Polychlorinated biphenyl exposure and corticosterone levels in seven polar seabird species

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Abstract

The role of polychlorinated biphenyls (PCBs) on exposure-related endocrine effects has been poorly investigated in wild birds. This is the case for stress hormones including corticosterone (CORT). Some studies have suggested that environmental exposure to PCBs and altered CORT secretion might be associated. Here we investigated the relationships between blood PCB concentrations and circulating CORT levels in seven free-ranging polar seabird species occupying different trophic positions, and hence covering a wide range of PCB exposure. Blood $\sum_7$PCB concentrations (range: 61-115632 ng/g lw) were positively associated to baseline or stress-induced CORT levels in three species and negatively associated to stress-induced CORT levels in one species. Global analysis suggests that in males, baseline CORT levels generally increase with increasing blood $\sum_7$PCB concentrations, whereas stress-induced CORT levels decrease when reaching high blood $\sum_7$PCB concentrations. This study suggests that the nature of the PCB-CORT relationships may depend on the level of PCB exposure.

Capsule: In polar seabird species, the relationship between PCB and CORT concentrations may be related to the levels of contamination.

Key-words: Arctic; Antarctic; birds; PCBs; glucocorticoids; stress

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INTRODUCTION

In Polar Regions, increasing attention has been directed towards environmental contaminants and their potentially hazardous effects on susceptible wildlife species (Bargagli 2008; Bustnes et al. 2003, 2007; Gabrielsen 2007; Verreault et al. 2010; Wania 2003; Letcher et al. 2010). Among environmental contaminants, several persistent organic pollutants (POPs) may exhibit endocrine disruptive properties, and may alter functions of several hormones (e.g. Amaral Mendes 2002). For example, a number of studies have reported significant relationships between concentrations of POPs and plasma levels of reproductive hormones such as steroids and some pituitary hormones in free-living birds and mammals (Giesy et al. 2003; Vos et al. 2000; Jenssen 2006; Gabrielsen 2007; Verreault et al. 2008; Verreault et al. 2010).

Relationships reported to date in a limited number of studies on wild bird species between POP levels and stress hormones (glucocorticoids) have been largely inconclusive: in black-legged kittiwakes *Rissa tridactyla* baseline CORT levels were positively associated to $\sum_{11}$PCB concentrations (Nordstad et al. 2012). Also, in the most PCB-exposed Arctic seabird species, the glaucous gull *Larus hyperboreus*, a higher POP burden (including 58 PCB congeners, organochlorine pesticides, brominated flame retardants and their metabolically-derived products) was associated with higher baseline CORT levels in both sexes (Verboven et al. 2010). Moreover, in studies of pre-laying female kittiwakes and incubating snow petrels *Pagodroma nivea*, which bear low to moderate PCB contamination, stress-induced CORT levels increased with increasing $\sum_{10}$PCB concentrations and $\sum$POPs (including 7 PCBs congeners and organochlorine pesticides), respectively (Tartu et al. 2014, Tartu et al. 2015). On the other hand, stress-induced CORT levels decreased with increasing POPs (58 PCB congeners, organochlorine pesticides, brominated flame retardants and their metabolically-derived products) in male glaucous gulls that accumulate the highest levels of these contaminants among Arctic species (Verboven et al. 2010). This suggests that the nature of the relationship between POPs, and CORT secretion may be related to the levels of contamination. The major POP detected in wildlife are still the PCBs despite their global ban more than 30 years ago. PCBs bio-accumulate in top predators such as polar seabirds (Letcher et al. 2010; Corsolini et al. 2011) and occasionally high levels of these compounds accumulate in lipid-rich tissues. Since PCB may be a good proxy for POPs in general, the link between PCB levels and stress hormones therefore deserves more attention especially because of the major role of stress hormones in allostasis (McEwen and Wingfield 2003; Angelier and Wingfield 2013). For example, in an experimental study conducted on captive American kestrels *Falco spaverinus* dosed with PCBs, decreased levels of baseline and stress-induced CORT were reported compared to levels measured in the control group (Love et al. 2003). CORT secretion is regulated through a number of physiological mechanisms. At the endocrine level, a stressful event will trigger the release of corticotropin-releasing hormone (CRH) from the hypothalamus; CRH will then stimulate the secretion of adrenocorticotropic hormone (ACTH) from the anterior pituitary, which in turn will activate the synthesis of glucocorticoids from the adrenal cortex (Sapolsky et al. 2000; Wingfield 2013). In birds, up to 90% of glucocorticoids released into the bloodstream will bind to corticosteroid-binding globulin (CBG) and will be transported to target cells. Concurrently, glucocorticoids will provide negative feedback signals for ACTH and CRH release (Wingfield 2013). This hormonal cascade may trigger an array of physiological and behavioural adjustments that shift energy investment away from reproduction, and redirect it towards survival (Wingfield and Sapolsky, 2003). Glucocorticoids are therefore considered as major mediators of reproductive decisions in birds (reviewed in Wingfield and Sapolsky, 2003) and have a strong connection with fitness in some seabird species (Angelier et al. 2010; Goutte et al. 2011;Schultner et al. 2014). It is thus crucial to determine how both baseline and stress-induced glucocorticoid secretion can be influenced by ubiquitous and abundant environmental contaminants including PCBs. Baseline and stress-induced CORT levels (i.e. CORT levels measured in response to a capture/handling stress), may depict different physiological status: baseline CORT mirrors energetic state (Landys et al. 2006), while stress-induced CORT can be used to infer on an individual's sensitivity to stress. The CORT release following a stress can be modulated (elevated or low release) in order to maximize either survival or reproduction (Lendvai et al. 2007; Bókony et al. 2009).

The aim of the present study was to investigate the relationships between $\sum_7$PCB concentrations, plasma baseline CORT levels and stress-induced CORT levels in seven polar seabird species. We selected seabird species occupying different trophic positions that encompassed a wide range of plasma PCB levels (Letcher et al. 2010). These include the glaucous gull, the black-legged kittiwake, the common eider *Somateria mollissima*, these three species were sampled in the Norwegian Arctic (Bear Island and Kongsfjorden, 74° 22'N, 19° 05'E and 78°54'N, 12°13'E, respectively) the snow petrel, the cape petrel *Daption capense*, the south polar skua *Catharacta maccormicki*, the three species were sampled in Antarctica (Adélie land, 66°40'S, 140°01'E) and the wandering albatross *Diomedea exulans* which was sampled at Crozet Island (46° 24′ S, 51° 45′E) a subantarctic French territory. All species were sampled within a short period of time during the breeding period, that is, from late incubation to early chick-rearing (corresponding to the month of June for Arctic species, and early to late December for Antarctic and subantarctic species). Based on the previous reports on PCB/CORT relationships (Verboven et al. 2010; Nordstad et al. 2012; Tartu et al. 2014), we predicted that the relationships between PCB and CORT levels would differ between species according to their blood PCB levels: 1) baseline CORT concentrations would increase with increasing PCB levels, whereas 2) stress-induced CORT levels would increase in moderately contaminated species and decline in highly contaminated bird species.

MATERIAL AND METHODS

Ethics statement

Animals were handled in accordance with the national guidelines for ethical treatment of experimental animals from the Governor of Svalbard, the Norwegian Animal Research Authority (NARA), and the ethic committee of the Institut Polaire Français Paul Emile Victor (IPEV): Governor of Svalbard (2004/00481-12 to G.W. Gabrielsen and J. Verreault, (2007/00165) to S.A. Hanssen and B Moe; NARA 2006/16056 to G.W. Gabrielsen and J. Verreault, (2007/6072) to S.A. Hanssen and B. Moe, FOTS id 2086, 3319 to O. Chastel and IPEV programs no. 109 to H. Weimerskirch and no. 330 to O. Chastel.

Sampling year, study site and species

Two hundred eighty-six blood samples were available from three high Arctic seabird species: the black-legged kittiwake (hereafter 'kittiwakes', N=25, 2011), the common eider (N=55 females, 2007) and the glaucous gulls (N=38, 2006) and four Antarctic species, the wandering albatross (N=75, 2008), the snow petrel (N=35, 2010), the cape petrel (N= 27, 2011), and the south polar skua (N=31, 2003). Main diet and average body mass during late incubation to early chick-rearing are reported for all species in **Table 1**. Wandering albatrosses were not weighed but the average body mass of wandering albatrosses during incubation is around 8403 ± 642 g for females and 10,720 ± 966 g for males (Weimerskirch 1995). Study sites, bird capture, and sampling protocols have been described in previous studies (Verboven et al. 2010; Bustnes et al. 2012, Angelier et al. 2013; Goutte et al. 2013; Tartu et al. 2014; Tartu et al. 2015; Goutte et al. 2014). Because in seabirds blood CORT and PCB levels may vary between breeding phases (Nordstad et al. 2012), we selected blood samples of birds collected during late incubation and early chick-rearing periods. Briefly, a first blood sample (*ca.* 0.3 mL) for baseline CORT analysis was collected immediately after capture from the alar vein using a heparinized syringe and a gauge needle (Romero and Reed 2005). Birds were then kept in opaque cloth bags during

30 min after which blood samples were collected immediately following previously described methods for stress-induced CORT analysis (e.g. Tartu et al. 2014). Stress-induced CORT levels were calculated by subtracting the baseline CORT concentrations from the CORT concentration following 30 min handling protocol: stress-induced CORT levels = ($CORT_{t=30min}$ –$CORT_{t=0min}$). Wandering albatrosses and south polar skuas were not subjected to a capture/handling stress protocol and only baseline CORT levels are available.

Molecular sexing and hormone assay

Whole blood samples were centrifuged and plasma was stored at −20°C until assayed. Red blood cells were kept at −20°C for molecular sexing (polymerase chain reaction amplification, Weimerskirch et al. 2005) at the CEBC, with the exception of common eiders (only females incubate) and glaucous gulls which were sexed based on morphometric measurements. Plasma concentrations of CORT were determined by radioimmunoassay for all species as described elsewhere (Lormée et al. 2003; Verboven et al. 2010). Radioimmunoassays were conducted at CEBC for all species except for glaucous gulls for which radioimmunoassays were conducted at the university of Glasgow veterinary school. For glaucous gull data, an inter-laboratory validation was conducted.

PCB analysis

For POPs, cross-validation between the different labs (EPOC/LPTC , NILU and the National Wildlife Research Centre) was not possible due to limited sample volumes, however, quality assurance and quality control procedures are performed routinely in NILU, the National Wildlife Research Centre and EPOC/LPTC using standard reference materials, method blanks, duplicate extractions and injections of authentic standards, and these labs met the established criteria for QA/QC (for details see Verboven et al. 2009; Goutte et al. 2014 and Tartu et al. 2014). POPs analyses for kittiwakes and common eiders were conducted on whole blood samples at NILU with the method described in Tartu et al. 2014 by gas chromatography coupled with a mass spectrometer (GC-MS). The same method (GC-MS) was used for glaucous gulls, POPs were measured in plasma at the National Wildlife Research Centre, a detailed method was described in Verboven et al. 2010. Finally, for wandering albatrosses, snow petrels, cape petrels and south polar skuas, POPs were measured in plasma at EPOC/LPTC as described in Goutte al. 2014 by gas chromatography coupled with electron capture detection. In the present study, we focused on 7 major PCB congeners (CB-28, -52, -101, -118, -138, -153 and -180) since they are the most abundant in the marine ecosystem and often the most bioaccumulative in a wide range of seabird species inhabiting the polar regions (Gabrielsen et al. 1995; Savinova et al. 1995). We used the $\sum_7$PCBs (i.e. $\sum$CB-28, -52, -101, -118, -138, -153 and -180) for further analyses.

Lipid determination

Lipids were determined in plasma on an aliquot of 10 µL by the *sulfo-phospho-vanillin* (*SPV*) reaction for colorimetric determination for cape petrels, snow petrels, south polar skuas and wandering albatrosses at EPOC/LPTC (Frings et al. 1972). For common eiders, kittiwakes and glaucous gulls, lipids were determined using a gravimetric method using the whole sample amount at NILU and National Wildlife Research Centre. In order to compare whole blood to plasma samples, PCB concentrations were converted to ng/g lipid weight (lw).

Statistics

We used generalized linear models (GLMs) with a gaussian error distribution to test whether CORT (baseline and stress-induced levels) and Σ_7PCB concentrations were different between males and females for each species. As consequences, using males and females separately, we used GLMs with a gaussian error distribution to test whether Σ_7PCB concentrations were related to 1) baseline CORT levels and 2) stress-induced CORT levels. Because our purpose was to describe a general pattern between Σ_7PCB concentrations and CORT levels, we calculated geometric means for Σ_7PCB concentrations, baseline and stress-induced CORT levels. Toxicity data are essentially lognormally distributed and the geometric mean is more appropriate (Posthuma et al. 2001). Following visual inspection of the data we tested whether Σ_7PCBs were 1) linearly related to baseline CORT or 2) quadratically or linearly related to stress-induced CORT levels, again by using a GLM with a gaussian error distribution. Dependent continuous variables were log-10 transformed when necessary to achieve normality. All statistical analyses were performed using R 2.13.1 (R Development Core Team 2008).

RESULTS

Sex differences in baseline and stress-induced CORT levels

Baseline CORT levels were not different between sexes in any species (GLM, $F<2.7$, $P>0.115$). In glaucous gulls, females had higher stress-induced CORT levels than males (GLM, $F_{1,36}=4.3$, $P=0.045$), in snow petrels, kittiwakes and cape petrels stress-induced CORT levels were not different between females and males (GLM, $F<1.2$, $P>0.289$).

Sex difference in Σ_7PCBs concentrations

In kittiwakes, south polar skuas and glaucous gulls Σ_7PCB concentrations were significantly higher in males than in females (GLM, kittiwakes: $F_{1,23}=8.7$, $P=0.007$; south polar skuas: $F_{1,29}=4.2$, $P=0.048$; glaucous gulls: $F_{1,36}=9.4$, $P=0.004$). In snow petrels, wandering albatrosses and cape petrels Σ_7PCBs concentrations were not different between females and males (GLM, $F<2.8$, $P>0.108$).

Relationships between Σ_7PCBs concentrations and CORT levels

Statistics on the relationships between CORT levels (baseline and stress-induced) and Σ_7PCB concentration are given in **Table 2**. In male kittiwakes, both baseline and stress-induced CORT levels significantly increased with increasing Σ_7PCB concentrations (**Table 2, Figure 1J-2G**). Positive trend were observed between baseline CORT levels and Σ_7PCB concentrations in female wandering albatrosses (**Table 2, Figure 1C**) as well as CORT stress-induced levels and Σ_7PCB concentrations in male snow petrels (**Table 2, Figure 2F**). Moreover, a significant negative relationship was found between stress-induced CORT levels and Σ_7PCB concentrations in male glaucous gulls (**Table 2, Figure 2I**). In common eiders, cape petrels and south polar skuas CORT (baseline and stress-induced) levels were not related to Σ_7PCB concentrations.

With regard to the global analysis using the geometric means for Σ_7PCB concentrations and CORT levels (one point per species and sex); in females, a positive trend associated Σ_7PCB concentrations and baseline CORT levels (GLM, Σ_7PCB, $F_{1,5}=4.2$, $P=0.095$, **Figure 3A**). Stress-induced CORT levels were not associated toΣ_7PCB concentrations, to $(\Sigma_7\text{PCB})^2$ nor to $(\Sigma_7\text{PCB}) \times (\Sigma_7\text{PCB})^2$, (GLM, $F_{1,3}=1.1$, $P=0.378$; $F_{1,3}=2.7$, $P=0.201$; $F_{1,3}=3.8$, $P=0.147$, respectively, **Figure 3B**). Significant relationships were observed in males only. Specifically, Σ_7PCB concentrations were positively associated to baseline CORT levels (GLM, $F_{1,4}=14.3$, $P=0.019$, **Figure 3C**) and negatively associated to stress-induced CORT levels (GLM, $(\Sigma_7\text{PCB})$: $F_{1,2}=59.1$, $P=0.016$ **;** $(\Sigma_7\text{PCB})^2$: $F_{1,2}=87.4$, $P=0.011$; $(\Sigma_7\text{PCB}) \times (\Sigma_7\text{PCB})^2$: $F_{1,2}=73.4$, $P=0.013$; **Figure 3D**).

DISCUSSION

This is, to our knowledge, the first study that comprehensively investigates the relationships between circulating CORT levels and PCB levels in multiple seabird species feeding at various trophic positions and thus exposed to various PCB concentrations. Baseline CORT levels significantly increased as a function of Σ_7PCB concentrations in male kittiwakes and a positive trend was observed in female wandering albatrosses. Stress-induced CORT levels were positively related to Σ_7PCB concentrations in male kittiwakes and a positive trend was observed in male snow petrels whereas stress-induced CORT levels were negatively related to Σ_7PCB concentrations in male glaucous gulls. Interestingly, Σ_7PCB concentrations were found to be unrelated to baseline or stress-induced CORT levels in common eiders, cape petrels and south polar skuas. The general pattern including all seven seabird species showed, in females, a positive trend between baseline CORT levels and Σ_7PCB concentrations whereas stress-induced CORT levels were unrelated to Σ_7PCB concentrations. In males, baseline CORT levels generally increase with increasing blood $\sum_7$PCB concentrations, whereas stress-induced CORT levels decrease when reaching high blood $\sum_7$PCB concentrations. However, caution should be made when interpreting this general pattern since only seven seabird species were included in this analysis. And several factors could not be taken into account such as species-specific differences in hormone regulation, diet composition, biotransformation of contaminants, but also individual nutritional status, phylogeny and life-history traits (which could also influence PCBs and CORT) and differences in methodology.

In mammals and fish, the modes of action of contaminants on glucocorticoids, including PCBs have been studied extensively (Odermatt et al. 2006). For example, certain methyl sulfone-containing PCB metabolites act as antagonists on human glucocorticoid receptors (GR, Johansson et al. 1998). Moreover, oral administration of a commercial PCB mixture resulted in a depression of the number of GR in the brain of Arctic charrs *Salvelinus alpinus* (Aluru et al. 2004).

The increase in baseline CORT levels with increasing Σ_7PCB concentrations in the present study may be explained based on cytochrome P450 (CYP)-mediated enzymes activity. Some contaminants including PCBs have been shown to inhibit/stimulate CYP enzymes in the steroidogenesis pathway. Hence, PCBs may inhibit the metabolism of CORT (to aldosterone), thus elevating CORT, or stimulate metabolism of desoxy-CORT (to CORT), thus also elevating CORT (Xu et al. 2006). However, in captive American kestrels, the reverse pattern was found:

oral PCB administration resulted in lower levels of baseline CORT concentrations compared to a control group, and liver PCB concentrations were associated with baseline CORT levels with an inverted U-shaped pattern (Love et al. 2003). Furthermore, baseline CORT declined when liver PCB concentrations reached 20 µg/g ww. Love et al. (2003) discussed this inverted U-shaped pattern as an apparent hormetic response of CORT to PCBs (Calabrese and Baldwin, 1999): adrenal monoxygenase (P-450 family) have the capacity to metabolize contaminants and may have produced toxic metabolites. The inverted U-shaped pattern may result in long-term damage of these toxic metabolites to the adrenal cortex: "remaining intact cells still produce a hermetic baseline CORT response in relation to liver PCB concentrations; however because the cortex has been damaged, there are fewer cells overall resulting in baseline levels depressed below those of controls" (Love et al. 2003).

For stress-induced CORT levels, we found in our study positive associations in two species (snow petrels and black-legged kittiwakes). Two possible explanations could support these positive relationships between PCB (and organochlorine pesticides) and stress-induced CORT levels: either they increase the ability of the adrenal glands to release CORT or they decrease the negative feedback capacity of CORT on the hypothalamus or the pituitary. In kittiwakes the capacity of CORT to decrease post-stress episode has been measured by dexamethasone injection (a potent CORT agonist), and the CORT concentrations measured following dexamethasone injection were not related to ∑PCB concentrations nor to ∑organochlorine pesticides (Tartu et al. unpublished data). However, the CORT levels measured in kittiwakes following an ACTH injection were positively associated to ∑PCBs but not to ∑organochlorine pesticides (Tartu et al. unpublished data). This suggests that in kittiwakes, increasing ∑PCB concentrations may increase the adrenal sensitivity. ACTH is one of the few polypeptide hormones having a positive trophic effect on its own receptors (Beuschlein et al. 2001; Penhoat et al. 1989). Thus, an increase of ACTH-R in the most PCB-exposed birds may be the consequence of an excess of ACTH stimulation to the adrenals. Alternatively, it may be possible that PCBs mimic ACTH and activate ACTH-R or increase ACTH secretion; both cases would result in an increase of ACTH-R, however we have no experimental support for such interpretation. An enhanced CORT stress response in adult birds may favour survival at the expense of parental investment (Wingfield and Sapolsky 2003). Indeed, in kittiwakes and wandering albatrosses, even relatively low POP exposure was associated with a reduction in long-term breeding success (Goutte et al. 2014; Goutte at al. unpublished data). In male glaucous gulls, we found a negative association between stress-induced CORT levels and $\sum_7$PCB concentrations. In the present study, glaucous gulls' mean baseline CORT concentrations (10.8 ng/mL) were almost as high as that of CORT levels attained following a stressful episode (16.5 ng/mL). The relatively low stress-induced CORT levels in the most PCB exposed male glaucous gulls may suggest a permanent saturation of ACTH-R as a result of chronic elevation of baseline CORT. Chronic elevation of baseline CORT may result in an array of deleterious biological effects (Sapolsky et al. 2000) which can explain the negative effects of POPs on adult survival which have been reported in glaucous gulls (Erikstad et al. 2013). Interestingly, the strongest associations between CORT (baseline and stress-induced)levels and Σ_7PCB concentrations were only observed in males, which often bear higher levels of PCB compared to females. As suggested earlier, more species would be required to corroborate these patterns, although these may be confounded by several factors (e.g. differences in hormone regulation, diet composition, biotransformation of contaminants, individual nutritional status, phylogeny, life-history traits, differences in methodology, etc.) that would be necessary to investigate in future studies. Regardless, present meta-analysis investigation provides valuable insights onto the associations between CORT levels and $\sum$PCB concentrations in polar seabirds.

Additional controlled studies using a mechanistic approach are warranted to verify whether there is a causal linkage between PCB exposure and perturbation in CORT homeostasis in present seabirds such as wandering albatrosses, kittiwakes, snow petrels and glaucous gulls. Although present study focused solely on PCBs and polar seabirds, other contaminants such as brominated flame retardants have been shown to impair CORT levels (Verboven et al. 2010) and some bird species including gulls feeding in urban environment or raptors may be exposed to substantially higher contaminant levels (Chen and Hale, 2010; Gentes et al. 2012; Guerra et al. 2012). It is therefore crucial to better understand the effects contaminant exposure may have on CORT regulation, which may significantly impact the adaptability of free-ranging bird species in such a changing environment.

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Figure caption:

Figure 1: Relationships between baseline CORT (ng/ml) and log-transformed $\sum_7$PCBs (ng/g lw) in female common eiders (A, COEI), female and male snow petrels (B, H; SNPE), wandering albatrosses (C, I; WAAL), kittiwakes (D, J; BLKI), cape petrels (E, K; CAPE), south polar skuas (F, L; SPSK) and glaucous gulls (G, M; GLGU). Solid line refers to a significant linear regressions ($P=0.008$) and dashed line to a regression close to statistical significance ($P=0.053$). Closed triangles denote males and open circles females.

Figure 2: Relationships between stress-induced CORT levels (ng/ml) and log-transformed $\sum_7$PCBs (ng/g lw) in female common eiders (A, COEI), female and male snow petrels (B, F; SNPE), kittiwakes (C, G; BLKI), cape petrels (D, H; CAPE) and glaucous gulls (E, I; GLGU). Solid lines refer to significant linear regressions ($P<0.031$) and dashed line to a regression close to statistical significance ($P=0.078$). Closed triangles denote males and open circles females.

Figure 3: Relationships between log-transformed $\sum_7$PCBs (ng/g lw), baseline CORT levels (ng/ml) in A) seven female and C) six male seabird species; and stress-induced CORT levels (ng/ml) in B) five female and C) and D) four male seabird species. Data represent geometric means for $\sum_7$PCBs, baseline CORT and stress-induced CORT levels. Solid line refers to significant relationship ($P<0.05$) and dashed line to linear regression close to statistical significance ($P<0.10$). Closed triangles denote males and open circles females. COEI = common eider, SNPE = snow petrel; WAAL = wandering albatross; BLKI = kittiwake; CAPE = cape petrel; SPSK = south polar skua and GLGU = glaucous gull.

Table 1: Diet (see footnote references 1 to 7), parental care behaviour, as well as mean blood/plasma lipid content, body mass, and plasma concentrations of Σ_7PCBs, baseline and stress-induced CORT levels in females and males of seven seabirds species. First row values are mean (geometric for Σ_7PCBs) ± standard deviation (sd) and 2nd row range (min – max). Non-available data are referred to as 'na'.

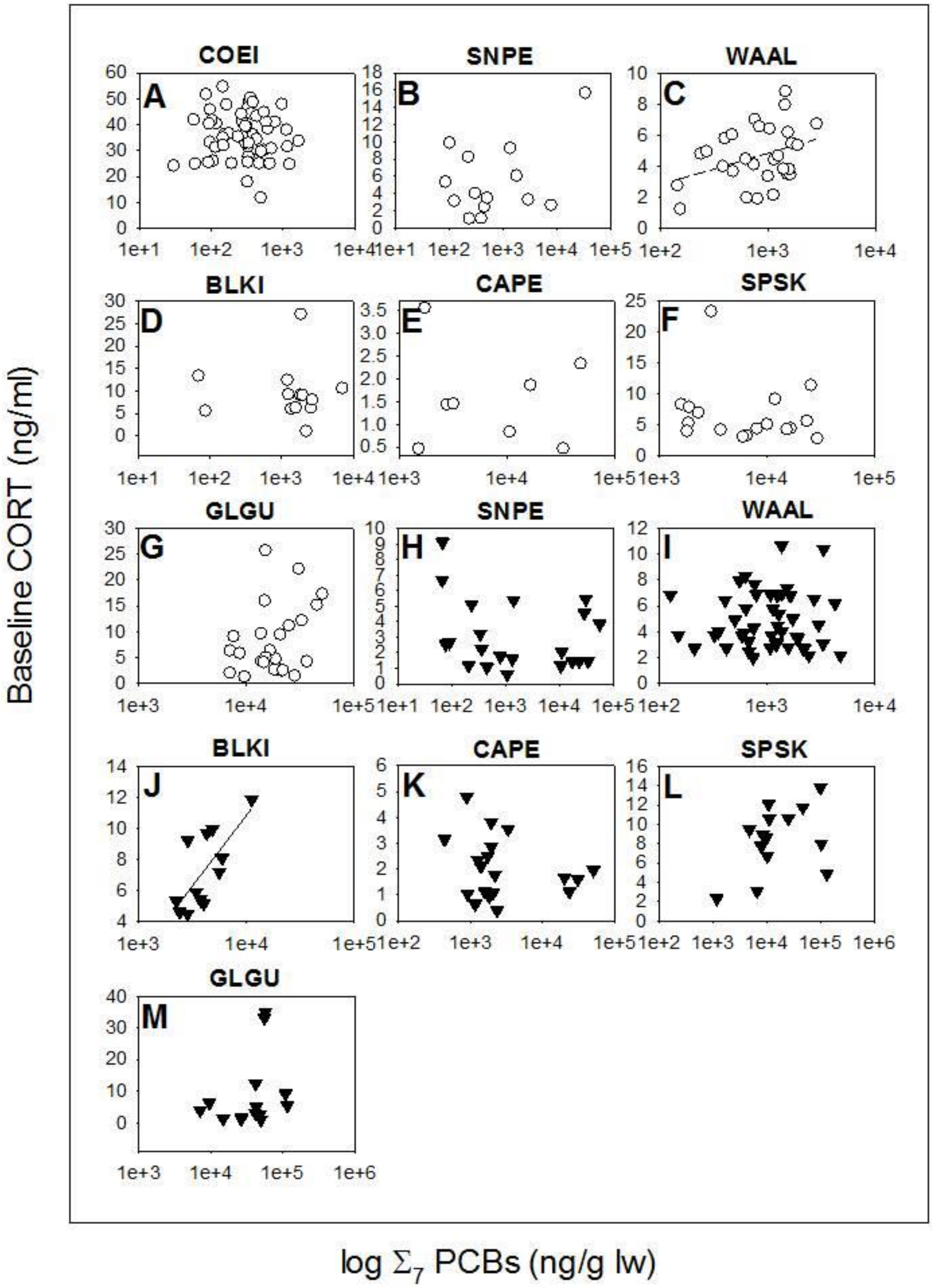
	Sex	GLGU	SPSK	SNPE	CAPE	WAAL	BLKI	COEI
Diet		Fish, other seabird species (adult, chicks, eggs) (1,5)	Fish, other seabird species (adult, chicks, eggs) (1,5)	Marine invertebrates, crustaceans, fish, carrion (1,3)	Marine invertebrates, crustaceans, fish, carrion (1,3)	Cephalopods, fish (6)	Marine invertebrates, fish (4,5)	Benthic mollusks, crabs, urchins (1,2,7)
Parental care		bi-parental	bi-parental	bi-parental	bi-parental	bi-parental	bi-parental	female only
Blood/plasma lipids (%)	Females	0.84 ± 0.22	0.6 ± 0.16	0.68 ± 1.11	0.2 ± 0.08	0.63 ± 0.12	0.26 ± 1.11	0.28 ± 0.09
		0.40 - 1.26	0.35 - 0.90	0.48 - 0.88	0.13 - 0.37	0.50 - 0.99	0.06 - 3.32	0.15 - 0.49
	Males	0.79 ± 0.15	0.49 ± 0.18	0.70 ± 0.12	0.21 ± 0.09	0.60 ± 0.11	0.12 ± 0.04	na
		0.54 - 1.0	0.24 - 0.78	0.50 - 0.94	0.10 - 0.51	0.38 - 0.94	0.07 - 0.23	na
Body mass (g)	Females	1397 ± 118.6	1495 ± 92.4	393 ± 55.3	433 ± 39.5	na	397 ± 15.9	1140 ± 112.9
		1180 - 1620	1325 - 1700	307 - 538	365 - 525	na	375 - 430	1274 - 1829
	Males	1755 ± 103.5	1342 ± 97.3	444 ± 47.4	510 ± 60.5	na	425 ± 20.7	na
		1530 - 1920	1140 - 1540	374 - 545	420 - 640	na	390 - 471	na
Σ_7PCBs (ng/g lw)	Females	17850 ± 11738	6358 ± 9113	660.2 ± 8904	7177 ± 17531	803.8 ± 622.1	2757 ± 3447	558.2 ± 668.4
		7089 - 51068	1604 - 29383	85.2 - 33666	1529 - 48695	144.0 - 2831	140.4 - 14125	60.6 - 3346
	Males	35357 ± 32392	15193 ± 42812	1531 ± 15406	2803 ± 13740	1017 ± 1055	7956 ± 4821	na
		7062 - 115,632	1162 - 128089	65.7 - 55119	432.1 - 51219	125.8 - 4769	4429 - 22165	na
Baseline CORT (ng/ml)	Females	13.6 ± 16.7	5.6 ± 4.9	4.1 ± 4.1	1.2 ± 1.0	4.3 ± 1.9	7.7 ± 6.2	6.0 ± 4.7
		1.2 - 25.7	2.7 - 23.2	1.1 - 15.7	0.5 - 3.6	1.2 - 8.8	1.0 - 27.0	0.6 - 27.0
	Males	10.8 ± 11.2	7.7 ± 3.3	4.9 ± 4.5	1.7 ± 1.2	4.4 ± 2.2	6.9 ± 2.5	na
		1.0 - 35.1	2.3 - 13.8	1.2 - 18.3	0.4 - 4.8	2.0 - 10.6	4.5 - 11.9	na
Stress-induced CORT (ng/ml)	Females	27.0 ± 11.7	na	39.1 ± 7.6	47.5 ± 7.6	na	40 ± 10.4	34.4 ± 9.2
		10.9 - 50.3	na	23.3 - 56.0	37.5 - 59.5	na	19.5 - 55.5	11.7 - 54.7
	Males	16.5 ± 15.4	na	38.2 ± 9.7	42.0 ± 11.5	na	37.6 ± 6.9	na
		1.3 - 59.5	na	22.6 - 56.4	20.7 - 61.2	na	23.7 - 48.5	na

(1)del Hoyo et al. 1992; (2)Guillemette et al. 1992; (3)Ainley et al. 1993; (4)Mehlum and Gabrielsen 1993; (5)del Hoyo et al. 1996; (6)Cherel and Klages 1998; (7)Varpe 2010. COEI = common eider, SNPE = snow petrel; WAAL = wandering albatross; BLKI = kittiwake; CAPE = cape petrel; SPSK = south polar skua and GLGU = glaucous gull.

Table 2: Relationships between log transformed $\sum_7$ PCB concentrations and A) baseline and B) stress-induced CORT levels in seven female and six male seabird species.

Independent variable: $\sum_7$ PCBs ng/g lw		Females				Males			
Dependent variable	Species	Df	F	P	Correlation	Df	F	P	Correlation
A) Baseline CORT	COEI	1,52	0.19	0.661		na	na	na	
	WAAL	1,27	4.10	0.053	(+)	1,44	0.02	0.886	
	BLKI	1,11	0.00	0.955		**1,10**	**10.83**	**0.008**	**(+)**
	CAPE	1,6	0.08	0.781		1,17	0.92	0.352	
	SNPE	1,12	2.11	0.172		1,19	1.33	0.263	
	SPSK	1,15	0.50	0.488		1,12	2.48	0.141	
	GLGU	1,22	2.89	0.103		1,12	1.23	0.290	
B) Stress-induced CORT	COEI	1,52	0.11	0.743		na	na	na	
	BLKI	1,11	1.00	0.339		**1,10**	**6.95**	**0.025**	**(+)**
	CAPE	1,6	0.43	0.535		1,17	0.00	0.970	
	SNPE	1,12	1.12	0.312		1,19	3.47	0.078	(+)
	GLGU	1,22	0.08	0.775		**1,12**	**5.95**	**0.031**	(-)

Numbers in bold are significant relationship ($P<0.05$). Directions are given for significant relationships and trends ($P<0.10$). COEI = common eider, SNPE = snow petrel; WAAL = wandering albatross; BLKI = kittiwake; CAPE = cape petrel; SPSK = south polar skua and GLGU = glaucous gull.



COEI
SNPE
WAAL
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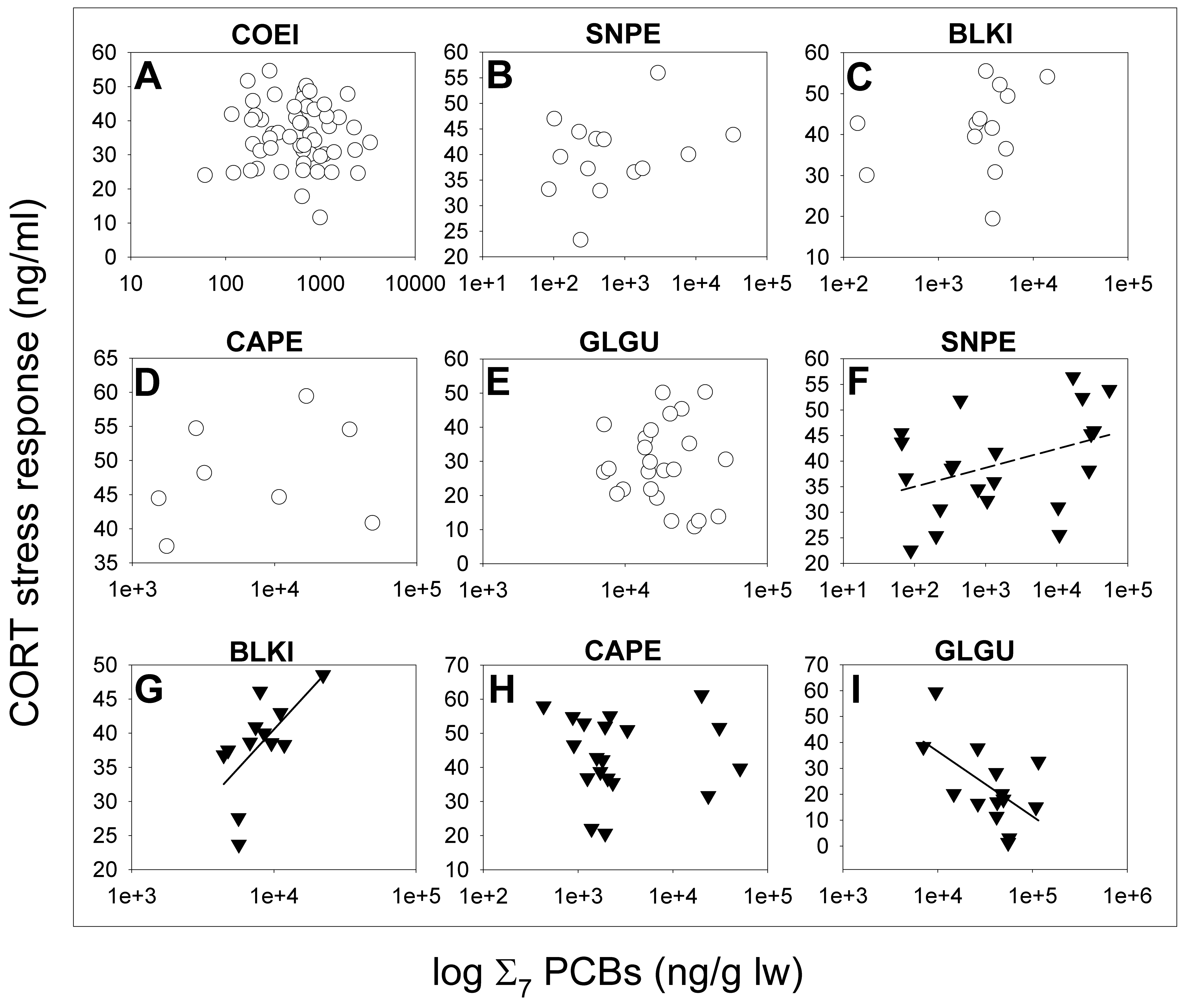
Baseline CORT (ng/ml)
log Σ7 PCBs (ng/g lw)

Figure 1



COEI
A
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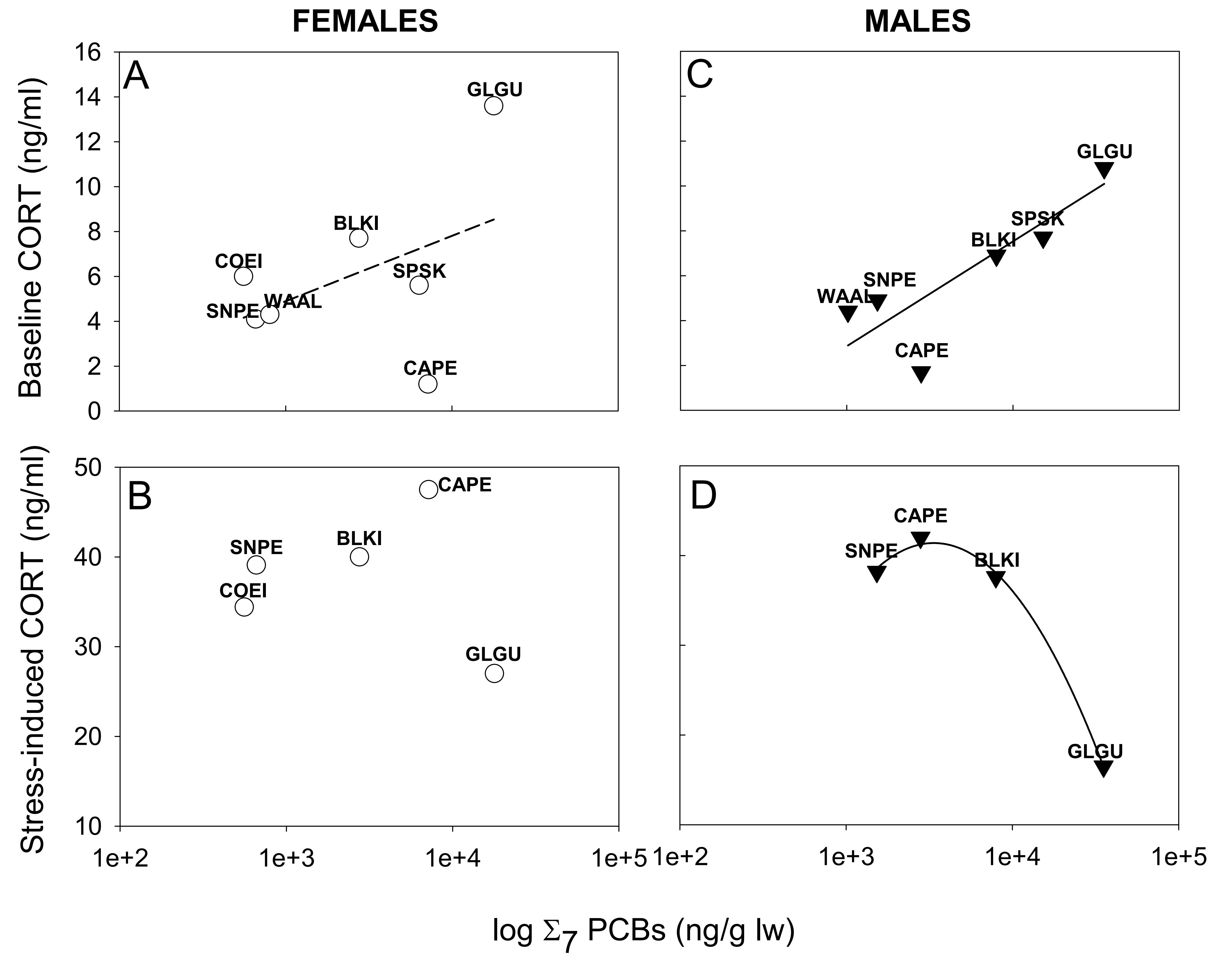
CORT stress response (ng/ml)
log Σ7 PCBs (ng/g lw)



FEMALES
MALES
Baseline CORT (ng/ml)
Stress-induced CORT (ng/ml)
A
B
C
D
GLGU
BLKI
COEI
SPSK
SNPE
WAAL
CAPE
1e+2
1e+3
1e+4
1e+5
0
2
4
6
8
10
12
14
16
10
20
30
40
50
log Σ7 PCBs (ng/g lw)