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Evidence for complex sources of persistent halogenated compounds in birds from the south China sea

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ABSTRACT

Persistent halogenated compounds (PHCs), including dichlorodiphenyltrichloroethane and its metabolites (DDTs), polybrominated diphenyl ethers (PBDEs), alternative brominated flame retardants (ABFRs), and dechlorane plus (DP), were analyzed in muscle of six bird species from the South China Sea. DDTs, with concentrations up to 19,000 ng/g lipid weight (lw), were the dominant contaminants contributing to 66–99% of PHCs in birds. Concentrations of PBDEs, ABFRs, and DP ranged from 1.1 to 130, 0.73–40, and 0.21–2.5 ng/g lw, respectively. Historically pollution of DDTs and flame retardants in surrounding Asian lands were the main sources for PHCs in birds. BDE 209 was the primary PBDE congener in all birds. 1,2-bis(2,4,6-tribromophenoxy) ethane (BTBPE) and decabromodiphenyl ethane (DBDPE) were the main ABFRs. *Anti*-DP and *p,p′*-DDE were the dominating compounds of DP and DDTs, respectively. Only concentrations of BDEs 153, 203, 196, and 207, *p,p′*-DDE, and *p,p′*-DDD showed significant and positive correlations with $\delta^{15}N$ values in samples. The resident birds, red-footed booby (*Sula sula*), had much lower levels of *p,p′*-DDE and most of PBDEs than those in migratory birds from the South China Sea. Results of stable isotope ratios of carbon suggest the highly variable food items for the five migratory bird species. The abundance of DBDPE in red-footed booby might be related with the ingestion of plastic debris, which still warrants further verification.

1. Introduction

Persistent halogenated compounds (PHCs), such as dichlorodiphenyltrichloroethane and its metabolites (DDTs), polybrominated diphenyl ethers (PBDEs), and dechlorane plus (DP) have been of international concern for several decades due to their persistence, bioaccumulation, toxicity and long-range transport (Batt et al., 2017; Sun et al., 2018). DDT was widely used as an insecticide during 1950s–1980s and has been banned since 1983 in China. Approximately 400,000 tons of DDT was used in China, contributing 20% to the total global production (Chen et al., 2006). PBDEs have been extensively used as non-reactive additive flame retardants in plastics, textiles, paints, furniture, electronics, automobiles, and construction materials (Cordner et al., 2013; Mo et al., 2018). Toxicological studies have suggested that PBDEs may impact thyroid, liver, and kidney morphology, fetal toxicity/teratogenicity, reproductive success, neurodevelopment, thyroid hormone levels, and liver ethoxyresorufin-O-deethylase (EROD) activity (Chen and Hale, 2010). Due to mounting environmental concerns, Penta- and Octa-BDEs have been globally phased out between 2004 and 2010, and Deca-BDE was also restricted in Europe in 2008 (Gentes et al., 2012). In order to meet flammability standards as defined by fire safety codes, many alternative brominated flame retardants (ABFRs), including pentabromotoluene (PBT), hexabromobenzene (HBB), 1,2-bis(2,4,6-tribromophenoxy) ethane (BTBPE), and decabromodiphenyl ethane (DBDPE), are being produced and used in consumer products. Several current-use ABFRs also showed bioaccumulation and biomagnification behaviors and undertook long-range atmospheric transport, which lead to ubiquitously occurrence of

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ABFRs in the environment (Wu et al., 2010; Gentes et al., 2012). DP, commercialized in the 1960s as a substitute for the pesticide Mirex, has also been suggested as a possible replacement product for Deca-BDE (Hoh et al., 2006). DP is now classified as a high production volume chemical in the United States (Gentes et al., 2012).

Birds have long been used as sentinel species to monitor spatiotemporal trends of PHCs in the environments, because they occupy high trophic positions in food webs and accumulate contaminants through food chains, making them especially sensitive to environmental contamination (Jaspers et al., 2006; Sakellarides et al., 2006; Voorspoels et al., 2006; Chen and Hale, 2010; Abbasi et al., 2016). Numerous studies have been conducted on contamination of PHCs in diverse bird species from different regions. Many factors, including dietary exposure, metabolic capability, migration pattern, age, sex, and nutritional state, can influence the levels of organic contaminants in birds. For example, concentrations of PBDEs in common kingfishers (*Alcedo atthis*) from e-waste recycling sites were 100- to 1000- folds greater than areas without e-waste recycling activities (Peng et al., 2019). Levels of PBDEs and DBDPE in light-vented bulbul (*Pycnonotus sinensis*) from urban regions were higher than those from rural areas (Sun et al., 2012). In passerine birds from three metropolises in China, DDTs dominated in Wuhan city whereas flame retardants dominated in Guangzhou and Beijing cities (Yu et al., 2014). Contamination profiles of PHCs in birds were valuable indicators reflective of local contamination in different regions.

South China Sea, which has an average depth of about 1200 m and an area of around 3.5 million km^2, is a marginal sea in Southeast Asia and is surrounded by many developing countries, including China, Philippines, Indonesia, Malaysia, Vietnam and Brunei (Zhang et al., 2007; Li et al., 2012). South China Sea is also surrounded by "hot spots" of PHCs. DDTs were historically used in many developing countries in Southeast Asia for agricultural and public health purpose (Chaiyarat et al., 2015; Ghani et al., 2017; Trinh Thi et al., 2019). Due to extensive e-waste recycling activities, these countries were considered to disposal 20–50 million tons of e-waste annually, accounting for about 70% of global production (Sun et al., 2014a). Intensive e-waste recycling processes can accelerate the release of large quantities of persistent and toxic chemicals, for instance, polychlorinated biphenyls (PCBs) and PBDEs, into the environment (Luo et al., 2009). PHCs in these regions may enter the South China Sea via rain wash, urban and agricultural runoff, and atmospheric deposition, then further accumulate in marine biota through food webs, posing long-term adverse impacts on marine ecosystems (Sun et al., 2014a). To date, only few investigations have reported the contamination status of PHCs in organisms, especially species with high trophic levels, from the South China Sea (Liu et al., 2011; Sun et al., 2017). To the best of our knowledge, there has been no report on PHCs in birds from the South China Sea yet.

In the present study, six bird species were collected from islands located in the South China Sea. We analyzed a series of PHCs (DDTs, PBDEs, ABFRs, and DP). The objectives were to gain knowledge about the levels and composition profiles of these PHCs in birds, and to find some evidence for PHC sources in birds from the South China Sea.

2. Materials and methods

2.1. Sample collection

A total of six bird species including red-footed booby (*Sula sula*, $n = 5$), Chinese pond heron (*Ardeola bacchus*, $n = 3$), intermediate egret (*Ardea intermedia*, $n = 3$), oriental pratincole (*Glareola maldivarum*, $n = 3$), pacific golden plover (*Pluvialis fulva*, $n = 3$), and common sandpiper (*Actitis hypoleucos*, $n = 4$) were collected from Xisha Islands (16°49′53″ N , 112°20′22″ E), South China Sea, between 2017 and 2019. All birds were found dead or stranded on the beaches of Xisha Islands, and were shipped frozen to Guangzhou Institute of Geochemistry, Chinese Academy of Sciences, where dissections were performed. Pectoral muscles excised from each bird were separately lyophilized, ground, and stored in glass bottles at −20 °C until chemical analysis. More information of these birds was documented in Table S1 (Supplementary Material).

2.2. Sample extraction and clean-up

After spiking with 10 μL of internal standards (CBs 24, 82 and 198 for DDTs; BDE 118, BDE 128, 4-F-BDE 67, and 3-F-BDE 153 for PBDEs, ABFRs and DP), pectoral muscle samples (4 g dry weight) were homogenized with diatomaceous earth and extracted using Accelerated Solvent Extraction (APLE-3500, Beijing Jitan Instruments Co., Ltd, China) with 66 mL of hexane/dichloromethane (1/1, v/v) for twice. The extract was concentrated to 1 mL using a rotary evaporator, and exchanged solvent to hexane (10 mL). An aliquot of the extract (1 mL) was used to determine lipids by gravimetric method. The remaining extract was treated with concentrated sulfuric acid (15 mL) to remove lipids and further purified by passing through a complex column (length, 30 cm; inner diameter, 10 mm) loaded with neutral silica (8 cm, 3% water deactivated), acid silica (8 cm, 44% sulfuric acid), and anhydrous sodium sulfate (1 cm) from the bottom to top. Target contaminants were eluted with 30 mL hexane/dichloromethane (v/v, 1:1), which were subsequently concentrated to near dryness under a gentle nitrogen flow and then dissolved in 100 μL of isooctane. Known amounts of recovery standards (BDEs 77, 181, and 205; CBs 30, 65 and 204) were added to the final extracts before instrumental analysis.

2.3. Instrumental analysis

All PHCs were analyzed with an Agilent 6890 GC equipped with an Agilent 5975 MS (GC-MS) in the selective ion-monitoring (SIM) mode. An electron impact (EI) source was used to analyze DDTs (*p,p′*-DDMU, *p,p′*-DDE, *p,p′*-DDD and *p,p′*-DDT), and a DB-5MS capillary column (60 m × 0.25 mm × 0.25 μm, J&W Scientific) was used to separate the peaks of DDTs. An electron capture negative ionization (ECNI) source was used to analyze PBDEs, ABFRs and DP. Tri-to hepta-BDE congeners (BDEs 28, 47, 66, 153 and 183), PBT, HBB and DP were separated on a DB-XLB (30 m × 0.25 mm × 0.25 μm, J&W Scientific) capillary column. Octa-to deca-BDEs (BDEs 202, 197, 203, 196, 208, 207, 206, and 209), BTBPE and DBDPE were separated on a DB-5HT (15 m × 0.25 mm × 0.10 μm, J&W Scientific) capillary column. Details of the GC conditions and MS parameters can be found elsewhere (Luo et al., 2009).

2.4. Stable isotope analysis

Pectoral muscle samples of birds for $\delta^{13}C$ and $\delta^{15}N$ analysis were lyophilized and ground into powder. The sample (about 0.5 mg dry weight) was placed into a tin capsule and analyzed by using a Flash EA 112 series elemental analyzer interfaced with a Finnigan MAT ConFlo III isotope ratio mass spectrometer. Duplicates of the samples were analyzed to confirm the results. Stable isotope abundances were calculated using the following formula:

$$\delta X = (R_{sample}/R_{s\tan dard} - 1) \times 1000 \ (in\ units\ of\ ‰)$$

where X represents ^{15}N or ^{13}C, and R_{sample} and R_{standard} are the $^{13}C/^{12}C$ or $^{15}N/^{14}N$ ratios of the sample and reference standard (Vienna Pee Dee Belemnite for $\delta^{13}C$ and nitrogen for $\delta^{15}N$), respectively. The precisions of this technique are ± 0.5‰ (two standard deviations) for $\delta^{15}N$ and ± 0.2‰ (two standard deviations) for $\delta^{13}C$ based on internal laboratory standard measurements taken every 20 samples.

2.5. Quality assurance and quality control

Procedural blanks were treated identically to the bird samples. One procedural blank was performed with each batch of 11 samples. Trace amounts of BDEs 153, 197, 208, 207, 206 and 209, and PBT were detected in blanks. The concentrations of contaminants in bird samples were corrected by those in blanks. The surrogate recoveries were 106 ± 9% for CB 30, 118 ± 6% for CB 65, 90 ± 7% for CB 204, 93 ± 13% for BDE 77, 96 ± 11% for BDE 181, 102 ± 8% for BDE 205 in all samples, respectively. The average recoveries of contaminants in matrix spikes and spiked blanks were 73–101% and 81–95%, respectively. The relative standard deviations (RSDs) of all targets were < 20% in the triplicate samples. For BDEs 153, 197, 208, 207, 206 and 209, and PBT, the method detection limits (MDLs) were defined as three times the standard deviation of concentrations of these compounds in blanks. For undetected targets in blanks, MDLs were defined as a signal of 10 times the noise level. Based on the lipid weight (lw) of the samples, MDLs for DDTs, PBDEs, ABFRs and DP ranged from 0.02 to 0.15 ng/g lw, 0.01–0.50 ng/g lw, 0.01–0.58 ng/g lw and 0.02–0.07 ng/g lw, respectively (Table S2).

2.6. Data analysis

Concentrations were expressed on a lw basis. Statistical analysis was performed with SPSS 25 (SPSS Inc., Illinois, USA). The level of significance was set as $p < 0.05$. For samples with concentrations below MDL, a value of 1/2 MDL was used in data analysis. The log transformed concentrations of PHCs followed normal distribution and were used in statistical analysis. The statistical analysis was only performed for chemicals detected in > 50% of samples. One-way analysis of variance (ANOVA) accompanied by Tukey's test was used to determine interspecific differences in concentrations of PHCs among bird species. Pearson correlation analysis was used to investigate the relationships between the log transformed concentrations of PHCs and $\delta^{15}N$ values in bird species.

3. Results and discussion

3.1. Residue levels of PHCs in birds

Median and range concentrations of DDTs, PBDEs, ABFRs, and DP in birds were summarized in Table 1. The concentrations of DDTs (sum of *p, p′*-DDT and its metabolites, *p, p′*-DDMU, *p, p′*-DDD and *p, p′*-DDE) ranged from 7.4 ng/g lw in red-footed booby to 19,000 ng/g lw in intermediate egret. The median concentrations of DDTs decreased in the following order: intermediate egret (5900 ng/g lw) > Chinese pond heron (2700 ng/g lw) > pacific golden plover (270 ng/g lw) and common sandpiper (270 ng/g lw) > oriental pratincole (180 ng/g lw) > red-footed booby (14 ng/g lw) (Table 1). The median concentration of DDTs in red-footed booby was significantly lower than those in other bird species in this study ($p < 0.05$). The median concentration of DDTs in Chinese pond heron was about two times higher than that in the same bird species from an e-waste recycling site (Luo et al., 2009). The median concentration of DDTs in common sandpiper was similar to that in common snipe from an e-waste recycling region (Luo et al., 2009). DDT concentrations (0.05–0.14 ng/g ww) in red-footed booby were much lower than those (3.0–10.0 ng/g ww) in brown booby from São Pedro and São Paulo Archipelago, Brazil (Dias et al., 2013).

Sum PBDE levels ranged from 1.1 ng/g lw in red-footed booby to 130 ng/g lw in common sandpiper. The median concentrations of PBDEs decreased in the following order: Chinese pond heron (75 ng/g lw) > intermediate egret (30 ng/g lw) > common sandpiper (14 ng/g lw) > oriental pratincole (13 ng/g lw) > pacific golden plover (5.1 ng/g lw) > red-footed booby (1.3 ng/g lw) (Table 1). The median concentrations of PBDEs in birds except for red-footed booby in this study were at the same magnitude as those in bird muscles from the Shimentai National Nature Reserve in South China (Peng et al., 2015), and were also at the similar levels as those in light-vented bulbul and long-tailed shrike (*Lanius schach*) from rural and urban areas in China (Sun et al., 2012). Several studies have investigated the levels and compositions of PBDEs in Sulidae. PBDE levels in red-footed booby in the present study were much lower than those in northern gannet (*Morus bassanus*) eggs from Bonaventure Island, Gulf of St. Lawrence (Champoux et al., 2017), and lower than those in the livers of brown booby (*Sula leucogaster*) from São Pedro and São Paulo Archipelago, Brazil (Dias et al., 2013). PBDE concentrations in Chinese pond heron in this study were much lower than those in muscles of the same bird species from an e-waste recycling region (Luo et al., 2009). Although both common sandpiper and common snipe belong to Scolopacidae, the median concentration of PBDEs in common sandpiper in the present study was one order of magnitude lower than that in common snipe from an e-waste recycling region (Luo et al., 2009).

The highest median concentration of ABFRs was found in Chinese pond heron (3.4 ng/g lw), while the lowest was found in pacific golden plover (1.3 ng/g lw). PBT, HBB, BTBPE, and DBDPE were detected in more than 76% of the samples. Median and range concentrations (ng/g lw) of PBT, HBB, BTBPE, and DBDPE in birds were showed in Table S3. The median concentrations of ABFRs decreased in the following order: Chinese pond heron (3.4 ng/g lw) > red-footed booby (3.1 ng/g lw) > common sandpiper (2.7 ng/g lw) > intermediate egret (2.3 ng/g lw) > oriental pratincole (1.8 ng/g lw) > pacific golden plover (1.3 ng/g lw) (Table 1). ABFR concentrations in birds except red-footed booby were much lower than PBDE concentrations in the present study. Similar results were found in waterbirds from an e-waste recycling region (Luo et al., 2009), and passerine birds from South China (Sun et al., 2012). The median concentrations of ABFRs in birds from this study were lower than those in waterbirds from an e-waste recycling region (Luo et al., 2009). For the sulidae birds, ABFR levels (sum of HBB and BTBPE) was also lower than PBDE levels in northern gannet eggs from Bonaventure Island, Gulf of St. Lawrence (Champoux et al., 2017), which was consistent with the results in this study. The concentrations of HBB in birds from the current study were lower than those in northern gannet eggs from Bonaventure Island, Gulf of St. Lawrence (Champoux et al., 2017).

Sum DP levels ranged from 0.21 ng/g lw in common sandpiper to 1.2 ng/g lw in Chinese pond heron. DP levels in birds from this study were much lower than those in kingfishers from e-waste sites and were lower than those in kingfishers from non-e-waste sites (Peng et al., 2019). The median concentrations of DP in birds in this study were lower than those in peregrine falcon (*Falco peregrinus*) eggs from the Western North Atlantic Regions (Liu et al., 2019).

The differences in concentrations of PHCs in bird species were evaluated by one-way ANOVA, followed by a Tukey's post hoc test.

Table 1
Median and range concentrations (ng/g lw) of PHCs in birds from South China Sea.

Species	Feeding	N	Lipid (%)	DDTs	PBDEs	ABFRs	DPs	Total PHCs
Red-footed Booby	Piscivorous	5	5.6 (1.7–6.0)	14 (7.4–71)	1.3 (1.1–3.6)	3.1 (1.8–40)	0.33 (0.29–1.1)	30 (12–81)
Chinese Pond Heron	Piscivorous	3	2.9 (1.3–3.2)	2700 (750–11000)	75 (36–97)	3.4 (1.5–17)	1.2 (0.60–2.5)	2700 (830–11000)
Intermediate Egret	Piscivorous	3	3.0 (2.4–3.8)	5900 (1800–19000)	30 (28–43)	2.3 (0.73–11)	0.66 (0.55–1.1)	5900 (1900–19000)
Oriental Pratincole	Insectivorous	3	3.3 (2.8–4.3)	180 (61–7900)	13 (11–14)	1.8 (1.5–2.0)	0.33 (0.30–0.63)	200 (77–7900)
Pacific Golden Plover	Insectivorous	3	3.5 (3.4–4.2)	270 (41–400)	5.1 (4.9–71)	1.3 (1.0–1.9)	0.60 (0.43–1.7)	280 (120–400)
Common Sandpiper	Insectivorous	4	3.6 (2.6–5.7)	270 (83–840)	14 (6.5–130)	2.7 (1.5–17)	0.34 (0.21–0.47)	290 (88–980)

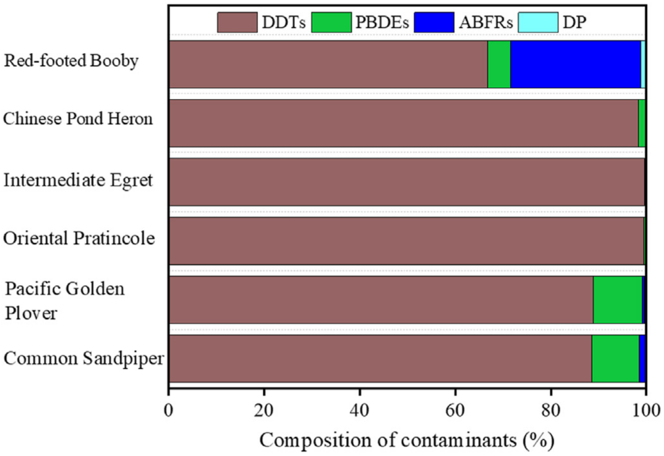

Fig. 1. Compositions of PBDEs, ABFRs, DP and DDTs in birds.

Significant differences were found in concentrations of BDEs 153, 183, 197, 203, 196, 207, 206, and 209, *syn*-DP, *anti*-DP, *p,p′*-DDE, and *p,p′*-DDD in six bird species ($p < 0.05$). The levels of *p,p′*-DDE and most of PBDEs (BDEs 153, 183, 197, 203, 196, 207, 206, and 209) were significantly lower in red-footed booby than in other bird species. The relatively lower levels of *p,p′*-DDE and PBDEs in red-footed booby can be attributed to the different PHC sources in different habitats for birds.

3.2. *Composition profiles of PHCs in birds*

DDTs were the predominant contaminants in birds, accounting for 66–99% of total PHCs (sum of DDTs, PBDEs, ABFRs, and DP) (Fig. 1). The contributions of PBDEs to total PHCs were higher than those of ABFRs and DP for all bird species except for red-footed booby, which contained higher fractions of ABFRs than PBDEs (Fig. 1). The predominance of DDTs in total PHCs was also observed in common magpie (*Pica pica*) and tree sparrow (*Passer montanus*) from China (Yu et al., 2014), and passerine birds from the Shimentai National Nature Reserve, South China (Peng et al., 2015). On contrary, higher levels of PBDEs compared with DDTs were found in passerine birds from an e-waste recycling region (Sun et al., 2012, 2014b), blood samples of magnificent frigatebird (*Fregata magnificens*) from Barbuda, great frigatebird (*Fregata minor*), masked booby (*Sula dactylatra*) and red-footed booby from Laysan Island, and brown booby (*Sula leucogaster*) and great frigatebird from Palmyra Atoll (Gilmour et al., 2019). The difference in composition patterns of PHCs could be attributed to different dietary sources for birds, such as the heavy pollution of PBDEs from e-waste recycling activities (Sun et al., 2012, 2014b), and the intensive agricultural activities in rural sampling areas (Yu et al., 2014; Peng et al., 2015).

Among the DDT and its metabolites (DDTs), *p,p′*-DDT and *p,p′*-DDMU were found in 24% and 43% of the birds, separately, while *p,p′*-DDE and *p,p′*-DDD were detected in all birds. *p,p′*-DDE was the dominating component of all DDTs in birds, contributing over 94% of the total DDTs (Fig. 2). Technical DDTs, generally contain 75% of *p,p′*-DDT, 15% of *o,p′*-DDT, 5% of *p,p′*-DDE, and others (Pan et al., 2016). DDT can be biodegraded to *p,p′*-DDE under aerobic conditions or *p,p′*-DDD under anaerobic conditions (Guo et al., 2009), and DDE tends to be more resistant to further degradation than DDD in the environment (Chen et al., 2006). Therefore, DDE was the predominant component of DDTs. This distribution pattern was similar to that in various birds of prey from northern China, where *p,p′*-DDE also accounted for more than 94% of DDTs in bird species (Chen et al., 2009). This pattern was also similar with those reported in other regions. For example, the concentrations of *p,p′*-DDE were significantly higher than other derivatives of DDTs in birds from Korea (Choi et al., 2001), Europe (Hela et al., 2006; Sakellarides et al., 2006), and North America (Fangstrom et al., 2005; Jimenez et al., 2005). Higher proportions of *p,p′*-DDE to total DDTs in birds indicated that DDTs were from historical residue of technical DDTs.

The predominant PBDE congeners in birds were BDEs 47, 153, and 209 (Fig. 2). BDE 209 was the most abundant component in all bird species, accounting for 32–63% (on average) of total PBDEs. High percentages of BDE 209 in birds reflected the extensive use of Deca-BDE flame retardant in surrounding countries of South China Sea in recent years. Deca-BDE technical mixtures rapidly become one of the major BFRs since Penta- and Octa-BDE were phased out from the market. Previous studies indicated that BDE 153 was the major chemical in terrestrial birds (Law et al., 2003; Lindberg et al., 2004), whereas PBDEs in aquatic birds were often dominated by BDE 47, followed by BDE 99 (Elliott et al., 2005; Norstrom et al., 2002; Sellstrom et al., 2003). All birds except common sandpiper had high fractions of BDE 153 in PBDEs, which was different from the previously results for aquatic birds (Law et al., 2003; Elliott et al., 2005). However, higher proportions of BDE 153 was observed in pectoral muscles of some aquatic bird species (Luo et al., 2009), which was consistent with this study. Moreover, Drouillard et al. (2007) found that BDE 47 had the lowest retention factor among Tetra-to Hepta-BDE congeners, but BDE 153 was most persistent in the American kestrel. Higher percentages of BDE 153 may be attributed to different metabolic abilities for PBDEs in different bird species. The investigated birds except for red-footed booby in the present study always migrated in islands and surrounding continents, and the habitats and food sources of these birds are not consistent. We assumed that birds in the present study could accumulate PBDE congeners via diverse food items, including terrestrial plants and insects in islands and aquatic organisms in marine environment.

The predominant ABFRs in birds were BTBPE and DBDPE. The percentages of BTBPE and DBDPE observed in birds were 28–60% and 29–58%, respectively. BTBPE and DBDPE were also reported in birds from China, with higher levels of DBDPE compared with this study. For instance, BTBPE and DBDPE were 100% detectable in the muscle, liver, and kidney of birds from an e-waste recycling site, South China, with ranges of 0.07–2.4 and 9.6–120 ng/g lw, respectively (Shi et al., 2009). BTBPE and DBDPE were detected in muscles of the five aquatic bird species from the e-waste recycling region in South China, with median levels of < 0.6–3.3 and 10–180 ng/g lw, respectively (Luo et al., 2009; Zhang et al., 2011a).

Anti-DP was the primary DP congener in pectoral muscles of birds (Fig. 2), accounting for 60–77% and 23–29% of the total DP, separately. The fractions of *anti*-DP concentrations (f_{anti}) to the sum of *syn*-DP and *anti*-DP were 0.71 ± 0.03 in red-footed booby, 0.68 ± 0.06 in Chinese pond heron, 0.71 ± 0.02 in intermediate egret, 0.77 ± 0.11 in oriental pratincole, 0.73 ± 0.04 in pacific golden plover, and 0.72 ± 0.03 in common sandpiper, respectively. The average f_{anti} values ranged from 0.68 to 0.77 in birds from this study, which were within the scope of DP technical mixture (0.65–0.80) (Chen et al., 2013). *Anti*-Cl_{11}-DP, mono-dechlorination product of *anti*-DP, was detected in 43% of the bird samples. However, it is unclear whether the *anti*-Cl_{11}-DP detected in organisms was derived from the biotransformation of *anti*-DP *in vivo* and/or bioaccumulation from the environment matrices (Sverko et al., 2008; Zhang et al., 2011b).

3.3. *Source attribution of PHCs in bird species*

3.3.1. *PCA analysis*

In the present study, PCA analysis was carried out on log-transformed concentrations of DDTs, PBDEs, ABFRs, and DP to elucidate the relationships among variables (Fig. 3). PHCs were clustered into three separate groups (Fig. 3a): ABFRs, including HBB, PBT, BTBPE, and DBDPE were in the 1st group and heavily loaded by PC 2; low-halogenated banned PHCs, including *p,p′*-DDD, BDEs 28, and 47, were in the 2nd group and also heavily loaded by PC 2; and high-halogenated PHCs, including BDEs 153, 197, 183, 203, 196, 207, 209, 208, and 206,

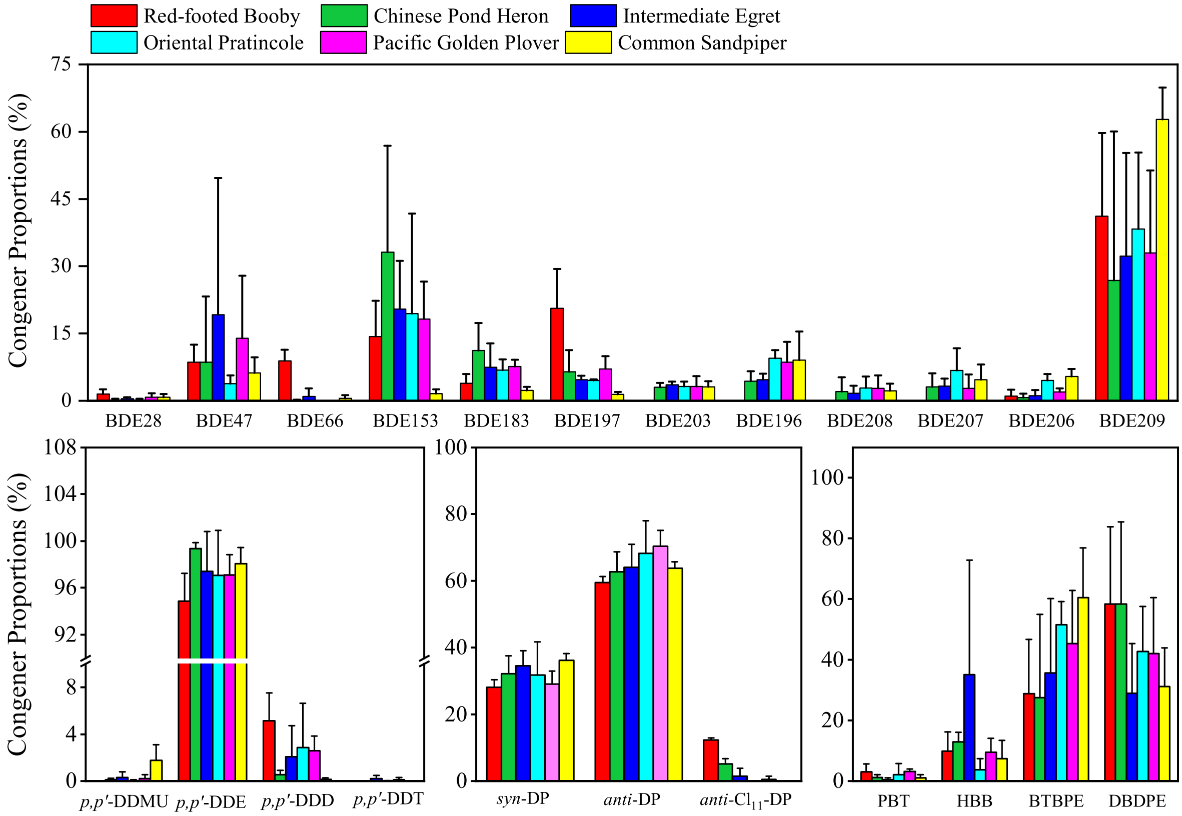

Fig. 2. Compositions of PBDE, ABFR, DP and DDT congeners in birds. Error bars represent standard deviations.

p,p′-DDE, *syn*- and *anti*-DP, were in the 3rd group and heavily loaded by PC 1. Red-footed booby was separated from the other bird species, indicating a different bioaccumulation pattern of PHCs for this species (Fig. 3b). The PCA results clearly indicate different sources for ABFRs, low-halogenated banned PHCs, and high-halogenated PHCs. Red-footed booby was separated from other bird species according to factor scores, as ABFRs were more preferentially accumulated in red-footed booby. Historical contamination of pesticides and halogenated flame retardants in Asian continents were the main sources of PHCs in migratory birds, while alternative brominated flame retardants in the South China Sea were the main sources of PHCs in resident birds.

3.3.2. Influence of trophic levels and dietary sources

Stable nitrogen isotope analysis was usually used to elucidate the trophic levels of organisms, while carbon isotope features were useful to identify different dietary sources (Liu et al., 2018). The results of δ^{15}N and δ^{13}C were provided in Fig. 4. Red-footed booby had a narrow range of δ^{15}N and δ^{13}C. In contrast, δ^{15}N and δ^{13}C values of other species were highly variable. Red-footed booby is a seabird mainly feeding on marine fish, especially Spanish mackerel. Red-footed booby is the only resident bird in the six invested bird species in the present study, which may explain the consistent δ^{15}N and δ^{13}C values in red-footed booby. The other bird species are migrant birds with multiple food items. Chinese pond heron and intermediate egret are wading

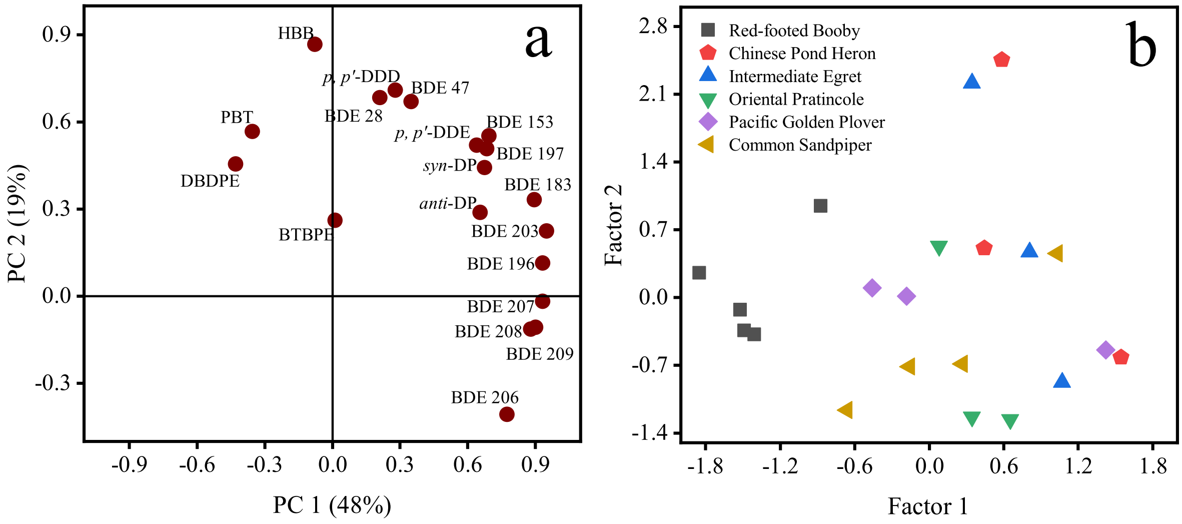

Fig. 3. Principal component analysis results based on the log transformed concentrations of PHC congeners (PC1, 48% variance; PC2, 19% variance) in birds from South China Sea. The figure legends represent the factor loadings (a) and factor scores (b).

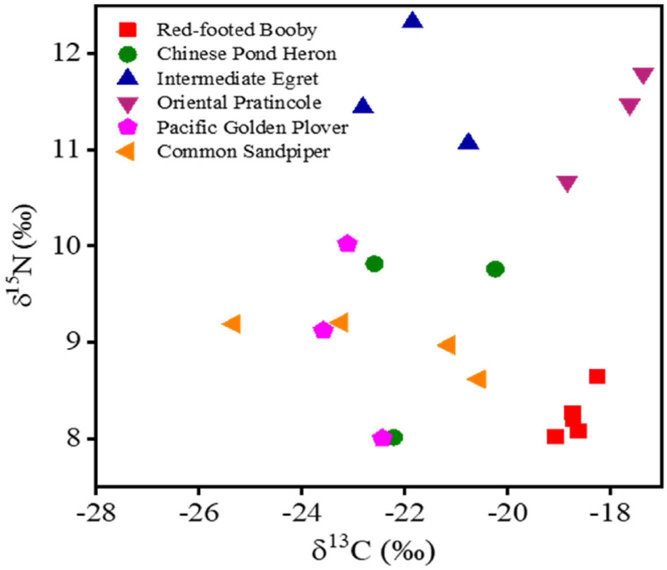

Fig. 4. Stable isotope ratios (‰) of nitrogen and carbon in birds.

birds, which mainly feed on fish, shrimp, frog, locust, mole cricket and other aquatic and terrestrial insects, and small invertebrates. Pacific golden plover, common sandpiper, and oriental pratincole are wading birds, that mainly feeds on coleoptera, orthoptera, lepidoptera, beetles and other insects, crustaceans and other small invertebrates.

In order to explore the influence of trophic levels on bioaccumulation of PHCs in birds, correlation analysis was performed between log-transformed concentrations of target compounds and δ^{15}N values (Table S4). The total concentrations of PHCs had a significant and positive correlation with δ^{15}N values in all bird samples ($p < 0.05$). Concentrations of *p,p′*-DDE, *p,p′*-DDD, BDEs 153, 203, 196, and 207 had significant and positive correlations with δ^{15}N values in all bird samples ($p < 0.05$). Mo et al. (2018) discovered that *p,p′*-DDE and most of PBDE congeners had significant positive correlations with δ^{15}N values in insectivorous resident birds, while that was not observed in granivorous resident birds. The results suggested that trophic levels cannot explain the bioaccumulation profiles of most individual chemicals in the present study.

Among the six species investigated in the present study, red-footed booby is the only resident species in South China Sea. The migratory birds may be exposed to higher levels of PHCs in Asian continents than South China Sea. It should be noted that DBDPE had higher contributions to PHCs in red-footed booby than other birds. Ingestion of plastics has been reported in plenty of studies (Votier et al., 2011; Lavers et al., 2013; Verlis et al., 2014), and could be a reason for the high proportions of DBDPE in red-footed booby. In our previous study, microplastics were found in 3 out of 5 red-footed boobies, and 1 out of 4 other marine birds in South China Sea (Zhu et al., 2019). PHCs with high molecular weight, such as BDE 209 and DBDPE were always considered as less bioaccumulative than most of PHCs (Yu et al., 2014). However, BDE 209 in ingested plastics was able to be leached into avian digestive fluids, and further accumulated in bird tissues (Tanaka et al., 2015; Guo et al., 2019). Overall, birds in South China Sea were threatened by PHCs via various exposure sources. Historical contamination of pesticides and halogenated flame retardants in Asian continents were the main sources of PHCs in migratory birds. Plastics and emerging PHCs related with plastics were important for PHC exposure to residential marine birds like red-footed booby.

4. Conclusions

This study investigated the occurrence and profiles of DDTs, PBDEs, ABFRs and DP in six bird species from South China Sea. DDTs were the predominant PHCs in all birds. BDE 209 was the primary PBDE congener in all birds. BTBPE and DBDPE were the main components of ABFRs. *Anti*-DP and *p,p′*-DDE were the dominating isomer and component of DP and DDTs, respectively. Only BDEs 153, 203, 196, and 207, *p,p′*-DDE, and *p,p′*-DDD showed significant positive correlations with δ^{15}N values in all bird samples. Red-footed booby, the only resident species, had different stable isotope ratios of nitrogen and carbon, and accumulation profiles of PHCs with those for other birds. The abundance of DBDPE in red-footed booby was most likely attributed to the ingestion of microplastics.

CRediT authorship contribution statement

Chunyou Zhu: Methodology, Formal analysis, Writing - original draft. **Yuxin Sun:** Methodology, Formal analysis, Writing - original draft. **Daning Li:** Methodology, Writing - original draft. **Xiaobo Zheng:** Methodology, Formal analysis, Writing - original draft. **Xianzhi Peng:** Writing - original draft. **Ting Zhu:** Formal analysis, Writing - original draft. **Ling Mo:** Methodology, Writing - original draft. **Xiaojun Luo:** Methodology, Writing - original draft. **Xiangrong Xu:** Methodology, Writing - original draft, Writing - review & editing. **Bixian Mai:** Methodology, Writing - original draft, Writing - review & editing.

Declaration of competing interest

All authors declare no conflict of interest.

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data related to this article can be found at https://doi.org/10.1016/j.envres.2020.109462.

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