

Concentrations of organochlorines in sperm whales (*Physeter macrocephalus*) from Southern Australian waters

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Abstract

Concentrations of DDTs, PCBs and HCHs were measured in sperm whales involved in two mass stranding events on the west coast of Tasmania, Australia in February 1998. DDTs and PCBs were present in all samples analysed, while only three contained HCHs. The relationships between organochlorines, sex, age and reproductive groups were marked by high variability. Differences in organochlorine concentrations were observed between animals from the two stranding sites and discussed in light of the ecology of this species. Concentrations of all pollutants were stratified throughout the vertical aspect of the blubber and possible reasons for and the implications of this are discussed.

Concentrations of compounds were higher than those documented in this species in the Southern Hemisphere previously, although were relatively lower than those documented in the Northern Hemisphere. However, comparisons were confounded by spatial and temporal differences. Continued monitoring of marine mammals throughout this region in a co-ordinated, standardized manner is essential for establishing definite temporal and spatial variations in pollutant concentrations.

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1. Introduction

Pollution is one of a number of contemporary environmental problems that face marine mammals (Johnston et al., 1996). While the majority of man-made toxic compounds are no longer produced in most developed countries, they are still produced in developing nations and are in use globally (Tanabe et al., 1994; Simonich and Hites, 1995). Many of these pollutants, particularly the organochlorines, are characterized by their chemical stability and resistance to metabolic degradation. As a result, they are readily taken up into food webs and accumulate with increasing trophic levels.

Organochlorines are transported globally as a function of atmospheric circulation, primarily involving movement from low latitudes to high latitudes, with the open ocean serving as their ultimate repository (Iwata et al., 1993; Tanabe et al., 1983). Their resilience and this

form of transportation suggest that at least in the near future, levels will not decline, but may eventually reach a global equilibrium (Wania and Mackay, 1995) as pollutants move away from source points. This has consequences for those areas that are removed from the sources of pollutant production and in the past have been characterized by low pollutant concentrations, such as the high latitudes and the Southern Hemisphere in general.

Some cetacean species bioaccumulate organochlorine pollutants as a result of their longevity, location towards the top of the food chain, the ability of their blubber layer to store up to 90% of whole body burdens and their limited ability to metabolize many of these compounds (Tanabe et al., 1981, 1988; Jepson et al., 1999). Of these compounds, the polychlorinated biphenyls (PCBs) and dichlorodiphenyltrichloroethane (DDT) and its metabolites dichlorodiphenyldichloroethylene (DDE) and dichlorodiphenyldichloroethane (DDD) present the largest problem to marine mammals (Borrell and Aguilar, 1993; Reijnders, 1994). These organochlorines have been associated with deleterious effects on the immune, endocrine and nervous systems of

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pinnipeds and cetaceans, resulting in disruption in growth, development, reproductive impairment and resistance to disease (Béland et al., 1993; Reijnders, 1994; Reijnders and Ruiters-Dijkman, 1995; Ross et al., 1996; Skaare et al., 2000). Deleterious effects on the immune system and subsequent lowering of the ability to resist disease have been suggested as possible factors involved in mass mortality events and strandings (Aguilar and Borrell, 1994a; Joiris et al., 1997; Jepson et al., 1999; Tilbury et al., 1999). However, directly associating contaminant concentrations and toxicity is difficult due to a number of confounding factors. Age, sex, the diet and condition of an individual, individual and species-specific ability to metabolize and excrete pollutants all have considerable effects on concentrations (Evans, 2003). Many samples used in pollutant studies have been derived from single strandings, bycaught animals or animals harvested in whaling operations. For a large proportion of single-stranded animals, the cause of death is unknown but may be associated with disease. Consequently, concentrations of pollutants in these samples may not reflect that of the healthy population (Aguilar et al., 1999). Sex ratios and age structure are often biased in bycaught animals and those from the whaling industry, thereby hampering the identification of patterns of pollutant concentrations throughout populations. Mass-stranded animals are thought to be largely free of biases associated with disease and therefore represent the best source of unbiased, comprehensive samples. However, pollutant studies utilizing mass-stranded animals (other than those associated with viral epidemics) are sparse.

Oceanic species of cetaceans such as sperm whales (*Physeter macrocephalus*), as part of open ocean ecosystems distanced from point sources of pollutants, can be regarded as removed from the short term changes in pollutant concentrations typical of inshore areas (Aguilar, 1985; Reijnders, 1986). Female sperm whales are distributed in waters from the equator to around 40° S (Rice, 1989), with individual groups ranging throughout areas in the order of 600 × 600 nautical miles. Within this area, they may move up to 55 nautical miles per day searching for food (Jaquet et al., 2000). Differences in the chemical profiles and concentrations of pollutants in different water masses should be reflected in the pollutant load of animals that feed in those areas (Aguilar, 1987; Aguilar et al., 1993). Therefore, concentrations of organochlorine compounds in sperm whales should be reflective of broad scale regional oceanic levels of pollution unbiased by variable, localised concentrations of pollutants close to point sources.

Three mass strandings of sperm whales on the west and north coasts of Tasmania, Australia in 1998 (Evans et al., 2002) provided a unique opportunity to investigate (i) levels of organochlorines within this species, thereby contributing to the sparse documentation of

these compounds in this region; (ii) variation in the levels of these compounds with age and sex as well as between stranding groups; (iii) the relationship between blubber lipid content and concentrations of compounds in this species and (iv) possible stratification of organochlorines within the blubber across females and males and the implications of any stratification to inter-study comparisons in this species.

2. Materials and methods

2.1. Sample collection

Samples ($n = 37$) of 50–100 g blubber encompassing the complete depth were collected from individuals involved in two mass strandings in February 1998 (STR1: Ocean Beach, Strahan, $n = 25$ and STR2: Greens Pt. Beach Marawah, $n = 12$). These were part of a series of three mass strandings of this species that occurred on the west and north coasts of Tasmania in 1998 (Evans et al., 2002; Fig. 1).

All samples were taken from the dorsal area in line with the posterior insertion of the flipper between 48–72 h after death. All samples were wrapped in aluminium foil, stored in ice on site and then frozen at -20°C on return to the University of Tasmania.

The sex and total length of all animals [tip of upper jaw to deepest notch in fluke taken in a straight line dorsally (Norris, 1961)] were recorded. Each individual

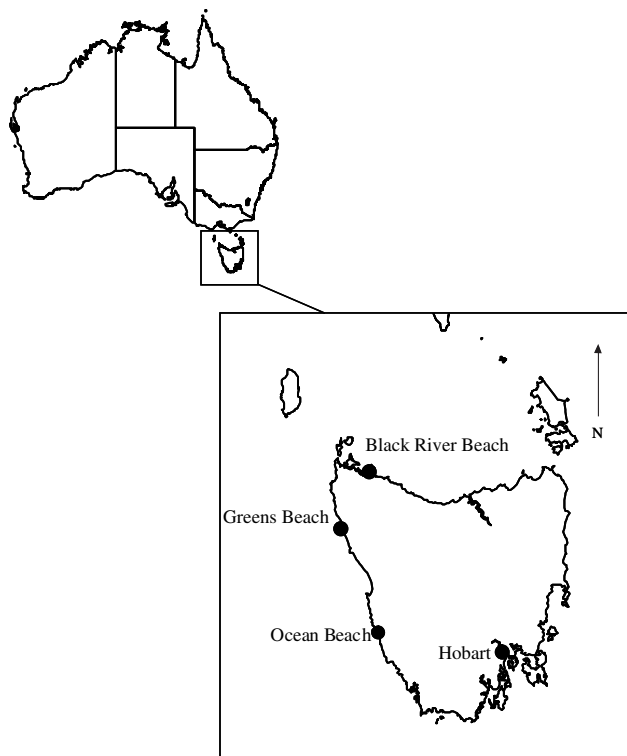


Fig. 1. Location of sperm whale strandings, Tasmania, Australia 1998.

was assigned an age based on counts of the number of growth layers in a tooth taken from that animal (Evans and Robertson, 2001; Evans et al., 2002). Lactation status was determined by applying pressure to teats and through the identification of the presence of milk via the in situ dissection of mammary glands.

2.2. Sample analyses

2.2.1. Stratification of samples

The lipid content of the blubber of sperm whales has been found to vary with depth (Lockyer, 1991; Evans et al., 2003). To investigate whether organochlorine concentrations followed the same stratification patterns as a consequence of their lipophilic nature, a subsample ($n = 10$) of blubber samples from STR2 encompassing the total depth of blubber were sectioned into three parts following Lockyer (1991). These were then treated as separate samples throughout the analysis.

2.2.2. Extraction of lipid fraction of blubber

Subsamples of each blubber sample were cleaned (outer surfaces of the sample were removed) and approximately three grams (mean: 2.9 ± 0.3 g) were weighed to ± 0.01 g. These were roughly chopped and ground in a mortar and pestle with anhydrous sodium sulphate and placed into a 125 ml Soxhlet apparatus to which 80 ml of *n*-hexane was added for 4 h. Extracts were transferred to a hot water bath and evaporated to 40 ml and then divided into two portions: 30 ml for pollutant analysis and 10 ml to determine lipid content (Evans et al., 2003).

2.2.3. Determination of blubber lipid content

The 10 ml extract was transferred to a previously tared vial and the solvent evaporated under a stream of N_2 . After returning to room temperature the total lipid content of the sample determined gravimetrically. Lipid content was expressed as a percentage of the wet weight of the tissue after accounting for the division of the fat extract volume.

2.2.4. Determination of organochlorine concentrations

Each 30 ml extract was cleaned using sulphuric acid following the procedures in Murphy (1972). After phase separation, the cleaned extract was concentrated to 1 ml and 100 μ l of an internal standard (tetrachlorobenzene for the PCBs and deuterated PAH for the HCHs and DDTs) was added. Each extract was analysed for *p,p'*-DDT, *p,p'*-DDE, *p,p'*-DDD, α -HCH (benzene hexachloride, also known as hexachlorocyclohexane), β -HCH, γ -HCH (lindane) δ -HCH and the seven PCB congeners identified on the ICES primary list (CBs 28, 52, 101, 118, 138, 153 and 180).

For the identification and quantification of PCBs, each sample was injected into a Varian 3400 gas chromatograph with a 1079 split/splitless injector (injector temperature 290 °C), equipped with an electron capture detector (temperature 300 °C). The column was a Chrompack CP58-60 column of 0.25 mm internal diameter, 30 m length and a stationary phase CP-SIL 8 CB low bleed with a film thickness of 0.25 μ m. Helium was used as the carrier gas and the injection volume was 1 μ l with the injector operating in the splitless mode. Temperature was programmed according to the following sequence: injection at 60 °C; oven held for the first minute and then increased by 10 °C min^{-1} to 300 °C. The oven was then held at this temperature for 10 min. The autosampler used was a Varian 8200CX. PCB congeners were selected from those present in Aroclor 1254 based on their relative peak size and isolation from interfering peaks. PCBs in the samples were identified using individual congeners and quantified using congeners in Aroclor 1254 and additionally with individual congeners.

A Varian CP-3800 gas chromatograph with a 1079 split/splitless injector (injector temperature 290 °C) and a Varian Saturn 2000 ion trap mass spectrometer were used for the identification and quantification of *p,p'*-DDT and its metabolites and the four HCH isomers. The column was a J&W DB5MS column of 0.25 mm internal diameter, 30 m length and a stationary phase DB5 low bleed with a film thickness of 0.25 μ m. Column temperature was initially 40 °C and was increased at 10 °C min^{-1} until it reached 290 °C where it was held for 8 min. There was a constant column flow of 1.0 ml min^{-1} and the autosampler used was a Varian 8200CX. Software used for data analysis and calculations was the Varian Saturn GC/MS workstation version 5.41. Detection limits were 0.1 μ g ml^{-1} .

Standards containing the analytes of interest were prepared at concentrations of 1, 10 and 100 μ g ml^{-1} . These were analysed at the same time as the samples and were used to quantify the analytes by relative retention times and mass spectral confirmation. Procedural blanks were included in each run during the course of analysis. Extraction efficiencies calculated by running a series of spiked samples for all analytes were above 85%.

All materials used during the analytical process were cleaned between each sample to prevent cross-contamination. All concentrations are presented on the basis of the extractable lipid content of each sample. Presenting results on this basis accounts for at least part of the heterogeneity associated with the nutritive condition of the animals sampled and the methods used for lipid extraction (Aguilar et al., 2002).

We found during analysis that the PCB congener 138 co-eluted with *p,p'*-DDT, and although longer running times helped to separate out compounds, final

concentrations were probably still biased to some degree by the co-elution. Consequently, concentrations of this congener were not used in statistical analyses.

2.2.5. Statistical analyses

Probability plots were used to test data for normality and due to non-normality all organochlorine concentration data were log-transformed. The relationships between organochlorine concentrations on a wet weight basis and blubber lipid content were investigated via linear regression. Body mass is thought to influence pollutant levels in individuals (Aguilar et al., 1999). In order to investigate this relationship, length data were used to calculate estimates of weight for all individuals ($W_i = 0.006648L_m^{3.18}$; Lockyer, 1981). Organochlorine concentrations were regressed against estimated weight and age to determine any possible relationships.

Because pollutants are known to accumulate in other cetaceans with age (Aguilar et al., 1999), it was important to account for possible age effects when testing for differences between groups of animals (e.g. stranding, sex). Differences between stranding groups were investigated using a multivariate general linear model (GLM) with age as a covariate. Similarities in concentrations between reproductive groups (non-lactating, lactating and immature females), age groups and sexes were tested using a one-way Analysis of Similarity (ANOSIM).

Animals were assigned to age groups based on approximate maturity of individuals (Lockyer, 1981; Rice, 1989). These corresponded to (1) juvenile or

immature (female: ≤ 13 years; male: < 19 years; $n = 5$); (2) sexually mature but not physically mature (female: $> 13 \leq 30$ years; male: $\geq 19 \leq 35$ years; $n = 9$) and (3) sexually and physically mature (female: > 30 years; male: > 35 years; $n = 20$). Three animals were not aged and all males were less than 23 years.

Concentrations of organochlorines were compared between stranding groups via backwards stepwise discriminant function analysis (DFA) using a Wilke's Lambda method at the 95% significance level.

To determine if pollutant concentrations were stratified with depth throughout the blubber layer, log-transformed data from the three strata were compared using a repeated measures ANOVA.

Where concentrations were presented as 'not detected' (ND) the midpoint between zero and the detection limit ($0.1 \mu\text{g ml}^{-1}$) was used for statistical analyses.

3. Results

DDTs and PCBs were present in all of the 37 samples analysed (Tables 1–3). Σ DDT concentrations ranged from 0.2 to $9.4 \mu\text{g g}^{-1}$ lipid weight (mean = 1.9 ± 2.2) while Σ PCB ranged from 0.3 to $3.3 \mu\text{g g}^{-1}$ lipid weight (mean = 0.9 ± 0.6). Σ HCH concentrations were substantially lower than DDT and PCB concentrations, ranging from below detection levels to $0.3 \mu\text{g g}^{-1}$ lipid weight (mean = 0.01 ± 0.04). Both β - and δ -HCH isomers were absent from or below detection limits in all

Table 1
Mean concentrations \pm SD of HCHs in southern Australian sperm whales

	<i>N</i>	Lipid content (%)	α -HCH	β -HCH	γ -HCH	δ -HCH	Σ HCH
All	37	49.2 \pm 17.9	0.003 \pm 0.01	ND	0.01 \pm 0.04	ND	0.01 \pm 0.04
STR1	25	55.3 \pm 18.0	0.003 \pm 0.01	ND	0.01 \pm 0.1	ND	0.01 \pm 0.1
STR2	12	36.5 \pm 8.6	0.002 \pm 0.01	ND	ND	ND	0.002 \pm 0.01
All females	32	50.4 \pm 18.1	0.003 \pm 0.01	ND	0.01 \pm 0.1	ND	0.01 \pm 0.1
Female	3	56.8 \pm 27.8	ND	ND	ND	ND	ND
age group 1							
Female	4	49.1 \pm 30.2	ND	ND	ND	ND	ND
age group 2							
Female	21	50.9 \pm 16.0	0.003 \pm 0.01	ND	0.01 \pm 0.1	ND	0.02 \pm 0.1
age group 3							
Non-lactating females	16	44.4 \pm 14.1	0.01 \pm 0.02	ND	ND	ND	0.01 \pm 0.02
Lactating females	2	41.8 \pm 9.9	ND	ND	ND	ND	ND
All males	5	41.3 \pm 15.6	ND	ND	ND	ND	ND
Male age group 1	2	44.6 \pm 26.9	ND	ND	ND	ND	ND
Male age group 2	3	42.3 \pm 12.1	ND	ND	ND	ND	ND
Range	37	16.2–89.3	ND–0.1	ND	ND–0.3	ND	ND–0.3

All values in $\mu\text{g g}^{-1}$ lipid weight.
ND: not detected.

Table 2
Mean concentrations \pm SD of DDTs in southern Australian sperm whales

	<i>N</i>	<i>p,p'</i> -DDD	<i>p,p'</i> -DDE	<i>p,p'</i> -DDT	Σ DDT	<i>p,p'</i> -DDE/ Σ DDT	Σ DDT/ Σ PCB
All	37	0.2 \pm 0.2	1.0 \pm 1.0	0.7 \pm 1.2	1.9 \pm 2.2	0.6 \pm 0.2	2.1 \pm 1.8
STR1	25	0.1 \pm 0.1	0.7 \pm 0.5	0.4 \pm 0.6	1.2 \pm 1.1	0.7 \pm 0.2	1.9 \pm 1.6
STR2	12	0.4 \pm 0.3	1.6 \pm 1.4	1.4 \pm 1.9	3.4 \pm 3.0	0.5 \pm 0.1	2.7 \pm 2.1
All females	32	0.2 \pm 0.2	0.9 \pm 0.7	0.7 \pm 1.3	1.7 \pm 1.9	0.6 \pm 0.2	2.1 \pm 1.9
Female	3	0.3 \pm 0.4	1.3 \pm 1.4	0.3 \pm 0.4	1.9 \pm 1.6	0.6 \pm 0.2	2.2 \pm 1.4
age group 1							
Female	4	0.1 \pm 0.1	1.1 \pm 0.8	0.2 \pm 0.2	1.4 \pm 0.8	0.7 \pm 0.2	1.4 \pm 0.5
age group 2							
Female	21	0.2 \pm 0.2	0.8 \pm 0.6	0.9 \pm 1.5	1.9 \pm 2.2	0.6 \pm 0.2	2.4 \pm 2.2
age group 3							
Non-lactating females	16	0.2 \pm 0.1	0.8 \pm 0.5	0.5 \pm 0.7	1.5 \pm 1.2	0.6 \pm 0.2	1.8 \pm 1.5
Lactating females	2	0.4 \pm 0.3	1.5 \pm 0.6	3.6 \pm 3.8	5.5 \pm 4.7	0.3 \pm 0.2	7.0 \pm 1.6
All males	5	0.3 \pm 0.4	1.8 \pm 2.1	1.0 \pm 1.1	3.1 \pm 3.6	0.7 \pm 0.2	2.1 \pm 1.0
Male age group 1	2	0.6 \pm 0.8	2.8 \pm 3.7	1.5 \pm 2.1	4.8 \pm 6.5	0.8 \pm 0.3	1.7 \pm 1.7
Male age group 2	2	0.2 \pm 0.02	1.2 \pm 0.7	0.7 \pm 0.1	2.1 \pm 0.6	0.6 \pm 0.2	2.6 \pm 0.4
Range	37	ND–1.1	0.2–5.4	ND–6.3	0.2–9.4	0.2–1.0	0.5–8.1

All values in $\mu\text{g g}^{-1}$ lipid weight.
ND: not detected.

Table 3
Mean concentrations \pm SD of PCBs in southern Australian sperm whales

	<i>N</i>	PCB28	PCB52	PCB101	PCB118	PCB153	PCB180	Σ PCB
All	37	0.03 \pm 0.02	0.1 \pm 0.04	0.003 \pm 0.02	0.3 \pm 0.2	0.3 \pm 0.2	0.2 \pm 0.1	0.9 \pm 0.6
STR1	25	0.03 \pm 0.03	0.1 \pm 0.03	ND	0.2 \pm 0.1	0.2 \pm 0.2	0.2 \pm 0.1	0.7 \pm 0.3
STR2	12	0.04 \pm 0.01	0.04 \pm 0.04	0.01 \pm 0.03	0.4 \pm 0.4	0.5 \pm 0.3	0.3 \pm 0.1	1.3 \pm 0.8
All females	32	0.03 \pm 0.03	0.1 \pm 0.04	0.004 \pm 0.02	0.2 \pm 0.2	0.3 \pm 0.2	0.2 \pm 0.1	0.8 \pm 0.4
Female	3	0.03 \pm 0.02	0.1 \pm 0.1	ND	0.4 \pm 0.5	0.4 \pm 0.4	0.2 \pm 0.1	1.0 \pm 1.1
age group 1								
Female	4	0.05 \pm 0.05	0.1 \pm 0.1	0.02 \pm 0.1	0.2 \pm 0.1	0.4 \pm 0.4	0.3 \pm 0.1	1.0 \pm 0.6
age group 2								
Female	21	0.03 \pm 0.02	0.04 \pm 0.03	0.001 \pm 0.01	0.2 \pm 0.1	0.3 \pm 0.1	0.2 \pm 0.1	0.8 \pm 0.3
age group 3								
Non-lactating females	16	0.04 \pm 0.03	0.1 \pm 0.04	0.01 \pm 0.02	0.2 \pm 0.1	0.3 \pm 0.2	0.2 \pm 0.1	0.8 \pm 0.4
Lactating females	2	0.03 \pm 0.01	0.02 \pm 0.03	0.02 \pm 0.02	0.2 \pm 0.2	0.3 \pm 0.2	0.2 \pm 0.1	0.7 \pm 0.5
All males	5	0.04 \pm 0.02	0.1 \pm 0.03	ND	0.4 \pm 0.5	0.4 \pm 0.4	0.3 \pm 0.2	1.3 \pm 1.2
Male age group 1	2	0.04 \pm 0.03	0.1 \pm 0.1	ND	0.8 \pm 0.9	0.7 \pm 0.8	0.3 \pm 0.3	1.9 \pm 2.1
Male age group 2	2	0.04 \pm 0.01	0.04 \pm 0.03	ND	0.2 \pm 0.03	0.3 \pm 0.1	0.3 \pm 0.2	0.8 \pm 0.4
Range	37	0.01–0.1	ND–0.1	ND–0.1	0.1–1.4	0.04–1.2	0.1–0.6	0.3–3.3

All values in $\mu\text{g g}^{-1}$ lipid weight.
ND: not detected.

samples analysed. Of the DDT isomers and PCB congeners, *p,p'*-DDE and the PCBs 28, 118, 153 and 180 were present in all samples analysed.

3.1. Concentrations of pollutants with respect to weight and age

Concentrations of *p,p'*-DDD, *p,p'*-DDE, the PCB congeners 118 and 153 and Σ PCB were found to be significantly negatively related to estimated weight only amongst animals involved in STR2 (*p,p'*-DDD: $r^2 = 0.5$, $F_{1,11} = 9.2$, $P = 0.01$; *p,p'*-DDE: $r^2 = 0.4$, $F_{1,11} = 6.2$, $P = 0.03$; PCB118: $r^2 = 0.7$, $F_{1,11} = 23.1$, $P = 0.001$; PCB153: $r^2 = 0.6$, $F_{1,11} = 18.0$; $P = 0.002$; Σ PCB:

$r^2 = 0.6$, $F_{1,11} = 16.9$, $P = 0.002$). Concentrations of all organochlorines were not related to estimated weight when only animals from STR1, all females and females of age group three were included.

Concentrations of the PCB 101 were found to be significantly negatively related to age amongst females in age group three ($r^2 = 0.3$, $F_{1,19} = 6.0$, $P = 0.03$). However, this organochlorine was only present above detectable levels in only one animal and as a result, this relationship cannot be regarded as accurate. In all other whales, concentrations of all HCHs, DDTs and PCBs were not related to age. An analysis of similarity with pairwise tests demonstrated no significant differences in organochlorine concentrations between age groups

Table 4

General linear model (with age as a co-variate) results from comparisons of pollutant concentrations between sperm whales from the two stranding sites

Compound	All (d.f. = 2)		All females (d.f. = 2)		Female age group 3 (d.f. = 2)	
	F-ratio	P	F-ratio	P	F-ratio	P
γ -HCH	0.4	ns	0.3	ns	0.3	ns
Σ HCH	0.4	ns	0.3	ns	0.3	ns
<i>p,p'</i> -DDD	1.9	ns	2.5	ns	0.5	ns
<i>p,p'</i> -DDE	5.0	0.01	1.2	ns	1.0	ns
<i>p,p'</i> -DDT	1.1	ns	0.9	ns	0.6	ns
Σ DDT	5.3	0.01	3.0	ns	2.0	ns
PCB28	2.2	ns	1.1	ns	0.7	ns
PCB52	0.1	ns	0.1	ns	0.04	ns
PCB101	3.3	0.05	3.7	0.04	3.7	0.04
PCB118	8.1	0.002	7.4	0.003	4.2	0.03
PCB153	6.5	0.005	5.0	0.02	3.7	0.04
PCB180	7.0	0.003	5.0	0.02	4.6	0.02
Σ PCB	8.9	0.001	7.7	0.003	4.9	0.02
<i>p,p'</i> -DDE/ Σ DDT	0.9	ns	1.7	ns	2.9	ns
Σ DDT/ Σ PCB	2.6	ns	0.3	ns	0.6	ns

ns: not significant.

(Global R: -0.06 ; Sample statistic: 78.8%; 5000 randomisations).

3.2. Concentrations of pollutants with respect to stranding groups

When comparing organochlorine concentrations between the two stranding sites, the age of individuals was found to have no effect ($F_{12,17} = 0.8$, $P = 0.6$). Individuals involved in STR2 contained significantly higher concentrations of the DDT isomer *p,p'*-DDE and Σ DDT and the PCB congeners 101, 118, 153, 180 and Σ PCB (Table 4). When only females were included, similar differences were observed in PCBs, however *p,p'*-DDE and Σ DDT were not significantly different between the two sites. Although γ -HCH was either absent or below detection limits in all animals involved in STR2, its presence amongst individuals from STR1 was also low: it was only detected in one individual. There were no significant differences in the ratios *p,p'*-DDE/ Σ DDT and Σ DDT/ Σ PCB between individuals from the two sites.

Discriminant function analysis correctly assigned 89.2% of all cases on the basis of stranding site. Cross-validation using a jackknife analysis also correctly assigned 89.2% of all cases to the correct stranding group.

3.3. Concentrations of pollutants with respect to sex and reproductive group

The blubber from male sperm whales differed from females in that no detectable concentrations of HCHs or the PCB 101 were present (Tables 1 and 3). HCHs were also not detected in the two lactating females (Table 1). An analysis of similarity with pairwise tests demonstrated no significant differences in organochlorine concentra-

tions between reproductive groups or sexes (Global R: -0.04 ; Sample statistic: 71.0%; 5000 randomisations).

3.4. Concentrations of pollutants in relation to blubber lipid content

Wet weight concentrations of organochlorines demonstrated different relationships with blubber lipid content across all groups of animals. The PCB congener 118 and Σ PCB demonstrated a positive relationship

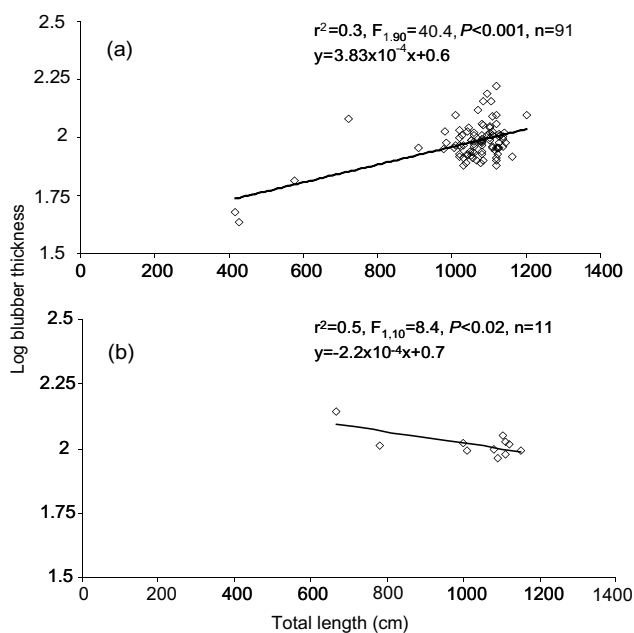


Fig. 2. The relationship between blubber thickness (mm) and total length (cm) in (a) female and (b) male southern Australian sperm whales.

Table 5
Mean concentrations \pm SD of HCHs and DDTs in the outer, middle and inner strata of blubber in southern Australian sperm whales from STR2

Strata	Group	N	Lipid content (%)	α -HCH	β -HCH	γ -HCH	δ -HCH	Σ HCH	<i>p,p'</i> -DDD	<i>p,p'</i> -DDE	<i>p,p'</i> -DDT	Σ DDT
Outer	All	10	35.7 \pm 14.2	ND	ND	ND	ND	ND	0.3 \pm 0.3	1.2 \pm 1.2	1.1 \pm 2.1	2.6 \pm 3.5
	All females	7	35.7 \pm 16.2	ND	ND	ND	ND	ND	0.3 \pm 0.3	1.2 \pm 1.4	1.3 \pm 2.5	2.9 \pm 4.2
	Non-lactating mature females	5	41.6 \pm 14.6	ND	ND	ND	ND	ND	0.2 \pm 0.1	0.7 \pm 0.2	0.4 \pm 0.2	1.2 \pm 0.4
	Lactating females	1	28.5	ND	ND	ND	ND	ND	0.2	0.9	0.5	1.61
	Males	3	35.8 \pm 10.6	ND	ND	ND	ND	ND	0.2 \pm 0.1	1.0 \pm 0.5	0.6 \pm 0.3	1.8 \pm 0.9
	Range			13.2–63.1	ND	ND	ND	ND	ND	0.1–1.1	0.4–4.4	0.1–7.0
Middle	All	10	47.7 \pm 10.7	ND	ND	ND	ND	ND	0.2 \pm 0.1	0.9 \pm 0.6	0.7 \pm 0.5	1.7 \pm 1.2
	All females	7	49.3 \pm 12.3	ND	ND	ND	ND	ND	0.1 \pm 0.1	0.7 \pm 0.4	0.6 \pm 0.4	1.4 \pm 0.8
	Non-lactating mature females	5	49.6 \pm 14.1	ND	ND	ND	ND	ND	0.2 \pm 0.1	0.7 \pm 0.4	0.5 \pm 0.2	1.3 \pm 0.6
	Lactating females	1	56.0	ND	ND	ND	ND	ND	0.1	0.3	0.2	0.51
	Males	3	43.9 \pm 6.1	ND	ND	ND	ND	ND	0.2 \pm 0.2	1.3 \pm 1.0	1.0 \pm 0.7	2.5 \pm 1.8
	Range			28.9–65.5	ND	ND	ND	ND	ND	ND–0.4	0.3–2.4	0.2–1.6
Inner	All	10	32.7 \pm 13.5	0.01 \pm 0.02	ND	ND	ND	0.01 \pm 0.02	0.4 \pm 0.5	1.4 \pm 1.2	2.3 \pm 5.6	4.0 \pm 7.2
	All females	7	30.6 \pm 10.7	0.01 \pm 0.03	ND	ND	ND	0.01 \pm 0.03	0.4 \pm 0.5	1.4 \pm 1.4	3.1 \pm 6.7	4.9 \pm 8.6
	Non-lactating mature females	5	32.5 \pm 11.7	0.02 \pm 0.04	ND	ND	ND	0.01 \pm 0.03	0.2 \pm 0.1	0.8 \pm 0.4	0.3 \pm 0.3	1.3 \pm 0.6
	Lactating females	1	19.8	ND	ND	ND	ND	ND	1.6	4.6	18.2	24.3
	Males	3	37.5 \pm 15.7	ND	ND	ND	ND	ND	0.1 \pm 0.1	1.3 \pm 0.8	0.4 \pm 0.3	1.8 \pm 0.4
	Range			19.8–61.2	ND–0.1	ND	ND	ND	ND–0.1	ND–1.6	0.4–4.6	ND–18.2

All values in $\mu\text{g g}^{-1}$ lipid weight.

Table 6
Mean concentrations \pm SD of PCBs in the outer, middle and inner strata of blubber in southern Australian sperm whales from STR2

Strata	Group	<i>N</i>	PCB 28	PCB52	PCB101	PCB118	PCB153	PCB180	Σ PCB
Outer	All	10	0.1 \pm 0.02	0.1 \pm 0.1	0.04 \pm 0.1	0.4 \pm 0.2	0.4 \pm 0.2	0.3 \pm 0.2	1.3 \pm 0.4
	All females	7	0.04 \pm 0.01	0.04 \pm 0.03	0.1 \pm 0.1	0.4 \pm 0.2	0.5 \pm 0.2	0.3 \pm 0.2	1.3 \pm 0.4
	Non-lactating mature females	5	0.04 \pm 0.01	0.1 \pm 0.03	0.1 \pm 0.1	0.3 \pm 0.2	0.4 \pm 0.1	0.3 \pm 0.2	1.1 \pm 0.4
	Lactating females	1	0.04	ND	0.1	0.5	0.4	0.3	1.4
	Males	3	0.1 \pm 0.02	0.1 \pm 0.1	ND	0.4 \pm 0.2	0.4 \pm 0.2	0.4 \pm 0.2	1.3 \pm 0.5
	Range			0.02–0.1	ND–0.2	ND–0.3	0.2–0.7	0.2–0.9	0.1–0.6
Middle	All	10	0.02 \pm 0.02	0.02 \pm 0.03	ND	0.2 \pm 0.1	0.2 \pm 0.1	0.3 \pm 0.2	0.8 \pm 0.4
	All females	7	0.02 \pm 0.02	0.02 \pm 0.03	ND	0.3 \pm 0.1	0.3 \pm 0.1	0.3 \pm 0.2	0.3 \pm 0.2
	Non-lactating mature females	5	0.03 \pm 0.02	0.03 \pm 0.03	ND	0.3 \pm 0.1	0.3 \pm 0.1	0.3 \pm 0.2	0.9 \pm 0.3
	Lactating females	1	0.01	ND	ND	0.2	0.3	0.1	0.7
	Males	3	0.02 \pm 0.02	0.01 \pm 0.01	ND	0.1 \pm 0.1	0.2 \pm 0.1	0.2 \pm 0.2	0.5 \pm 0.4
	Range			ND–0.1	ND–0.1	ND	0.01–0.5	0.01–0.4	0.1–0.7
Inner	All	10	0.03 \pm 0.03	0.01 \pm 0.02	ND	0.3 \pm 0.1	0.4 \pm 0.7	0.2 \pm 0.1	0.9 \pm 0.4
	All females	7	0.04 \pm 0.03	0.01 \pm 0.02	ND	0.3 \pm 0.1	0.4 \pm 0.2	0.2 \pm 0.1	1.0 \pm 0.4
	Non-lactating mature females	5	0.04 \pm 0.04	0.01 \pm 0.02	ND	0.3 \pm 0.2	0.5 \pm 0.2	0.3 \pm 0.1	1.1 \pm 0.4
	Lactating females	1	0.04	ND	ND	0.4	0.6	0.3	1.2
	Males	3	0.03 \pm 0.01	0.02 \pm 0.03	ND	0.2 \pm 0.1	0.3 \pm 0.1	0.3 \pm 0.2	0.8 \pm 0.3
	Range			0.01–0.1	ND–0.1	ND	0.1–0.5	0.1–0.7	0.1–0.5

All values in $\mu\text{g g}^{-1}$ lipid weight.
ND: not detected.

with blubber lipid content, while *p,p'*-DDD and *p,p'*-DDE demonstrated a negative relationship with blubber lipid content. Concentrations of *p,p'*-DDD demonstrated a significant negative relationship with blubber lipid content in all animals pooled ($r^2 = 0.1$; $F_{1,36} = 4.9$; $P = 0.03$). Individuals from STR1 demonstrated significant positive relationships between the PCB 118 ($r^2 = 0.3$; $F_{1,24} = 10.3$; $P = 0.004$) and Σ PCB ($r^2 = 0.2$; $F_{1,24} = 4.8$; $P = 0.04$; Fig. 2) and blubber lipid content and a significant negative relationship with *p,p'*-DDD ($r^2 = 0.2$; $F_{1,23} = 6.2$; $P = 0.02$; Fig. 2), while those from STR2 demonstrated a significant negative relationship with *p,p'*-DDE ($r^2 = 0.4$; $F_{1,11} = 5.8$; $P = 0.04$). Females of age group three demonstrated a significant positive relationship between PCB 118 ($r^2 = 0.2$; $F_{1,28} = 8.2$; $P = 0.008$) and Σ PCB ($r^2 = 0.1$; $F_{1,28} = 4.4$; $P = 0.05$) and blubber lipid content, while all females pooled demonstrated a significant negative relationship between *p,p'*-DDD and blubber lipid content ($r^2 = 0.1$; $F_{1,31} = 4.0$; $P = 0.05$).

3.5. Stratification of pollutant concentrations

The distribution of pollutant concentrations across the vertical aspect of the blubber layer varied between the groups of pollutants (Tables 5 and 6). While there were exceptions, the general pattern of distribution for

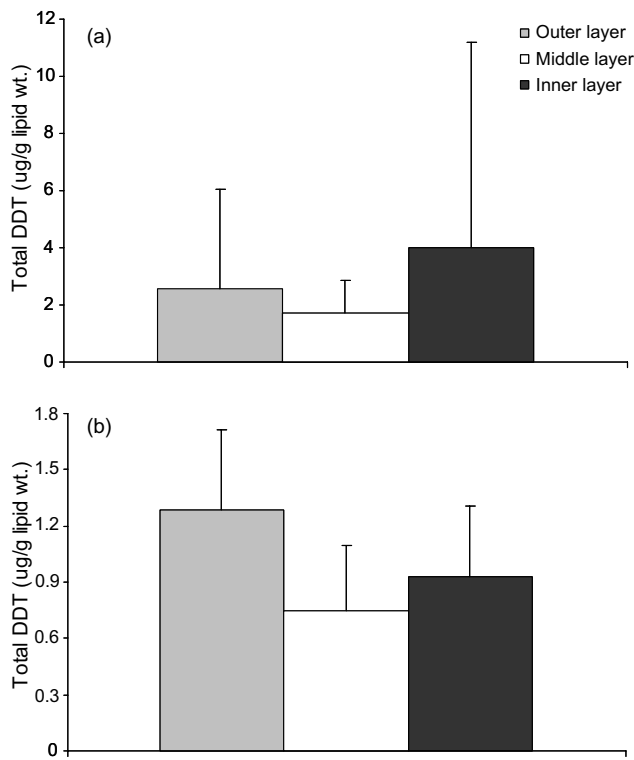


Fig. 3. Average concentration of (a) Σ DDT and (b) Σ PCB in the outer, middle and inner strata of blubber in southern Australian sperm whales.

Table 7

Results of repeated measures ANOVA on the distribution of pollutant concentrations throughout the blubber layer of southern Australian sperm whales

Compound	F-ratio (d.f. = 9)	P
α -HCH	1.0	ns
Σ HCH	1.0	ns
<i>p,p'</i> -DDD	72.0	<0.001
<i>p,p'</i> -DDE	1.0	ns
<i>p,p'</i> -DDT	5.5	0.04
Σ DDT	10.9	0.01
PCB28	318.2	<0.001
PCB52	32.4	<0.001
PCB101	2.0	ns
PCB118	139.3	<0.001
PCB153	76.9	<0.001
PCB180	131.3	<0.001
Σ PCB	2.0	ns

ns: not significant.

DDTs was inner strata > outer strata > middle strata and the pattern for PCBs: outer strata > inner strata > middle strata (Fig. 3). HCHs when present, were only present in the inner most strata of the blubber layer and the PCB congener 101 when present was only present in the outermost blubber stratum. Distributions of most compounds were significantly different between the three strata (Table 7). Concentrations of α -HCH, Σ HCH, *p,p'*-DDE, the PCB congener 101 and Σ PCB were not significantly different between the three strata.

Small sample sizes prevented an investigation into potential differences in stratification on the basis of reproductive condition, age or sex.

4. Discussion

The results of this study highlight the complexity of organochlorine accumulation in sperm whales. High variability was associated with concentrations of all organochlorines. Differences between stranding groups support the potential for organochlorines as means of discriminating social and thereby foraging groups of sperm whales, and highlight diet as a primary influence on organochlorine concentrations in this species.

4.1. Concentrations of with respect to weight and age

Typically across marine mammals, both males and females tend to accumulate organic pollutants rapidly during juvenile stages. As males age, accumulation slows and attains a plateau in adults. Females, however demonstrate a sharper slowing in accumulation, with burdens stabilizing and even decreasing in older individuals (Aguilar et al., 1999). This difference in accumulation has been attributed to the capacity for lipophilic pollutants to be passed across the placental membrane to the foetus during pregnancy, and through

the transfer to dependent young via milk during lactation. Transfer of pollutants to the foetus varies between species and has been reported to be in the order of 5–15% of the total body burden of the mother (Duinker and Hillebrand, 1979; Tanabe et al., 1980). Transfer during lactation increases substantially and has been reported to be in the order of 72–98% of the mother's body burden (Tanabe et al., 1980; Cockcroft and Ross, 1989). This increase is primarily associated with the depletion of body reserves of fat, protein and minerals to supply precursors for milk synthesis (Pond, 1984). The magnitude of organochlorine transfer and therefore overall patterns in lifetime accumulation, thereby depends on the reproductive rate of the species and individual, and the intensity of reproductive transfer (Aguilar et al., 1999).

Relationships between pollutant concentrations and age were absent amongst sperm whales in this study. Large numbers of adult females, a lack of adult males and high individual variation may serve mask any relationships present across the animals in this study. This may be further enhanced by the reproductive history of females in this study. Aguilar and Borrell (1994b) proposed that the highest transfer of pollutants from a mother to calf would occur during a female's first reproductive cycle. With continuing transfer of concentrations during successive reproductive cycles, the overall burden in the female decreases and therefore, the amount transferred also decreases. In such a scenario, the first calf from a female would contain the highest pollutant burdens of all of her offspring. The mean age of females sampled was 34.6 ± 14.8 year (0.75–60 year). Female sperm whales are considered to be sexually mature (but not physically mature) at around 10–13 years, although have been found to be pregnant as early as 7 year (Best et al., 1984). Females in this study were, for the majority, older than 20 year (90%) and therefore are likely to have already given birth to their first calf, thereby offloading the largest portion of their pollutant burdens. Low numbers of juveniles and high individual variation may serve to confound any distinct differences between sexually immature and mature females and further any variation in pollutant burdens within these two groups of females.

Female sperm whales are well known for their gregariousness, forming socially cohesive groups of 10–30 adult females and immature individuals. These groups are themselves composed of dynamic associations between mostly permanent units of 12–13 individuals that may or may not be related to each other (Whitehead et al., 1991; Whitehead and Kahn, 1992; Mesnick et al., 2003). Preliminary genetic analyses support the presence of smaller units comprised of related and unrelated individuals in the stranding groups studied here (Mesnick, 2001; Mesnick et al., 2003). The complex social structure of sperm whales is fundamentally asso-

ciated with optimizing the survival of offspring through communal defense and communal care of young (Best, 1979; Richard et al., 1996). These strategies serve to free lactating mothers from supervisory roles over young, allowing continual foraging and the costs of reproduction to largely be met by feeding thereby minimising demands on energy reserves. A reproductive strategy involving the continuous acquisition of energy throughout the reproductive period, allowing adjustments in energy acquisition in response to changes in energy demands, and a prolonged lactation period, is typical of that of income breeders (Jönsson, 1997). The opposite strategy is the reliance on endogenous energy stores during a reproduction period involving fasting and is typical of the reproductive strategies of capital breeders such as baleen whales and a number of phocid species (Jönsson, 1997). The reproductive strategy of capital breeders often involves high intensity, relatively short lactation periods involving high energy transfer to young, therefore allowing rapid growth and shorter dependence periods (Ofstedal, 1997). It could be assumed that the two reproductive strategies would ultimately result in different patterns of pollutant intake, metabolism and offloading. Offloading of pollutants from income breeding mothers to their young would be gradual over a prolonged lactation period and would also involve continual intake of further concentrations of pollutants with feeding during the reproductive period. The ultimate result of an income breeding reproductive strategy and low fecundity as observed in sperm whales may be essentially stable pollutant loads across adult females within a group for which pollutant intake is effectively the same (e.g., individuals in the same foraging group).

Differences in these two reproductive strategies may result in differing patterns of pollutant intake, metabolism and offloading. Offloading of pollutants from income breeding mothers to their young may not become significant until times during which continual foraging cannot meet higher energetic demands associated with lactation and stored energy reserves are therefore required to meet these demands. If body burdens of pollutants are not substantial, fecundity is low and foraging strategies serve to minimize demands placed on energy reserves during reproduction, the overall effect of offloading of pollutants in females may not be obvious. Without detailed information on the reproductive history of individuals, it is difficult to assess the effect of reproduction on the relationships between age and pollutant concentrations observed in this study.

Individuals from STR2 demonstrated a decrease in the concentration of *p,p'*-DDD, *p,p'*-DDE, the PCB congeners 118 and 153 and Σ PCB with estimated weight. This was largely influenced by relatively high concentrations in two immature animals (one female, one male) in this dataset, particularly by a male aged five

years, and much lower concentrations in the adult females involved in this stranding. The results observed here therefore may be more associated with age, rather than differences in the size of individuals.

4.2. Concentrations of pollutants with respect to sex

Males were distinctly different from females in their lack of detectable quantities of HCHs and the PCB 101. This may reflect differences in the intake, metabolism and excretion of individual organochlorines between sexes. However, both pollutants were not common in detectable concentrations throughout all animals in this study: HCHs were only detected in three animals and the PCB 101 detected in two animals. Larger sample sizes are required to establish whether this is indeed an indicator of differential deposition of these organochlorines between males and females. Aguilar (1983) reported higher concentrations of *p,p'*-DDT, *p,p'*-DDE, *p,p'*-DDD, Σ DDT and Σ PCB in female sperm whales but noted that age, reproductive status and fattening condition could mask differences between sexes. Sexual differences in diving ability, migratory habits and diet have been suggested to account for different pollutant inputs and subsequent patterns in pollutant burdens in this species (Aguilar, 1983).

While females are largely restricted to waters north of 40° S, males range into higher latitudes, traveling as far as the ice edge. Movements of males into polar, less polluted waters (due to their greater distance from point sources) and the confinement of females to more temperate and tropical and more highly polluted waters (due to their closer proximity to point sources; see Iwata et al. (1993) for an overview of global organochlorine distributions) would result in differing inputs and consequently, differing overall burdens between sexes. Differences in the diet of males and females in this area in late summer have been observed (Evans and Hindell, unpublished data) and may be reflective of different foraging habitats between sexes. However, dietary assessments were marked by high individual variability. This variability, coupled with small sample sizes and a lack of adult males makes it difficult to draw any conclusions on potential differences in organochlorine concentrations between sexes.

4.3. Concentrations of pollutants with respect to stranding group

As most persistent pollutants are incorporated into the bodies of marine mammals via food (Aguilar et al., 1999), differences in the concentrations of organochlorines between the two stranding groups may reflect differences in the composition of diet or in the location of feeding grounds between the two groups. Analysis of the recent diet of individuals revealed differences in the diet

between the two stranding groups (Evans and Hindell, unpublished data), but was characterized by intra-group variation, possibly the result of the presence of discrete foraging units within each stranding group. The nature of the associations between female sperm whale groups has been postulated to be reflective of foraging associations, and the dynamics of these associations vary with prey distributions and densities (Whitehead et al., 1991).

Although not statistically significant, differences in the *p,p'*-DDE/ Σ DDT (STR1: 0.7 ± 0.2 ; STR2: 0.5 ± 0.2) and Σ DDT/ Σ PCB (STR1: 1.9 ± 1.6 ; STR2: 2.7 ± 2.1) ratios of the two stranding groups may indicate potential differences in the longer-term diet and movement patterns of the two groups. The Σ DDT/ Σ PCB ratio is regarded as one of the most important for use in discriminating different populations of animals because of its sensitivity to distance from land and to trophic level (Aguilar, 1984). This ratio is higher in water masses closer to agricultural areas and lower in waters close to industrialised areas. The DDE/ Σ DDT ratio is based on the process of conversion of DDT through a chief metabolic pathway via dehydrochlorination to its metabolite DDE (Aguilar, 1984). This ratio therefore, is dependent on the amount of time since the release of DDT into the ocean and is indicative of differing release chronologies between different areas (Aguilar, 1987).

The recent diet of these sperm whales demonstrated that the diet of individuals from the two stranding groups differed and suggests that there may have been some segregation in the foraging of the two stranding groups (Evans and Hindell, unpublished data). However, these differences were marked by high individual variability and may be influenced by the distribution of males throughout the dataset. Cephalopods distributed throughout tropical and subtropical waters contributed to the diet of individuals from STR1 to a greater extent than they did to the diet of individuals from STR2, while those distributed throughout more southerly waters contributed to a larger extent to the diet of individuals from STR2. If this is a reflection of the longer-term diet of these animals, then individuals from STR1 may have been exposed to more recent inputs of DDT due to closer point source proximity. In the Southern Hemisphere, the vast majority of the populated area is in tropical and subtropical regions and levels of pollutants in both the air and surface water reflect this distribution (Iwata et al., 1993). Assessment of longer-term dietary components through the use of techniques such as fatty acid analysis may provide further insight into the diet of individuals and in association, potential differences in organochlorine accumulation.

The mean blubber lipid content of individuals involved in STR1 (55.3 ± 18.0) was significantly higher than that of individuals involved in STR2 (36.5 ± 8.6 ; $t_{35} = 3.32$, $P = 0.002$). It is possible that mobilisation of lipids, resulting in lower blubber lipid contents may have

also contributed to the observed higher concentrations of pollutant compounds remaining in blubber tissue of individuals involved in STR2.

4.4. Concentrations of pollutants in relation to blubber lipid content

Changes to pollutant concentrations as a result of the mobilisation of lipids are thought to involve a combination of two processes: (1) pollutants leave storage sites with the lipids to which they are bound, and pass into the blood and onto to other tissues, or are excreted resulting in tissue concentrations remaining constant; (2) pollutants remain in the tissues (lipids are more readily mobilised than lipophilic compounds), resulting in an increase in concentrations relative to the amount of lipids in the tissue (Aguilar, 1987; Aguilar et al., 1999). These two processes result in an overall increase in concentrations, but at a lower level than that which a purely concentrative model would produce.

Different organochlorines were observed to demonstrate different relationships between concentrations and blubber lipid content. DDTs decreased with increasing blubber lipid content, following the general pattern of an increase in concentration with mobilisation of lipids (Aguilar et al., 1999; see above), while PCBs increased with blubber lipid content amongst individuals from STR1 and in adult females. The relationship between PCB concentrations and blubber lipid content observed in this study is the opposite of that observed in cetaceans elsewhere (Aguilar et al., 1999). Although this relationship was statistically significant in adult females and individuals from STR1 (which were dominated by adult females), it was not a strong relationship and may have been biased by small samples sizes and high individual variability (Fig. 2).

Differences in the behaviour of individual compounds to changes in lipid content may be the result of: (i) differing affinities of individual organochlorines to different lipid groups, (ii) differing energetic utilization of lipid groups and (iii) differences in the ability to metabolise specific pollutant compounds. Concentrations of DDTs and highly chlorinated PCBs have been reported to be related to levels of triglycerides and non-esterified fatty acids, while concentrations of less chlorinated PCBs are reported to be related to levels of the highly polar phospholipids (Aguilar, 1985; Kawai et al., 1988). Differences in the rate and extent to which these lipids are mobilised would result in differing behaviours of pollutant concentrations associated with those lipids.

Both the diet and blubber lipid content of sperm whales from these strandings was typified by high individual variability (Evans and Hindell, unpublished data; Evans et al., 2003), suggesting differences in the feeding intensity and foraging success of individuals. Associated with this, if differing foraging strategies are employed by

individuals (i.e. differences in the spatial distribution of foraging individuals or differences in dive profiles), this may be reflected in differing metabolic demands on energy stores and therefore, individual variation in the utilization of energy. Both factors would ultimately result in high individual variability in energy stores and subsequently, on concentrations of organochlorines and the exchange in these between the blubber, blood and other organs.

Further studies involving larger number of animals across all age and reproductive groups would serve to establish the relationships of individual organochlorines with lipid content and additionally the effects of gender and individual variation on these relationships.

4.5. Stratification of pollutant concentrations

Differential deposition, or mobilisation, of pollutants throughout blubber tissue appears to occur in this species. Stratification of pollutant concentrations throughout the blubber layer has been suggested in fin (*Balaenoptera physalus*) and sei (*Balaenoptera borealis*) whales (Aguilar, 1985; Aguilar and Borrell, 1990), but has not been documented for this species.

Two interacting factors may contribute to the stratification of pollutant concentrations: (1) lipid composition varies with depth throughout the blubber layer of cetaceans, with inner layers containing longer-chain unsaturated fatty acids and outer layers containing medium-chain fatty acids (Ackman et al., 1975; Koopman et al., 1996). Individual organochlorines are reported to demonstrate different affinities for different lipids (Kawai et al., 1988; Guitart et al., 1996). As a result, the distribution of pollutants may therefore reflect any differential distribution in concentrations of individual lipids throughout the blubber layer. (2) Pollutants are absorbed and released into the blood from organs such as the liver and blubber following changes in the ingestion and excretion rates of compounds (Moriarty, 1984). The magnitude of this exchange is dependent on the size of the vascular network supplying each organ. The innermost layer of the blubber is exposed to a much larger circulatory network than the middle and outer layers. As a result, pollutants present in the inner layer of blubber are more easily transferred than those in the middle and outer layers and consequently, are sites of greater pollutant concentration variability (Aguilar and Borrell, 1994b). This exchange may also be enhanced or diminished by the polarity of individual compounds (Aguilar and Borrell, 1991).

Investigations into the stratification of pollutant concentrations throughout the blubber layer found that concentrations of DDTs were highest in the inner layer of blubber, while those of Σ PCB were highest in the outer layer of blubber. It must be noted that differences in PCB concentrations between layers were small and

Table 8
Mean concentrations \pm SD (range) of organochlorines in sperm whales elsewhere

Location	Year	Sex	<i>n</i>	DDT	DDE	DDD	Σ DDT	Σ PCB	Σ HCH	γ -HCH	<i>p,p'</i> -DDE/ Σ DDT	Σ DDT/ Σ PCB
South Africa ^a	1974	F	1	ND	0.4	ND	0.4	ND	–	–	1.0	–
		M	11	0.4 \pm 0.4 (ND–1.3)	0.4 \pm 0.1 (0.2–0.7)	0.01 \pm 0.02 (ND–0.1)	0.8 \pm 0.5 (0.3–1.9)	ND	–	–	0.6 \pm 0.2 (0.4–1.0)	–
South Africa ^{b*}	1976–1981	F	1	0.1	0.1	0.04	0.13	ND	–	–	0.7	–
		M	1	ND	0.2	ND	0.2	ND	–	ND	1.0	–
South Africa ^{c^A}	1978	U	1	–	–	–	1.0	–	–	–	–	–
South Africa ^{d*}	1986	F [#]	1	–	–	–	0.4	0.1	–	–	–	3.0
Australia ^{e*}	U	U	1	–	–	–	8.6	0.2	0.2	–	–	53.8
Australia ^{f^A}	U	U	1	0.6	0.1	0.6	1.2	–	–	–	0.1	–
		F	1	0.2	0.1	0.5	0.8	–	–	–	0.1	–
California ^g	1968	F	4	0.01 \pm 0.01 (0.0–0.02)	3.8 \pm 0.5 (3.3–4.4)	0.5 \pm 0.1 (0.5–0.6)	4.3 \pm 0.6 (3.8–5.0)	–	–	–	–	–
		M	2	1.7 \pm 1.2 (0.9, 2.6)	3.4 \pm 3.7 (0.7, 6.0)	0.5 \pm 0.4 (0.2, 0.8)	5.6 \pm 5.4 (1.8, 9.4)	–	–	–	–	–
West Indies ^{h*}	1971–1975	F	1	4.0	9.9	1.6	15.5	4.0	–	–	0.6	3.9
		M	1	0.2	0.8	0.1	1.1	0.7	–	–	0.7	1.6
Massachu- setts ^{h*}	1971–1975	F	1	3.7	4.6	0.6	8.9	2.1	–	–	0.5	4.2
Eastern North Atlantic ⁱ	1979–1980	F	6	2.7	4.0	0.5	7.7	15.6	–	–	–	–
		M	8	1.2	2.9	0.5	5.1	9.9	–	–	–	–
Iceland ^j	1982	M	10	2.5 \pm 1.1	4.2 \pm 1.0	1.1 \pm 0.2	7.8 \pm 1.5	10.5 \pm 2.1	–	–	0.5 \pm 0.1	0.8 \pm 0.1
Orkney Islands ^k	1994	M	3	3.3 \pm 0.9 (2.3–3.4)	7.1 \pm 1.9 (5.3–9.1)	0.8 \pm 0.2 (0.5–0.9)	11.2 \pm 2.9 (8.1–13.9)	3.5 \pm 1.0 (2.4–4.4)	0.1 \pm 0.02 (0.1–0.14)	0.1 \pm 0.02 (0.07–0.1)	0.6 \pm 0.03 (0.61–0.66)	3.2 \pm 0.2 (3.0–3.4)
		M	1	3.7	5.3	0.7	9.7	3.9	0.2	0.2	0.6	2.5
Wadden Sea ^k	1994	M	1	4.8 \pm 1.0 (3.6–5.5)	8.3 \pm 1.7 (6.4–9.7)	1.2 \pm 0.3 (0.9–1.4)	14.3 \pm 3.0 (10.9–16.5)	4.8 \pm 1.2 (3.5–5.7)	0.2 \pm 0.01 (0.2–0.2)	0.2 \pm 0.01 (0.18–0.19)	0.6 \pm 0.01 (0.58–0.59)	3.0 \pm 0.1 (2.9–3.2)
Netherlands ^k	1995	M	3	–	–	–	2.7	1.1	–	–	–	–
Orkney Islands ^{l+}	1994	M	11	–	–	–	–	–	–	–	–	–
								(1.2–15.5)	(0.3–6.3)			

Scotland ^{l+}	1993–1995	M	3	–	–	–	11.5 (11.4–11.6)	5.3 (3.9–5.6)	–	–	–	–
Belgium ^{l+}	1994	M	4	–	–	–	6.9 (5.3–12.7)	4.6 (3.0–16.4)	–	–	–	–
Netherlands ^l	1995	M	3	–	–	–	–	4.8 (2.8–4.9)	–	–	–	–
Belgium ^m	1994	M	4	3.1 ± 0.4 (2.7–3.5)	4.0 ± 0.7 (3.0–4.6)	0.7 ± 0.04 (0.6–0.7)	7.7 ± 1.1 (6.4–8.9)	2.9 ± 0.3 (2.5–3.2)	0.03 ± 0.01 (0.02–0.03)	–	0.5 ± 0.03 (0.47–0.54)	2.7 ± 0.4 (2.2–3.1)
Netherlands ^m	1995	M	3	2.6 ± 1.0 (1.6–3.6)	4.4 ± 1.4 (2.9–5.7)	0.8 ± 0.2 (0.5–1.0)	7.8 ± 2.6 (5.0–10.3)	3.2 ± 1.0 (2.1–4.2)	0.02 ± 0.01 (0.02–0.03)	–	0.6 ± 0.02 (0.55–0.58)	2.4 ± 0.03 (2.4–2.5)

All concentrations $\mu\text{g g}^{-1}$ lipid weight except * $\mu\text{g g}^{-1}$ wet weight and ^ where not stated. + median instead of mean given. F: female; M: male; U: not stated. # uncertainty to identification of sex as total length stated as 16.0 m.

^a Henry and Best (1983). ΣDDT derived from *p,p'* isomers of DDTs, ΣPCB quantified from four major peaks in Aroclor 1254 (congeners not identified).

^b Cockcroft and Ross (1991). Does not state whether DDTs *p,p'* or *o,p'* isomers or how ΣDDT and ΣPCB were quantified.

^c Van Dyk et al. (1982). Does not state how ΣDDT was quantified.

^d de Kock et al. (1994). Does not state how ΣDDT was quantified. ΣPCB was derived from the identification of congeners from standard congener mixtures (all congeners not identified).

^e Kemper et al. (1994). Does not state whether DDTs *p,p'* or *o,p'* isomers or how ΣDDT and ΣPCB were quantified.

^f Anderson (1991). Does not state whether DDTs *p,p'* or *o,p'* isomers or how ΣDDT and ΣPCB were quantified.

^g Wolman and Wilson (1970). ΣDDT derived from *o,p'* and *p,p'* isomers of DDTs.

^h Taruski et al. (1975). Does not state whether DDTs *p,p'* or *o,p'* isomers or how ΣDDT was quantified. ΣPCB was derived from the sum of the heights of those peaks in the sample that matched Aroclor 1254 and 1260 (congeners not identified).

ⁱ Aguilar (1983). DDTs shown are *p,p'* isomers. ΣDDT derived from the sum of both *p,p'* and *o,p'* isomers. ΣPCB was derived from the sum of the heights of those peaks in the sample that matched Aroclor 1254 and 1260 (congeners not identified).

^j Borrell (1993). DDTs shown are *p,p'* isomers. ΣPCB was derived from the sum of the heights of those peaks in the sample that matched Aroclor 1254 and 1260 (congeners not identified).

^k Law et al. (1996). DDTs shown are *p,p'* isomers. ΣDDT derived from the sum of both *p,p'* and *o,p'* isomers. ΣPCB was derived from the sum of the ICES7 congeners.

^l Wells et al. (1997). DDT derived from the sum of both *p,p'* and *o,p'* isomers. ΣPCB was derived from the sum of the ICES7 congeners.

^m Holsbeek et al. (1999). ΣDDT derived from the sum of both *p,p'* and *o,p'* isomers. ΣPCB was derived from the sum of the ICES7 congeners.

not as distinct as those observed in DDT concentrations. Overall concentrations of both DDTs and PCBs were observed to be lowest in the middle layer of blubber of individuals.

Stratification of lipids and constituent fatty acids in harbour porpoises and fin whales suggests that the inner region of the blubber is a more active site for lipid deposition and mobilisation, while the outer layers of blubber are more stable and lipids contained in these layers are not readily mobilized (Ackman et al., 1975; Koopman et al., 1996). Preliminary analysis into the stratification of fatty acids in the blubber of sperm whales in this study suggests that constituent fatty acids follow similar patterns to those documented in other species (Bedard, 1998). If the majority of lipid deposition occurs in the inner layers of blubber and the outer layers are relatively stable sites, pollutants stored in the outer layers of the blubber may not necessarily undertake the same patterns in concentration changes demonstrated by those pollutants stored in the inner layers of the blubber. If lipids are not readily mobilized from the outer layers of blubber and PCBs concentrate in these areas due to constituent lipid preferences, the outer layers of blubber may ultimately serve as pollutant reservoirs, concentrating PCBs. Further studies into the distribution of organochlorines throughout the blubber layer are required to ascertain whether greater concentrations of PCBs accumulate in the outer layer of blubber and that therefore, DDTs and PCBs are distributed differently throughout the blubber layer in sperm whales.

The presence of stratification in organochlorine concentrations suggests that no single section of the blubber layer of sperm whales is necessarily more representative of overall pollutant concentrations in this organ. This must be considered when comparing concentrations of pollutants between animals from different studies, particularly those based on samples from live whales obtained with the use of biopsy darts. These sampling methods do not penetrate the whole blubber depth in large whales such as sperm whales, taking only a fraction of the outer blubber layer, and therefore, may not provide a comprehensive indication of the presence and concentration of pollutants in blubber tissue. Comparisons from or with such studies must be undertaken with care, in light of possible biases due to stratification.

4.6. Concentrations of pollutants in comparison to sperm whales elsewhere

The overall mean concentrations of organochlorines were relatively low compared to those documented in this species in the Northern Hemisphere (Table 8). However, some individuals demonstrated concentrations of organochlorines comparable to, or above levels found in a number of studies of sperm whales in the

Northern Hemisphere. Mean concentrations of compounds were comparable or higher than those previously reported for this species in the Southern Hemisphere. Differences in analytical techniques, lack of gender specification, biases to adult males and presentation of results without relation to lipid amounts confound comparisons. It has been well established that differences in analytical techniques, the presentation of results and differences in the life history and biology (e.g. size, sex, age, diet, nutritive condition, health) of individuals can have significant effects on the ability to make inter-study comparisons (Aguilar et al., 1999). Additionally, as observed in this study, different groups of sperm whales from the same region can demonstrate differences in organochlorine concentrations. Differences in the dietary composition and foraging areas of groups are likely to result in differing intakes of organochlorines and as a result, it is difficult to positively identify temporal changes in organochlorine concentrations across large regions.

The highest concentrations of organochlorines in sperm whales were those of DDT and its metabolites, while concentrations of PCBs were considerably lower. This is also been observed in the majority of sperm whales studied elsewhere (Table 8). Concentrations of PCBs exceeded those of DDTs only in animals from the eastern North Atlantic and around Iceland (Aguilar, 1983; Borrell, 1993). This may be a reflection of global pollutant inputs, atmospheric and oceanic transport of pollutants and the metabolic capabilities of this species. Striped and bottlenose dolphins and fin whales have also been reported to have higher body loads of DDTs than PCBs. Subsequently, it has been hypothesized that DDT compounds are less easily metabolized than PCBs in cetaceans (Aguilar and Borrell, 1994b). Of the concentrations of individual PCB congeners, individuals in this study contained higher concentrations of the congener 153. This congener is highly bio-persistent (Van den Berg et al., 1998) and a similar dominance of this congener in PCB concentrations has been reported in a number of cetacean species elsewhere including sperm whales (Law et al., 1996; Holsbeek et al., 1999; Minh et al., 2000). The position of and degree of halogenation in PCBs and species-specific differences in the reaction of metabolic enzyme systems to these pollutants determine the rate and degree of metabolism of PCBs. This therefore determines the excretion or bioaccumulation of these compounds (Van den Berg et al., 1998). Preferential metabolism and elimination of PCBs has been inferred in a number of cetacean species from occurrence patterns in pollutants between predators and their prey, and the reactions of enzyme systems associated with the metabolism of toxic compounds such as the cytochrome P450 1A enzyme system (Kannan et al., 1989; Tanabe et al., 1988; Ross et al., 2000). It is possible that this also occurs in sperm whales. Further

research investigating the enzyme systems responsible for the metabolism of organochlorines, such as the cytochrome P450 family would provide greater insight into congener specific metabolism in this species.

DDT and PCB concentrations in southern Australian sperm whales were on the whole, less than those reported to be linked with deleterious effects in other marine mammal species (Béland et al., 1993; Ross et al., 1996; Jepson et al., 1999). However, the intake, metabolism, excretion and physiological reaction to pollutant concentrations are species-specific (Reijnders, 1994) and as a result, it is difficult to extrapolate the effects observed in one species to concentrations of organochlorines in another species. Additionally, a number of organochlorines are reported to undergo multiple non-additive interactions producing responses in mammalian systems at lower concentrations than if introduced individually (e.g. synergistic interactions between PCBs and dioxins in the development of porphyria and induction of cytochrome P450 enzyme systems and thyroid hormone levels in rodents) (Van den Berg et al., 1998). In vitro studies on sperm whale tissues could serve to provide some indication of the effects of changes in concentrations of individual organochlorines and any interactive effects in organ systems.

Continued monitoring of concentrations of organochlorines in oceanic species is essential in establishing temporal trends in regional organochlorine concentrations in cetaceans in the Southern Ocean. This can only be realised through the coordinated execution of a program in which associated life history, distribution and migratory data (to account for local and regional pollution influences) are collected. This information, linked with toxicokinetic studies, will provide a better understanding of the effects of such concentrations both in the short and long-term on populations of cetaceans in this area.

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References

- Ackman, R.G., Hingley, J.H., Eaton, C.A., Logan, V.H., Odense, P.H., 1975. Layering and tissue composition in the blubber of the northwest Atlantic sea whale (*Balaenoptera borealis*). *Canadian Journal of Zoology* 53, 1340–1344.
- Aguilar, A., 1983. Organochlorine pollution in sperm whales, *Physeter macrocephalus*, from temperate waters of the eastern North Atlantic. *Marine Pollution Bulletin* 14 (9), 349–352.
- Aguilar, A., 1984. Relationship of DDE/ΣDDT in marine mammals to the chronology of DDT input into the ecosystem. *Canadian Journal of Fisheries and Aquatic Science* 41, 840–844.
- Aguilar, A., 1985. Compartmentation and reliability of sampling procedures in Organochlorine pollution surveys of cetaceans. *Residue Reviews* 95, 91–114.
- Aguilar, A., 1987. Using organochlorine pollutants to discriminate marine mammal populations: a review and critique of the methods. *Marine Mammal Science* 3 (3), 242–262.
- Aguilar, A., Borrell, A., 1990. Patterns of lipid content and stratification in the blubber of fin whales (*Balaenoptera physalus*). *Journal of Mammalogy* 71 (4), 544–554.
- Aguilar, A., Borrell, A., 1991. Heterogeneous distribution of organochlorine contaminants in the blubber of baleen whales: implications for sampling procedures. *Marine Environmental Research* 31, 275–286.
- Aguilar, A., Borrell, A., 1994a. Abnormally high polychlorinated biphenyl levels in striped dolphins (*Stenella coeruleoalba*) affected by the 1990–1992 Mediterranean epizootic. *The Science of the Total Environment* 154, 237–247.
- Aguilar, A., Borrell, A., 1994b. Reproductive transfer and variation of body load of organochlorine pollutants with age in fin whales (*Balaenoptera physalus*). *Archives of Environmental Contamination and Toxicology* 27, 546–554.
- Aguilar, A., Borrell, A., Pastor, T., 1999. Biological factors affecting variability of persistent pollutant levels in cetaceans. *Journal of Cetacean Research and Management (special issue 1)*, 83–116.
- Aguilar, A., Jover, L., Borrell, A., 1993. Heterogeneities in organochlorine profiles of Faroese long-finned pilot whales: indication of segregation between pods? Report of the International Whaling Commission (special issue 14), 359–367.
- Aguilar, A., Borrell, A., Reijnders, P.J.H., 2002. Geographical and temporal variation in levels of organochlorine contaminants in marine mammals. *Marine Environmental Research* 53, 425–452.
- Anderson, G.R.V., 1991. Australia. Progress report on cetacean research, May 1989 to May 1990. Report of the International Whaling Commission 41, 223–229.
- Best, P.B., 1979. Social organisation in sperm whales, *Physeter macrocephalus*. In: Winn, H.E., Olla, B.E. (Eds.), *Behaviour in Marine Mammals. Current Perspectives in Research*. Plenum Press, New York, 3, pp. 227–289.
- Best, P.B., Canham, P.A.S., Macleod, N., 1984. Patterns of reproduction in sperm whales *Physeter macrocephalus*. Report of the International Whaling Commission (special issue 6), 51–79.
- Béland, P., DeGuisé, S., Girard, C., Lagacé, A., Martineau, D., Michaud, R., Muir, D.C.G., Norstrom, R.J., Pelletier, É., Ray, S., Shugart, L.R., 1993. Toxic compounds and health and reproductive

- effects in St. Lawrence beluga whales. *Journal of Great Lakes Research* 19 (4), 766–775.
- Borrell, A., 1993. PCB and DDTs in blubber of cetaceans from the northeastern North Atlantic. *Marine Pollution Bulletin* 26 (3), 146–151.
- Borrell, A., Aguilar, A., 1993. DDT and PCB pollution in blubber and muscle of long-finned pilot whales from the Faroe Islands. Report of the International Whaling Commission (special issue 14), 351–358.
- Cockcroft, V.G., Ross, G.J.B., 1989. Age, growth and reproduction of bottlenose dolphins *Tursiops truncatus* from the east coast of Southern Africa. *Fishery Bulletin* 88, 289–302.
- Cockcroft, V.G., Ross, G.J.B., 1991. Occurrence of organochlorines in stranded cetaceans and seals from the east coast of southern Africa. UN Environmental Program Marine Technical Report 3, 271–276.
- de Kock, A.C., Best, P.B., Cockcroft, V., Bosma, C., 1994. Persistent organochlorine residues in small cetaceans from the east and west coasts of southern Africa. *The Science of the Total Environment* 154, 153–162.
- Duinker, J.C., Hillebrand, M.T.J., 1979. Mobilization of organochlorines from female lipid tissue and transplacental transfer to fetus in a harbour porpoise (*Phocoena phocoena*) in a contaminated area. *Bulletin of Environmental Contamination and Toxicology* 23, 728–732.
- Evans, K., 2003. Pollution and marine mammals in the Southern Hemisphere: present or potential threat?. In: Gales, N.J., Hindell, M.A., Kirkwood, R. (Ed.), *Marine Mammals and Humans: Fisheries, Tourism and Management*. CSIRO Publishing, Melbourne.
- Evans, K., Robertson, K., 2001. A note on the preparation of sperm whale teeth (*Physeter macrocephalus*) for age determination. *Journal of Cetacean Research and Management* 3 (1), 101–107.
- Evans, K., Morrice, M., Hindell, M., Thiele, D., 2002. Three mass standings of sperm whales (*Physeter macrocephalus*) in southern Australian waters. *Marine Mammal Science* 18 (3), 622–643.
- Evans, K., Hindell, M.A., Thiele, D., 2003. Body fat and condition in sperm whales, *Physeter macrocephalus*, from southern Australian waters. *Comparative Biochemistry and Physiology A* 134, 847–862.
- Guitart, R., Guerrero, X., Silvestre, A.M., Gutiérrez, J.M., Mateo, R., 1996. Organochlorine residues in tissues of striped dolphins affected by the 1990 Mediterranean epizootic: relationships with fatty acid composition. *Archives of Environmental Contamination and Toxicology* 30, 79–83.
- Henry, J., Best, P.B., 1983. Organochlorine residues in whales landed at Durban, South Africa. *Marine Pollution Bulletin* 14 (6), 223–227.
- Holsbeek, L., Joiris, C.R., Debacker, V., Ali, I.E., Roose, P., Nellissen, J., Gobert, S., Bouquegneau, J., Bossicart, M., 1999. Heavy metals, organochlorines and polycyclic aromatic hydrocarbons in sperm whales stranded in the southern North Sea during 1994/1995 winter. *Marine Pollution Bulletin* 38 (4), 304–313.
- Iwata, H., Tanabe, S., Sakai, N., Tatsukawa, R., 1993. Distribution of persistent organochlorines in the oceanic air and surface seawater and the role of the ocean on their global transport and fate. *Environmental Science and Technology* 27, 1080–1098.
- Jaquet, N., Dawson, S., Slooten, E., 2000. Seasonal distribution and diving behaviour of male sperm whales off Kaikoura: foraging implications. *Canadian Journal of Zoology* 78, 407–419.
- Jepson, P.D., Bennett, P.M., Allchin, C.R., Law, R.J., Kuiken, T., Baker, J.R., Rogan, E., Kirkwood, J.K., 1999. Investigating potential associations between chronic exposure to polychlorinated biphenyls and infectious disease mortality in harbour porpoises from England and Wales. *The Science of the Total Environment* 243/244, 339–348.
- Johnston, P.A., Stringer, R.L., Santillo, D., 1996. Cetaceans and environmental pollution: the global concerns. In: Simmonds, M.P., Hutchinson, J.D. (Eds.), *The Conservation of Whales and Dolphins*. John Wiley and Sons Ltd, Chichester, pp. 219–261.
- Joiris, C.R., Holsbeek, L., Bossicart, M., Tapia, G., 1997. Mercury and organochlorines in four sperm whales stranded on the Belgian coast, November 1994. *Bulletin de L'Institut Royal des Sciences Naturelles de Belgique* 67, 69–74.
- Jönsson, I., 1997. Capital and income breeding as alternative tactics of resource used in reproduction. *Oikos* 78, 57–66.
- Kannan, N., Tanabe, S., Ono, M., Tatsukawa, R., 1989. Critical evaluation of the polychlorinated biphenyl toxicity in terrestrial and marine mammals: increasing impact of non-ortho and mono-ortho coplanar polychlorinated biphenyls from land to ocean. *Archives of Environmental Contamination and Toxicology* 18, 850–857.
- Kawai, S., Fukushima, M., Miyazaki, N., Tatsukawa, R., 1988. Relationship between lipid composition and organochlorine levels in the tissues of striped dolphin. *Marine Pollution Bulletin* 19 (3), 129–133.
- Kemper, C., Gibbs, P., Obendorf, D., Marvanek, S., Lenghaus, C., 1994. A review of heavy metal and organochlorine levels in marine mammals in Australia. *The Science of the Total Environment* 154, 129–139.
- Koopman, H.N., Iverson, S.J., Gaskin, E., 1996. Stratification and age-related differences in blubber fatty acids of the male harbour porpoise (*Phocoena phocoena*). *Journal of Comparative Physiology B* 165, 628–639.
- Law, R.J., Stringer, R.L., Allchin, C.R., Jones, B.R., 1996. Metals and organochlorines in sperm whales (*Physeter macrocephalus*) stranded around the North Sea during the 1994/1995 winter. *Marine Pollution Bulletin* 32 (1), 72–77.
- Lockyer, C., 1981. Estimates of growth and energy budget for the sperm whale, *Physeter catadon*. *Mammals in the sea*, FAO, Fisheries 3, 489–504.
- Lockyer, C., 1991. Body composition of the sperm whale *Physeter catadon*, with special reference to the possible functions of fat depots. *Rit Fiskideildar* 12, 1–24.
- Mesnick, S.L., 2001. Genetic relatedness in sperm whales: evidence and cultural implications. *Behavioural and Brain Sciences* 24 (2), 346–347.
- Mesnick, S.L., Evans, K., Taylor, B.L., Hyde, J., Escorza-Treviño, S., Dizon, A.E., 2003. Sperm whale social structure: why it takes a village to raise a child. In: Wall, B.M.D., Tyack, P.L. (Eds.), *Animal Social Complexity. Intelligence, Culture and Individualized Societies*. Harvard University Press, New York, pp. 170–174.
- Minh, T.B., Nakata, H., Watanabe, M., Tanabe, S., Miyazaki, N., Jefferson, T.A., Prudente, M., Subramanian, A., 2000. Isomer-specific accumulation and toxic assessment of polychlorinated biphenyls, including coplanar congeners, in cetaceans from the North Pacific and Asian coastal waters. *Archives of Environmental Contamination and Toxicology* 39, 398–410.
- Moriarty, F., 1984. Persistent contaminants, compartmental models and concentration along food chains. *Ecological Bulletins* 36, 35–45.
- Murphy, P.G., 1972. Sulfuric acid cleanup of animal tissues for analysis of acid-stable chlorinated hydrocarbon residues. *Journal of the AOAC* 55 (6), 1360–1362.
- Norris, K.S., 1961. Standardized methods for measuring and recording data on the smaller cetaceans. *Journal of Mammalogy* 42 (4), 471–476.
- Oftedal, O.T., 1997. Lactation in whales and dolphins: evidence of divergence between baleen- and toothed-species. *Journal of Mammary Gland Biology and Neoplasia* 2 (3), 205–230.
- Pond, C.M., 1984. Physiological and ecological importance of energy storage in the evolution of lactation: evidence for a common pattern of anatomical organisation of adipose tissue in mammals. *Symposia of the Zoological Society of London* 51, 1–32.

- Reijnders, P.J.H., 1986. Perspectives for studies of pollution in cetaceans. *Marine Pollution Bulletin* 17 (2), 58–59.
- Reijnders, P.J.H., 1994. Toxicokinetics of chlorobiphenyls and associated physiological responses in marine mammals with particular reference to their potential for ecotoxicological risk assessment. *The Science of the Total Environment* 154, 229–236.
- Reijnders, P.J.H., Ruiter-Dijkman, E.M.D., 1995. Toxicological and epidemiological significance of pollutants in marine mammals. In: Blix, A.S., Walløe, L., Ulltang, Ø. (Eds.), *Whales, Seals, Fish and Man*. Elsevier Science, B.V., Amsterdam, pp. 575–587.
- Rice, D.W., 1989. Sperm whale *Physeter macrocephalus* Linnaeus, 1758. In: Ridgeway, S.H., Harrison, R. (Eds.), *Handbook of Marine Mammals, Volume 4: River Dolphins and the Larger Toothed Whales*. Academic Press, San Diego, pp. 177–233.
- Richard, K.R., Dillon, M.C., Whitehead, H., Wright, J.M., 1996. Patterns of kinship in groups of free-living sperm whales (*Physeter macrocephalus*) revealed by multiple molecular genetic analyses. *Proceedings of the National Academy of Sciences USA* 93, 8792–8795.
- Ross, P., de Swart, R., Addison, R., van Loveren, H., Vos, J., Osterhaus, A., 1996. Contaminant-induced immunotoxicity in harbour seals: wildlife at risk? *Toxicology* 112, 157–169.
- Ross, P.S., Ellis, G.M., Ikonomou, M.G., Barrett-Lennard, L.G., Addison, R.F., 2000. High PCB concentrations in free-ranging Pacific killer whales, *Orcinus orca*: effects of age, sex and dietary preference. *Marine Pollution Bulletin* 40, 504–515.
- Simonich, S.L., Hites, R.A., 1995. Global distribution of persistent organochlorine compounds. *Science* 269, 1851–1854.
- Skaare, J.U., Bemhoft, A., Derocher, A., Gabrielsen, G.W., Goksøyr, A., Henriksen, E., Larsen, H.J., Lie, E., Wiig, Ø., 2000. Organochlorines in top predators at Svalbard—occurrence, levels and effects. *Toxicology Letters* 112–113, 103–109.
- Tanabe, S., Tanaka, H., Maruyama, K., Tatsukawa, R., 1980. Elimination of chlorinated hydrocarbons from mother striped dolphins (*Stenella coeruleoalba*) through parturition and lactation. In: Fujiyama, T. (Ed.), *Studies on the Levels of Organochlorine Compounds and Heavy Metals in the Marine Organisms*. University of the Ryukus, Okinawa, pp. 115–121.
- Tanabe, S., Tatsukawa, R., Tanaka, H., Maruyama, K., Miyazaki, N., Fujiyama, T., 1981. Distribution and total burdens of chlorinated hydrocarbons in bodies of striped dolphins (*Stenella coeruleoalba*). *Agricultural and Biological Chemistry* 45 (11), 2569–2578.
- Tanabe, S., Mori, T., Tatsukawa, R., 1983. Global pollution of marine mammals by PCBs, DDTs and HCHs (BHCs). *Chemosphere* 12 (9), 1269–1275.
- Tanabe, S., Watanabe, S., Kan, H., Tatsukawa, R., 1988. Capacity and mode of PCB metabolism in small cetaceans. *Marine Mammal Science* 4, 103–124.
- Tanabe, S., Iwata, H., Tatsukawa, R., 1994. Global contamination by persistent organochlorines and the ecotoxicological impact on marine mammals. *The Science of the Total Environment* 154, 163–177.
- Taruski, A.G., Olney, C.E., Winn, H.E., 1975. Chlorinated hydrocarbons in cetaceans. *Journal of the Fisheries Research Board of Canada* 32, 2205–2209.
- Tilbury, K.L., Adams, N.G., Krone, C.A., Meador, J.P., Early, G., Varanasi, U., 1999. Organochlorines in stranded pilot whales (*Globicephala melaena*) from the coast of Massachusetts. *Archives of Environmental Contamination and Toxicology* 37, 125–134.
- Van den Berg, M., Bimbaum, L., Bosveld, A.T.C., Brunström, B., Cook, P., Feeley, M., Giesy, J.P., Hanberg, A., Hasegawa, R., Kennedy, S.W., Kubiak, T., Larsen, J.C., van Leeuwen, F.X.R., Liem, A.K.D., Nolt, C., Peterson, R.E., Poellinger, L., Safe, S., Srenk, D., Tillit, D., Tysklind, M., Younes, M., Wærn, F., Zacharewski, T., 1998. Toxic equivalency factors (TEFs) for PCBs, PCDDs, PCDFs for humans and wildlife. *Environmental Health Perspectives* 106, 775–792.
- Van Dyk, L.P., Wiese, I.H., Mullen, E.C., 1982. Management and determination of pesticide residues in South Africa. *Residue Reviews* 82, 37–124.
- Wania, F., Mackay, D., 1995. A global distribution model for persistent organic chemicals. *The Science of the Total Environment* 160/161, 211–232.
- Wells, D.E., McKenzie, C., Ross, H.M., 1997. Patterns of organic contaminants in marine mammals with reference to sperm whale strandings. *Bulletin de L'Institut Royal des Sciences Naturelles de Belgique* 67 (supplement), 91–104.
- Whitehead, H., Kahn, B., 1992. Temporal and geographic variation in the social structure of female sperm whales. *Canadian Journal of Zoology* 70, 2145–2149.
- Whitehead, H., Waters, S., Lyrholm, T., 1991. Social organization of female sperm whales and their offspring: constant companions and casual acquaintances. *Behavioural Ecology and Sociobiology* 29, 385–389.
- Wolman, A.A., Wilson, A.J., 1970. Occurrence of pesticides in whales. *Pesticides Monitoring Journal* 4 (1), 8–10.