THE EFFECT OF MIGRATION AND FASTING ON ORGANOCHLORINE CONTAMINANT BURDENS IN ANTARCTIC HUMPBACK WHALES

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Introduction

Persistent Organic Pollutants (POPs) were first detected in Antarctic wildlife in the 1960s[1] and continue to be found today [2]. Global fractionation of semi-volatile POPs has been proposed as the primary mechanistic pathway supplying polar regions with contaminant input [3, 4].

The dependence of polar species on lipid rich diets to survive the temperature and productivity extremes of high latitude environments makes them particularly susceptible to bioaccumulation of these often highly lipophilic chemicals. Marine mammals are at the greatest risk of accumulating toxic levels of these chemicals due to their high trophic level, longevity and high proportion of body fat.

Whilst Arctic marine mammals have come under scientific scrutiny, there has been a comparative lack of POP research on Antarctic biota [5]. Of notable significance are the few reports concerning Antarctic baleen whales [6, 7].

Southern hemisphere humpback whales (*Megaptera novaeangliae*), feed in the Southern Ocean and overwinter in tropical breeding grounds, seasonally undergoing the longest migration and associated fasting events known in any mammal. No peer-reviewed reports currently exist on levels of POPs in southern hemisphere populations, necessitating baseline data. The extreme life history behavior of these populations provide a unique opportunity to study the toxicokinetics of POPs during a period of chronic energy deficit. Previously, medical research has evidenced the toxic effects associated with rapid weight loss and concomitant mobilization of POP burdens [8, 9] Seasonal mobilization of blubber POP fractions during prolonged periods of lipid depletion, may place humpback whales in a higher chemical risk category to that commonly attributed baleen whales [10].

This study provides a comprehensive baseline for monitoring accumulation of POPs in the species, whilst providing insight into the toxicokinetics of POP burdens in wildlife during prolonged periods of fasting and migration.

Materials and methods

Sample collection. Biopsy sampling occurred in Moreton Bay Marine Park, North Stradbroke Island, South East Queensland, Australia (approximately 27° 26 S, 153° 34 E). Forty-one skin and blubber biopsies were collected from free-swimming males as described in detail elsewhere [11]. Biopsies were collected at two time points during the annual migration and are categorized into two cohorts: 1) "early migration" (n=21): individuals sampled on the northward leg of the journey, travelling to breeding grounds (sampled in June/July) and; 2) "late migration" (n=20): individuals on the southward leg of the journey, returning to Antarctic feeding grounds following 4-9 months of migration associated fasting (sampled September/October). Sampling Samples were stored at -20^{o} C in pre-cleaned amber vials until the time of analysis. Samples from stranded animals (n=9) were opportunistically collected between 2006 and 2009 as described in [12].

Chemical analysis. All blubber samples were analyzed for 32 polychlorinated biphenyls (PCBs), and 25 organochlorine pesticides including: chlorobenzenes; the dichlorodiphenyltrichloroethane (DDT) group; hexachlorocyclohexanes (HCHs); chlordanes (Chlordane (cis-, trans-, oxy-), chlordene, Nonachlor (trans- and cis-)) and toxaphenes [12]). Eleven, of the total 41, blubber samples were also analysed for the additional cyclodines: Heptachlor-exo epoxide, Heptachlor-endo epoxide, Dieldrin, Eldrin, Isodrin, Endrin Endosulfan I, Endosulfan II, and Endosulfane-sulfate. Contaminant values are reported in nanograms per gram (ng/g) lipid. Samples were extracted and underwent a series of clean up steps, quantification was carried out via high

resolution gas chromatography coupled to a high resolution mass spectrometer, and quality control measures were adhered to. The procedure is described in full elsewhere [12].

Statistical analysis. Differences in absolute POP levels were examined by comparing POP concentrations expressed on a lipid-normalized basis. POP data was log transformed to maintain the homogeneity of variance and Principal Component Analysis was performed (PCA). Concentrations of POPs in the humpback whales were grouped by migratory status (i.e. early or late) and the group means were analysed by a one-way Analysis of Variance (ANOVA). For all tests statistical significance was set at $p < 0.05$.

Results and discussion

Levels of POPs in Antarctic humpback whales.

A summary of analytical findings are presented in Table 1. Hexachlorobenzene (HCB) had the highest average concentration (ng/g lipid; mean ± SD) (64.4 ± 7.7) followed by ΣDDT (25.7 ± 3.6) > ΣToxaphenes (22 ± 2.8) > CHLs (18.8 ± 5) > ΣHCHs (15.5 ± 7.9) > ΣPCBs (3.08 ± 0.7). HCB was detected in all samples analysed. *para,para'*-dichlorodiphenylethene (*p,p*'DDE), the main metabolite of DDT, accounted for 53.3% of total DDT and was detected at an average concentration of 13.8 ± 3.5 ng/g lw and was detected in 92.7% of samples analysed. Of the 32 PCB congeners analyzed 15 were detected in free-swimming humpback whales (n = 41) at levels above 0.05 ng/g lw. The greatest contributions were from PCB 153 (0.6 ± 0.1 ng/g lw; n=41) and PCB 101 (0.6 ± 0.1 ng/g lw; n=41), which accounted for 17.9% and 18.4% of total PCB burden respectively. Overall ΣPCBs were found at an average concentration of 3.08 ± 0.7 ng/g lw (n=41). Cyclodienes were not quantified in all samples but were analyzed in seven free-swimming males with notable levels of dieldrin (38.4 ± 15.7 ng/g lw), endrin (38.3 ± 9.2) and high levels of heptachlor-endo-epoxide (90.7 ± 12 ng/g lw).

Stranded animals generally had higher levels of POPs than free-swimming animals. One free-swimming calf was also opportunistically sampled. Levels found in the calf were consistently higher than the average level found in adults. Notable were the higher concentrations of HCB (110 ng/g lw) and heptachlor-endo-epoxide (219.3 ng/g lw).

Table 1: Organochlorine contaminant burdens detected in humpback whale blubber (ng/g lipid; mean ± SD)

Compound	Free-swimming (*n* = 41)	Stranded (*n* = 9)	Calf (*n* = 1)
ΣPCB_{32}	3.08 ± 0.7	5.6 ± 0.3	3.6
ΣHCHs (α-, β-, ϒ-)	13.9 ± 0.3	19.6 ±3.4	n.d.
ΣDDT_6	25.7 ±3.5	77.7 ± 27.4	11.4
HCB	64.4± 7.7	144.5± 51.9	110.1
ΣCHL_7	18.7 ± 5.2	79.3 ± 2.1	29.3
$\Sigma Toxaphene_7$	22 ± 2.8	330.7 ± 31.9	66.0
Dieldrin[a]	38.4 ± 15.7	57.8 ± 24.6	28.9
Aldrin[a]	4.9 ± 1.7	29.4 ± 10.2	13.8
Isodrin[a]	4.8 ± 1.6	21.8 ± 9.7	11.9
Endrin[a]	38.3 ± 9.2	101.4 ± 67.7	89.4
Heptachlor-exo-epoxide[a]	3.1 ± 0.5	3.4 ± 3	4.6
Heptachlor-endo-epoxide[a]	90.5 ± 12	1.6 ± 1.4	219.3
Endosulfan-I[a]	5.6 ± 2.6	18.5 ± 4.3	6.1
Endosulfan-II[a]	4.6 ± 0.5	n.d	11.1
Endosulfan-sulphate[a]	7.7 ± 3.4	n.d.	97.3

The Effect of Migration and Fasting on POPs

Principal Component Analysis (PCA) represented a comprehensive approach to exploring the toxicokinetics of POPs during the fasting event. The clustering of the two groups (i.e. early [n = 21] and late [n= 20] migration) evidenced a significant difference between groups and demonstrated the observed lipid depletion and lipid normalized concentration effect of contaminants in the late migration cohort (Figure 1). One-way ANOVAs showed that the two dominant congeners, *p,p'*DDE and HCB, exhibited a significant concentration effect in late migrating cohorts (ng/g lipid; mean ± SD) (*p,p'* DDE: early 10.8 ± 1.8, late 17.8 ±3.2, $F(1, 40) = 4.1$, $P < 0.05$; HCB: early 52.0 ± 7.9, late 81.7 ±13.1; $F(1, 40) = 3.8$, $P < 0.05$; Figure 2). No significant concentration effect was evidenced for any other compound. As neither recapture of single individuals, nor age determination is feasible for this species and population, concentration effects for compounds detected only at minor levels would likely have been obscured by inter-individual variability.

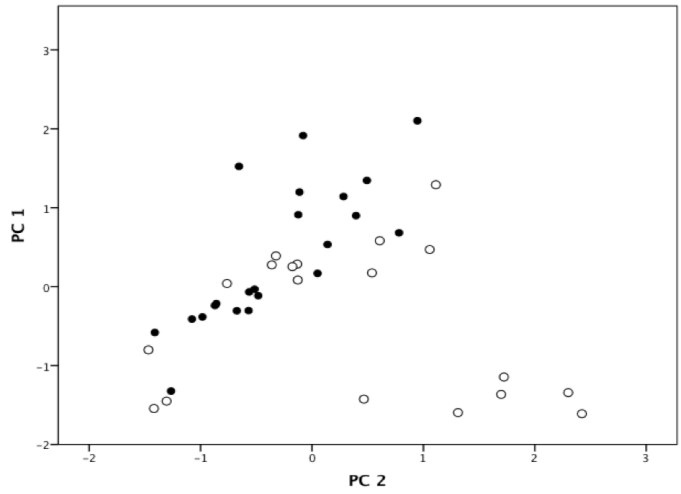

Figure 1: Principal Component Analysis (PCA) of early migration (black circles) and late migration (white circles) cohorts of humpback whales.

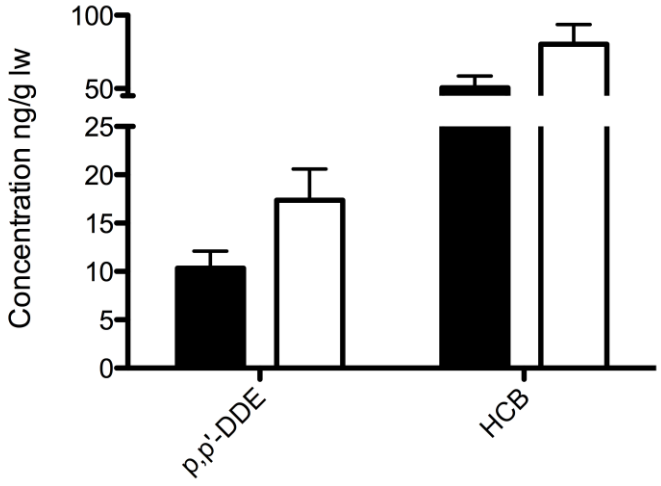

Figure 2: Concentration (ng/g lipid weight; Mean ± SD) of *p,p'*-DDE and HCB in humpback whale blubber at two time points on the annual migration (early: black bars; late:white bars).

This study provides the first evidence of fluctuating blubber organochlorine concentrations in Antarctic humpback whales as a function of seasonal migration and fasting events. The increasing blubber concentrations and concomitant mobilization of lipophilic chemicals during times of energy deficit demonstrates the need for careful consideration of nutritional adaptations or stress in the assessment of chemical risks posed to wildlife.

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