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The use of blue tit eggs as a biomonitoring tool for organohalogenated pollutants in the European environment

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ABSTRACT

In the present study, large scale geographical variation in the occurrence of organohalogenated pollutants (OHPs) was investigated throughout Europe using eggs of a terrestrial resident passerine species, the blue tit (*Cyanistes caeruleus*). Blue tit eggs from 10 sampling locations, involving suburban, rural and remote areas, in 7 European countries were collected and analysed. Sum polychlorinated biphenyl (PCB) levels ranged from 150 ng/g lipid weight (lw) to 2003 ng/g lw. Sum polybrominated diphenyl ethers (PBDEs) ranged from 3.95 ng/g lw to 114 ng/g lw. As expected, PCB and PBDE concentrations were significantly higher in the sampled suburban locations compared to the rural and remote locations. Sum organochlorine pesticides (OCPs) ranged from 122 ng/g lw to 775 ng/g lw. OCP concentrations were, against the expectations, found to be lower in the rural sampling locations compared to the other locations. Contamination profiles of PCBs, PBDEs and OCPs differed also among the sampling locations, which may be due to local contamination sources. Finally, we compared the results of this study with previously reported OHP concentrations in the eggs of a closely related species, the great tit (*Parus major*), from the same sampling locations in Europe. We found no differences in concentrations between the species. In addition, we found a significant, positive correlation between the sum PCB concentrations in blue tit eggs and great tit eggs, suggesting similar exposure pathways, mechanisms of accumulation and maternal transfer of PCBs. In conclusion, our results suggest the usefulness of eggs from passerine birds as a biomonitoring tool for OHPs on a large geographical scale.

1. Introduction

Organohalogenated pollutants (OHPs), such as polychlorinated biphenyls (PCBs), polybrominated diphenyl ethers (PBDEs) and organochlorine pesticides (OCPs), are lipophilic substances that are a great cause of concern because of their persistent and bioaccumulative nature. They are also known to cause adverse health effects in humans and wildlife (Jones and de Voogt, 1999). Furthermore, there is evidence for long-range transport of these substances to regions where they have never been used or produced (Scheringer, 2009). As a consequence these pollutants are now distributed worldwide and are even found in remote locations, such as the polar regions (Braune, 2007). Although many of these pollutants have been banned, high concentrations still exist in the environment. Therefore, monitoring studies are essential to assess the current levels and to evaluate health risks of different OHPs in the environment.

Bird eggs have been used successfully to monitor OHPs in numerous studies (Donaldson et al., 1999; Norstrom et al., 2002; Elliott et al., 2005; Jaspers et al., 2005; Van den Steen et al., 2006; 2008). Eggs of most bird

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species can easily be collected and the removal of a single egg from a clutch has only a minor effect at the population level (Furness, 1993; Henny and Kaiser, 1996; Henny et al., 2004). An important advantage of the use of birds is that they can be used both as indicators of exposure and to assess potential effects on condition, reproduction and survival (Yoccoz et al., 2009). Because eggs can readily be sampled from the same location each year, long-term monitoring studies using eggs are also feasible. Moreover, studies on widespread bird species enable monitoring on a large spatial scale (Van den Steen et al., 2009a). Large scale geographical studies are very valuable to obtain more information about the local usage, emission patterns and spatial distribution of OHPs. However, to date, few studies have monitored concentrations of OHPs in birds' eggs on a broad geographic scale. For example, double crested cormorant (*Phalacrocorax auritus*) and herring gull (*Larus argentatus*) eggs have been used to assess the spatial distribution of OHPs in the Great Lakes, which have a total surface of about 244,000 km^2 (Hebert et al., 1994; Ryckman et al., 1998). In a previous study, we used eggs of a terrestrial resident passerine species, the great tit (*Parus major*), as a biomonitoring tool for OHPs in the European environment (Van den Steen et al., 2009a). Many resident passerine bird species spend their entire adult life in relatively small home ranges, territories and foraging areas. They are therefore particularly useful for monitoring local contamination with OHPs, because, in contrast to migratory species, residues in eggs will mostly reflect local contamination (Moore, 1966; Dauwe et al., 2006).

The first aim of this study was to investigate concentrations and profiles of PCBs, PBDEs and OCPs in eggs of another terrestrial passerine species, the blue tit (*Cyanistes caeruleus*), from different sampling locations in Europe. Blue tits are small, insectivorous passerine birds that are widely distributed throughout most of Europe (Cramp and Perrins, 1993). Great and blue tits are primarily insectivorous, feeding mostly on beetles, spiders and larvae of butterflies and moths. Outside the breeding season, both species also feed on fruits and seeds. Great tits are found foraging on the ground, while blue tits are much more arboreal, frequently feeding high in the crowns (Cramp and Perrins, 1993). They have previously been used as a biomonitor for heavy metals (Eens et al., 1999; Dauwe et al., 2000; Eeva et al., 2009). Their ubiquity permits sampling of almost any wooded area in Europe. Because blue tits are cavity-nesting birds and nest sites are often a limiting resource, they will readily nest in man-made boxes. Therefore, breeding populations can be rapidly established and monitored, and samples can be easily collected. Moreover, blue tits are resident or non-migratory in many populations and they have small home ranges, making them useful to monitor local contamination. Our second aim was to investigate differences in pollutant concentrations and profiles among the different types of sampling locations, involving suburban, rural and remote locations. Based on previous studies (Jaward et al., 2004; Jaspers et al., 2009; Van den Steen et al., 2009a), we expected the highest concentrations of PCBs and PBDEs in suburban sampling locations and the highest concentrations of OCPs in the rural sampling locations. Finally, we compared the results of this study with previously reported OHP concentrations in great tit eggs from the same sampling locations in Europe (Van den Steen et al., 2009a) and evaluated the usefulness of both tit species for monitoring OHPs in terrestrial environments. Although great tits and blue tits are closely related, they differ in several life-history parameters such as body size, clutch size and metabolic rate and in foraging behaviour (Cramp and Perrins, 1993), which may affect accumulation and maternal transfer of OHPs to eggs.

2. Materials and methods

2.1. Sample collection

In the breeding season of 2006 (April–May), researchers from 7 European countries (Table 1, Fig. 1) collected blue tit eggs in 10 existing nest box populations. In all populations, one random egg per clutch (3–10 eggs per sampling site, see Table 1) was collected. All eggs were collected before incubation had started, labeled individually and stored in a freezer (−20 °C) until transport to the laboratory. Eggs were transported on dry ice and stored frozen until analysis. In total, 65 blue tit eggs were analysed for PCBs, PBDEs and OCPs. A questionnaire was sent to the collectors in order to characterise the sampling sites and potential contamination sources of OHPs. Sampling sites were located in suburban, rural or remote areas (Table 1, Fig. 1). Suburban sampling locations were closely located to a city or densely populated area. Rural sampling locations were characterised by agricultural activities (e.g. crop cultivation, fruit trees). Remote locations were not in the vicinity of a city, industrial sites or agricultural activities.

Table 1
Sampling locations of blue tit eggs together with the type, number of analysed eggs, habitat type and the presence of industrial and/or agricultural activities.

	Country	Type	Number of analysed eggs	Habitat type	Industrial and/or agricultural activities
D	Germany	Rural	10	Mixed forest	Agricultural activities
E1	Spain	Suburban	7	Natural forest close to Barcelona	Intensive industrial activities
E2	Spain	Remote	7	Deciduous forest and pine plantations	–
F	France	Suburban	6	Forest and garden near to Toulouse	Intensive agriculture
FIN1	Finland	Rural	7	Pine dominated mixed forests	Copper smelter and related industry (heavy metals and sulphuric oxides)
FIN2	Finland	Suburban	5	Mixed forest near Oulu	Chemical industry, wood and paper industry, agriculture
I1	Italy	Suburban	7	Oak wood and small forest patches close to Rome	Intensive agriculture
I2	Italy (Sicily)	Remote	3	Deciduous forest	Extensive agriculture
N	Norway	Suburban	7	Woodland plots close to Oslo	–
P	Portugal	Suburban	7	Mixed forest	Agricultural activities

2.2. Contaminant analysis

Further sample treatment and analysis were performed according to previously described methods (Jaspers et al., 2005; Van den Steen et al., 2006). A homogenised sample of approximately 0.5 g whole egg content was weighed, mixed with anhydrous Na_2SO_4 and spiked with internal standards (ε-HCH, CB 46 and 143, BDE 77 and 128). Extraction was carried out with 100 ml hexane/acetone (3:1, *v*/*v*) in an automated Soxhlet extractor (Büchi, Flawil, Switzerland) in hot extraction mode for 2 h. The lipid content was determined gravimetrically on an aliquot of the extract (105 °C, 1 h), while the rest of the extract was cleaned on a column filled with ~8 g acidified silica and eluted with 15 ml hexane and 10 ml dichloromethane. The eluate was concentrated to 100 µl under a gentle nitrogen stream and transferred to an injection vial. In all samples, concentrations of 22 PCB congeners (CB 28, 31, 74, 95, 99, 101, 105, 110, 118, 128, 138, 149, 153, 156, 163, 170, 180, 183, 187, 194, 196 and 199), 7 PBDE congeners (BDE 47, 49, 99, 100, 153, 154 and 183), dichlorodiphenyltrichloroethane (*p*,*p*′- and *o*,*p*′-DDT) and metabolites (*p*,*p*′-DDE and *p*,*p*′-DDD), hexachlorocyclohexanes (HCHs; α-, β- and γ-HCHs), chlordanes (CHLs; *cis*-chlordane (CC), *trans*-chlordane (TC), *trans*-nonachlor (TN) and oxychlordane (OxC)), and hexachlorobenzene (HCB) were determined.

For the PCB analysis, an Agilent 6890 gas chromatograph (GC) connected to an Agilent 5973 mass spectrometer (MS) operated in electron

Fig. 1. Map of Europe with sampling locations. Different types of sampling locatuibs have different symbols: suburban (●), rural (*) and remote (+). Scale 1:25,000.

ionisation (EI) mode was equipped with a 25 m × 0.22 mm × 0.25 μm HT-8 capillary column (SGE, Zulte, Belgium). The ion source, quadrupole and interface temperatures were set at 230, 150 and 300 °C, respectively. The MS was used in the selected ion-monitoring (SIM) mode with two ions monitored for each PCB homologue group.

For the analysis of the OCPs and PBDEs, an Agilent 6890 GC connected to an Agilent 5973 MS operated in electron capture negative ionisation (ECNI) mode was equipped with a 25 m × 0.22 mm × 0.25 μm HT-8 capillary column (SGE, Zulte, Belgium). Methane was used as moderating gas and the ion source, quadrupole and interface temperatures were set at 160, 150 and 300 °C, respectively. The MS was used in the SIM mode with two ions monitored for each pesticide in specific windows, while ions $m/z = 79$ and 81 were monitored for PBDEs during the entire run.

Multi-level calibration curves in the linear response interval of the detector were created for the quantification, and good correlation ($r^2 > 0.999$) was achieved. The identification of OHPs was based on the relative retention times to the internal standard used for quantification, ion chromatograms and intensity ratios of the monitored ions. A deviation of the ion intensity ratios within 20% of the mean values obtained for calibration standards was considered acceptable. The quality control was performed by regular analyses of procedural blanks, by random injection of standards and solvent blanks. A standard reference material SRM 1945 (PCBs, PBDEs and OCPs in whale blubber) was used to test the method accuracy. Determined concentrations were within 10% of the certified values. The quality control scheme is also assessed through regular participation in interlaboratory comparison exercises organized by the Arctic Monitoring and Assessment Programme (AMAP) and the National Institute of Standards and Technology (NIST). For each analyte, the mean procedural blank value was used for subtraction. BDE 47 and 99 had blank levels which were lower than 5% of the values found in the samples. Nevertheless, the blank levels were subtracted from the sample values. After blank subtraction, the limit of quantification (LOQ) was set at 3 times the standard deviation of the procedural blank. For analytes that were not detected in procedural blanks, LOQs were calculated for a signal-to-noise ratio equal to 10. LOQs for the analysed compounds ranged between 0.5 and 4.0 ng/g lipid weight (lw).

2.3. Statistical analysis

Statistical calculations were performed using Statistica for Windows (Statsoft, 1997). The level of significance was set at $\alpha = 0.05$ throughout this study. The data were normally distributed (Kolmogorov–Smirnov test: $p > 0.05$ for all cases) and therefore parametric tests were used. One-way ANOVAs were used to test for differences in contamination levels among the sampling locations and to investigate whether there was a difference in contamination levels among the suburban, rural and remote sampling locations. Post hoc tests (Tukey HSD) were performed if there were significant differences among the sampling locations. Pearson correlations were performed to test whether PCB, PBDE and OCP concentrations were intercorrelated and to test whether a correlation existed between the mean egg concentrations per sampling location in blue tits and great tits. Differences in concentrations between blue tit and great tit eggs were investigated using unpaired *t*-tests. After standardization, we conducted a principal component analysis (PCA) to compare the congener profiles among the sampling sites. Principal components (PCs) with eigenvalues above 1 were considered to account for a significant contribution to the total variance according to the latent root criterion (Hair et al., 1998). Factor loadings and factor scores were determined and used to interpret PC patterns. Compounds with factor loadings greater than 0.65 on any PC were considered significant. The first two PCs were used for the statistical analyses.

3. Results

3.1. Egg concentrations of PCBs, PBDEs and OCPs

Sum PCB concentrations ranged from 150 ± 13 ng/g lw in E2 (Spain) to 2003 ± 312 ng/g lw in E1 (Spain; Fig. 2a). Sum PCB concentrations differed significantly among the sampling locations (One-way ANOVA: $F_{9,55} = 4.86$, $p < 0.001$). Concentrations in eggs from E1 were significantly higher compared to D, FIN2, FIN1, P, I2 and E2 (Tukey HSD: $p < 0.04$; Fig. 2a). No other significant differences were found among the sampling locations (Tukey HSD: $p > 0.13$).

Sum PBDEs concentrations ranged from 3.95 ± 1.70 ng/g lw in E2 (Spain) to 114 ± 40 ng/g lw in P (Portugal; Fig. 2b). Sum PBDE concentrations differed significantly among the sampling locations (One-way ANOVA: $F_{9,55} = 3.14$, $p = 0.004$). Concentrations in eggs from E1 and P were significantly higher than E2 (Tukey HSD: $p < 0.04$; Fig. 2b). No other significant differences were found among the sampling locations (Tukey HSD: $p > 0.05$).

Sum OCP concentrations ranged from 122 ± 9.11 ng/g lw in FIN1 (Finland) to 775 ± 190 ng/g lw in F (France; Fig. 2c). Sum OCP concentrations differed significantly among the sampling locations (One-way ANOVA: $F_{9,55} = 5.03$, $p < 0.001$). Sum OCP concentrations in eggs of *F* were significantly higher than most of the other sampling locations (Tukey HSD: $p < 0.02$; Fig. 2c), except E2 (Tukey HSD: $p = 0.08$) and I2 (Tukey HSD: $p = 0.29$). No other significant differences were found among the sampling locations (Tukey HSD: $p > 0.08$).

Sum PCB and sum PBDE concentrations in eggs from suburban sampling locations were significantly higher compared to those in eggs from remote and rural locations (Sum PCBs: One-way ANOVA: $F_{2,62} = 7.23$, $p = 0.002$; Tukey HSD: $p < 0.04$; Sum PBDEs: One-way ANOVA: $F_{2,62} = 8.27$, $p = 0.0007$; Tukey HSD: $p < 0.006$). Sum OCP concentrations in eggs from rural sampling locations were significantly smaller compared to those from remote and suburban locations (One-way ANOVA: $F_{2,62} = 4.40$, $p = 0.02$; Tukey HSD: $p = 0.03$).

The sum PCB and sum PBDE concentrations were positively correlated, albeit not quite significant (Pearson correlation: $n = 10$, $r = 0.58$, $p = 0.08$). Sum OCPs were not correlated with either the sum PCBs (Pearson correlation: $n = 10$, $r = 0.16$, $p = 0.66$) or the sum PBDEs (Pearson correlation: $n = 10$, $r = -0.20$, $p = 0.58$).

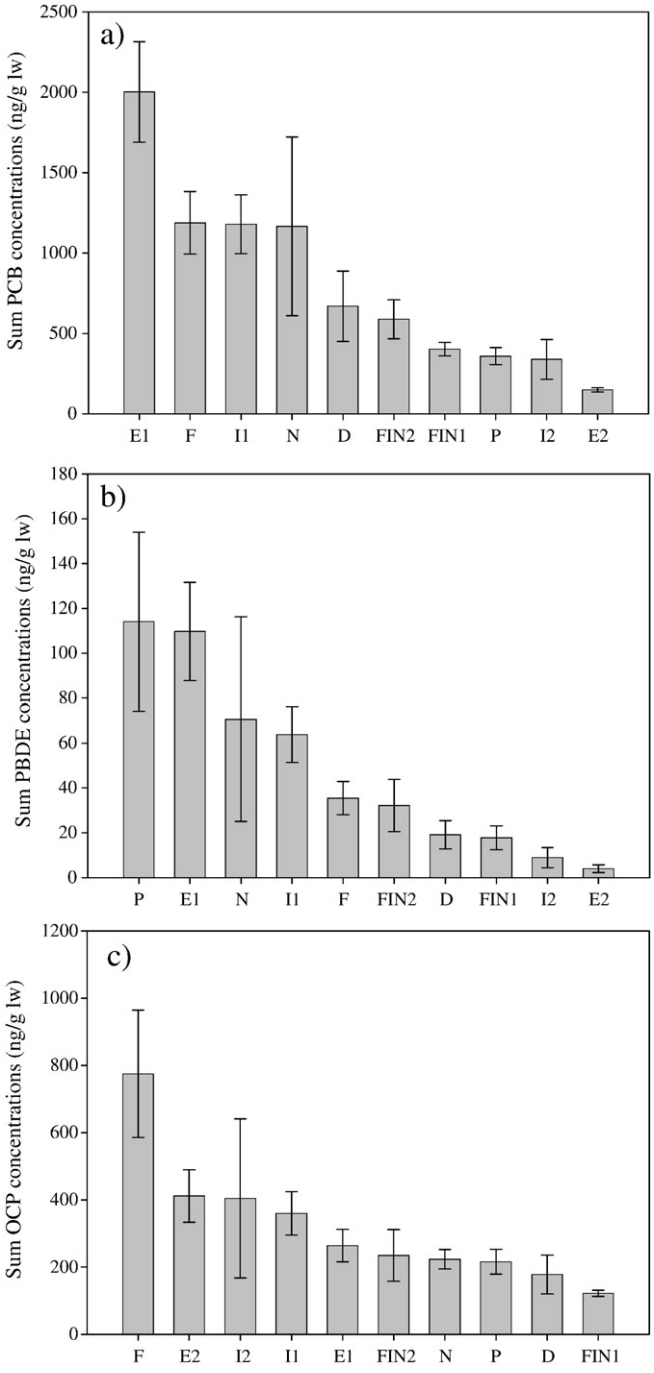

Fig. 2. Concentrations of (a) sum PCBs (b) sum PBDEs and (c) sum OCPs with standard errors: concentrations are expressed per gram lipid weight (lw). See Table 1 for sampling locations.

3.2. Profiles of PCBs, PBDEs and OCPs

CB 153, CB 180 and CB 138 were the most abundant PCB congeners and accounted for 24%, 19% and 11% of the sum PCBs, respectively. PCA revealed two PCs which accounted for 26% and 17% of the total variance, respectively (Fig. 3a). Significant differences among the sampling locations were found for both PC1 and PC2 (One-way ANOVAs: PC1: $F_{9,55} = 14.65$, $p < 0.001$; PC2: $F_{9,55} = 5.58$, $p < 0.001$). PC1 was positively correlated with CB 18, CB 52, CB 49, CB 44 and CB 209, and negatively with CB 118. PC2 was positively correlated with CB 199 and negatively with CB 99. PC1 of E2 differed significantly from all other sampling locations (Tukey HSD: $p < 0.001$; Fig. 3a), except I2

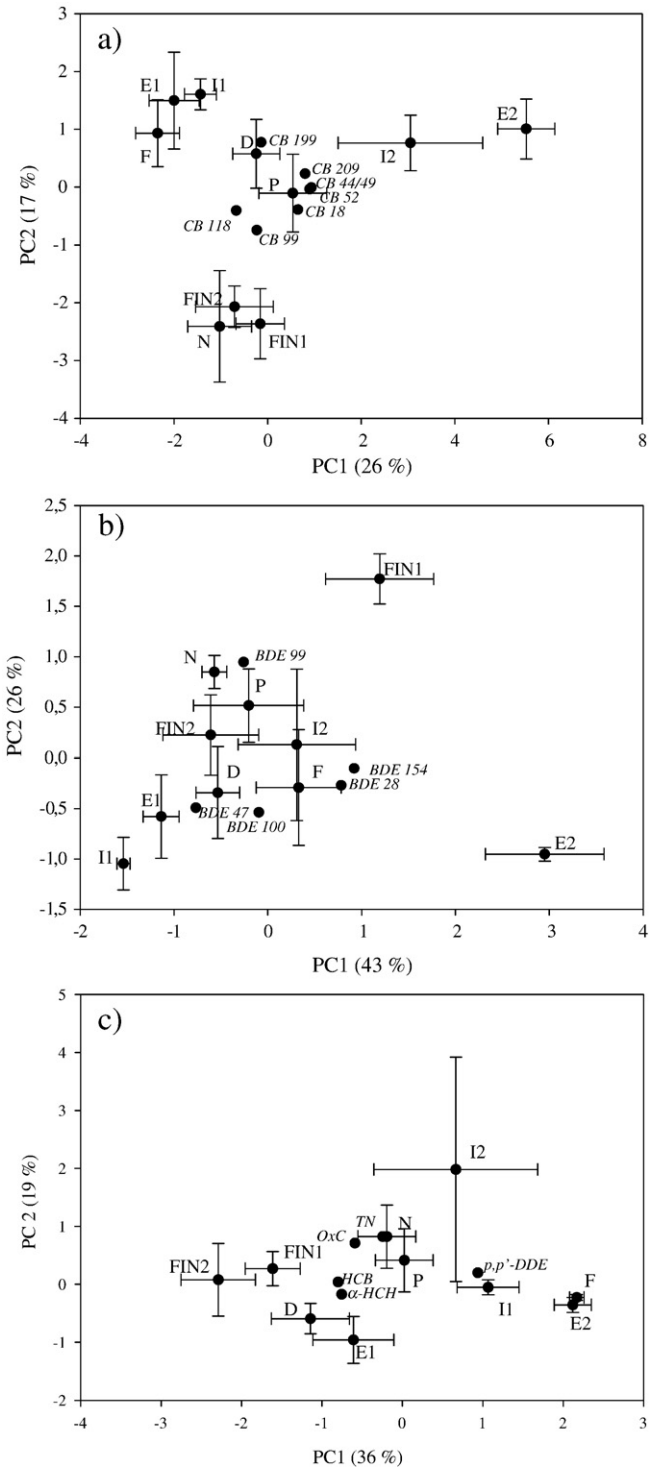

Fig. 3. Plots of factor scores with standard errors and factor loadings from the Principal Component Analysis (PCA) for the (a) sum PCBs, (b) sum PBDEs and (c) sum OCPs. Compounds with factor loadings greater than 0.65 on any PC were considered significant.

(Tukey HSD: $p=0.43$). PC1 of I2 differed significantly from E1, F, I1 and N (Tukey HSD: $p<0.01$; Fig. 3a). PC2 of FIN1 and N was significantly lower compared to D, E1, E2, F and I1 (Tukey HSD: $p<0.03$; Fig. 3a). PC2 of FIN2 was significantly different from E1 and I1 (Tukey HSD: $p<0.05$; Fig. 3a). No other significant differences were found among the sampling locations (Tukey HSD: $p>0.05$).

BDE 99, BDE 47 and BDE 100 were the most abundant PBDE congeners and accounted for 34%, 31% and 16% of the sum PBDEs, respectively. PCA revealed two PCs which accounted for 43% and 26% of the total variance, respectively (Fig. 3b). There were significant differences among the sampling locations for both PC1 and PC2 (One-way ANOVAs: PC1: $F_{9,55}=9.93$, $p<0.001$; PC2: $F_{9,55}=5.45$, $p<0.001$). PC1 was positively correlated with BDE 28 and BDE 154, while it was negatively correlated with BDE 47. PC2 was positively correlated with BDE 99. PC1 of E2 differed significantly from the other sampling locations (Tukey HSD: $p<0.03$; Fig. 3b), except from FIN1 (Tukey HSD: $p=0.09$). PC1 of FIN1 was significantly higher than E1 and I1 (Tukey HSD: $p<0.006$; Fig. 3b). PC2 of FIN1 was significantly higher compared to D, E1, E2, F and I1 (Tukey HSD: $p<0.01$; Fig. 3b). PC2 of N was significantly higher than E2 and I1 (Tukey HSD: $p<0.03$; Fig. 3b). No other significant differences were found among the sampling locations (Tukey HSD: $p>0.05$).

p,p′-DDE was the most abundant OCP congener and accounted for 85% of the sum OCPs. PCA revealed two PCs which accounted for 36% and 19% of the total variance, respectively (Fig. 3c). There were significant differences among the sampling locations for both PC1 and PC2 (One-way ANOVAs: PC1: $F_{9,55}=11.63$, $p<0.001$; PC2: $F_{9,55}=2.56$, $p=0.02$). PC1 was positively correlated with *p,p′*-DDE, while it was negatively correlated with α-HCH and HCB. PC2 was positively correlated with OxC and TN. PC1 of both E2 and F were significantly higher compared to D, E1, FIN1, FIN2, N and P (Tukey HSD: $p<0.03$; Fig. 3c). PC1 of I1 was significantly higher compared to D, FIN1 and FIN2 (Tukey HSD: $p<0.005$; Fig. 3c). PC2 of I2 was significantly higher than D and E1 (Tukey HSD: $p<0.03$; Fig. 3c). No other significant differences were found among the sampling locations (Tukey HSD: $p>0.05$).

3.3. Comparison between blue tit and great tit

There were no differences in mean sum PCB, sum PBDE and sum OCP concentrations between the blue tit and great tit eggs from the same sampling sites (Paired *t*-tests: sum PCBs: $n=10$, $t=0.007$, $p=0.99$; sum PBDEs: $n=10$, $t=0.72$, $p=0.49$; sum OCPs: $n=10$, $t=1.31$, $p=0.22$).

Mean sum PCB concentrations in blue tit and great tit eggs from the same sampling sites were strongly positively correlated (Pearson correlation: $n=10$, $r=0.86$, $p=0.001$; Fig. 4), whereas sum PBDE and OCP concentrations were not correlated between the two species (Pearson correlation: sum PBDEs: $n=10$, $r=-0.26$, $p=0.47$; sum OCPs: $n=10$, $r=0.51$, $p=0.14$).

4. Discussion

4.1. Concentrations of PCBs, PBDEs and OCPs

For PCBs, PBDEs and OCPs, concentrations differed significantly among the sampling locations. Sum PCB and sum PBDE concentrations were significantly higher in eggs from the suburban sampling locations compared to the remote and rural locations. PCBs and PBDEs have previously been linked to industrialization and urbanization (Lovett et al., 1998; Jaward et al., 2004; Van den Steen et al., 2008, 2009a). Both the lowest and highest concentrations of sum PCBs were found in Spain (E2 and E1, respectively). Similar to the PCBs, lowest concentrations of sum PBDEs were also found in the remote location E2 in Spain. Highest PBDE concentrations were found in P (Portugal) and E1 (Spain). Heavy industrial activities were reported in E1 (Table 1, personal communication J.C. Senar) which is located near Barcelona (ca. 2,000,000 inhabitants), but not for P, also a suburban sampling location (Table 1, personal communication A.C. Norte). The higher range concentrations of PCBs and PBDEs in the present study are similar compared to a previous study in blue tits from a suburban sampling location in Belgium (maximum sum PCBs: 2282 ng/g lw; maximum sum PBDEs: 77 ng/g lw; Van den Steen et al., 2009b).

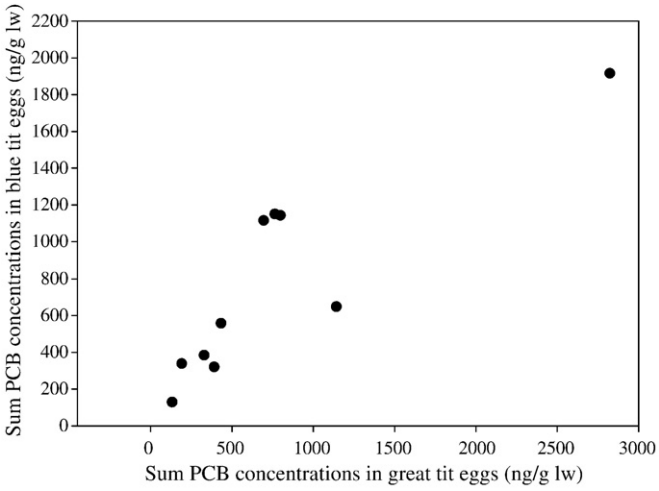

Fig. 4. Correlation between mean sum PCB concentrations in great tit and blue tit eggs from 10 sampling locations in Europe.

In contrast to our expectations, OCP concentrations were significantly lower in the rural sampling locations compared to the suburban and remote locations. This is in contradiction with previous studies in which great tit eggs were used as a biomonitoring tool for OHPs in the European environment (Van den Steen et al., 2008, 2009a). Studies using air samplers have also reported higher levels of OCPs in rural locations (Jaward et al., 2004; Harner et al., 2004). This discrepancy may be due to the low number of rural sampling locations ($n=2$) in the present study. The highest OCP concentrations were found in the sampling locations in France (F) which has been considered to be a suburban location. However, the location has also been characterized by the presence of intensive agriculture (Table 1, personal communication P. Heeb). This shows the importance of the definition and classification of the type of sampling location, which is inevitably somewhat subjective and not always straightforward. In addition, potential contamination sources, which can influence the results, may be overlooked.

4.2. Profiles of PCBs, PBDEs and OCPs

CB 153, CB 180 and C138 were the most abundant PCB congeners. A previous study, in which the maternal transfer of OHPs in blue tits was investigated, showed a similar congener profile in the eggs (Van den Steen et al., 2009b). However, significant differences in congener profile were found among the sampling locations. A higher contribution of CB 118 was found in F, E1, I1 and N. This profile is probably due to contamination with PCB mixture Aroclor 1254 which contains high concentrations of CB 118 (Frame et al., 1996). In addition, local contamination sources may also be responsible for the different profiles among the sampling locations (Ormerod et al., 2000).

Similar to a previous study on blue tits eggs (Van den Steen et al., 2009b), BDE 99, BDE 47 and BDE 100 were the most important PBDE congeners. BDE 47 and BDE 99 are the major congeners in the Penta-BDE commercial mixture (WHO, 1994). Although the Penta-BDE mixture has been withdrawn from the market in Europe in 2004 (Directive EEC, 2003), the congeners present in this mixture are still ubiquitous in the environment (Law et al., 2006). Moreover, the presence of BDE 47 and BDE 99 may also be the result of debromination of higher brominated congeners in the environment or in biota (Soderström et al., 2004; Van den Steen et al., 2007). Except for E2 (Spain) and FIN 1 (Finland), PBDE profiles did not differ much among the sampling sites, which is probably due to the fact that contamination with PBDEs is more widespread and originates from diverse sources (Siddiqi et al., 2003).

p,p′-DDE was the most abundant OCP and accounted for 85% of the sum OCPs, which is also in accordance with a previous study in blue tits (Van den Steen et al., 2009b). Since DDE is the major breakdown product of DDT, the accumulation profile of DDTs suggests a historical input rather than contribution from recent sources. DDT and its metabolites can still be found in the environment and in biota, although it has been banned in Europe for more than 25 years. A higher contribution of the highly bioaccumulative *p,p′*-DDE was observed in E2, F and I1, while γ-HCH and HCB were less abundant at these sites. The local historical usage and specific applications of different OCPs may be responsible for the different profiles among the sampling locations.

4.3. Comparison between blue tit and great tit

Both bleu tits and great tits have been suggested to be useful as biomonitors of local contamination with OHPs (Van den Steen et al., 2009a,b), because of their small home ranges, territories and foraging areas (Moore, 1966; Dauwe et al., 2006). Although blue tits and great tits do not migrate long distances, they often move between habitats from summer to winter (Ulfstrand, 1976), which can have an effect on contamination levels. Despite clear behavioural and life-history differences between the two tit species (Cramp and Perrins, 1993), no differences in overall concentrations were found between both species. In addition, a significant, positive correlation was found between the sum PCB concentrations in blue tit eggs and great tit eggs, suggesting similar exposure pathways, mechanisms of accumulation and maternal transfer. For the OCPs and PBDEs, no correlations were found between the concentrations in blue tit and great tit eggs. Differences in diet, accumulation and/or degradation between the two species may be responsible for this lack of correlation. It has already been shown that both species can be useful as a biomonitoring tool for OHPs (Van den Steen et al., 2006, 2008, 2009a,b,c). However, OHP concentrations in eggs decreased with laying order in both blue tits and great tits (Van den Steen et al., 2009b,c). Despite the observed laying order effects, the variance in concentrations was larger among clutches than within clutches for both species (Van den Steen et al., 2009a,b). When both species are present in a sampling location, we suggest using the eggs of great tits for biomonitoring OHPs for the following reasons (Hollamby et al., 2006). First of all, great tits are about 20% larger than blue tits (Cramp and Perrins, 1993). They also have a higher life expectancy compared to blue tits (Cramp and Perrins, 1993). Great tits have, therefore, the potential to accumulate more pollutants in comparison to blue tits. Larger sample volumes and sizes can be obtained, because great tits have larger eggs and are more abundant than blue tits (Cramp and Perrins, 1993). Although both tit species are resident, female great tits showed higher site fidelity compared to female blue tits (Könczey et al., 1997). Therefore, great tits are expected to reflect local contamination sources even better than blue tits.

In conclusion, our results suggest that eggs of blue tits, just like those of great tits, are useful as a biomonitoring tool of OHPs on a large geographical scale. However, when the eggs of both species are available in a sampling location, great tits are preferred because of several characteristics which are favourable for monitoring purposes.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.scitotenv.2009.12.028.

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