Selective bioaccumulation of chlorinated pesticides and metabolites in Arctic seabirds

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Avian chlorinated pesticide accumulation is highly species-specific.

Abstract

Chlorinated pesticides and metabolites (CPs) were quantified in the seabird species: little auk (*Alle alle*), Brünnich's guillemot (*Uria lomvia*), black guillemot (*Cepphus grylle*) and black-legged kittiwake (*Rissa tridactyla*). The purpose was to evaluate avian accumulation of selected CPs based on their concentrations and relative patterns, their relation to dietary descriptors (stable isotopes of carbon and nitrogen), to enzymes involved in biotransformation, as well as CPs' accumulation potential relative to the recalcitrant polychlorinated biphenyl PCB-153. In all species, the CP pattern was dominated by *p,p′*-dichlorodiphenyltrichloroethane (DDE) and hexachlorbenzene (HCB). Except for HCB, concentrations were not related to trophic position. Most CPs were quantified in black guillemot, indicating a slower elimination compared to other seabird species. Brünnich's guillemot showed efficient elimination of chlordanes, whereas the opposite was found for little auk. Kittiwake showed higher accumulation of persistent CP and metabolites than auks, whereas accumulation of less recalcitrant CPs was low.
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1. Introduction

Arctic seabirds tend to accumulate high concentrations of organochlorine contaminants (OCs) (Bourne and Bogan, 1972; Gabrielsen et al., 1995; Fisk et al., 2001a). OC concentrations in birds occupying high trophic positions, such as glaucous gulls (*Larus hyperboreus*), are high enough to cause concern about effects (Gabrielsen et al., 1995; Bustnes et al., 2005). In Arctic seabirds occupying lower trophic positions, the OC concentrations are lower than threshold levels for possible effects found in other seabird species (Braune et al., 2001; Borgå et al., 2005; Fisk et al., 2005).

Seabirds, as other vertebrates, accumulate OCs mainly from the diet, usually leading to biomagnification; increasing OC concentrations with increasing trophic position in the food web (e.g. Fisk et al., 2001b). Several marine food web studies, spanning over a broad range of trophic positions, have demonstrated relationships between OC concentrations and dietary descriptors such as stable carbon and nitrogen isotope ratios ($\delta^{13}C$ and $\delta^{15}N$, respectively) (Broman et al., 1992; Ruus et al., 1999; Fisk et al., 2001b; Hop et al., 2002; Hoekstra et al., 2003). Due to elimination of the lighter isotopes ^{12}C and ^{14}N, the heavier isotopes ^{13}C and ^{15}N are enriched and may be used to distinguish animals based on their carbon source ($\delta^{13}C$) and trophic position ($\delta^{15}N$) (Hobson and Welch, 1992; Hobson et al., 1995). However, within a species, or when comparing species covering a narrow trophic range, $\delta^{13}C$ and $\delta^{15}N$ may not help explaining the variation in OC

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concentrations (Fisk et al., 2001a; Sagerup et al., 2001; Borgå et al., 2005).

Recent contaminant studies of Arctic seabirds have demonstrated that the OC accumulation is not uniform among the seabird species, but that it varies with differences in diet and biotransformation abilities among seabird species (Fisk et al., 2001a; Moisey et al., 2001; Borgå et al., 2005). For example, the Brünnich's guillemot (*Uria lomvia*) has more efficient chlordane metabolism than the other auks (Fisk et al., 2001a), whereas it is a poor metabolizer of *ortho-meta* unsubstituted congeners of polychlorinated biphenyls (*om*-PCBs) (Borgå et al., 2005). Based on their PCB pattern, the little auk (*Alle alle*) seems to efficiently metabolize non-persistent congeners, whereas the black guillemot (*Cepphus grylle*) is a poor metabolizer of *meta-para* unsubstituted (*mp*) PCB congeners.

The cytochrome P450 enzyme system (CYP) is involved in the first oxidative step of OC biotransformation (Walker, 1998). The presence and activity of CYP isoforms determine the ability and capacity to biotransform contaminants and thus OC levels and patterns (Murk et al., 1994; Walker, 1998). As the contribution of *om*-PCBs to $\sum$PCBs decreased with increasing CYP1A activity (ethoxyresorufin-*O*-deethylation [EROD]), there seems to be a direct link between OC pattern in Arctic seabirds and their biotransformation abilities (Borgå et al., 2005). Similarly, black guillemot showed lowest CYP2B/3A activity (testosterone-6β-hydroxylation), and poor *mp*-PCBs elimination.

The aim of the present study was to investigate potential selective bioaccumulation of chlorinated pesticides (CPs) in four different seabird species from two families (Auks: little auk, Brünnich's guillemot, and black guillemot. Gulls: black-legged kittiwake [*Rissa tridactyla*]). Of the seabirds included in the present study, little auk occupies the lowest trophic position based on δ^{15}N, followed by Brünnich's guillemot and kittiwake, with black guillemot occupying the highest trophic position (Borgå et al., 2005). The δ^{13}C range is narrower than reported for birds with distinct benthic or pelagic feeding (Dahl et al., 2003).

The CYP enzyme activity is low in all species compared to birds from industrialized areas, but with higher EROD activity in little auk than the other species (Borgå et al., 2005). Little auk and Brünnich's guillemot migrate south-west into the northern Atlantic Ocean and to the southern region of Greenland, whereas kittiwake migrates the same route in a broader and more southward band, with some individuals overwintering in central Europe (Anker-Nilssen et al., 2000). Black guillemot, on the other hand, is an Arctic resident throughout the year (Anker-Nilssen et al., 2000).

Although several factors such as sex and age may influence exposure and accumulation of CPs in avian wildlife, the focus of the present study is on dietary exposure and biotransformation, with control for variation due to sex. More specifically, when evaluating the influence of exposure, the avian CP bioaccumulation is related to the seabirds' dietary descriptors (δ^{13}C, δ^{15}N) and dietary CP content. Furthermore, to investigate the influence of biotransformation, the seabirds' CP

bioaccumulation is compared to enzymatic activities. Finally, the above was combined with the bioaccumulation potential of each CPs (relative to the recalcitrant PCB-153, controlled for exposure) to categorize the seabirds as efficient or non-efficient metabolizers. A final categorization was based also on earlier chlordane (Fisk et al., 2001a) and HCHs (Moisey et al., 2001) studies from the Canadian Arctic, and on PCB results from the European Arctic (Borgå et al., 2005). The present study thereby aimed to confirm the bioaccumulation categorization of CPs between the two regions of the Arctic.

2. Materials and methods

2.1. Sampling and analyses

Five females and five males of each seabird species were shot using a shotgun with steel pellets in the marginal ice zone of the north-central (76°08′−76°96′N, 32°52′−33°31′E) and north-western (76°46′−77°45′N, 27°00′−28°13′E) Barents Sea in May 1999 (total $n = 40$). Within 15 min after death, the seabirds were dissected, and livers were stored in liquid nitrogen for later analyses of CYP enzyme activities determined by liver microsome EROD rates (Wolkers et al., 1998, and references therein), and testosterone hydroxylation activities (Wortelboer et al., 1992). Liver and muscle samples for CP and stable isotope analyses, respectively, were dissected and stored frozen at −20 °C in containers of polypropylene or aluminium foil, respectively. Body mass (g), sex, age (juvenile/adult based on plumage) were registered. Values of δ^{13}C and δ^{15}N were determined in the seabirds' muscle at the Institute for Energy Technology, Kjeller, Norway, as described in detail by Hobson et al. (1995) and Hop et al. (2002). The seabirds' enzyme activities and stable isotope ratios, and their interrelations were presented in detail by Borgå et al. (2005), based on the same seabird samples.

The seabirds' main prey were collected simultaneously as the seabirds, assuming a diet of 100% copepods (*Calanus glacialis* and *Calanus hyperboreus*) in little auk, 20% euphausiids (*Thysanoessa inermis*), 20% amphipods (*Themisto libellula*) and 60% polar cod (*Boreogadus saida*) in Brünnich's guillemot, and 100% polar cod in black guillemot and kittiwake, based on previous dietary analysis of these species in the Barents Sea (Lønne and Gabrielsen, 1992; Mehlum and Gabrielsen, 1993; Weslawski et al., 1999). Zooplankton were collected by net hauls from 200 to 240 m to the surface using nets with 1.00 and 1.55 mm mesh (WP3 net, Macrozooplankton net, Tucker trawl), and sorted into species and size/stage specific pooled samples; copepods 580−1135 individuals per sample, euphausiids 100−115 individuals per sample, amphipods 7−16 individuals per sample. Polar cods in the size range eaten by the seabirds (12−18 cm, Erikstad, 1990) were collected with benthic trawls at approximately 300 m depth, and analysed whole individually.

The CP concentrations of seabird liver and whole prey were quantified using methods based on Brevik (1978) with slight modifications for extraction of lipids and CPs with cyclohexane and acetone, and lipid clean-up with sulphuric acid from Bernhoft and Skaare (1994). In brief, an internal standard (PCB-29, -112, and -207) was added after an initial coarse homogenization using an Ika Ultra Turrax™, followed by further homogenization and extraction of CPs and lipids in cyclohexane and acetone (3:2, v:v) using a Cole-Parmer ultrasonic homogenizer (4710 series). A portion of the lipid extract was used to gravimetrically determine the content of extractable organic matter, mainly neutral lipids. Separation and identification of the CPs in the lipid-free extract was done by a high-resolution gas chromatography (HRGC, Agilent 6890 Plus GC system, Agilent Technologies) equipped with two fused silica capillary columns of different polarity (SPB-5 and SPB-1701; 60 m, 0.25 mm ID, 0.25 μm film; Supelco Inc.) and ^{63}Ni-micro electron capture detector (Agilent Technologies). The seabird samples were analysed for the content of α-, β- and γ-hexachlorocyclohexane ($\sum$HCHs), hexachlorobenzene (HCB), *cis*-chlordane, oxychlordane and *trans*-nonachlor ($\sum$Chlordanes), the dichlorodiphenyltrichloroethane compounds *p,p'*-DDE, *p,p'*-DDD, *o,p'*-DDD, and *p,p'*-DDT ($\sum$DDTs), and Mirex. Zooplankton and fish were additionally analysed for *trans*-chlordane, whereas *o,p'*-DDD and Mirex were omitted. The range of

CP recoveries for the seabird analyses was 77–105% (mean 88%), whereas it was 72–123% (mean 97%), and 90–160% (mean 115%) for CPs in zooplankton and PCBs polar cod, respectively. The compound dependent quantification limit (QL), defined as $3 \times$ detection limit (DL), ranged from 0.06 to 0.29 ng g^{-1} wet weight (ww) (mean 0.19 ng g^{-1} ww) for seabirds, and from 0.01 to 0.23 ng g^{-1} ww for zooplankton (mean 0.05 ng g^{-1} ww) and from 0.06 to 0.41 ng g^{-1} ww (mean 0.15 ng g^{-1} ww) for polar cod. The reproducibility, precision, linearity and sensitivity of the analyses were within the accredited requirements of the performing laboratory (Environmental Toxicology Laboratory at the Norwegian School of Veterinary Science).

2.2. Data treatment

The seabirds' CP pattern is presented as proportion of each CP relative to $\sum$CPs, including HCB, β-HCH, oxychlordane, *cis*-chlordane, *trans*-nonachlor, *p,p′*-DDE, and Mirex, with $0.5 \times$ DL replacing three Mirex and five *cis*-chlordane non-detections. The other CPs (α- and γ-HCH, *o,p′*-DDD, *p,p′*-DDD and *p,p′*-DDT) were below DL and/or QL in more than 25% of the samples, and were not included in graphs or statistics. They are, however, listed in Table 1, with only samples above QL included in the descriptive statistics.

The bioaccumulation potential was evaluated by calculating metabolic indices (MI), where each CP is compared to the highly bioaccumulative PCB-153 while controlling for the contribution from the respective prey (Tanabe et al., 1988):

$$MI = \frac{[CP]_{seabird}}{[PCB-153]_{seabird}} \Big/ \frac{[CP]_{prey}}{[PCB-153]_{prey}} \tag{1}$$

Metabolic index was only calculated for CPs quantified in both prey (Table 2) and seabirds (Table 1).

Table 1
Seabird descriptors (from Borgå et al., 2005) and chlorinated pesticide concentrations in seabirds from the Barents Sea May 1999 (mean ± SE, range)

Species	Little auk	Brünnich's guillemot	Black guillemot	Kittiwake
Chlorinated pesticide concentrations (ng g^{-1} lipid weight)				
HCB	464 ± 66.2	721 ± 67.4	592 ± 65.6	590 ± 89.5
	195–892	422–1164	320–983	291–1184
α-HCH[a]	nd	4.7 ± 0.7	5.1 ± 0.8	nd
		3.7–5.9	2.9–7.8	
β-HCH	40.8 ± 6.8	24.0 ± 2.9	28.7 ± 5.5	21.8 ± 3.5
	15.0–75.5	14.3–47.1	14.5–74.2	9.5–42.2
γ-HCH[b]	nd	nd	nd	nd
Oxychlordane	175.8 ± 30.5	165 ± 23.8	186 ± 21.9	471 ± 103
	66.4–373.1	105–339	92.3–290	36.3–1090
cis-Chlordane[c]	54.6 ± 9.6	7.5 ± 0.9	18.6 ± 2.0	45.2 ± 4.9
	17.3–108	4.6–13.1	8.5–30.0	19.5–71.6
trans-Nonachlor	367 ± 56.1	11.4 ± 1.4	107 ± 11.9	83.1 ± 14.1
	156–655	7.5–23.7	61.2–187	35.9–179
p,p′-DDE	784 ± 88.9	850 ± 141	608 ± 43	1168 ± 231
	499–1426	427–2024	368–880	296–2896
o,p′-DDD[d]	nd	nd	14.1 ± 2.3	nd
			7.9–19.1	
p,p′-DDD[e]	nd	nd	11.7 ± 1.4	nd
			7.1–16.4	
p,p′-DDT[f]	nd	nd	nd	nd
Mirex[g]	59.6 ± 9.2	23.6 ± 2.5	29.5 ± 2.9	156 ± 32.3
	30.9–113	13.5–39.8	15.9–47.4	55.1–371
Seabird descriptors				
Lipid %	4.1 ± 0.2	3.6 ± 0.4	5.3 ± 1.0	7.7 ± 1.3
	3.2–4.8	2.5–6.0	2.5–13.8	3.0–14.5
δ^{15}N (‰)	10.5 ± 0.1	13.1 ± 0.1	14.2 ± 0.1	13.5 ± 0.1
	9.5–11.0	12.8–13.6	13.7–15.0	12.9–14.2
δ^{13}C (‰)	−21.4 ± 0.1	−21.0 ± 0.1	−21.9 ± 0.1	−21.3 ± 0.1
	−22.0 to −20.8	−21.3 to −20.6	−22.1 to −21.6	−21.9 to −20.8
EROD (pmol min^{-1} mg protein^{-1})	36.9 ± 3.2	8.2 ± 1.5	10.1 ± 0.7	12.0 ± 1.4
	21.3–51.3	3.0–16.2	7.0–13.4	4.7–20.3
Testosterone-6β-hydroxylation (pmol min^{-1} mg protein^{-1})	118.5 ± 7.8	139.7 ± 14.8	89.8 ± 9.7	129.1 ± 8.3
	87.2–175.9	78.3–211.4	59.9–160.5	94.2–167.2

nd = concentrations below detection limit (DL) or quantification limit (QL) in all samples.

[a] α-HCH was below QL in 75% of the samples, and only above QL in six black guillemots and three Brünnich's guillemots. The numbers given are only based on the samples above QL.

[b] γ-HCH was below QL in four black and one Brünnich's guillemots and not detected in the rest of the samples.

[c] *cis*-Chlordane was above the QL in all samples, except five Brünnich's guillemots which were replaced by $0.5 \times$ DL, and included in statistics and graphs as they constituted less than 25% of the total samples.

[d] *o,p′*-DDD was below DL in more than 75% of the samples, and only above QL in four black guillemots. The numbers given are only based on the samples above QL.

[e] *p,p′*-DDD was below DL in all samples except for six black guillemots. The numbers given are only based on the samples above QL.

[f] *p,p′*-DDT was not detected in any samples.

[g] Mirex was above the QL in all samples, except for one little auk and two Brünnich's guillemots which were replaced by $0.5 \times$ DL, and included in statistics and graphs as they constituted less than 25% of the total samples.

Table 2
Lipid adjusted concentrations (mean ± SE) in the dominating prey of the seabirds[a]

	Copepods ($n = 15$)	Euphausiids ($n = 3$)	Amphipods ($n = 9$)	Polar cod ($n = 12$)
Lipid %	2.6 ± 0.2	1.8 ± 0.2	1.9 ± 0.4	3.8 ± 0.4
HCB	1.4 ± 0.3	39.4 ± 4.8	31.6 ± 5.9	58.6 ± 10.4
α-HCH	7.5 ± 0.6	14.7 ± 2.2	11.8 ± 2.3	12.8 ± 1.9
β-HCH	1.7 ± 0.2	3.2 ± 0.5	6.1 ± 1.1	2.6 ± 0.3
γ-HCH	4.4 ± 0.3	7.8 ± 1.2	7.0 ± 0.9	5.1 ± 0.7
Oxychlordane	5.5 ± 0.6	7.1 ± 1.1	12.3 ± 1.8	9.4 ± 1.7
cis-Chlordane	12.3 ± 0.8	24.8 ± 2.8	41.5 ± 8.3	64.7 ± 11.8
trans-Nonachlor	9.1 ± 0.6	22.4 ± 2.5	44.2 ± 10.9	54.5 ± 10.2
p,*p*′-DDE	4.6 ± 0.3	21.2 ± 2.0	40.5 ± 11.3	81.9 ± 15.8

[a] Details on seabirds' prey composition, as well as species names are in Section 2.

Univariate relationships between individual CPs and the dietary and enzyme activities were analysed by Type III Sum of Squares Analysis of Variance (ANOVA) in SAS 8.0 for Windows (SAS Institute Inc., 1989). Non-significant explanatory variables were backward selected from the initial model, with a α-level 0.05:

$$CP = \delta^{13}C + \delta^{15}N + EROD + \text{testosterone-6}\beta\text{-hydroxylation} + \text{lipid} \quad (2)$$

Lipid content was included as a covariate in the ANOVA of CP concentrations, and both CP concentrations and enzyme activities were logarithmically transformed to reduce variance heterogeneity and skewness. In the ANOVA of CP pattern, the relative contribution of each CP to $\sum$CP was the response, whereas explanatory variables were entered as above.

Indirect ordination analysis (principal component analysis PCA, CANOCO 4.5 for Windows) was used to analyse interrelationships among the various CPs and the seabird samples (Ter Braak and Šmilauer, 1998). The PCA was applied on logarithmically transformed wet weight concentrations with lipid content as a covariable, and on standardized concentrations (sample-standardized by norm, which is similar to CP pattern where each CP is a proportion of the total). The PCA assigns scores to the individual samples and CPs that are linear combinations presented by uncorrelated principal components (PCs). PCs are extracted that minimize the residual sum of

squares among all response variables (here CPs), where PC1 accounts for the majority of the variance followed by PC2, etc. The CPs are presented as arrows pointing to the direction of increasing value. Dietary and enzymatic variables as well as sex are displayed in the PCA diagram as passive explanatory variables. Further rules of interpretation of the PCA diagram are described by Ter Braak (1995), Van Wijngaarden et al. (1995), and Van den Brink and Ter Braak (1999).

Stable isotope values of $\delta^{15}N$ and $\delta^{13}C$ (SI) were expressed as:

$$SI = \left(\left(\frac{R_{sample}}{R_{standard}} \right) - 1 \right) 1000 \quad (3)$$

where R is the corresponding ratio of $^{15}N/^{14}N$ or $^{13}C/^{12}C$ related to standard values in atmospheric air (IAEA-N-1 and 2) or PeeDee Belemnite (PDB: USGS 24), respectively. Seabirds' $\delta^{15}N$ values were converted into trophic positions (TP) according to Fisk et al. (2001a):

$$TP = 3 + \frac{(\delta^{15}N - 10.1)}{3.8} \quad (4)$$

3. Results

3.1. Concentrations, patterns and bioaccumulation potential of chlorinated pesticides

HCB, β-HCH, oxychlordane, *trans*-nonachlor and *p*,*p*′-DDE were quantified in all seabird samples, whereas Mirex and *cis*-chlordane were below QL in a few samples (Table 1). Black guillemot was the species in which most of the CPs were quantified above the QL (Table 1). Lipid adjusted concentrations (lw) of the individual CPs ranged from 2.5 to 2900 ng g^{-1} lw, depending on species and CP (Table 1). The *p*,*p*′-DDE followed by HCB contributed most to $\sum$CP in all species (Fig. 1A). Within each pesticide group, β-HCH and *p*,*p*′-DDE were the only compounds above the QL of the HCHs and DDTs, respectively. Oxychlordane contributed most to $\sum$Chlordane, except in little auk in which *trans*-nonachlor dominated (Fig. 1B).
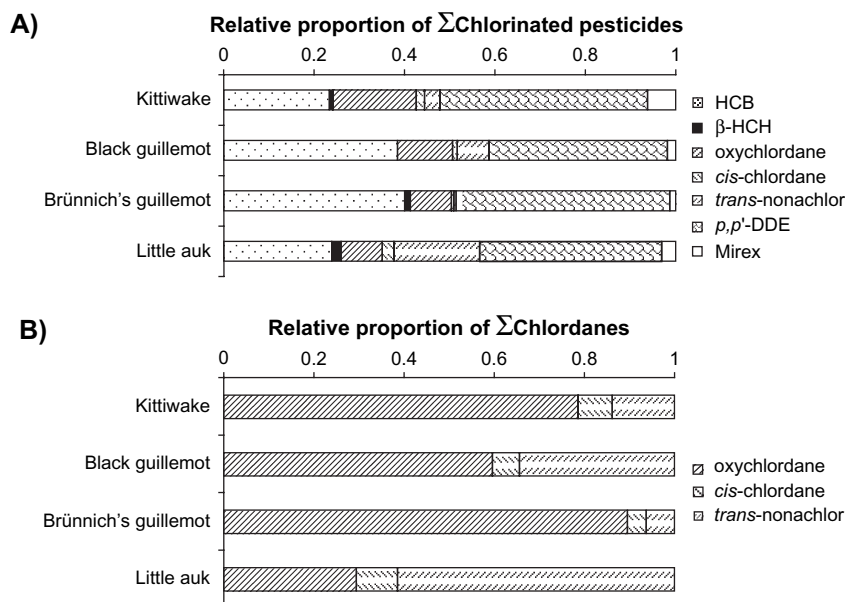

Fig. 1. Compositional pattern of chlorinated pesticides (CP) in Arctic seabirds. (A) Mean contribution of each CP to $\sum$CP. (B) The relative contribution of each chlordane compound to $\sum$Chlordanes.

Although there was high variation in CP concentrations within each species, as illustrated by the large spread of samples in the ordination diagram, the samples from each species were grouped (Fig. 2A). PC1 described 65% of the variation in CP concentrations among the samples due to high concentrations of β-HCH, *cis*-chlordane and *trans*-nonachlor in little auk compared to the other species. Kittiwake was separated from the auks along PC2, which accounted for 21% of the variation, due to high concentrations of oxychlordane, *p,p'*-DDE, HCB and Mirex compared to the other species. Brünnich's guillemot had lowest concentrations of all CP, except for HCB, compared to the other species (Fig. 2A). The CP pattern was relatively similar among the species, and the samples greatly overlapped along PC1, which accounted for 50% of the variation (Fig. 2B). HCB and *p,p'*-DDE were the CPs that were most responsible for the separation of samples along PC1 (Fig. 2B). These substances were also the CPs with highest contribution in all samples (Fig. 1A). Little auk was separated from the other seabirds along PC2, which accounted for 25% of variation among the samples, due to high relative contribution of β-HCH, *trans*-nonachlor and *cis*-chlordane (Fig. 2B). Although oxychlordane and Mirex separated the samples along PC2 due to higher absolute concentrations in kittiwake (Fig. 2A), they had low variation in standardized concentrations (pattern) among the samples. Therefore, oxychlordane and Mirex did not contribute to separation of the

samples in the ordination diagram of CP pattern (i.e. positioned close to the origin) (Fig. 2B).

The bioaccumulation potential of β-HCH, *cis*-chlordane and *trans*-nonachlor was low relative to PCB-153 for all species, whereas the other CPs varied more depending on species (Fig. 3). Brünnich's guillemot had bioaccumulation potential of HCB, oxychlordane and *p,p'*-DDE close to PCB-153, with values around 1. *p,p'*-DDE was close to 1 also in little auk, whereas the bioaccumulation potential of the other CPs in little auk was low compared to PCB-153. Both black guillemot and kittiwake had low bioaccumulation of *p,p'*-DDE compared to little auk and Brünnich's guillemot. Although *trans*-nonachlor had low bioaccumulation potential in all species compared to PCB-153, it was distinctly higher in little auk than the other species. The opposite was seen for oxychlordane, which had high bioaccumulation potential (>0.5) in all species except for little auk (0.25) (Fig. 3). Kittiwake had low bioaccumulation of all CPs compared to PCB-153, with highest value (0.5) for oxychlordane.

3.2. Relationship between CPs and dietary descriptors and enzymatic parameters

Sex was close to the origin in both the concentration and the pattern PCA, and was therefore not important for the CP bioaccumulation amongst the samples. As illustrated by the
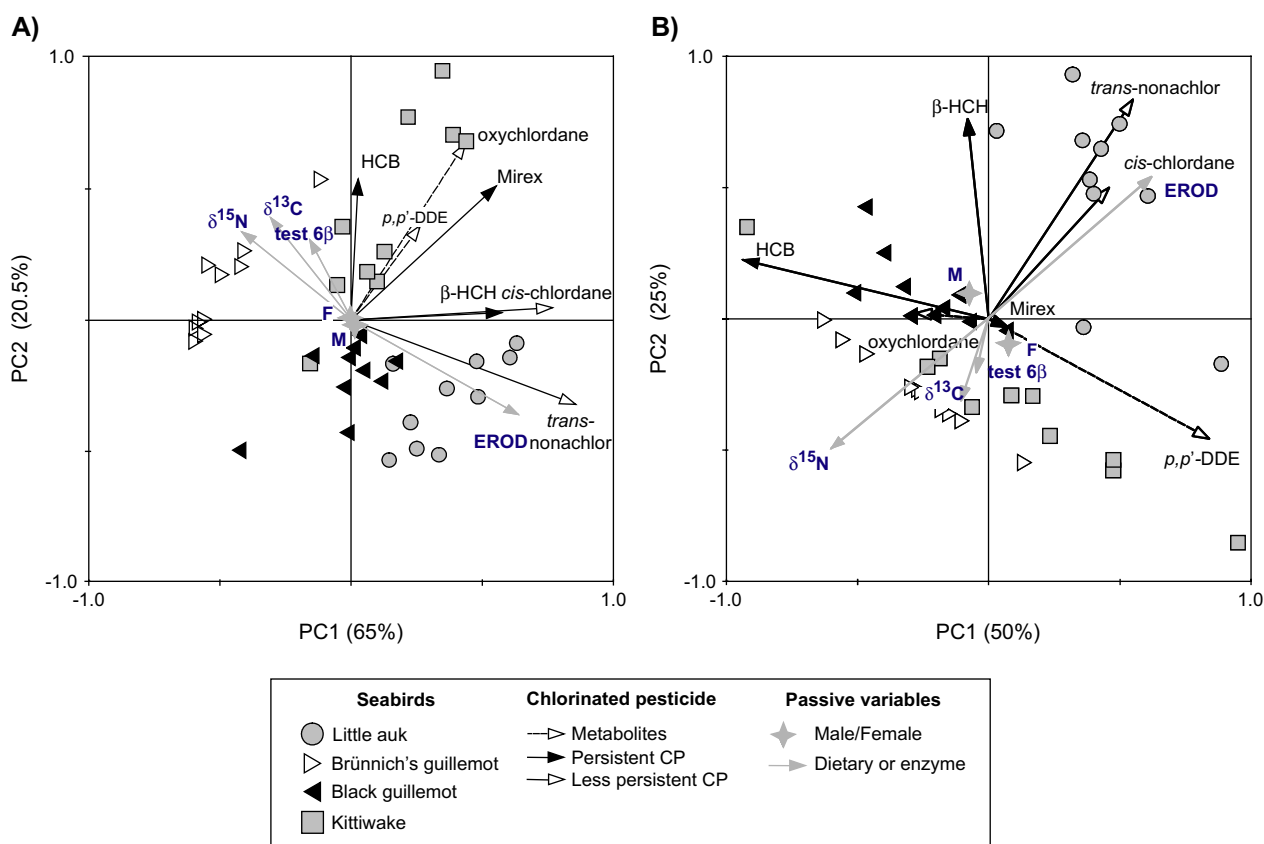

Fig. 2. Principal component analysis of (A) absolute concentrations (logarithmically transformed with lipid content as covariable, and (B) standardized concentrations (pattern) of chlorinated pesticides in four arctic seabirds. Arrows are the respective CPs pointing to the direction of increasing value, whereas symbols represent the individual samples. Dietary (δ^{13}C, δ^{15}N) and enzymatic variables (EROD, testosterone-6β-hydroxylation) in addition to sex (*M*ale/*F*emale) were included as passive variables (grey) to visualise their values in the seabirds.
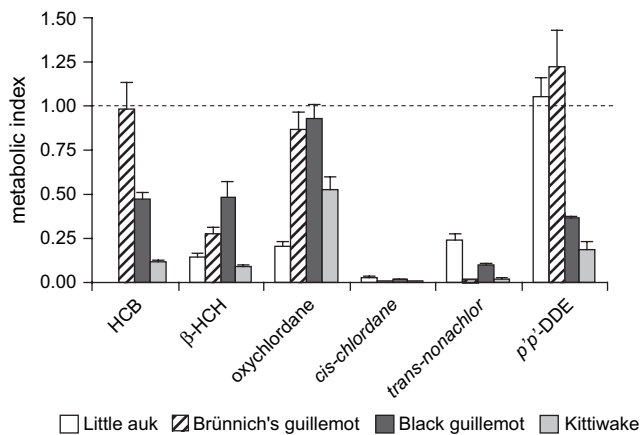
Fig. 3. Metabolic indices of chlorinated pesticides in Arctic seabirds from the Barents Sea. Compounds >1 are accumulated relative to the persistent PCB-153, whereas compounds <1 are eliminated. HCB was not quantified in little auk's main prey of calanoid copepods.

PCA, $\delta^{15}N$ and EROD are pointing in opposite directions, and are therefore negatively correlated (Fig. 2).

Thus, inclusion of both $\delta^{15}N$ and EROD in the ANOVA renders one of them non-significant for CPs where they have an influence, such as for β-HCH, HCB and *cis*-chlordane. For these CPs, the respective ANOVA was therefore run including only one of these parameters at the time. None of the lipid adjusted CP concentrations were related to the dietary parameters, except for HCB which increased with increasing $\delta^{15}N$, and *trans*-nonachlor and β-HCH which decreased with increasing $\delta^{13}C$ values (Table 3). The lipid adjusted concentrations of β-HCH, *cis*-chlordane, *trans*-nonachlor and Mirex increased with increasing EROD activity, whereas the other CP concentrations were not related to the CYP activities.

The relative contribution of β-HCH and *trans*-nonachlor to $\sum$CP decreased with increasing $\delta^{13}C$, whereas the relative contribution of HCB and oxychlordane to $\sum$CP increased, whereas β-HCH, *trans*-nonachlor and *cis*-chlordane decreased

with increasing $\delta^{15}N$ (Table 3). The relative contribution of *cis*-chlordane, *trans*-nonachlor, and β-HCH to $\sum$CP increased, whereas HCB decreased, with increasing EROD activity. None of the CP proportions or absolute concentrations were related to testosterone-6β-hydroxylation.

4. Discussion

Earlier studies have considered HCH and chlordane bioaccumulation from the same seabird species from the Canadian Arctic (Fisk et al., 2001a; Moisey et al., 2001), and PCBs from the same seabirds from the Barents Sea (Borgå et al., 2005). The present study focuses on differences in bioaccumulation of selected chlorinated pesticides in the Barents Sea seabirds. The aim was to confirm the earlier studies from another region, and to evaluate the biotransformation abilities among the various seabird species based on the accumulation pattern of CPs and similar information for PCBs. Of the analysed CPs, α-, β-, and γ-HCH, HCB, *cis*-chlordane, *trans*-nonachlor, Mirex, and *p,p′*-DDT, *p,p′*- and *o,p′*-DDD are parent compounds, whereas oxychlordane and *p,p′*-DDE are biotransformation products (Buser et al., 1992; de March et al., 1998). Although several factors influence CP bioaccumulation, the present study focuses on the influence of exposure from diet and elimination by biotransformation.

4.1. Relationships between CP accumulation, diet and enzyme activities

There were only few significant relationships between CP concentration, or pattern, and dietary parameters. Of the exceptions was HCB, which was the only CP which increased in concentration with $\delta^{15}N$. This weak relationship between trophic level and CP concentration has also been shown for chlordanes and HCHs in Canadian Arctic seabirds (Fisk et al., 2001a; Moisey et al., 2001), and for PCBs in central

Table 3

Relationship between concentrations and patterns of individual chlorinated pesticides and increasing values of stable isotopes and EROD based on ANOVA Type III SS[a]

	$\delta^{15}N$		$\delta^{13}C$		EROD	
Concentrations						
HCB	↑	$F = 10.4, p = 0.0026$				
β-HCH			↓	$F = 4.91, p = 0.0031$	↑	$F = 8.47, p = 0.0062$
Oxychlordane						
cis-Chlordane					↑	$F = 37.91, p < 0.0001$
trans-Nonachlor			↓	$F = 25.8, p < 0.0001$	↑	$F = 62.51, p < 0.0001$
p,p′-DDE						
Mirex					↑	$F = 6.51, p = 0.0150$
Pattern						
HCB	↑	$F = 11.9, p = 0.0014$			↓	$F = 20.2, p < 0.0001$
β-HCH	↓	$F = 7.4, p = 0.0098$	↓	$F = 10.0, p = 0.0031$	↑	$F = 4.7, p = 0.0365$
Oxychlordane	↑	$F = 4.53, p = 0.0399$				
cis-Chlordane	↓	$F = 10.67, p = 0.0023$			↑	$F = 22.2, p < 0.0001$
trans-Nonachlor	↓	$F = 39.5, p < 0.0001$	↓	$F = 44.0, p < 0.0001$	↑	$F = 17.8, p = 0.0015$
p,p′-DDE						
Mirex						

[a] Arrows indicate positive or negative relationships (i.e. directions in PCA plot, Fig. 2). Non-significant relationships are left blank.

Barents Sea seabirds (Borgå et al., 2005). It is probably related to the trophic position proximity of the seabirds. In contrast to the food web studies showing CP enrichment from trophic position 1.6 to 5.0 (e.g. Fisk et al., 2001b; Hop et al., 2002), the present study ranged over only 1.5 trophic positions, from 2.7 to 4.2.

Low HCB and high HCH concentrations in little auk compared to the other species correspond to the concentrations in its major prey, calanoid copepods. Surprisingly, most CP concentrations were high in little auk, which occupied the lowest trophic position. The CP levels were generally low in black guillemot, which had the highest trophic position. High CP concentrations in kittiwake are probably a combination of its high trophic position, higher field metabolic rate in gulls as compared to auks (Ellis and Gabrielsen, 2002), and migration to more contaminated over-wintering areas (Anker-Nilssen et al., 2000), as discussed for PCBs (Borgå et al., 2005).

The relative contribution of oxychlordane and HCB to $\sum$CP increased with increasing δ^{15}N, whereas β-HCH, *cis*-chlordane and *trans*-nonachlor decreased with increasing δ^{15}N. Increasing relative contribution of oxychlordane and HCB to $\sum$CP with trophic position corresponds with earlier findings, and reflects the biomagnification potential of these CPs (Fisk et al., 2001a). Likewise, decreasing *cis*-chlordane and *trans*-nonachlor contributions reflect lower biomagnification due to more rapid elimination. Other significant relationships between CPs and dietary descriptors were *trans*-nonachlor and β-HCH which decreased in concentrations and relative contribution to $\sum$CP with increasing δ^{13}C.

The concentrations of *trans*-nonachlor, *cis*-chlordane and β-HCH increased with EROD activity, mainly due to elevated EROD activity in little auk since it did not differ among the other species. Also the relative contribution of these compounds to $\sum$CPs decreased with δ^{15}N. As EROD activity and δ^{15}N are highly correlated (highest EROD in species with lowest trophic position) it is difficult to decipher whether diet or EROD activity is responsible for the present accumulation pattern. Also other species-specific factors correlated with these may be important. Nevertheless, the lack of relationship between testosterone-6β-hydroxylation and any of the CP concentrations or proportions indicates that enzymes through the CYP2B/3A system are not highly relevant for the CP accumulation. However, the opposite was suggested based on the chlordane pattern in northern Baffin Bay seabirds (Fisk et al., 2001a). Furthermore, the lack of relationship between CP concentration and δ^{15}N for all CPs, except for HCB, suggests that trophic position is not a major determinant for the difference in CP accumulation among the present seabirds.

4.2. Bioaccumulation potential

In contrast to the other species, little auk has a high relative contribution of *cis*-chlordane and *trans*-nonachlor, both of which can be metabolized to oxychlordane. Similarly, the bioaccumulation potential of oxychlordane was lower in little auk than the other species, whereas those of *cis*-chlordane and *trans*-nonachlor were higher. This implies poor biotransformation of *cis*-chlordane and *trans*-nonachlor to oxychlordane by little auk. However, for all seabirds the elimination is higher for chlordanes than for PCB-153, resulting in a low bioaccumulation potential (<0.25) for both *cis*-chlordane and *trans*-nonachlor.

The compositional pattern of CPs, dominated by HCB, *p,p'*-DDE and oxychlordane is similar to Brünnich's guillemot eggs from the Alaskan Arctic (Vander Pol et al., 2002). High relative proportions of CP metabolites (oxychlordane, *p,p'*-DDE) and persistent CPs (HCB), and a low relative proportion of less persistent CPs (*trans*-nonachlor, *cis*-chlordane), in Brünnich's guillemot compared to the other species is consistent with previous studies suggesting a higher biotransformation ability towards *trans*-nonachlor, *cis*-chlordane and HCHs in this species than for other auks (Fisk et al., 2001a; Moisey et al., 2001). The present study adds DDTs to this list. The similarity in CP pattern to seabirds from northern Baffin Bay illustrates that a species' metabolic ability is consistent across regions, and that the CP pattern depends more on species-specific and physiologically determined biotransformation abilities than on contaminant exposure. The findings also support that *om*-PCBs and CPs are metabolized by different enzyme groups, as Brünnich's guillemot poorly eliminated HCB and *om*-PCB compared to the other seabirds (Borgå et al., 2005), whereas *cis*-chlordane, *trans*-nonachlor, HCH and DDT are rather efficiently eliminated.

α-HCH is not considered persistent in seabirds, but is eliminated slower in auks than in gulls (Moisey et al., 2001). This is also reflected in the present study, as α-HCH was above QL only in black guillemot and some Brünnich's guillemots. Furthermore, *o,p'*-DDD and *p,p'*-DDD were quantified in low levels in black guillemot, but were below the QL in the other species, corresponding with the suggestion that black guillemot has a rather poor metabolic activity, based on their PCB pattern (Borgå et al., 2005).

Low bioaccumulation potential for all CPs in kittiwake compared to the auks illustrates its higher biotransformation and elimination ability compared to auks, as suggested for chlordanes, HCHs and PCBs (Fisk et al., 2001b; Moisey et al., 2001; Borgå et al., 2005). The high bioaccumulation potential of metabolites (oxychlordane and *p,p'*-DDE) in combination with low bioaccumulation potential, and a high degree of non-detection, of parent compounds is a strong indication of biotransformation of parent CPs and subsequent accumulation of persistent metabolites.

In summary, based on the quantified concentrations and interrelationships among CPs, little auk is classified as an efficient metabolizer of HCH and DDT compounds, but a rather poor metabolizer of chlordanes (Table 4). Brünnich's guillemot seems to efficiently metabolize HCHs, chlordanes and DDT isomers, but accumulate HCB at the same degree as the persistent PCB-153. Black guillemot seems to be a poor metabolizer of all CPs, except for chlordane. The high bioaccumulation potential of β-HCH suggests transformation of the other isomers into β-HCH, although other HCH isomers were also quantified in black and Brünnich's guillemot. Since the bioaccumulation potential is higher in black guillemot that in other seabirds, black guillemot probably has a slower HCH elimination than the

Table 4
The four arctic seabirds classified as Good or Poor in terms of elimination and/or biotransformation based on comparison to the other seabirds, and to the other organochlorines from the present study, given in Fisk et al. (2001a), Moisey et al. (2001) and Borgå et al. (2005)

Species	Little auk	Brünnich's guillemot	Black guillemot	Kittiwake
Chlordanes	Poor[a]	Good	Good	Good
HCHs	Good	Good	Poor	Good
HCB		Poor[b]		
DDTs	Good	Good	Poor	Good
om-PCBs	Good	Poor	Good	Good
mp-PCBs	Good	Good	Poor	Good

[a] Poor relative to the other seabirds, but still more efficient than PCB-153.
[b] High bioaccumulation potential when compared to PCB-153.

others. Also, the relative contribution of *trans*-nonachlor to $\sum$CP was high in black guillemot compared to kittiwake and Brünnich's guillemot. Kittiwake can be classified as efficient in biotransformation and elimination of all the CPs, both parent compounds and metabolites. The present study adds evidence that industrial PCBs and agricultural CPs are metabolized through different biotransformation pathways in avian wildlife, although the CP elimination is highly species-specific.

Acknowledgements

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