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Nonchemical Stressors

HEPATIC ETHOXYRESORUFIN-*O*-DEETHYLASE INDUCTION IN THE COMMON KINGFISHER FROM AN ELECTRONIC WASTE RECYCLING SITE

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Abstract: The health effects of exposure to electronic waste (e-waste)-derived pollutants are an important issue. The authors explored the association between the hepatic levels of e-waste–derived halogenated contaminants (including polychlorinated biphenyls [PCBs], polybrominated diphenyl ethers [PBDEs], and polybrominated biphenyls [PBBs]) and hepatic ethoxyresorufin-*O*-deethylase (EROD) activity of the common kingfisher (*Alcedo atthis*) from an e-waste site and 2 reference sites in South China. The summed concentrations of PCBs, PBDEs, and PBBs ranged from 620 ng/g to 15 000 ng/g, 25 ng/g to 900 ng/g, and 14 ng/g to 49 ng/g wet weight, respectively, in the kingfishers from the e-waste site, and these values were significantly greater (2–3 orders of magnitude) than those obtained at the 2 reference sites. Correspondingly, significant hepatic EROD induction was observed in the kingfishers from the e-waste site compared with the reference sites. The EROD activity was significantly correlated to the levels of most of the PCB and PBDE congeners examined as well as PBB 153, suggesting that EROD induction may be evoked by these e-waste–derived pollutants. *Environ Toxicol Chem* 2016;35:1594–1599. © 2015 SETAC

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INTRODUCTION

The recycling of unwanted and obsolete electronic and electrical equipment (commonly known as e-waste) has become a global issue [1,2]. Concerns focus not only on the growing quantity and diversity of e-waste generated, but also on the occurrence and potential risks of chemicals derived from e-waste. There is increasing evidence of environmental contamination with compounds such as polychlorinated biphenyls (PCBs), polybrominated diphenyl ethers (PBDEs), and polybrominated biphenyls (PBBs) at e-waste recycling sites in some developing countries (e.g., China, India, and some African countries) [2,3]. Most of these chemicals are persistent, bioaccumulative, and toxic, ultimately posing serious risks to the health of wildlife and humans [4–6].

Although the wildlife species that inhabit e-waste sites are exposed to a wide range of contaminants [7], little is known about their actual health impacts. To our knowledge, only a single study has reported adverse effects on organisms (fish) from an e-waste site; the fish showed thyroid endocrine disorder and erythrocyte DNA damage, both of which are significantly associated with PBDE exposure [8]. Wildlife that occupy high trophic levels, such as fish-eating birds, appear to be particularly susceptible to the bioaccumulation of e-waste–derived compounds and thus the potential effects of these chemicals [9,10]. The potential effects of these pollutants on piscivorous birds that are resident in e-waste sites, however, remain largely unknown.

Both laboratory and field studies have demonstrated that PCBs and PBDEs can cause a variety of adverse health effects on avian species [10–15]. More specifically, some of these chemicals, such as certain coplanar PCBs (co-PCBs), can induce cytochrome P450 (CYP) monooxygenases (the CYP1A subfamily, specifically), which are mediated through the aryl hydrocarbon receptor (AhR) pathway [10–12]. Certain PBDEs might also act as agonists for AhR, leading to CYP induction [14–16]. The CYP induction is not a toxic response in itself, but it does indicate AhR activation, which is linked with toxicity [17]. It is becoming clear that this induction can affect the signaling pathway regulated by CYP endogenous substrates and the generation of reactive oxygen species (ROS), leading to various secondary toxicities including carcinogenicity, hormonal alterations, embryotoxicity, histopathological changes, and immunotoxicity [17]. Therefore, CYP induction can serve as an indicator of both exposure to and the effects of certain pollutants [17].

Birds, especially the piscivorous birds, have been successfully used as bioindicators for the contamination and effects of pollutants [18]. The common kingfisher (*Alcedo atthis*) (hereafter referred to as kingfisher), a fish-eating bird, is one of the most common and widely distributed resident birds in South China [19]. Our previous investigations revealed that the kingfisher collected from an e-waste site in South China contained elevated levels of PBDEs and other organohalogen compounds [19,20]. However, the effects of these pollutants on the critical physiological functions required for optimal health is unknown.

In the present study, hepatic CYP1A-like enzyme induction based on ethoxyresorufin *O*-deethylase (EROD) activity was examined in kingfishers from an e-waste site and 2 reference sites in South China. Subsets of liver samples also

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were analyzed for selected PCBs, PBDEs, and PBBs. Dichlorodiphenyltrichloroethane (DDT) had been extensively used in the sampling area [21], and DDT and its metabolites chlorodiphenyldichloroethylene (DDE) and dichlorodiphenyl\dichloroethane (DDD) were also examined in the current liver samples. We explored the possible relation between pollutant exposure and EROD activities in the kingfishers. The present work could be valuable for assessing the potential effects of e-waste–derived pollutants on the wild bird population.

MATERIALS AND METHODS

Sample collection

In total, 37 kingfishers (*A. atthis*) were collected from a major e-waste site in South China (the Qingyuan e-waste recycling site; $n = 14$ [9 males and 5 females]) and 2 reference sites (reference site 1, $n = 10$ [6 males and 4 females]; and reference site 2, $n = 13$ [6 males and 7 females]) between August 2010 and March 2011. All sampled birds were adult, and the sampling was conducted outside the breeding season of this species (April to July). The kingfishers were caught using a net, under a license from the Forestry Bureau of Guangdong Province, China. The number of kingfisher samples was limited to <15 at each sampling site (Figure 1). The e-waste site is located in Qingyuan City, Guangdong Province, where several types of e-waste—including televisions, monitors, circuit boards, and electric cable—were recycled for recovery of valuable metals. The e-waste was recycled by primitive techniques, such as physical dismantling, burning, and acid baths, in the open air or in small workshops. Traditional agricultural practices, including rice planting and fish farming, are conducted near the e-waste site [22]. Reference site 1 is a national nature reserve, and reference site 2 is located in an agricultural area where rice planting and fish farming are practiced; no known e-waste recycling activity had been performed at the 2 sites.

Captured birds were euthanized with N_2 and decapitated immediately in the field. The posterior portion of the left lobe liver was flash frozen in liquid nitrogen for EROD assay. The remaining liver from each bird was stored in liquid nitrogen for chemical analysis. All the samples were later transferred from liquid nitrogen to an ultracold freezer (–80 °C) for storage until processing.

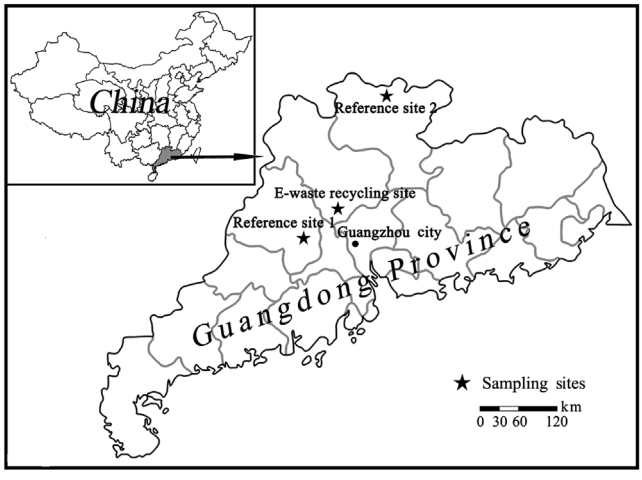

Figure 1. Map of sampling sites.

Microsomal preparation and EROD assay

Hepatic microsomes of the kingfishers were prepared by fractional ultracentrifugation. Approximately 0.2 g of liver samples was minced and homogenized in a glass homogenizer containing a sodium phosphate buffer (0.1 M KH_2PO_4 and 0.1 M Na_2HPO_4, pH 7.4). The homogenate was centrifuged at 11 000 *g* for 20 min at 4 °C to yield a postmitochondrial supernatant (PMS). The PMS was further centrifuged at 105 000 *g* for 1 h at 4 °C. After removal of the supernatant, the microsomal pellet was resuspended in an equivalent volume of resuspension buffer (50 mM Tris-HCl, 1 mM EDTA, 1 mM dithiothreitol, pH 7.4–7.5, dissolved in a 20% glycerol solution). The microsomes were flash frozen in liquid nitrogen, and stored at –80 °C until EROD activity determination.

We evaluated EROD activity using a fluoresence-based EROD assay kit (Genmed Scientifics). All samples were analyzed in duplicate. The EROD activities were measured in 96-well microplates using a spectrofluorometer, with an excitation wavelength of 530 nm and an emission wavelength of 590 nm. Protein concentration was determined separately using a protein assay kit (Bio-RAD), with bovine serum γ-globulin as reference.

Chemical analysis

An aliquot of each liver was analyzed for PCBs, PBDEs, PBBs, and DDT and its metabolites DDD and DDE (the sum of DDT, DDE, and DDD is designated as "DDTs"), using previously described methods [19,20]. Briefly, after spiking with surrogate standards (BDEs 77, 181, and 205 and ^{13}C-BDE 209 for PBDEs and PBBs; CBs 30, 65, and 204 for PCBs and DDTs), the samples were homogenized with anhydrous sodium sulfate and Soxhlet-extracted with an acetone/hexane mixture for 48 h. The extract was passed through a gel permeation chromatography column to remove bulk lipids and interfering coextractables and was further purified with a silica column containing neutral activated silica and 40% sulfuric acid silica gel. Known amounts of internal standards (10 ng of BDEs 118 and 128 for PBDEs and PBBs; 50 ng of CBs 24, 82, and 198 for PCBs and DDTs) were spiked into the final extracts before instrumental analysis. All samples were analyzed on an Agilent 7890A-5975C gas chromatography–mass spectrometer, with an electron-impact ion source for PCBs and DDTs and an electron-capture negative ionization ion source for the other target compounds.

Quality assurance and quality control

For EROD activity assay, all the operations were performed on ice with precooled equipment and solutions. The correlation coefficients yielded from resorufin and protein standard curves were >0.998. Coefficients of variation of the duplicates were <15%. Finally, blind triplicate samples were processed for 1 kingfisher sample, generating relative standard deviations of 15% and 10% for EROD activity and protein concentration, respectively.

For chemical analysis, procedural blanks, blind triplicate samples, and triplicate spiked blanks (20 PCB congeners, 10 PBDE congeners, and DDTs spiked into solvents) were processed. Trace levels (<1% of the levels detected in the bird samples) of BDEs 47, 153, and 209 and CBs 118 and 153 were detected in the procedural blanks, and the mean values were subtracted from samples. The mean recoveries for surrogate standards ranged from 81% to 95%. The mean recoveries of chemicals spiked in blanks ranged from 86% to

106%. The final concentrations were corrected by the surrogated standard recoveries. The relative standard deviation percentages were <10% in the triplicate spiked blanks and <15% in the blind triplicate samples.

Calculation of toxic equivalents

Toxic equivalents (TEQs) calculated with the major co-PCBs, including CBs 77, 81, 105, 114, 118, 123, 126, 156, 167, and 169, were estimated by multiplying the congener concentrations by the congener-specific avian toxic equivalency factor proposed by the World Health Organization [23].

Statistical analysis

The chemical concentration and EROD activity data in the kingfisher from certain sampling sites were tested for normality using a Kolmogorov–Smirnov test. The data were found to be in violation of this assumption, and therefore logarithmic transformations were employed. To evaluate the possible differences in chemical concentrations and EROD activity between the e-waste site and the reference sites, and between the males and females from certain sampling sites, 2-way analysis of variance (ANOVA) was used. No sex-specific differences in the concentration of pollutants or EROD activity was found in the kingfishers from the 3 sampling sites, and the effect of sex was not controlled for in subsequent analyses because of the low sample size. Pearson correlation analyses were performed to illustrate the relationship between EROD activity and the concentrations of $\sum$TEQs, $\sum$PCBs, $\sum$PBDEs, individual PCB and PBDE congeners, or PBB 153, and between $\sum$PBDEs and $\sum$TEQs, $\sum$PCBs, or BB 153. If any of these chemical concentrations was significantly correlated, partial correlation analyses were conducted to identify differences in the contributions of the various chemicals to the increased EROD activity. The criterion for significance in all statistical tests was set at $p < 0.05$.

RESULTS

The concentrations of the major congeners of PCBs, PBDEs, and PBBs, and DDTs in liver of the kingfishers are presented in Table 1. Residues of the $\sum$PCBs, $\sum$PBDEs, $\sum$PBBs, and $\sum$DDTs were consistently detected in the kingfishers from the e-waste site, at median concentrations of 3000 ng/g, 140 ng/g, 22 ng/g, and 43 ng/g wet weight, respectively. The hepatic levels of e-waste–derived chemicals (i.e., PCBs, PBDEs, and PBBs) in the kingfishers from the e-waste site were 2 to 3 orders of magnitude greater than those from the 2 reference sites (p values < 0.0001). In contrast, no significant difference ($p > 0.3$) was observed between the e-waste site and the reference sites in the hepatic levels of DDTs, which were pesticide-originated contaminants. The $\sum$TEQs in the kingfishers from the e-waste site ranged from 420 pg/g to 2530 pg/g wet weight, which were 2 orders of magnitude greater than those observed at the reference sites ($p < 0.0001$; Table 1).

Concentrations of $\sum$PBDEs and $\sum$PCBs were significantly correlated in these birds ($r = 0.98$, $p < 0.001$). However, no significant relation was found between the $\sum$PBDEs and $\sum$TEQs or BB 153 (r values < 0.35, p values > 0.05).

The hepatic EROD activity in the kingfishers from the e-waste site and the 2 reference sites is shown in Figure 2A. A significant increase ($p < 0.005$) in EROD activity was observed in the kingfishers from the e-waste site compared with the 2 reference sites. The EROD activity did not differ between the 2 reference sites ($p = 0.68$). The EROD activities were positively correlated with the concentrations of $\sum$TEQs in the kingfishers from the 3 sites (Figure 2B). They also were significantly positively correlated with the concentrations of $\sum$PCBs and $\sum$PBDEs (Figure 2C and D), most of the individual PCB and PBDE congeners and PBB 153, but not DDTs (Supplemental Data, Table S1). A partial correlation analysis, controlling for $\sum$PCBs, showed that the concentration of $\sum$PBDEs was significantly correlated with EROD activity ($r = 0.31$, $p = 0.04$).

DISCUSSION

The high lipophilicity and persistence of PCBs, PBDEs, and PBBs result in biomagnification in the food web, facilitating greater accumulation in higher trophic animals [24]. The accumulation of PCBs, PBDEs, and PBBs in organisms from e-waste sites in China has been previously reported in several species, including aquatic invertebrates and fish [7], but information on the contamination status of these chemicals in higher trophic level organisms such as fish-eating birds is limited [19,20]. The present study found substantially greater (2–3 orders of magnitude) PCB, PBDE, and PBB concentrations in the liver of the kingfishers from an e-waste site compared with 2 reference sites in South China, a result that is consistent with recent reports on PBDEs in kingfishers and other waterbirds from the same e-waste site [19,20]. The PCB, PBDE, and PBB loadings, which originated primarily from e-waste, likely were consumed locally by the kingfishers through their diet of various fish species, as noted by Mo et al. [19].

Although a large body of evidence indicates that wildlife at e-waste sites has been heavily contaminated by pollutants, little research has focused on the potential effects of these contaminants on the wild organisms inhabiting e-waste regions, and no such study has been conducted in wild avian species. The EROD activity of liver microsomes has, over the years, proved to be a useful biomarker for both exposure to and toxic effects of xenobiotics that can induce CYP1A mono-oxygenases and perhaps other CYP isoforms [17]. The induction of EROD activity has also been reported in diverse bird species that have been exposed to a considerable variety of environmental contaminants, including organohalogen compounds, both in the laboratory and in field studies [10–15,25]. Similarly, in the present study, the kingfishers from the e-waste site had significantly higher EROD activities than did those from the 2 reference sites, suggesting high AhR-mediated stress in the e-waste site kingfishers. The elevated EROD activity in the kingfishers from the e-waste site is of concern because the EROD induction is associated with a series of biological effects, such as the disruption of oxidative homeostasis, the potential formation of DNA adducts that can lead to carcinogenesis, reproductive effects, and increased liver somatic index [17]. The potential detrimental effects resulting from EROD induction in the e-waste site kingfishers, and perhaps other avian species, should be further investigated.

Induction of EROD caused by specific PCB congeners has been well documented in birds [25]. A planar conformation is requisite for AhR binding; as a result, the co-PCBs demonstrate EROD-inducing potential in several bird species [25], which could explain the significant correlations among EROD activity and the concentrations of co-PCBs or $\sum$TEQs in the current kingfishers. Similar results were also reported for wild fish-eating birds, including common cormorants (*Phalacrocorax carbo*) [10,26], double-crested cormorants (*Phalacrocorax auritus*) [27], and black-footed albatross (*Phoebastria nigripes*) [11], as well as many other avian

Table 1. Hepatic concentrations (ng/g wet wt) of electronic waste (e-waste)–derived organohalogen compounds and DDTs in the common kingfisher from an e-waste site and 2 reference sites in South China[a]

	E-waste recycling site ($n = 14$)	Reference site 1 ($n = 10$)	Reference site 2 ($n = 13$)
Concentrations of e-waste–derived organohalogen compounds			
PCBs[b]			
CB 28/31	190 (36−530)	0.67 (0.25−1.92)	0.41 (0.11−1.5)
CB 52	130 (19−430)	0.17 (0.11−0.28)	0.54 (0.03−1.4)
CB 101	180 (17−1100)	0.28 (0.06−0.82)	0.21 (0.01−2.6)
CB 118	360 (22−2000)	1.9 (0.63−5.1)	1.8 (0.15−5.1)
CB 138	220 (20−1200)	1.5 (0.39−3.5)	1.2 (0.12−4.0)
CB 153	230 (21−1300)	1.6 (0.40−5.0)	1.4 (0.17−4.1)
CB 180/193	71 (8.2−250)	0.97 (0.22−3.0)	0.49 (0.07−1.6)
∑TEQs[c]	1.1 (0.42−2.5)	0.06 (0.03−0.09)	0.02 (0.01−0.05)
∑PCBs[d]	3000 (620−15 000)	21 (7.1−45)	19 (5.0−70)
PBDEs[e]			
BDE 28	2.1 (0.78−6.3)	0.02 (0.002−0.22)	0.12 (0.03−0.37)
BDE 47	41 (4.6−330)	0.22 (0.07−1.5)	0.31 (<0.005−1.43)
BDE 99	11 (1.9−128)	0.34 (0.03−0.49)	0.06 (<0.001−0.12)
BDE 100	14 (1.3−93)	0.6 (0.17−3.7)	0.53 (0.07−0.72)
BDE 153	20 (2.6−110)	0.49 (0.16−1.8)	0.24 (0.04−0.81)
BDE 154	23 (2.4−112)	0.68 (0.23−3.0)	0.27 (0.001−0.69)
BDE 209	4.8 (1.8−26)	0.45 (0.02−1.1)	0.13 (0.07−0.54)
∑PBDEs[f]	140 (25−900)	3.2 (2.3−4.5)	2.0 (1.3−4.4)
PBBs			
BB 153	19 (14−35)	0.12 (0.01−0.47)	<0.003
BB 209	2.0 (<0.003−13)	<0.003	<0.003
∑PBBs[g]	22 (14−49)	0.12 (0.01−0.47)	<0.003
Concentrations of DDTs			
p,p′-DDE	40 (16−190)	67 (21−170)	85 (18−180)
p,p′-DDD	1.5 (0.39−8.0)	0.55 (0.005−1.5)	1.8 (0.33−8.8)
p,p′-DDT	0.78 (<0.001−2.8)	0.17 (0.007−0.98)	1.1 (0.31−7.3)
∑DDTs[h]	43 (17−200)	67 (22−170)	88 (21−190)

[a]Values are median (range in parentheses), except where below (<) the detection limit.
[b]The 7 indicator PCBs are listed.
[c]Sum of TEQs calculated from 10 non-ortho or mono-ortho PCB congeners based on bird toxic equivalency factors proposed by the World Health Organization [23].
[d]Sum concentrations of the 82 PCB congeners detected.
[e]Major congeners are listed.
[f]Sum concentrations of the 16 PBDE congeners investigated.
[g]Sum of BBs 153 and 209.
[h]Sum concentrations of p,p′-DDE, DDD, and DDT.
PCBs = polychlorinated biphenyls; PBDEs = polybrominated diphenyl ethers; PBBs = polybrominated biphenyls; DDE = chlorodiphenyldichloroethylene; DDD = dichlorodiphenyldichloroethane; DDT = dichlorodiphenyltrichloroethane.

species [12,25]. Aside from induction by the co-PCBs, certain nonplanar PCBs may evoke an EROD response in some avian species [28]. In our kingfishers, EROD activity was significantly correlated with the concentrations of most of the nonplanar PCBs and ∑PCBs, suggesting that an EROD induction was associated with the birds' exposure to these PCB congeners. Another explanation is that the significant correlations may be coincident with co-PCB exposure because their concentrations are often related to co-PCBs. Furthermore, the kingfishers used in the present study were coexposed to several other chemicals, such as PBDEs and PBBs. The possibility of additive and/or synergistic effects of these chemicals in increasing EROD activity of the kingfishers may also lead to the PCB–EROD correlation. Previous avian studies have also reported positive correlations between EROD activity and the concentrations of some nonplanar PCBs or ∑PCBs [28,29]. The nonplanar PCBs were associated with the induction of CYP of Family 2 (especially 2b) in mammals. The induction of EROD by nonplanar PCBs may indicate that the response of avian species is rather different from that of mammals [25,30].

Very little is currently known about the PBDE-induced EROD activity in avian species. Nevertheless, the limited studies suggest that certain PBDE congeners or the technical mixture may evoke EROD responses in some avian species, such as chicken (*Gallus gallus*) [14,16] and American kestrel (*Falco sparverius*) [15]. In the kingfishers used in the present study, the hepatic EROD activities were significantly correlated with the concentrations of most of the PBDE congeners examined and with ∑PBDEs. Because the concentration of ∑PBDEs was significantly correlated with ∑PCBs in these birds, we conducted a partial correlation analysis, controlling for ∑PCBs, to rule out the possibility that the ∑PBDEs–EROD correlation simply reflected a ∑PCBs–EROD correlation. Although the partial correlation ($r = 0.31$) was reduced compared with the Pearson correlation ($r = 0.64$), it remained significant ($p = 0.04$). Chen et al. [16] assessed the capacity to induce EROD activity by a number of PBDE congeners using chick hepatocytes, showing that the induction potencies follow the order of BDEs 77, 100, 119, and 126 > BDEs 153 and 183 > BDEs 66 and 85 > BDEs 28, 47, 99, and 154. Interestingly, the Pearson correlation coefficients of the EROD–PBDE relationships in the kingfishers used in the present study generally followed this trend (Supplemental Data, Table S1). Strong correlations between EROD activity and circulating plasma levels of BDEs 208, 206, and 197 and the hepatic concentration of BDE 208 also were observed in American kestrel (*F. sparverius*) dosed with BDE 209 [15]. However, no EROD–PBDE correlation has been found in young American kestrels (*F. sparverius*) dosed with lower

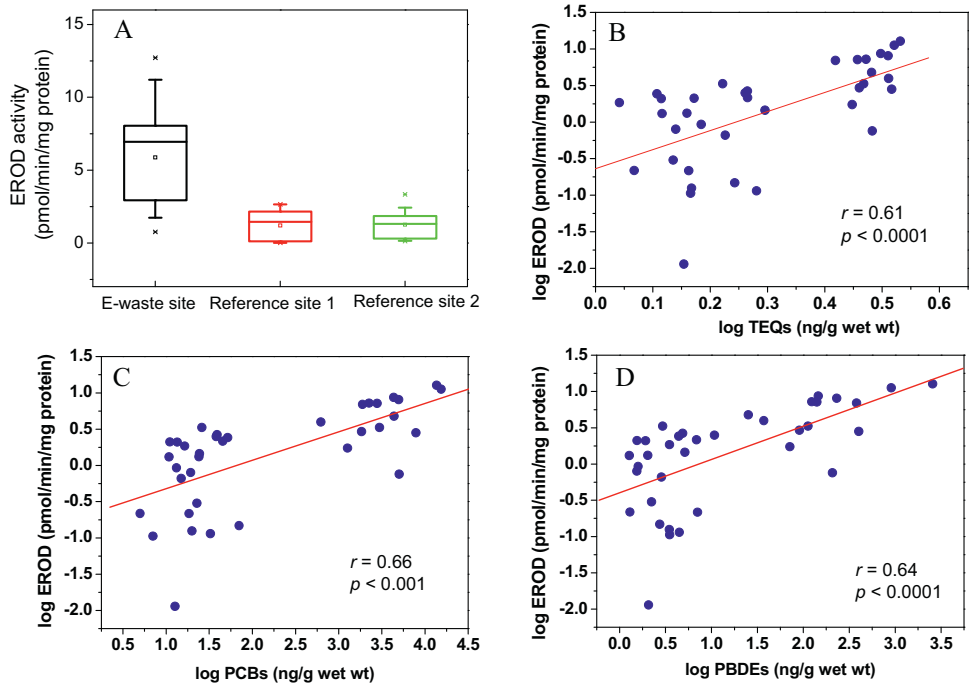

Figure 2. The hepatic ethoxyresorufin-*O*-deethylase (EROD) activities of the kingfishers from an electronic waste site and reference sites in South China (**A**), and the relationships between the hepatic EROD activities and the concentrations of ∑TEQs (**B**), ∑PCBs (**C**), and ∑PBDEs (**D**). Box plots are defined as follows: center line, median; hollow square, mean; box plot edges, the 25th and 75th percentiles. ∑TEQs = the summed concentrations of toxic equivalents, which were estimated by multiplying the major co-PCB concentrations by the congener-specific avian toxic equivalency factor proposed by the World Health Organization [23]; ∑PCBs = the summed concentrations of 82 polychlorinated biphenyl congeners examined; ∑PBDEs = the summed concentrations of 16 polybrominated diphenyl ether congeners examined.

brominated PBDEs in the laboratory [14] or in breeding ring-billed gulls (*Larus delawarensis*) [31], Forster's terns (*Sterna forsteri*) [32], Caspian terns (*Hydroprogne caspia*) [32], and surf scoters (*Melanitta perspicillata*) [33] that were exposed to several organohalogen compounds, including PBDEs, in the field. The results indicate that the EROD–PBDE correlation in birds appears to be species-specific, possibly resulting from the differences in the sequence of the AhR [34]. It should be noted that, to date, serious doubt exists on the agonistic properties of PBDEs on CYP1A. It has been reported that PBDEs having at least 4 bromine atoms (e.g., BDE 47) generally are poor inducers of EROD activity resulting from gene expression via AhR because their conformation is bulkier and more globular [35]. A previous study reported that some PBDE congener can even inhibit EROD activity, although these phenomena were observed in fish [36]. In addition, chlorinated/brominated dioxins and dienzofurans, which are cocontaminants with PBDEs and PCBs, may also be responsible for the observed EROD induction in the field studies, even though we have not measured these compounds.

Despite the long period of restrictions on the use and production of PBB flame retardants (PBBs were discontinued in 1978) [5], PBB 153, a major congener of PBB flame retardants, was detected at elevated levels in the birds used in the present study, and the levels of this congener were significantly correlated with hepatic EROD activities (Supplemental Data, Table S1). Although there is, to our knowledge, no evidence demonstrating PBB-induced EROD activity in avian species, experiments in fish have suggested that some PBB congeners or the commercial PBB mixture are EROD inducers [17].

CONCLUSIONS

The health effects of exposure to e-waste–derived pollutants are an important issue. In the present study, hepatic EROD induction in response to e-waste–derived pollutant accumulation was found in the wild kingfishers living in an e-waste site in South China. Although the correlations between the EROD induction and chemical exposure do not implicate causal relationships per se, the significant correlations are of concern because most of these chemicals have been demonstrated to be involved in EROD induction in laboratory-exposed animals. A need for further examination is warranted to determine the potential adverse effects resulting from EROD induction (e.g, oxidative damage and histopathological changes) in the kingfishers and other wildlife that inhabit e-waste sites.

Supplemental Data—The Supplemental Data are available on the Wiley Online Library at DOI: 10.1002/etc.3294.

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