Sandala et al., 2004). In our study, only 4-OH-CB146 and 4-OH-CB187 were detected in higher concentrations than the co-eluting isomers 4′-OH-CB172/Cl_7-OH-PCB. In other polar bear studies examining OH-PCBs in blood (plasma or whole blood), significant concentrations of 4′-OH-CB120, 3-OH-CB153, 3-OH-CB187, 4-OH-CB193, 4-OH-CB163, 4-OH-CB199, and 4′-OH-CB202, 4,4′-diOH-CB202, and 4′-OH-CB208 have been reported (Sandau, 2000; Sandala et al., 2004; Gebbink et al., 2008), but these metabolites were not analysed in our study.

There were significant lower levels of the most prevalent PCB congeners and Σ_{21}PCBs in mothers from 2008 compared to in mothers from 1998 (Table 3; Fig. 2). In contrast, several mono-*ortho* PCBs (PCB-74, -105, -118, and -167) and PCB-187 did not differ between the years (Table 3) resulting in PCB-pattern differences in the mothers from the two sampling years. As discussed for plasma levels, also the plasma pattern of PCBs may be affected by sampling location, ecological and physiological variables and thereby influence on the observed differences between the years. However, because the polar bears with the lowest PCB-levels (2008) had a higher proportion of PCBs known to be easily biotransformed by polar bears (PCB-74, -105, -118, -167 and PCB-187) (Norstrom et al., 1988; Norstrom and Muir, 1994; Bernhoft et al., 1997), our findings indicate biotransformation differences between the years. Similar findings have been observed in free-living seals (Boon et al., 1997; Sørmo et al., 2003b; Routti et al., 2008; Wolkers, 2008) and humans (Brown et al., 1989), and this phenomenon has been explained by a lower total load of CYP-enzyme inducing contaminants in the animals resulting in a reduced biotransformation particularly of the less persistent PCBs. The suggested explanation related to biotransformation differences between the years is supported by the significantly lower ratios of Σ_6OH-PCBs/Σ_{21}PCBs, 4-OH-CB107/PCB-105 + PCB-118 and 4-OH-CB187/PCB-187 in mothers from 2008 compared to mothers sampled in 1998 (Fig. 3).

3.2. Maternal transfer of PCBs and OH-PCBs

Maternal transfer of PCBs and OH-PCBs was investigated using the CM-ratios. Although CM-ratios based on plasma concentration in mother–cub-pairs may be influenced by toxicokinetical factors in both mothers and cubs, the ratios may give valuable insight into the mother–cub transfer and differences in exposure levels of PCBs and OH-PCBs between mothers and cubs. The CM-ratios for mother–cub pairs from 1998 and 2008 show that suckling cubs were exposed to higher concentrations of $\sum_{21}$PCBs (1998: 2.7 ± 0.6, 2008: 2.6 ± 0.8, 1998 and 2008: 2.7 ± 0.7) and lower concentrations of $\sum_6$OH-PCBs (1998: 0.43 ± 0.04, 2008: 0.67 ± 0.18, 1998 and 2008: 0.53 ± 0.16) compared to their mothers ($p \leq 0.001$) (Fig. 4). The difference in OH-PCB levels in mothers and cubs is mainly due to the significantly lower plasma concentration of the dominating metabolite 4-OH-CB187 in cubs compared to in mothers (Table 3; Fig. 4). The mother–cub differences was less prominent for most of the remaining OH-PCBs (4-OH-CB107, 3′-OH-CB138, 4-OH-CB146 and 3′-OH-CB180), where the levels were either slightly lower, equal, or for the metabolites found in the lowest concentrations, higher in cubs than in their mothers (Fig. 4). These findings are in accordance with studies reporting higher levels of PCBs and lower levels of OH-PCBs in cubs-of-the-year ($n = 3$) and older cubs (1–2 years of age) compared to in adult female polar bears (Bernhoft et al., 1997; Sandau, 2000; Polischuk et al., 2002). However, those particular reports were based on average plasma concentrations in the age-groups, and not on matched mother–cub pairs as in our study.

The CM-ratio for Σ_{21}PCBs in polar bears sampled in spring (April) from Svalbard (1998: 2.7 ± 0.6, 2008: 2.6 ± 0.8) is comparable to and somewhat higher than previously reported in polar bears from Canada during summer (July and August: 2.2) and fall (September to November: 1.5), respectively (Polischuk et al., 2002). The difference between the two studies, and particularly the comparison with the Canadian results from fall, may be due to age-related differences, season related dietary and physiological differences such as a relative lower intake of maternal milk in the older cubs and growth-dilution effects (Yakushiji et al., 1984; Olsen et al., 2003; Dietz et al., 2004; McKinney et al., 2010). Furthermore, the results from the Canadian bears may be confounded because ratios were estimated using mean plasma values for cubs and mothers and not data from mother–cub pairs.

Due to low lipid content in maternal blood of polar bears (~1%) and most likely also in cord blood compared to in milk (20–25%) (Bernhoft et al., 1997; Polischuk et al., 2002), it is likely that polar bear foetuses are exposed to lower levels of PCBs than suckling polar bear cubs. This assumption is supported by previous findings of lower transfer ratios from maternal blood to cord blood compared to from maternal blood to milk in humans where the milk also is the matrix with the highest lipid content (Maternal blood: 0.7%, Cord blood: 0.2%, Milk: 1.9%) (Guvenius et al., 2003; Park et al., 2008; Needham et al., 2010). The role of mother's milk in the maternal transfer of PCBs is reflected by the positive mother–cub correlations for the PCB congeners and Σ_{21}PCBs (Table 4; Fig. 5A), as well as Σ_{21}PCBs in cubs being explained by Σ_{21}PCBs in mothers (Table 2). Thus, and in accordance with previous studies in polar bears (Bernhoft et al., 1997; Polischuk et al., 2002), we suggest that the main exposure route for PCBs in cubs is via the lipid-rich mother's milk (Bernhoft et al., 1997; Polischuk et al., 2002). In the perspective of potential toxic effects, both the foetal stage and the time when newborn polar bears start to nurse may be critical periods for potential PCB-related developmental effects (Peterson et al., 1993; Brouwer et al., 1995, 1998; Ulbrich and Stahlmann, 2004). While the exposure concentrations most likely are lower at the foetal stage compared to after birth, prenatal exposure may cause a direct exposure of vital organs because of the minor capacity polar bear foetus have to store hydrophobic contaminants because of their small lipid reservoir (Blix and Lentfer, 1979). After birth, the small cubs experience a considerable shift from lower to higher PCB exposure as they start to suckle.

With respect to transfer efficiency of individual PCBs from mothers to cubs, the results indicate that PCB-47, -99, -128, -137, -138, -153, -156, -157, -170, -180, -183, and -189 were most efficiently transferred (Fig. 4). Based on the mean CM-ratios (± SD) and in decreasing order, the PCB congeners most efficiently transferred in 1998 were: PCB-138 (3.11 ± 0.68) > PCB-99 (3.08 ± 0.54) > PCB-47 (3.05 ± 1.19) > PCB-157 (3.02 ± 0.61) > PCB-153 (2.97 ± 0.67) > PCB-137 (2.94 ± 0.74) > PCB-156 (2.87 ± 0.54) > PCB-128 (2.75 ± 0.61) > PCB-170 (2.63 ± 0.60) > PCB-183 (2.56 ± 0.99) > PCB-180 (2.43 ± 0.58) > PCB-189 (2.31 ± 0.53). In 2008, the PCB congeners most efficiently transferred were: PCB-157 (3.74 ± 1.26) > PCB-99 (3.73 ± 1.08) > PCB-47 (3.30 ± 1.00) > PCB-156 (3.26 ± 0.98) > PCB-153 (3.11 ± 1.07) > PCB-138 (2.78 ± 0.93) > PCB-137 (2.76 ± 0.82) > PCB-128 (2.69 ± 2.10) > PCB-170 (2.48 ± 1.00) > PCB-180 (2.19 ± 0.90) > PCB-

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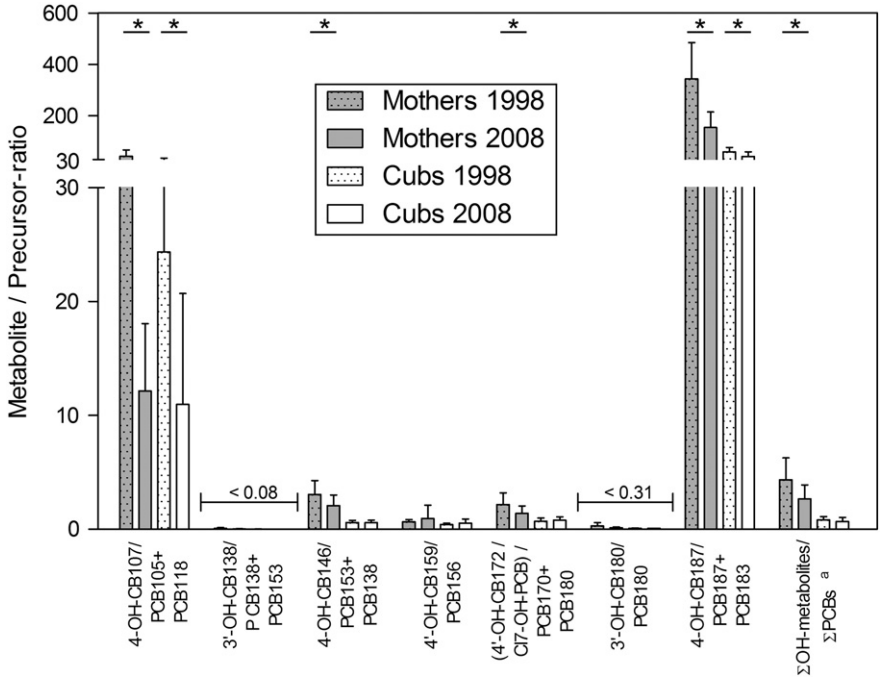

Fig. 3. Metabolite/precursor-ratios of various OH-PCB/PCB-combinations and Σ_6OH-PCBs/Σ_{21}PCBs in plasma samples of polar bear mothers in 1998 ($n=16$), mothers in 2008 ($n=10$), cubs in 1998 ($n=16$), and cubs in 2008 ($n=10$) from Svalbard presented as mean bars with standard deviation (SD) error bars. Ratios are based on wet weight (ww) concentrations of known or suggested metabolite–precursor combinations (Letcher et al., 2000; Sjödin et al., 2000; Verreault et al., 2008). a = 4′-OH-CB172/Cl_7-OH-PCB is not included in Σ_6OH-PCBs. * Significant differences in metabolite/precursor-ratios between 1998 and 2008.

189 (2.17 ± 0.70) > PCB-183 (1.92 ± 0.91). Thus, polar bears are exposed to particularly high concentrations of these persistent PCBs early in life. Furthermore, the results shows that the maternal transfer partly depend on the PCB congeners' degree of chlorination (Fig. 4). This was particularly prominent for the most heavily chlorinated congeners, PCB-170 to PCB-206 (except for PCB-187), where the transfer efficiency seems to decrease with increasing chlorination. These trends are in accordance with earlier studies on PCB transfer from

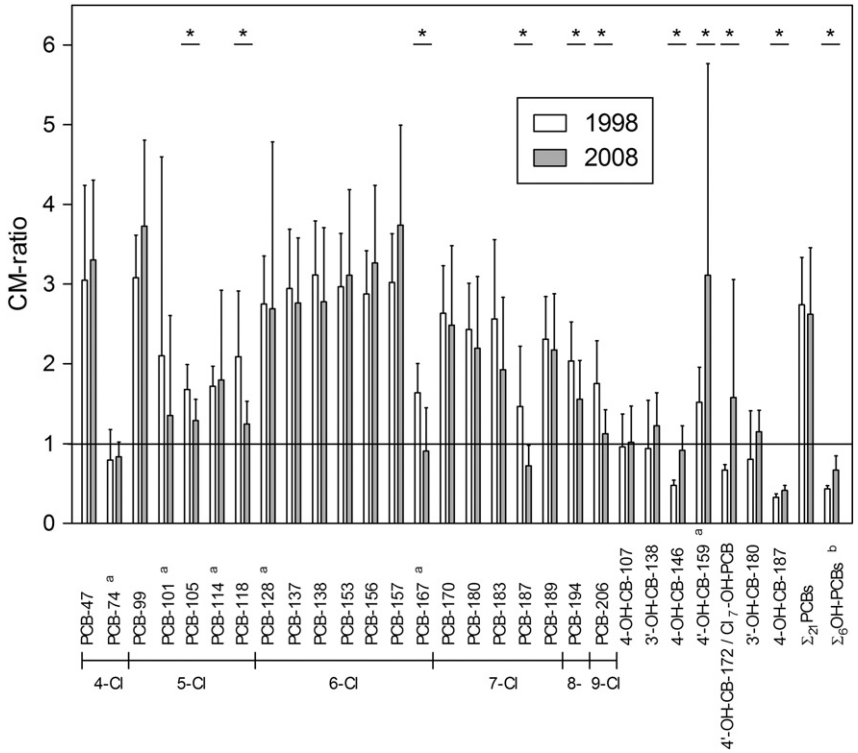

Fig. 4. Cub–mother-ratios (CM-ratios) of PCBs, OH-PCBs, $\sum_{21}$PCBs, and $\sum_6$OH-PCBs in mother–cub pairs of polar bear from Svalbard (Norway) sampled in 1998 ($n=16$) and 2008 ($n=10$) presented as mean bars with standard deviation (SD) error bars. CM-ratios are based on plasma lipid weight (lw) concentrations for PCBs and plasma wet weight (ww) concentrations for OH-PCBs. Y > 1 means the plasma concentration of the current compound is higher in the cubs compared to their corresponding mothers. a = detected concentrations are close to the detection limit. b = 4′-OH-CB172/Cl_7-OH-PCB is not included in Σ_6OH-PCBs. * Significant differences in CM-ratios between 1998 and 2008.

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Table 4
Correlations between levels of PCBs (ng/g lw), OH-PCBs (ng/g ww), $\sum_{21}$PCBs (ng/g lw), and $\sum_{6}$OH-PCBs (ng/g ww) in plasma of polar bear mother–cub pairs sampled in Svalbard (Norway) in 1998 and 2008 given as correlation coefficients (r) and p-values. For significant correlations, r and p-values are given in bold.

	Mother–cub 1998 ($n=16$)		Mother–cub 2008 ($n=10$)	
	r	p	r	p
PCB-47	**0.752**	**0.001**	**0.888**	**0.001**
PCB-74[a]	0.307	0.307	**0.680**	**0.030**
PCB-99	**0.876**	**≤0.001**	**0.869**	**0.001**
PCB-101[a]	0.289	0.328	0.604	0.065
PCB-105	**0.833**	**≤0.001**	**0.855**	**0.002**
PCB-114[a]	**0.809**	**0.001**	**0.729**	**0.017**
PCB-118	**0.588**	**0.016**	**0.904**	**≤0.001**
PCB-128[a]	**0.863**	**≤0.001**	0.524	0.120
PCB-137	**0.701**	**0.002**	**0.901**	**≤0.001**
PCB-138	**0.795**	**≤0.001**	**0.942**	**≤0.001**
PCB-153	**0.861**	**≤0.001**	**0.805**	**0.005**
PCB-156	**0.792**	**0.001**	**0.763**	**0.010**
PCB-157	**0.899**	**≤0.001**	**0.732**	**0.016**
PCB-167[a]	0.275	0.363	0.541	0.106
PCB-170	**0.918**	**≤0.001**	**0.718**	**0.019**
PCB-180	**0.884**	**≤0.001**	**0.663**	**0.036**
PCB-183	**0.658**	**0.006**	**0.814**	**0.004**
PCB-187	0.453	0.078	**0.916**	**≤0.001**
PCB-189	**0.912**	**≤0.001**	**0.739**	**0.015**
PCB-194	**0.811**	**≤0.001**	**0.874**	**≤0.001**
PCB-206	**0.666**	**0.005**	**0.818**	**0.004**
4-OH-CB107	**0.791**	**≤0.001**	**0.931**	**≤0.001**
3′-OH-CB138	**0.592**	**0.016**	**0.780**	**0.008**
4-OH-CB146	**0.929**	**≤0.001**	**0.889**	**0.001**
4′-OH-CB159[a]	**0.827**	**≤0.001**	0.297	0.405
3′-OH-CB180	0.431	0.142	**0.848**	**0.002**
4′-OH-CB172/Cl_7-OH-PCB[b]	**0.940**	**≤0.001**	**0.760**	**0.011**
4-OH-CB187	**0.851**	**≤0.001**	**0.976**	**≤0.001**
Σ_{21}PCB	**0.861**	**≤0.001**	**0.804**	**0.005**
Σ_{6}OH-PCBs	**0.938**	**≤0.001**	**0.942**	**≤0.001**

[a] Detected concentrations are close to the detection limit.
[b] Not included in Σ_{6}OH-PCBs.

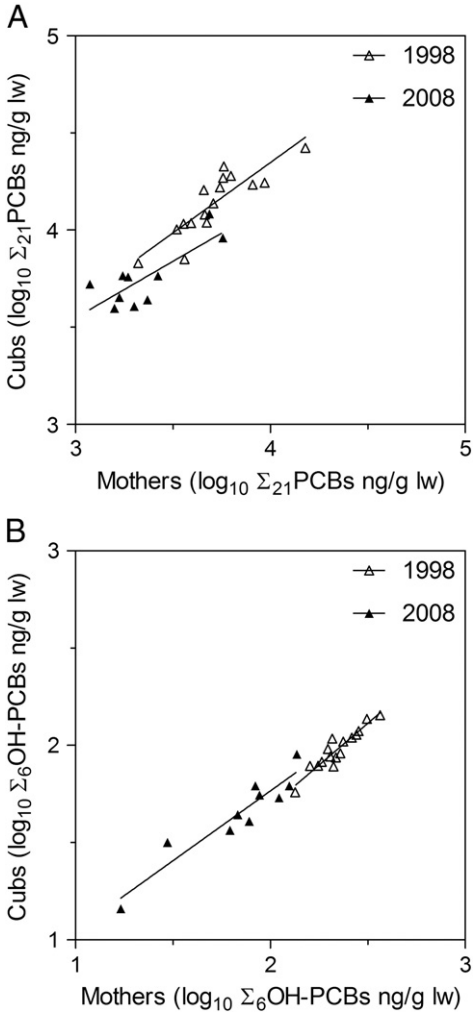

Fig. 5. Correlation between plasma concentrations of A) $\sum_{21}$PCB ($\log_{10}$ ng/g lw) in polar bear mother–cub pairs sampled in 1998 ($n=16$, $Y=0.724\pm0.112$, $R^2=0.75$) and 2008 ($n=10$, $Y=0.581\pm0.154$, $R^2=0.64$), and B) $\sum_{6}$OH-PCBs (log ng/g ww) in polar bear mother–cub pairs sampled in 1998 ($n=16$, $Y=0.858\pm0.085$, $R^2=0.88$) and 2008 ($n=10$, $Y=0.718\pm0.090$, $R^2=0.89$). Slope is significantly non-zero for all curves.

polar bear mothers to 1 year old cubs (yearlings) based on concentrations in subcutaneous tissue (Bernhoft et al., 1997).

With respect to OH-PCBs in cubs, these may originate from maternal transfer across the placenta and the umbilical cord, through the mother's milk (Guvenius et al., 2003; Park et al., 2008), and possibly from endogenous biotransformation of PCB-precursors in the cubs. Regarding endogenous biotransformation, several previous studies have reported that foetal and young mammals, including humans, have low and fluctuating levels of Phase I enzymes (e.g. CYP-enzymes), which metabolically biotransform PCBs into OH-PCBs (Eltom and Schwark, 1999; Miller et al., 2002; Blake et al., 2005; Czekaj et al., 2006; Bonfanti et al., 2009). The Σ_{6}OH-PCBs/Σ_{21}PCBs-ratio ($p\leq0.001$) and the individual metabolite/precursor-ratios ($p\leq0.014$), except from 4-OH-CB107/PCB-105 + PCB-118 and 4′-OH-CB159/PCB-156 in 2008 ($p\geq0.40$), were lower in cubs compared to mothers (Fig. 3). This indicates that polar bear cubs have a lower endogenous capacity to biotransform PCBs to OH-PCBs than polar bear mothers. Furthermore, the levels of OH-PCBs in cubs were explained by the levels of OH-PCBs in mothers (Table 2) and there were positive relationships between the OH-PCBs in mothers and their cubs (Table 4; Fig. 5B). This may reflect that the relative pattern of CYP-enzyme activities is similar in mothers and cubs, even though the activities per se are lower in cubs. However, it is also possible that these mother–cub associations reflect transfer of OH-PCBs via milk. Unfortunately, there appears to be no information on levels of OH-PCBs in polar bear milk. However, the levels of OH-PCBs are low in human milk (Guvenius et al., 2003; Needham et al., 2010), and it has been suggested that the low transfer of protein-binding compounds such as OH-PCBs to the offspring via human milk is due to the low protein content in human milk (approx. 1% during the first year) as compared to in the human plasma (approx. 7.5% during the first year) (Anderson, 1991; Nommsen et al., 1991; Karrman et al., 2007; Fromme et al., 2010). Although it is likely that the transfer of OH-PCBs through milk also is low in polar bears, the high protein content in polar bear milk (approx. 10% during the first year) and the minor difference in protein content between milk and plasma (approx. 7.5%) (Derocher et al., 1993; Tryland et al., 2002) may to a greater extent than in humans favour the transfer of OH-PCBs to offspring in polar bears. Further studies examining levels of OH-PCBs in polar bear milk and biotransformation of PCBs in cubs are needed to understand the relative importance of maternal transfer and endogenous biotransformation as potential sources to OH-PCBs in polar bear cubs.

3.3. Risk assessment and toxicological implication

The toxicological effects of PCBs and OH-PCBs have been documented in laboratory animals, humans and wildlife (Colborn et al., 1993; Safe, 1994; Brouwer et al., 1995, 1998; AMAP, 2004; Gauger et al., 2004; Meerts et al., 2004; Sonne, 2010). In polar bears from Svalbard, studies on adults indicated that the levels of organochlorines (e.g. PCBs) experienced during the 1990s had endocrine,

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immunosuppressive and reproductive effects (Skaare et al., 2001; Derocher et al., 2003; Haave et al., 2003; Braathen et al., 2004; Lie et al., 2004, 2005; Letcher et al., 2010). It should be noted that in 2008, the plasma concentration of Σ_{21}PCBs in cubs corresponded to the levels found in polar bear mothers in 1998 (Fig. 2). Thus, when taking into account that developing mammals (i.e. cubs) are particularly susceptible to contaminant-induced effects (Colborn et al., 1993), there is still a cause of concern about the health effects of PCBs and their metabolites in polar bear cubs at Svalbard. For instance, in 2008, the plasma concentration of Σ_6OH-PCBs in the cubs (49 ± 21 ng/g ww) was much higher (90–170 times) than the concentration of Σ_{14}OH-PCBs in cord blood plasma (0.286–0.553 ng/g ww) associated with thyroid hormones effects in human neonates (Sandau et al., 2002). The plasma Σ_6OH-PCBs in the cubs (2008) were also much higher (70–140 times) than plasma concentration of Σ_5OH-PCBs associated with thyroid effects in hooded seal pups (Gabrielsen et al., 2011). Furthermore, the concentration of 4-OH-CB107 in the polar bear cubs in 2008 (7.7 ± 7.6 ng/g ww) was approximately 300 times higher than the cord blood concentrations associated with effects on cognitive development in children (0.028 ± 0.045 ng/g ww) (Park et al., 2009). Because plasma levels of PCBs and OH-PCBs in polar bear cubs still appear to be above levels associated with health effects, we suggest that studies related to potential effects of PCBs and OH-PCBs on biological end-points in cubs, such as thyroid hormone variables should be conducted.

4. Conclusions

There was a significant transfer of PCBs from polar bear mothers to suckling cubs-of-the-year. In contrast, the maternal transfer of OH-PCBs was lower than for PCBs. Furthermore, our findings indicate that the cubs have a lower endogenous capacity to biotransform PCBs into OH-PCBs compared to their mothers. Plasma levels of Σ_{21}PCBs and Σ_6OH-PCBs in polar bear mothers and their suckling cubs from Svalbard were significantly lower in 2008 than in 1998. Nevertheless, the levels of PCBs and OH-PCBs in cubs from 2008 are still above levels associated with health effects in humans and wildlife. Thus, studies related to potential effects of PCBs and OH-PCBs on biological end-points in cubs, such as thyroid hormone variables should be conducted.

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References

AMAP. AMAP Assessment 2002: Persistent Organic Pollutants in the Arctic. Arctic Monitoring and Assessment Programme (AMAP), Oslo, Norway. 2004, xvi + 310 pp.
Andersen M, Lie E, Derocher AE, Belikov SE, Bernhoft A, Boltunov AN, et al. Geographic variation of PCB congeners in polar bears (*Ursus maritimus*) from Svalbard east to the Chukchi Sea. Polar Biol 2001;24:231–8.
Anderson PO. Drug use during breast-feeding. Clin Pharm 1991;10:594–624.
Berg V, Lyche JL, Gutleb AC, Lie E, Skaare JU, Aleksandersen M, et al. Distribution of PCB 118 and PCB 153 and hydroxylated PCB metabolites (OH-CBs) in maternal, fetal and lamb tissues of sheep exposed during gestation and lactation. Chemosphere 2010;80:1144–50.
Bernhoft A, Wiig O, Skaare JU. Organochlorines in polar bears (*Ursus maritimus*) at Svalbard. Environ Pollut 1997;95:159–75.
Blake MJ, Castro L, Leeder JS, Kearns GL. Ontogeny of drug metabolizing enzymes in the neonate. Semin Fetal Neonatal Med 2005;10:123–38.
Blix AS, Lentfer JW. Modes of thermal protection in polar bear cubs — at birth and on emergence from the den. Am J Physiol 1979;236:R67–74.
Bonfanti P, Colombo A, Villa S, Comelli F, Costa B, Santagostino A. The effects of accumulation of an environmentally relevant polychlorinated biphenyl mixture on cytochrome P450 and P-glycoprotein expressions in fetuses and pregnant rats. Chemosphere 2009;75:572–9.
Boon J, van der Meer J, Allchin C, Law R, Klunsoyr J, Leonards P. Concentration-dependent changes of PCB patterns in fish-eating mammals: structural evidence for induction of cytochrome P450. Arch Environ Contam Toxicol 1997;33:298–311.
Braathen M, Derocher AE, Wiig O, Sormo EG, Lie E, Skaare JU, et al. Relationships between PCBs and thyroid hormones and retinol in female and male polar bears. Environ Health Perspect 2004;112:826–33.
Braune BM, Outridge PM, Fisk AT, Muir DCG, Helm PA, Hobbs K, et al. Persistent organic pollutants and mercury in marine biota of the Canadian Arctic: an overview of spatial and temporal trends. Sci Total Environ 2005;351:4-56.
Brevik EM. Gas-chromatographic method for determination of organochlorine pesticides in human milk. Bull Environ Contam Toxicol 1978;19:281–6.
Brouwer A, van den Berg KJ. Binding of a metabolite of 3,4,3′,4′-tetrachlorobiphenyl to transthyretin reduces serum vitamin-a transport by inhibiting the formation of the protein complex carrying both retinol and thyroxine. Toxicol Appl Pharmacol 1986;85:301–12.
Brouwer A, Ahlborg UG, van den Berg M, Birnbaum LS, Boersma ER, Bosveld B, et al. Functional-aspects of developmental toxicity of polyhalogenated aromatic-hydrocarbons in experimental-animals and human infants. Eur J Pharmacol, Environ Toxicol Pharmacol Sect 1995;293:1-40.
Brouwer A, Morse DC, Lans MC, Schuur AG, Murk AJ, Klasson-Wehler E, et al. Interactions of persistent environmental organohalogens with the thyroid hormone system: mechanisms and possible consequences for animal and human health. Toxicol Ind Health 1998;14:59–84.
Brown JF, Lawton RW, Ross MR, Feingold J, Wagner RE, Hamilton SB. Persistence of PCB congeners in capacitor workers and Yusho patients. Chemosphere 1989;19:829–34.
Calvert W, Ramsay MA. Evaluation of age determination of polar bears by counts of cementum growth layer groups. Ursus 1998;10:449–53.
Cattet MRL, Caulkett NA, Obbard ME, Stenhouse GB. A body-condition index for ursids. Can J Zool (Rev Can Zool) 2002;80:1156–61.
Cheek A. Potential mechanisms of thyroid disruption in humans: interaction of organochlorine compounds with thyroid receptor, transthyretin, and thyroid-binding globulin. Environ Health Perspect 1999;107:273–8.
Christensen-Dalsgaard SN, Aars J, Andersen M, Lockyer C, Yoccoz NG. Accuracy and precision in estimation of age of Norwegian Arctic polar bears (*Ursus maritimus*) using dental cementum layers from known-age individuals. Polar Biol 2010;33:589–97.
Colborn T, Saal F, Soto A. Developmental effects of endocrine-disrupting chemicals in wildlife and humans. Environ Health Perspect 1993;101:378–84.
Czekaj P, Wiaderkiewicz A, Wiaderkiewicz R, Palasz A. Expression of constitutive and inducible cytochromes P450 in fetal and newborn rat liver. Pol J Environ Stud 2006;15:699–708.
Derocher AE, Wiig Ø. Postnatal growth in body length and mass of polar bears (*Ursus maritimus*) at Svalbard. J Zool 2002;256:343–9.
Derocher AE, Andriashek D, Arnould JPY. Aspects of milk-composition and lactation in polar bears. Can J Zool (Rev Can Zool) 1993;71:561–7.
Derocher AE, Wolkers H, Colborn T, Schlabach M, Larsen TS, Wiig O. Contaminants in Svalbard polar bear samples archived since 1967 and possible population level effects. Sci Total Environ 2003;301:163–74.
Derocher AE, Andersen M, Wiig O. Sexual dimorphism of polar bears. J Mammal 2005;86:895–901.
Dietz R, Riget FF, Sonne C, Letcher R, Born EW, Muir DCG. Seasonal and temporal trends in polychlorinated biphenyls and organochlorine pesticides in East Greenland polar bears (*Ursus maritimus*), 1990–2001. Sci Total Environ 2004;331:107–24.
Eltom SE, Schwark WS. CYP1A1 and CYP1B1, two hydrocarbon-inducible cytochromes P450, are constitutively expressed in neonate and adult goat liver, lung and kidney. Pharmacol Toxicol 1999;85:65–73.
Fein GG, Jacobson JL, Jacobson SW, Schwartz PM, Dowler JK. Prenatal exposure to polychlorinated-biphenyls — effects on birth size and gestational age. J Pediatr 1984;105:315–20.
Fromme H, Mosch C, Morovitz M, Alba-Alejandre I, Boehmer S, Kiranoglu M, et al. Pre- and Postnatal exposure to perfluorinated compounds (PFCs). Environ Sci Technol 2010;44:7123–9.
Gabrielsen KM, Villanger GD, Lie E, Karimi M, Lydersen C, Kovacs KM, et al. Levels and patterns of hydroxylated polychlorinated biphenyls (OH-PCBs) and their associations with thyroid hormones in hooded seal (*Cystophora cristata*) mother–pup pairs. Aquat Toxicol 2011;105:482–91.
Gauger KJ, Kato Y, Haraguchi K, Lehmler HJ, Robertson LW, Bansal R, et al. Polychlorinated biphenyls (PCBs) exert thyroid hormone-like effects in the fetal rat brain but do not bind to thyroid hormone receptors. Environ Health Perspect 2004;112:516–23.
Gebbink WA, Sonne C, Dietz R, Kirkegaard M, Riget FF, Born EW, et al. Tissue-specific congener composition of organohalogen and metabolite contaminants in East Greenland polar bears (*Ursus maritimus*). Environ Pollut 2008;152:621–9.
Grandjean P, Landrigan PJ. Developmental neurotoxicity of industrial chemicals. Lancet 2006;368:2167–78.
Greig DJ, Ylitalo GM, Hall AJ, Fauquier DA, Gulland FMD. Transplacental transfer of organochlorines in California sea lions (*Zalophus californianus*). Environ Toxicol Chem 2007;26:37–44.
Gutleb AC, Cenijn P, van Velzen M, Lie E, Ropstad E, Skaare JU, et al. In vitro assay shows that PCB metabolites completely saturate thyroid hormone transport capacity in blood of wild polar bears (*Ursus maritimus*). Environ Sci Technol 2010;44:3149–54.

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Guvenius DM, Aronsson A, Ekman-Ordeberg G, Bergman A, Noren K. Human prenatal and postnatal exposure to polybrominated diphenyl ethers, polychlorinated biphenyls, polychlorobiphenylols, and pentachlorophenol. Environ Health Perspect 2003;111:1235–41.

Haave M, Ropstad E, Derocher AE, Lie E, Dahl E, Wiig O, et al. Polychlorinated biphenyls and reproductive hormones in female polar bears at Svalbard. Environ Health Perspect 2003;111:431–6.

Henriksen E. Monitoring PCBs in polar bears: lessons learned from Svalbard. J Environ Monit 2001;3:493–8.

Huisman M, Koopman-Esseboom C, Fidler V, Hadders-Algra M, van der Paauw CG, Tuinstra LGMT, et al. Perinatal exposure to polychlorinated biphenyls and dioxins and its effect on neonatal neurological development. Early Hum Dev 1995;41:111–27.

Jenssen BM. Endocrine-disrupting chemicals and climate change: a worst-case combination for arctic marine mammals and seabirds? Environ Health Perspect 2006;114:76–80.

Karrman A, Ericson I, van Bavel B, Darnerud PO, Aune M, Glynn A, et al. Exposure of perfluorinated chemicals through lactation: levels of matched human milk and serum and a temporal trend, 1996–2004, in Sweden. Environ Health Perspect 2007;115:226–30.

Kester MHA, Bulduk S, Tibboel D, Meinl W, Glatt H, Falany CN, et al. Potent inhibition of estrogen sulfotransferase by hydroxylated PCB metabolites: a novel pathway explaining the estrogenic activity of PCBs. Endocrinology 2000;141:1897–900.

Lans MC, Klasson-Wehler E, Willemsen M, Meussen E, Safe S, Brouwer A. Structure-dependent, competitive interaction of hydroxy-polychlorobiphenyls, hydroxy-dibenzo-p-dioxins and hydroxy-dibenzofurans with human transthyretin. Chem Biol Interact 1993;88:7-21.

Lans MC, Spiertz C, Brouwer A, Koeman JH. Different competition of thyroxine-binding to transthyretin and thyroxine-binding globulin by hydroxy-PCBs, PCDDs and PCDFs. Eur J Pharmacol - Environ Toxicol Pharmacol Sect 1994;270:129–36.

Letcher RJ, Norstrom RJ, Lin S, Ramsay MA, Bandiera SM. Immunoquantitation and microsomal monooxygenase activities of hepatic cytochromes P4501A and p4502B and chlorinated hydrocarbon contaminant levels in polar bear (*Ursus maritimus*). Toxicol Appl Pharmacol 1996;137:127–40.

Letcher RJ, Klasson-Wehler E, Bergman Å. Chap. 11: methyl sulfone and hydroxylated metabolites of polychlorinated biphenyls. In: Paasivirta J, editor. The handbook of environmental chemistry, Vol. 3. Berlin: Springer -Verlag; 2000.. Part K.

Letcher RJ, Bustnes JO, Dietz R, Jenssen BM, Jorgensen EH, Sonne C, et al. Exposure and effects assessment of persistent organohalogen contaminants in Arctic wildlife and fish. Sci Total Environ 2010;408:2995–3043.

Lie E, Larsen HJS, Larsen S, Johnsen GM, Derocher AE, Lunn NJ, et al. Does high organochlorine (OC) exposure impair the resistance to infection in polar bears (*Ursus maritimus*)? Part I: effect of OCs on the humoral immunity. J Toxicol Environ Health A 2004;67:555–82.

Lie E, Larsen HJS, Larsen S, Johansen GM, Derocher AE, Lunn NJ, et al. Does high organochlorine (OC) exposure impair the resistance to infection in polar bears (*Ursus maritimus*)? Part II: possible effect of OCs on mitogen- and antigen-induced lymphocyte proliferation. J Toxicol Environ Health A 2005;68:457–84.

Løken KB, Lie E, Lundanes E, Skaare JU. Extension of a multicomponent method to include the determination of OH-PCBs and OH-PBDEs in biological matrices. Organohalogen Compd 2006;68:2430–3.

Lydersen C, Wolkers H, Severinsen T, Kleivane L, Nordoy ES, Skaare JU. Blood is a poor substrate for monitoring pollution burdens in phocid seals. Sci Total Environ 2002;292:193–203.

Machala M, Blaha L, Vondracek J, Trosko JE, Scott J, Upham BL. Inhibition of gap junctional intercellular communication by noncoplanar polychlorinated biphenyls: inhibitory potencies and screening for potential mode(s) of action. Toxicol Sci 2003;76:102–11.

Machala M, Blaha L, Lehmler HJ, Pliskova M, Majkova Z, Kapplova P, et al. Toxicity of hydroxylated and quinoid PCB metabolites: inhibition of gap junctional intercellular communication and activation of aryl hydrocarbon and estrogen receptors in hepatic and mammary cells. Chem Res Toxicol 2004;17:340–7.

Maervoet J, Vermeir G, Covaci A, Van Larebeke N, Koppen G, Schoeters G, et al. Association of thyroid hormone concentrations with levels of organochlorine compounds in cord blood of neonates. Environ Health Perspect 2007;115:1780–6.

McKinney MA, Stirling I, Lunn NJ, Peacock E, Letcher RJ. The role of diet on long-term concentration and pattern trends of brominated and chlorinated contaminants in western Hudson Bay polar bears, 1991–2007. Sci Total Environ 2010;408:6210–22.

McKinney MA, Letcher RJ, Aars J, Born EW, Branigan M, Dietz R, et al. Flame retardants and legacy contaminants in polar bears from Alaska, Canada, East Greenland and Svalbard, 2005–2008. Environ Int 2011;37:365–74.

Meerts I, Lilienthal H, Hoving S, van den Berg JHJ, Weijers BM, Bergman A, et al. Developmental exposure to 4-hydroxy-2,3,3 ′,4 ′,5-pentachlorobiphenyl (4-OH-CB107): long-term effects on brain development, behavior, and brain stem auditory evoked potentials in rats. Toxicol Sci 2004;82:207–18.

Miller MD, Marty MA, Arcus A, Brown J, Morry D, Sandy M. Differences between children and adults: implications for risk assessment at california EPA. Int J Toxicol 2002;21:403–18.

Needham LL, Grandjean P, Heinzow B, Jørgensen PJ, Nielsen F, Patterson DG, et al. Partition of environmental chemicals between maternal and fetal blood and tissues. Environ Sci Technol 2010;45:1121–6.

Nommsen LA, Lovelady CA, Heinig MJ, Lonnerdal B, Dewey KG. Determinants of energy, protein, lipid, and lactose concentrations in human-milk during the 1st 12 months of lactation — the Darling study. Am J Clin Nutr 1991;53:457–65.

Norstrom RJ, Muir DCG. Chlorinated-hydrocarbon contaminants in Arctic marine mammals. Sci Total Environ 1994;154:107–28.

Norstrom RJ, Simon M, Muir DCG, Schweinsburg RE. Organochlorine contaminants in Arctic marine food-chains — identification, geographical distribution, and temporal trends in polar bears. Environ Sci Technol 1988;22:1063–71.

Norstrom RJ, Belikov SE, Born EW, Garner GW, Malone B, Olpinski S, et al. Chlorinated hydrocarbon contaminants in polar bears from eastern Russia, North America, Greenland, and Svalbard: biomonitoring of Arctic pollution. Arch Environ Contam Toxicol 1998;35:354–67.

Olsen GH, Mauritzen M, Derocher AE, Sormo EG, Skaare JU, Wiig O, et al. Space-use strategy is an important determinant of PCB concentrations in female polar bears in the barents sea. Environ Sci Technol 2003;37:4919–24.

Oskam IC, Ropstad E, Dahl E, Lie E, Derocher AE, Wiig O, et al. Organochlorines affect the major androgenic hormone, testosterone, in male polar bears (*Ursus maritimus*) at Svalbard. J Toxicol Environ Health A 2003;66:2119–39.

Oskam IC, Ropstad E, Lie E, Derocher AE, Wiig O, Dahl E, et al. Organochlorines affect the steroid hormone cortisol in free-ranging polar bears (*Ursus maritimus*) at Svalbard, Norway. J Toxicol Environ Health A 2004;67:959–77.

Park JS, Bergman A, Linderholm L, Athanasiadou M, Kocan A, Petrik J, et al. Placental transfer of polychlorinated biphenyls, their hydroxylated metabolites and pentachlorophenol in pregnant women from eastern Slovakia. Chemosphere 2008;70:1676–84.

Park HY, Park JS, Sovcikova EJ, Kocan A, Linderholm L, Bergman A, et al. Exposure to hydroxylated polychlorinated biphenyls (OH-PCBs) in the prenatal period and subsequent neurodevelopment in eastern Slovakia. Environ Health Perspect 2009;117:1600–6.

Peterson RE, Theobald HM, Kimmel GL. Developmental and reproductive toxicity of dioxins and related compouds — cross-species comparisons. Crit Rev Toxicol 1993;23:283–335.

Polischuk SC, Letcher RJ, Norstrom RJ, Ramsay MA. Preliminary-results of fasting on the kinetics of organochlorines in polar bears (*Ursus maritimus*). Sci Total Environ 1995;161:465–72.

Polischuk SC, Norstrom RJ, Ramsay MA. Body burdens and tissue concentrations of organochlorines in polar bears (*Ursus maritimus*) vary during seasonal fasts. Environ Pollut 2002;118:29–39.

Ptak A, Ludewig G, Lehmler HJ, Wojtowicz AK, Robertson LW, Gregoraszczuk EL. Comparison of the actions of 4-chlorobiphenyl and its hydroxylated metabolites on estradiol secretion by ovarian follicles in primary cells in culture. Reprod Toxicol 2005;20:57–64.

Rickenbacher U, McKinney JD, Oatley SJ, Blake CCF. Structural specific interaction of halogenated dioxin and biphenyl derivates with iodothyrenine-5′-deiodinase in rat liver. J Med Chem 1986;29:641–8.

Routti H, Letcher RJ, Arukwe A, van Bavel B, Yoccoz NG, Chu SG, et al. Biotransformation of PCBs in relation to phase i and ii xenobiotic-metabolizing enzyme activities in ringed seals (*Phoca hispida*) from Svalbard and the Baltic Sea. Environ Sci Technol 2008;42:8952–8.

Roze E, Meijer L, Bakker A, Van Braeckel K, Sauer PJJ, Bos AF. Prenatal exposure to organohalogens, including brominated flame retardants, influences motor, cognitive, and behavioral performance at school age. Environ Health Perspect 2009;117:1953–8.

Rylander L, Stromberg U, Hagmar L. Dietary intake of fish contaminated with persistent organochlorine compounds in relation to low birthweight. Scand J Work Environ Health 1996;22:260–6.

Safe SH. Polychlorinated biphenyls (PCBs) — envionmental impact, biochemical and toxic responses, and implications for risk assessment. Crit Rev Toxicol 1994;24:87-149.

Sandala GM, Sonne C, Dietz R, Muir DCG, Valters K, Bennett ER, et al. Hydroxylated and methyl sulfone PCB metabolites in adipose and whole blood of polar bear (*Ursus maritimus*) from East Greenland. Sci Total Environ 2004;331:125–41.

Sandau CD. Analytical Chemistry of Hydroxylated Metabolites of PCBs and Other Halogenated Phenolic Compounds in Blood and their Relationship to Thyroid Hormone and Retinol Homeostasis in Humans and Polar Bears. Ph.D. Dissertation. Carleton University. Ottawa, 2000.

Sandau CD, Meerts IATM, Letcher RJ, McAlees AJ, Chittim B, Brouwer A, et al. Identification of 4-hydroxyheptachlorostyrene in polar bear plasma and its binding affinity to transthyretin: a metabolite of octachlorostyrene? Environ Sci Technol 2000;34:3871–7.

Sandau C, Ayotte P, Dewailly E, Duffe J, Norstrom R. Pentachlorophenol and hydroxylated polychlorinated biphenyl metabolites in umbilical cord plasma of neonates from coastal populations in Quebec. Environ Health Perspect 2002;110:411–7.

Schantz SL, Widholm JJ, Rice DC. Effects of PCB exposure on neuropsychological function in children. Environ Health Perspect 2003;111:357–76.

Schuur AG, Brouwer A, Bergman A, Coughtrie MWH, Visser TJ. Inhibition of thyroid hormone sulfation by hydroxylated metabolites of polychlorinated biphenyls. Chem Biol Interact 1998;109:293–7.

Schuur AG, Bergman A, Brouwer A, Visser TJ. Effects of pentachlorophenol and hydroxylated polychlorinated biphenyls on thyroid hormone conjugation in a rat and a human hepatoma cell line. Toxicol In Vitro 1999;13:417–25.

Sjödin A, Hagmar L, Klasson-Wehler E, Bjork J, Bergman A. Influence of the consumption of fatty Baltic Sea fish on plasma levels of halogenated environmental contaminants in Latvian and Swedish men. Environ Health Perspect 2000;108:1035–41.

Skaare JU, Bernhoft A, Wiig O, Norum KR, Haug E, Eide DM, et al. Relationships between plasma levels of organochlorines, retinol and thyroid hormones from polar bears (*Ursus maritimus*) at Svalbard. J Toxicol Environ Health A 2001;62:227–41.

Sonne C. Health effects from long-range transported contaminants in Arctic top predators: an integrated review based on studies of polar bears and relevant model species. Environ Int 2010;36:461–91.

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Sørmo EG, Skaare JU, Lydersen C, Kovacs KM, Hammill MO, Jenssen BM. Partitioning of persistent organic pollutants in grey seal (*Halichoerus grypus*) mother–pup pairs. Sci Total Environ 2003a;302:145–55.

Sørmo EG, Skaare JU, Jussi I, Jussi M, Jenssen BM. Polychlorinated biphenyls and organochlorine pesticides in Baltic and Atlantic gray seal (*Halichoerus grypus*) pups. Environ Toxicol Chem 2003b;22:2789–99.

Stirling I, Spencer C, Andriashek D. Immobilization of polar bears (*Ursus maritimus*) with telazol in the Canadian Arctic. J Wildl Dis 1989;25:159–68.

Tryland M, Brun E, Derocher AE, Arnemo JM, Kierulf P, Olberg RA, et al. Plasma biochemical values from apparently healthy free-ranging polar bears from Svalbard. J Wildl Dis 2002;38:566–75.

Ulbrich B, Stahlmann R. Developmental toxicity of polychlorinated biphenyls (PCBs): a systematic review of experimental data. Arch Toxicol 2004;78:252–68.

van den Berg KJ. Interaction of chlorinated phenols with thyroxine binding sites of human transthyretin, albumin and thyroid binding globulin. Chem Biol Interact 1990;76:63–75.

Verreault J, Muir DCG, Norstrom RJ, Stirling I, Fisk AT, Gabrielsen GW, et al. Chlorinated hydrocarbon contaminants and metabolites in polar bears (*Ursus maritimus*) from Alaska, Canada, East Greenland, and Svalbard: 1996–2002. Sci Total Environ 2005a;351:369–90.

Verreault J, Gabrielsen GV, Chu SG, Muir DCG, Andersen M, Hamaed A, et al. Flame retardants and methoxylated and hydroxylated polybrominated diphenyl ethers in two Norwegian Arctic top predators: glaucous gulls and polar bears. Environ Sci Technol 2005b;39:6021–8.

Verreault J, Dietz R, Sonne C, Gebbink WA, Shahmiri S, Letcher RJ. Comparative fate of organohalogen contaminants in two top carnivores in greenland: captive sledge dogs and wild polar bears. Comp Biochem Physiol Part C Toxicol Pharmacol 2008;147:306–15.

Wiig O, Derocher AE, Cronin NM, Skaare JU. Female pseudohermaphrodite polar bears at Svalbard. J Wildl Dis 1998;34:792–6.

Wolkers H, Krafft BA, van Bavel B, Helgason LB, Lydersen C, Kovacs K. Biomarker responses and decreasing contaminant levels in ringed seals (*Pusa hispida*) from Svalbard, Norway. J Toxicol Environ Health A 2008;71:1009–18.

Yakushiji T, Watanabe I, Kuwabara K, Tanaka R, Kashimoto T, Kunita N, et al. Rate of decrease and half-life of polychlorinated-biphenyls (PCBs) in the blood of mothers and their children occupationally exposed to PCBs. Arch Environ Contam Toxicol 1984;13:341–5.

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