

# National Dioxins Program

**Technical Report No. 7**  
Dioxins in Fauna in Australia

**A consultancy funded by the Australian Government  
Department of the Environment and Heritage**

**Prepared by Dr Ray Corell and Dr Jochen Müller**



**Australian Government**

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**Department of the Environment and Heritage**

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2. Dioxins emissions from Motor Vehicles in Australia
3. Inventory of Dioxins emissions in Australia, 2004
4. Dioxins in Ambient Air in Australia
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7. **Dioxins in Fauna in Australia**
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## Foreword

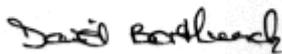
When the Australian Government established the four year National Dioxins Program in 2001, our knowledge about the incidence of dioxins in Australia was very limited.

The aim of the program was to improve this knowledge base so that governments were in a better position to consider appropriate management actions. Starting in mid 2001, a range of studies were undertaken which involved measuring emissions from sources such as bushfires, as well as dioxin levels in the environment, food and population. The findings of these studies were used to shed light on the risk dioxins pose to our health and the environment.

This work has been completed and the findings are now presented in a series of twelve technical reports.

Having good information is essential if there is to be timely and effective action by governments; these studies are a start. Our next step is to foster informed debate on how we should tackle dioxins in Australia, as this is an obligation under the Stockholm Convention on Persistent Organic Pollutants. The Department of the Environment and Heritage will be working closely with other Australian Government, State and Territory agencies to take this step.

Ultimately, the effective management of dioxins will be the shared responsibility of all government jurisdictions with the support of the community and industry.



David Borthwick

Secretary

Department of the Environment and Heritage

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- the Department of Agriculture, Fisheries and Forestry, who assessed the levels of dioxins in agricultural commodities
- Food Standards Australia New Zealand and the Department of Health and Ageing and who assessed the levels of dioxins in foods and assessed the health effects of dioxins
- officers of the Chemical Assessment Section in DEH who assessed the ecological effects of dioxins
- members of the National Dioxins Project Team which included representatives from the State and Territory environment protection agencies, the Australian Health Ministers Conference and the Primary Industries Ministers Council
- members of the National Dioxins Consultative Group which included representatives from industry and agricultural sectors, environment and public health groups and research institutions.

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## Contributors

The analytical data on the sperm whales was kindly made available for this report by AGAL and remains the property of AGAL.

This project was made possible through the cooperation of collaborators who supplied animal material. A complete list of the collectors is given in Appendix F. Particular thanks are extended to Mr Ron Haering of NSW National Parks and Wildlife Service, Ms Sally Troy of Parks Victoria and Dr Peter Mawson of the Department of Conservation and Land Management, Western Australia for their roles in coordinating regional sample collection.

## Executive Summary

### Description of survey

This study was a component of the National Dioxins Program tasked to quantify and assess the concentrations and relative chemical compositions of dioxin-like chemicals in Australian fauna.

The project involved the collection of several hundred fauna samples (primarily roadkill for terrestrial animals and stranded animals for marine mammals) with emphasis on spatial and biological diversity. The collected specimens were pooled into 66 samples covering all States as well as the Northern Territory, see table below.

<b>Fauna Class</b>	<b>NSW</b>	<b>NT</b>	<b>QLD</b>	<b>SA</b>	<b>TAS</b>	<b>VIC</b>	<b>WA</b>
Bird		3	3	10			3
Dingo				2			
Macropod	2	3	4	5		4	4
Marine mammal		2		2	9		
Monotreme				2	3		
Other marsupial		2		2			
Reptile				1			

Chemical analysis of the fauna samples was conducted by the Australian Government Analytical Laboratories, and a series of quality assurance/quality control (QA/QC) procedures were incorporated into the study, including replicate field sampling, replicate analysis and an interlaboratory comparison of analyses using an overseas laboratory highly regarded for its experience in the analysis of dioxin-like chemicals in fauna samples. The QA/QC procedure suggested that chemical analysis reproducibility was high, and that the identification of individual dioxin-like chemicals and quantification of their concentrations in fauna samples was reliable. The analysis of sampling replicates, or samples collected at different sites within the regions, demonstrated that the greatest uncertainty of the results related to variability in chemical concentrations between different individuals from a given species.

The concentrations of dioxin-like chemicals in the fauna samples were assessed both in terms of actual concentrations and their toxic equivalents (TEQs). In addition, the patterns of component chemicals were evaluated, and assessments of concentration patterns were made with respect to species and considering their respective food. An overview of the middle bound (incl. ½ LOD values) TEQ levels found in Australian fauna is given in the table below.

<b>Fauna class</b>	<b>No of samples</b>	<b>Minimum TEQ<sup>1</sup></b>	<b>Median TEQ</b>	<b>Maximum TEQ</b>
Bird	19	0.64	300	3,900
Dingo	2	1.7	2.0	2.3
Macropod	22	0.14	0.71	25
Marine mammal	13	1.1	28	590
Monotreme	5	9.3	23	60
Other marsupial	4	0.95	2.0	13
Reptile	1		0.65	

<sup>1</sup> Units are pg g<sup>-1</sup> lipid.

Dioxin-like chemicals were detectable in all samples and the levels expressed as toxicity equivalencies ranged from the limit of detection to 3,900 pg TEQ g<sup>-1</sup>. Overall the survey found highest concentrations in birds of prey (sparrowhawks, goshawks, falcons, eagles etc.) with a maximum level of 3,900 pg TEQ g<sup>-1</sup> lipid (middle bound). Piscivorous marine mammals also had high levels with a dolphin from the Port River in South Australia having a level of 590 pg TEQ g<sup>-1</sup> lipid. In contrast, levels were generally low in herbivorous animals such as macropods, a galah and a dugong (marine mammal that feeds exclusively on seagrass).

Concentrations of dioxin-like chemicals in the 22 macropod samples (mostly pooled samples of several animals) that were analysed in the study were relatively low with a median concentration of 0.71 pg g<sup>-1</sup> lipid. The highest concentrations of dioxin-like chemicals in macropods were detected in a sample (pool of three kangaroos) that was collected from the Para Wirra National Park located 25 km north north east of Adelaide; this sample had a TEQ of 25 pg g<sup>-1</sup> lipid.

Of the three groups of compounds (PCDDs, PCDFs and PCBs), the main contributors to the TEQ were the PCDDs – in the case of the galah (a primary feeder) this was dominated by OCDD, but in other bird samples (for example, black-shouldered kites) the main components were PeCDD and HxCDDs. PCBs contributed significantly to the TEQ load of birds. An example was a collared sparrowhawk from South Australia that had a TEQ of 3,900 pg g<sup>-1</sup> lipid with a contribution of 2,200 pg TEQ g<sup>-1</sup> lipid of the total TEQ coming from PCB 126.

Contribution of dioxin-like PCBs to TEQs was most dominant in marine mammals and the congener profiles of the PCDD/PCDFs and dioxin-like PCBs were similar for all marine mammals sampled. There was, however, an indication of elevated concentration of 1,2,3,7,6-PeCDD relative to the PCBs in both the dugong and dolphin samples from Darwin. It was noted that the bottlenosed dolphin from Port River South Australia had much higher PCB levels than those from a previous study in Spencer Gulf.

The TEQ of the other marsupials (possum, koala and bandicoot) were low and comparable to that of the macropods. The relative contributions of PCDD/PCDFs and PCBs were similar to those of the macropods but were very variable from sample to sample.

The TEQ of the monotremes were intermediate between those of birds and the macropods, with typically PCDD/PCDFs (especially PeCDD) making the greatest contribution. The highest value was 60 TEQ pg g<sup>-1</sup> lipid that was found in an echidna from Port Elliot, South Australia. There was one sample of platypus from Tasmania where PCBs contributed more than PCDD/PCDFs to the TEQ.

### **Field variability**

Estimates of field variability were obtained as part of the study. Coefficients of variation of 80% were typical between observations from similar animals from a similar location. The highly variable levels of analytes indicate that caution should be applied when comparisons are made between individual samples. Furthermore, there is evidence of local anomalies. Together these factors present a challenge to the interpretation of the data.

### **Comparison with overseas studies**

In general the levels of dioxin-like compounds were low by overseas standards.

The predatory birds from Australia generally had lower TEQ than did comparable birds from North America, Europe, India and Japan.

The macropods generally had a low TEQ (median  $0.71 \text{ pg g}^{-1}$  lipid) which was at the lower end of the range of values reported from caribou in Yukon ( $0.7\text{-}6.4 \text{ pg g}^{-1}$  lipid) and less than sika deer from Japan  $3.2\text{-}330 \text{ pg g}^{-1}$  lipid). Six of the 22 macropod samples had a TEQ greater than  $3 \text{ pg g}^{-1}$  lipid, which is the EU maximum permissible limit in meat for human consumption. However it is noteworthy that on a fresh weight basis the levels of dioxin-like chemicals are relatively low and the high levels recorded may be the result of the relatively low fat content of macropod meat.

The TEQs in the marine mammals reported in this survey are also low by world standards.

## Glossary/Abbreviations

AGAL	Australian Government Analytical Laboratories (Sydney).
ANOVA	Analysis of Variance.
ANZECC	Australian and New Zealand Environmental Conservation Council, now replaced by the Environment Protection and Heritage Council; representation includes environment Ministers from Australia and New Zealand.
Class	This is used to define a grouping of animal species. The exact definition is given in Table 2.1. The term class does not necessarily refer to a taxon.
Congener	Closely related chemicals derived from the same parent compound.
CSIRO	Commonwealth Scientific and Industrial Research Organisation.
CV	Coefficient of variation (see Box 1).
DAFF	Department of Agriculture, Fisheries and Forestry.
DEH	Department of the Environment and Heritage.
Dioxin	Polychlorinated dibenzo- <i>p</i> -dioxin; also the term dioxin is commonly used for polychlorinated dioxin-like chemicals in general.
FSANZ	Food Standards Australia and New Zealand.
Furan	Polychlorinated dibenzofuran.
Homologue	A group of structurally related chemicals that have the same degree of chlorination.
I-TEQ	Toxicity equivalencies using NATO-CCMS (1988) toxicity equivalency factors. Most data prior to 1998 reported in I-TEFs usually did not include PCBs.
IUPAC	International Union of Pure and Applied Chemistry.
Lipid	Fat found in living tissue.
LOD	Limit of detection.
Lower bound TEQ	Toxic equivalencies (TEQ) for which concentration of a non-detected congener assumed to be equal zero.
microgram	$\mu\text{g} = 10^{-6}$ gram (0.000 001g).
Middle bound TEQ	Toxic equivalencies (TEQ) for which concentration of a non-detected congener assumed to be equal to half the non detect value.
min	Minute or Minimum.
nanogram	$\text{ng} = 10^{-9}$ gram (0.000 000 001g).
NDP	National Dioxins Program.
ENTOX / NRCET	National Research Centre for Environmental Toxicology (Queensland).
PCB	Polychlorinated biphenyl[s].
PCDD/PCDF	Polychlorinated dibenzo- <i>p</i> -dioxins and furans – these typically will be in a mixture.
picogram	$\text{pg} = 10^{-12}$ gram (0.000 000 000 001g).

ppm, ppb, ppt	Parts per million, parts per billion, parts per trillion. These measures correspond to $\mu\text{g g}^{-1}$ , $\text{ng g}^{-1}$ and $\text{pg g}^{-1}$ when the density is $1 \text{ kg L}^{-1}$ .
psi	Pounds per square inch.
$\text{pg g}^{-1}$	Picogram ( $10^{-12} \text{ g}$ ) per gram. Equal to nanogram per kilogram ( $\text{ng kg}^{-1}$ ).
TEQ	Toxic Equivalency (for this report WHO <sub>98</sub> -TEQ used, including PCB).
TEF	Toxic equivalency factor of a specific dioxin, furan, or PCB. TEF indicates the toxicity of each congener with dioxin-like biochemical and toxic responses, relative to the toxicity of the dioxin 2,3,7,8-TCDD (van den Berg et al. 1998).
WHO <sub>98</sub> -TEQ	World Health Organization toxic equivalent: the quantified level of each individual congener multiplied by the corresponding TEF. TEQs of each congener are summed to achieve an overall toxic equivalent for a sample (van den Berg et al. 1998). In this document WHO <sub>98</sub> -TEQ is abbreviated to TEQ.
WHO <sub>98</sub> -TEQ <sub>DF</sub>	WHO <sub>98</sub> -TEQ for dioxins and furans.
WHO <sub>98</sub> -TEQ <sub>P</sub>	WHO <sub>98</sub> -TEQ for PCBs.
WHO <sub>98</sub> -TEQ <sub>DFP</sub>	WHO <sub>98</sub> -TEQ for all dioxin-like congeners and is equivalent to WHO <sub>98</sub> -TEQ or TEQ.
WHO	World Health Organization.
wwt	Wet weight.



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# 1 Introduction

## 1.1 Background

The first Australian inventory of dioxin emissions to air (Sources of Dioxins and Furans in Australia: Air Emissions) was published in 1998 (Environment Australia 1998). Few data exist on ambient levels of dioxins in Australia, therefore, the preparation of that inventory relied heavily on overseas data, using release estimation methodology. The limited monitoring data available indicated that environmental concentrations were generally low, but that there was insufficient information to assess the impact of dioxins in Australia.

At its meeting in December 2000, the Australian and New Zealand Environment and Conservation Council (ANZECC<sup>1</sup>) requested the development of a discussion paper on dioxins for use in consultation with stakeholders. In April 2001, public meetings were held in several cities across Australia to seek public input into the development of a possible national dioxins program. These workshops noted the lack of information on dioxins in Australia and recommended that data be obtained on levels in the environment and the population. Following on from these consultations, a proposal for a national dioxins program was tabled at the meeting of ANZECC in June 2001. At this meeting, Council noted that the Australian Government would fund a National Dioxins Program (NDP) with \$5 million over four years and that this program would generate data over the following two years which could be used to determine whether a specific regulatory approach would be required to manage dioxins.

The Department of the Environment and Heritage (DEH) is implementing the National Dioxin Program, in three phases:

- Information gathering about the current levels of dioxins in Australia
- Risk assessment using the information gathered as a basis to assess the potential risks of dioxins to the environment and human health
- Development of measures to reduce, and where feasible, to eliminate the release of dioxins in Australia.

Under the information gathering phase DEH commissioned organisations to undertake the following studies:

- Determination of ambient environmental levels (ambient air, aquatic, soils and fauna) of dioxins in Australia
- Determination of the levels of dioxins emissions from bushfires in Australia
- Determination of the levels of dioxin emissions from motor vehicles in Australia
- Determination of the levels of dioxins in the Australian population by analysis of blood serum
- Dioxins and dioxin-like compounds in pooled human milk samples.

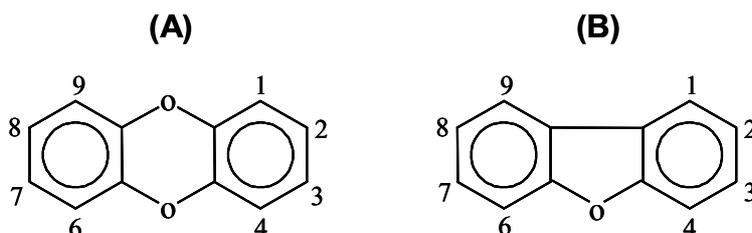
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<sup>1</sup> The Environment Protection and Heritage Council has replaced ANZECC and representation includes environment ministers from Australia and New Zealand.

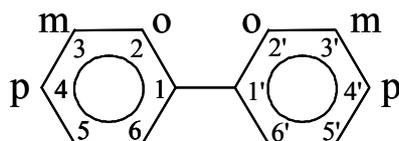
Studies of dioxins in food by Food Standards Australia and New Zealand (FSANZ) and dioxins in agricultural commodities under the National Residues Survey by the Department of Agriculture, Fisheries and Forestry (DAFF) also contributed valuable information on dioxins.

## 1.2 Dioxins

Polychlorinated dibenzo-p-dioxins (PCDDs) and polychlorinated dibenzo-furans (PCDFs) are a group of persistent chlorinated chemical compounds that share certain similar chemical structures, properties and biological characteristics. For the purposes of the National Dioxin Program the term “dioxins” is often used in the broader sense to include both PCDDs and PCDFs, and dioxin-like PCBs or co-planar polychlorinated biphenyls (PCBs). Several hundred of these compounds - or congeners<sup>2</sup> exist, of which 29 are considered to have significant toxicity (WHO (1998), Van Leeuwen and Younes (2000); and van den Berg et al. (1998)). It is these 29 closely related toxic chemicals that are the subject of this report. The general formulae for each of these compounds are presented in Figure 1.1 and 1.2.



**Figure 1.1** The structures of polychlorinated (A) dibenzo-p-dioxins and (B) dibenzofurans.



**Figure 1.2** The structures of polychlorinated biphenyls (PCB).

Since dioxins occur as complex mixtures of congeners in most environmental media (air, water, soil), the concept of toxic equivalents (TEQs) has been developed. This concept allows the toxicity of a complex mixture to be expressed as a single number.

Available animal-based toxicological data have been used to generate a set of weighting factors, each of which expresses the toxicity of a specific congener relative to the mass of is 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD), the most studied and most toxic polychlorinated dibenzo-p-dioxin. Multiplication of the mass of the congener by its

<sup>2</sup> Closely related chemicals derived from the same parent compound.

weighting factor (or toxic equivalents factor, TEF) yields the corresponding TCDD mass (or TEQ). The total toxicity of any mixture is then expressed as the sum of the individual congener TEQs.

The most widely adopted system of TEQs is that proposed by the North Atlantic Treaty Organisation (NATO) and known as the International Toxic Equivalents Factors (I-TEFs). This system has been updated and expanded by the WHO into a scheme that includes factors for humans and other mammals, birds and fish. The World Health Organization TEFs from 1998 for the 29 closely related chemicals that are the subject of this report are listed in Appendix A.

The different TEFs for birds and mammals limit the usefulness of comparisons of TEQs across groups of different animal classes. Furthermore, there are no data on the relative toxicity of many of the congeners for Australian fauna, especially for the monotremes. Where comparisons have been made the TEQs were based on the TEFs for mammals.

### **1.3 Objectives**

This study formed part of the Ambient Environmental Levels section of the Data Gathering and Consolidation phase of the NDP. The overall objective of the study was to determine ambient environmental levels of dioxins in Australian fauna through characterisation of the levels of dioxin-like chemicals (PCBs, dioxins and furans) in terms of concentrations and toxic equivalents in fauna across a range of urban, agricultural and remote reference areas.

Specific aims of the study were to:

1. Consolidate the current state of knowledge on dioxin levels in Australian terrestrial fauna
2. Gain a greater understanding of dioxin levels in Australian terrestrial fauna by either direct sampling or by sampling from archives, or both.

## 2 Project Design

### 2.1 Collection strategy

The Australian Government Department of Environment and Heritage (DEH) requested that there should be no sampling of live fauna; therefore priority was given to collecting samples from recently deceased animals. The strategy for the collection of deceased Australian native fauna utilised a national network of remote collectors who provided invaluable collection assistance. Organisations and individuals providing this network service included project team members, National Parks and Wildlife Service personnel, relevant State Government employees in natural resource management occupations, wildlife rescue and recovery organisations and individuals from educational institutions.

For the majority of cases, tissue samples were obtained opportunistically for analysis from the carcasses of recently (less than two days) deceased fauna caused by vehicle or other accidents. Samples were also obtained from a limited amount of archived material. Sample collectors were provided with a form (see Appendix E) to collate known data on the specimen forwarded for analysis. Payment of all expenses incurred in forwarding samples to AGAL was offered to all collectors. A complete list of collectors, locations and specimens obtained for analyses is presented in Appendix F.

Dioxin-like chemicals are persistent semi-volatile and hydrophobic organic pollutants. Their physical-chemical properties result in their tendency to accumulate from the abiotic into the biotic compartments such as the lipids of animals (bio-concentrate). Furthermore food is an important indirect pathway of these chemicals into animals and humans and they have a tendency to increase in levels with increasing trophic level (biomagnify) in particular the terrestrial food chain or marine mammals. For the above reasons sampling concentrated on the fatty tissue of animals and the results expressed on per gram of lipid basis.

#### 2.1.1 Sampling sites

In accordance with the priority air sheds and catchments within the three broad regions defined by the Australian Government Department of the Environment and Heritage (DEH) (i.e. northern, south eastern and south western Australia), samples were sought from each region from sites classified as:

- Industrial
- Urban
- Agricultural
- Remote.

Samples were not available for some of the sites, so samples that were available were used and allocated to one of the above classifications.

For the assessment of fauna, a total of 66 specimens were collected from 44 sites around Australia. The sampling was initially aimed at:

1. Assessing the spatial variation of dioxins across Australia by obtaining a geographic spread of macropod samples across Australia

2. Investigating biomagnification in samples found in close proximity to sampling sites nominated in other studies but within the framework of the present study 'Determination of ambient levels of dioxins in Australia'.

It became apparent, early in the study, that macropods contained relatively low concentrations of dioxins. Therefore, it was decided that a wider range of animals should be included as part of the investigation of ambient concentrations of dioxins in fauna. However, the availability of additional samples was limited, in part due to the widespread drought in 2002 and also because some samples could not be provided in the regions requested. In a number of cases, our reliance on opportunistic collection and the use of archived material precluded more detailed site information from being collected.

External collectors were identified and enlisted through the efforts of regional fauna coordinators in the states and territories. In other cases contact was made directly through professional contacts in target regions. Sample collectors were requested to complete the collector's form provided in duplicate and return one copy to the Study team and include one with the sample sent to the Australian Government Analytical Laboratories (AGAL). Where possible, fauna collection sites coincided with soil and ambient air sampling sites in a number of regions common to the aquatic environments, soils, air and/or fire studies.

### **2.1.2 Sample collection**

An extensive sampling strategy was designed to provide a national perspective of dioxin levels in members of the Family Macropodidae (macropods), which are a group of common native herbivores. This group was chosen as they are all primary feeders, and members can be found across Australia. A second and more intensive sampling effort was undertaken to collect and analyse a variety of animals representing different trophic levels throughout the target regions in order to assess spatial variability and bioaccumulation within ecosystems.

Despite intensive effort to obtain a greater range and coverage of fauna samples within NSW, the state was relatively poorly represented. The extreme weather and bushfire conditions in many areas of south-eastern Australia late in 2002 and in early 2003 was a contributing factor. The collection strategy was heavily dependent on the cooperation of National Parks and Wildlife staff in New South Wales and many members of this staff were engaged in fire fighting operations during the collection phase of this study and were thus unable to concentrate efforts on the collection of deceased fauna for this research program.

Several samples from the ACT were collected but could not be used through lack of fatty tissue. The lack of fatty tissue was attributed in part to the poor condition of the animals due to the prevailing drought conditions during the sampling period.

Composite sampling was used where practical. In some cases this was essential to obtain enough lipid for the analysis. In other cases (e.g. the macropod samples from Para Wirra), samples were pooled across three animals to obtain a more representative sample of the local macropod population. It was considered that the exact coincidence of individual animals was not crucial, as animal movement would tend to integrate the conditions over an animal's home range. The pooled sample will yield a result that is

an average of the subsamples and will represent a greater range of the fauna than would individual samples even if there are differences between the subsamples.

Deceased animals were obtained from a number of sources. The most readily available source was from vehicle accidents. Sample collected from culled animals provided another opportunity; for example western grey kangaroo (*Macropus fuliginosus*) samples were obtained following a cull in the Para Wirra reserve in South Australia. In other cases samples were obtained from trapped animals, as occurred with the collection of dingos (*Canis familiaris dingo*).

**Table 2.1 Classification of fauna included in this study**

<b>Faunal class</b>	<b>Comment</b>	<b>Number of samples</b>
Birds	various birds	19
Dingo	placental mammal	2
Macropods	kangaroo and wallaby	22
Marine mammals	dugong, Australian sea lion, dolphin and whale	13
Monotremes	echidna and platypus	5
Other marsupials	possum, koala and bandicoot	4
Reptiles	Goanna	1
<b>Total</b>		<b>66</b>

Composite samples from several animals have been treated as a single sample

### 2.1.3 Range of fauna collected

The project aimed to provide an estimate of background levels of dioxin-like compounds in fauna across Australia. Many factors could influence the concentration of these contaminants in fauna, such as local and regional spatial variation as well as phylogenetic and trophic level. Firstly spatial variation was investigated, which required examining a single taxon and trophic level across a range of spatially separated sites. The second aim was to investigate a broad range of species and trophic levels, which required the collection of a variety of Australian fauna. The range of fauna collected was limited by the laboratory requirement of 10 g of lipid per sample, which effectively excluded most invertebrates and amphibians.

The final range of samples used in the study is shown in Table 2.1 and Figure 2.1.

### Assessment of spatial variation using macropods

To achieve the first objective, locally common members of the macropod family were selected for the national assessment to provide data on a consistent but low trophic level taxon. In particular macropods generally have a restricted home range (approximately 20-150 hectares). This would allow an assessment of spatial variability in dioxin levels that may be explained by region and land use. It was envisaged that macropod samples could be obtained from all land uses (remote, agricultural, urban and industrial). However, obtaining suitable kangaroo carcasses in urban and industrial areas was difficult. Consequently, the majority of specimens were obtained from remote and agricultural areas. Exceptions to this included samples collected from metropolitan areas near Melbourne and Perth. The range of locations of the macropod samples analysed is given in Table 2.2. Further field details of these samples are given in Appendix H.

It is pertinent to mention Para Wirra in South Australia due to the uniqueness of this particular site. The site is approximately 15 km from a town and is surrounded by native vegetation, however, it is less than 30 km from Adelaide, and therefore not strictly remote. In addition the site was not urban, industrial or agricultural, and is well away from any heavy industry. Therefore, Para Wirra was classified as (following the description of the collectors) as remote for the purposes of this study.

As previously mentioned, there were few samples available from NSW and ACT due to unforeseen weather conditions and low lipid content. For example, the samples collected in Canberra, ACT failed to yield sufficient lipid for analysis.

A list of the samples that were dissected but yielded insufficient lipid is given in Appendix G.

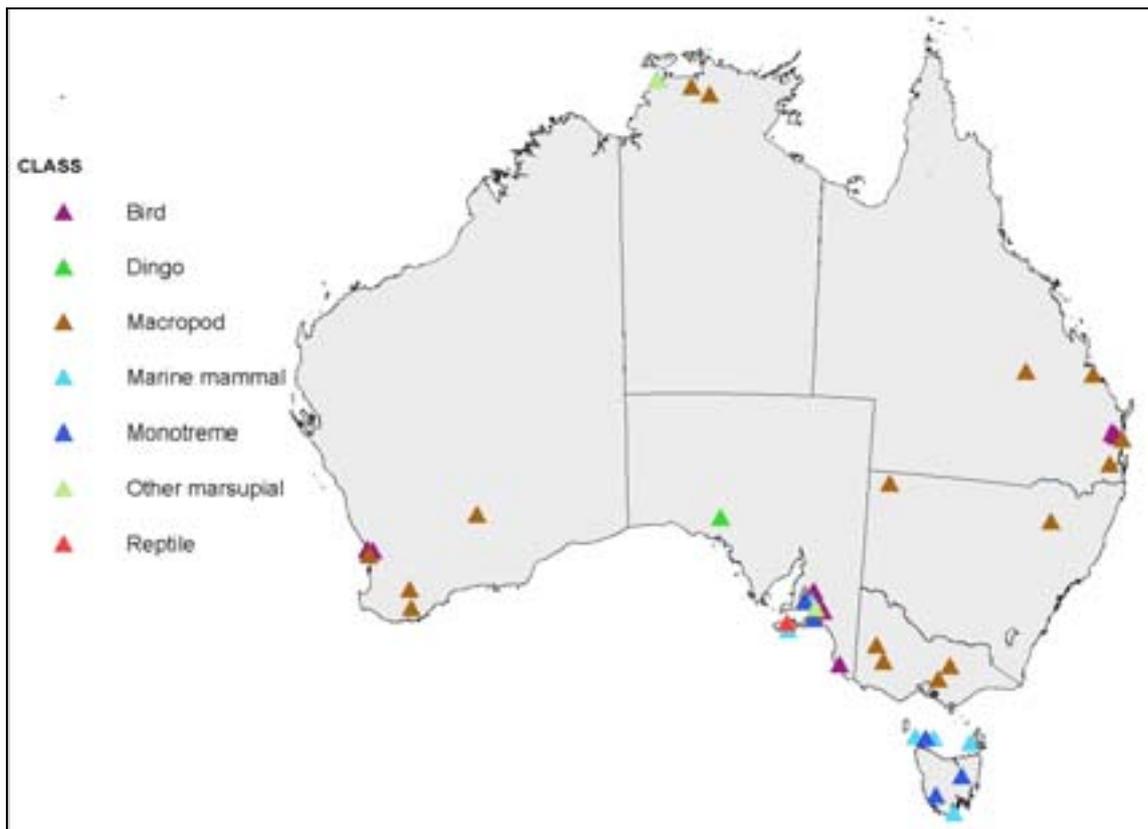
**Table 2.2 Number of Macropod samples by location and land use type**

Region	State	Urban/			State Total	Region Total
		Industrial	Agricultural	Remote		
South West	WA	1	3		4	
	Region	1	3			4
North	NT			3	3	
	Qld	1	3		4	
	Region	1	3	3		7
South East	NSW		1	1	2	
	Vic	2	2		4	
	SA		1	4	5	
	Region	2	4	5		11
Total by land use		4	10	8		22

### 2.1.3.1 Effects of animal class and trophic levels

The second objective of the sampling campaign was to measure the concentration of dioxin-like compounds in numerous species representing a range of fauna and trophic levels (from low-level herbivores to high level predators and carnivores). It was anticipated that this data would allow the quantification of any apparent biomagnification throughout the food chain. This could only be achieved in very broad terms because of the significant variation between specimens from the same site and because the samples were not coincident.

The range of fauna collected is shown in Table 2.1, whereas Table 2.2 and Figure 2.1 indicate the location of the various specimens collected.



**Figure 2.1** Location of fauna sampling sites

A number of Australian marsupials were included as a part of this study to enable a comparison between different types of marsupials. These included koala (*Phascolarctos cinereus*), possum (*Trichosurus vulpecula*) and bandicoot (*Isodon macrourus*). No carnivorous marsupials were available/received for this study, however two dingo (*Canis familiaris dingo*) samples were included to represent carnivorous placental mammals. Australia's two monotremes – the echidna (*Tachyglossus aculeatus*) and the platypus (*Ornithorhynchus anatinus*) – were also included in this study. Both are of a similar trophic level (i.e. they consume invertebrates), although the echidna is terrestrial and the platypus is aquatic.

In addition to the above, samples were obtained from species of marine mammals including whales (that had beached themselves), an Australian sea lion and two species of dolphins (all carnivores). A dugong, which is a herbivore, was also included to provide a contrast to the higher trophic levels. Birds were targeted because they cover a range of trophic levels, including the herbivorous galah (*Cacatua roseicapilla*), the pheasant coucal (*Centropus phasianinus*) that consumes invertebrates and small reptiles and birds of prey such as the sparrowhawk (*Accipiter cirrhocephalus*), which is at the top of the food chain. It was anticipated that the birds with a higher trophic level would have higher concentrations of dioxin-like substances due to biomagnification. The large birds of prey such as the wedge-tail eagle were collected opportunistically. Finally, two goannas were submitted to AGAL but only the heath goanna (*Varanus rosenbergi*) had sufficient lipid for analysis.

#### **2.1.4 Sampling, storage and handling methods**

AGAL technical staff advised the field collection coordinator, and in turn the remote collectors, of the most suitable methods of dissection and fat removal for the range of species to be analysed. Quantities of tissue from target species were collected opportunistically from large deceased animals in pre-selected locations. Typically, the tissue was from the base of the tail for large animals. For small animals typically the entire carcass was sent to AGAL for dissection.

It was requested by members of the project team that material should be as fresh as possible with no sign of decomposition, to permit handling and freight to AGAL and subsequent laboratory dissection and analysis. In the majority of cases, adequate quantities of fat or high-fat flesh were excised from the carcasses before being wrapped in aluminium foil, sealed in a plastic bag and deep-frozen. Once frozen, the fauna collection coordinator was advised of its availability and arrangements (including necessary transport permits) were made to facilitate prompt delivery to AGAL.

#### **2.1.5 Relevant protocols**

Sample collection, tissue extraction, preparation and forwarding protocols were distributed to all personnel assisting with the fauna collection program upon receipt of permits for the collection program. A copy of the field collection sheet is provided in Appendix E.

#### **2.1.6 Permit / approvals requirements**

Permit requirements for each State and Territory were arranged through consultation with the relevant State government agencies. Scientific Purposes Permits were obtained to allow for essential interference with deceased Australian fauna. In addition, permits were required for the transport of fauna tissue from the State/Territory of origin to AGAL, Sydney. AGAL obtained an import permit to allow material to be received for analysis<sup>3</sup>.

### **2.2 Methods used for chemical determinations**

The following method was used for determination of tetra- through octa-chlorinated dibenzo-p-dioxins (PCDDs), dibenzofurans (PCDFs) and polychlorinated biphenyl congeners (PCBs) in biological matrices by high-resolution gas chromatography/high resolution mass spectrometry (HRGC/HRMS).

This method provided data on all toxic 2,3,7,8-chlorinated PCDD (seven) and PCDF (ten) isomers, plus the 12 'dioxin-like' PCB congeners designated as toxic by the World Health Organization (WHO). The PCDD/PCDFs and PCBs were determined by the isotope dilution quantification technique. This technique allows determination of the dioxin toxicity equivalent ( $TEQ_{DF}$ ) as well as the PCB toxicity equivalent ( $TEQ_P$ ) for the 'dioxin-like' PCBs in a sample using WHO98 toxicity equivalency factors (TEFs). The total toxic equivalents ( $TEQ_{DFP}$ ) were calculated as the sum of  $TEQ_{DF}$  and  $TEQ_P$ .

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<sup>3</sup> As a condition of licence approval, the relevant State and Territory government departments administering the permits require a copy of the final fauna assessment report.

The detection limits and quantification levels in this method were usually dependent on the level of interferences rather than instrumental limitations. The analytical methodologies for the determination of PCDD/PCDFs and PCBs were based on USEPA methods 1613B and 1668A, respectively.

Subsamples from a given animal were combined to produce a representative sample of a particular tissue type. Samples were analysed by AGAL which is accredited for this type of analysis. In brief: a rendered or extractable portion of lipid was removed and spiked with a range of isotopically labelled surrogate standards. Clean up included partitioning with sulfuric acid then distilled water. Further purification was performed using column chromatography on acid and base modified silica gels, neutral alumina and carbon dispersed on celite. After clean up, the extract was concentrated to near dryness. Immediately prior to injection, internal standards were added to each extract, and an aliquot of the extract was injected into the gas chromatograph. The analytes were separated by the GC and detected by a high-resolution ( $\geq 10,000$ ) mass spectrometer. The quality of the analysis was assured through reproducible calibration and testing of the extraction, clean up, and GC/MS systems.

Further details are given in Appendix B.

## **2.3 Data quality**

The sampling scheme did not control for several variables, including different ages of animals, different collectors and biases in animal selections. These variables are discussed below.

There was no convenient method for aging the animals. This may be of consequence, as the dioxin-like compounds would accumulate in the fatty tissue of animals. Initially, when the animal is in a growth phase, the development of fatty tissue could potentially dilute the concentration of the dioxin-like congeners, but this effect will cease when the animal achieves its full size. Older animals would therefore be expected to carry a higher load of dioxin-like congeners than would younger animals. The sampling scheme would have provided an average across ages so the scheme would not be biased. The effect of age would therefore contribute to the uncertainty of the sampling scheme.

The manner of animal collection may have caused a bias. For example, road kills by definition must be near roads. The concentration of people into cities meant that there was more likelihood of a dead animal in the city being sampled than there would have been for a dead animal at a remote site. A further type of bias would arise from the possible increased likelihood of animals with a heavy load of contaminants to die. No allowance was made for these types of biases.

There was some variability in the condition of the animals. For example, the dingoes had been in a trap for a day before sampling. However, the dioxin-like compounds are very stable and, hence, are unlikely to decompose during handling.

The study included replicate laboratory analyses, replicate field samples and inter-laboratory controls to ensure the quality of the data. Between animal variation in the field was the largest source of variation. Details of this are given in Appendix J.

The field variability is of interest in its own right. This prompts questions as to what other factors are contributing to this variability. These factors may include age, breeding status or variability in the food chain.

Finally, this study of the variability of the samples from similar environments indicates that caution is required in the interpretation of the results presented in this study. From Appendix J it can be seen that the average CV is 79%. The CV for TEQ (including LODs as 0.5 the LOD) was 85%. This large CV means that a doubling of concentration between samples could well be attributable to sampling variation.

## 2.4 Error Bars

Error bars have been included on many of the barcharts presented in this report. The error bars give a measure of the within group variability and represent an estimate of the sample standard deviation estimated from the raw data. The standard deviation in this case includes not only the observed field variation but it may also include differences among species within the group and perhaps regional variation.

The standard deviation of necessity was based on only a few observations and is, therefore, itself very much subject to sampling variation. The presence of outliers also contributes uncertainty to the estimate of the standard deviation (see Cochran 1963, p44).

Despite the above shortcomings, bars representing standard deviation have been included to provide some measure of within group variability.

## 2.5 Below the limit of detection

There were many cases where the concentrations are less than the limit of detection (LOD). There are several ways of replacing the <LOD values with some estimate. Ideally some modeling technique can be used to obtain this estimate. However, in the current case there were too many unknowns and too few samples to produce a meaningful model that could be used to estimate the <LOD values.

The LOD varied between samples partly because of the amount of lipid that was available for the analysis varied between samples, but also some congeners may be masked by high concentrations of other components. In some cases the LOD was elevated due to high concentrations of related compounds. For example, the concentration of PCB 167 was quoted by AGAL as <40,000 pg g<sup>-1</sup> lipid.

Frequently <LOD values are quoted as 0.5 LOD. The rationale for this is the implicit assumption that the true value could occur with equal likelihood between zero and LOD – the central point of such a distribution is  $0.5 \times \text{LOD}$ . Often the choice of method for estimating the value does not matter to a large extent. However, in some cases, where the TEF is moderate, the <LOD values can have a relatively large effect on the TEQ. In such cases the TEQ can be artificially inflated due to the masking of one congener by another, and using the  $0.5 \times \text{LOD}$  rule.

The alternative extreme is to treat the <LOD values as zero. This method underestimates the TEQ.

The approach adopted in this report was to treat the <LODs as  $0.5 \times \text{LOD}$ .

### 3 Dioxin Levels in Australian Fauna

#### 3.1 PCDD/PCDFs and dioxin-like PCBs in fauna

An initial analysis of the data was undertaken by classifying the fauna into seven classes based on their taxonomy and ecology (Table 2.1)<sup>4</sup>. The average, minimum and maximum middle bound (incl. ½ LOD values) concentrations of PCDD/PCDFs and dioxin-like PCBs (on a TEQ<sub>DFP</sub> basis) across each of the fauna classes is summarised in Table 3.1. The distribution of PCDD/PCDFs and dioxin-like PCBs within the individual fauna classes, in particular for the bird, macropod and marine mammals, were skewed. This is indicated by the relatively large difference between the maximum and average values.

**Table 3.1 Comparison of average, minimum and maximum TEQ<sup>5</sup> for each class of fauna**

Fauna class				Marine	Other		
	Bird	Dingo	Macropod	mammal	Monotreme	marsupial	Reptile
Number of Samples	19	2	22	13	5	4	1
Average	730	2	2.7	80	30	4.4	0.51
Minimum	0.63	1.7	0.14	1.1	9.3	0.95	0.51
Maximum	3,900	2.3	25	590	60	13	0.51

The fauna concentration data are further presented in Figure 3.1 and clearly display some outlying concentrations. Concentrations positioned outside the “whiskers” were attributed to the wide range of species and locations used in this study. For example, within the bird class, a species with a relatively low concentration of PCDD/PCDFs and dioxin-like PCBs was the galah. Within the marine mammal class the species with a low concentration was the dugong sampled from Darwin, which like the galah, is a low trophic consumer (seed and seagrass grazers, respectively).

<sup>4</sup> Note that the term ‘class’ is not used in the taxonomic sense in this report. There are instances where the members of each fauna class are not trophically similar. For example, the galah (*Cacatua roseicapilla*), which is a primary feeder, is grouped with birds of prey. A compromise was necessary between having internally homogenous classes as opposed to too many classes.

<sup>5</sup> TEQs (pg TEQ<sub>DFP</sub> g<sup>-1</sup> lipid) were calculated with mammalian TEFs for dingos, macropods, marine mammals, monotremes and other marsupials. TEQs for reptiles and birds were calculated using avian TEFs as discussed in Section 1.2

### Box 1: Box and whisker plots

Box and whisker plots are a widely accepted way of presenting environmental data. They show where the data points are concentrated (the box) and the outlying values (the whiskers, open and closed circles). Box plots are often used to compare several sets of data.

Here a plot is used, where the boxes represent the 25<sup>th</sup> and 75<sup>th</sup> percentiles (1<sup>st</sup> and 3<sup>rd</sup> quartiles). The top of the box in these plots is the 75<sup>th</sup> percentile (75 % of the data fall below this line), while the bottom of the box represents the 25<sup>th</sup> percentile (25 % of the data fall below this line). The line in the middle of the box represents the median (50 percent of the data fall above and 50 percent below this number).

The whiskers on the box extend to data points that are up to 1½ times the Inter Quartile Range (IQR). The IQR is defined as the difference between the 75<sup>th</sup> and the 25<sup>th</sup> percentiles, and is equal to the range of about half the data. Outliers which are less than three times the IQR are shown as open circles, while those greater than three times the IQR are shown as closed circles. The statistical and graphical package XLSTAT was used to produce all box plots and calculate percentiles.

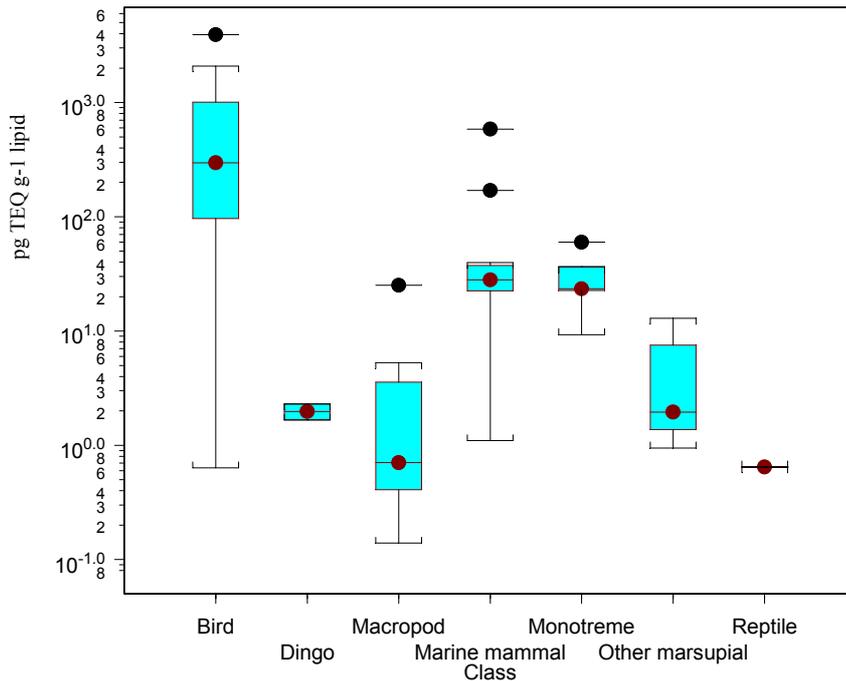
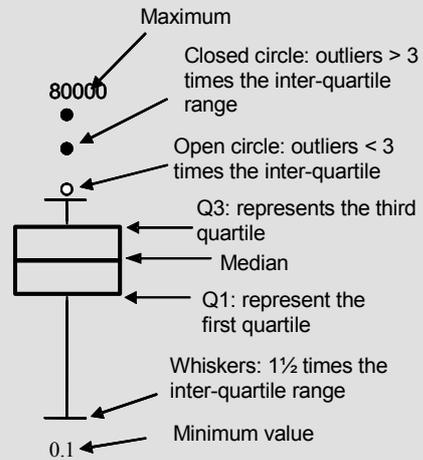
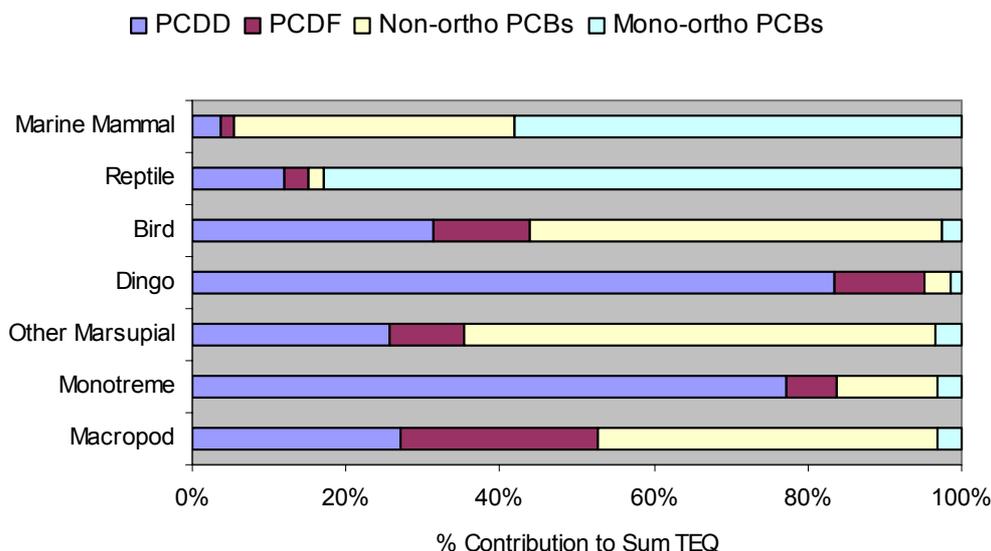


Figure 3.1 Concentrations of PCDD/PCDFs and PCBs

The TEQ for each sample was calculated according to the method described in Section 1.2. This TEQ consists of three components – TEQ<sub>D</sub>, TEQ<sub>F</sub> and TEQ<sub>P</sub>, which together make the TEQ<sub>DFP</sub>. TEQ<sub>DFP</sub> is usually abbreviated to TEQ. The TEQ<sub>P</sub> may be partitioned into non-ortho and mono-ortho components.

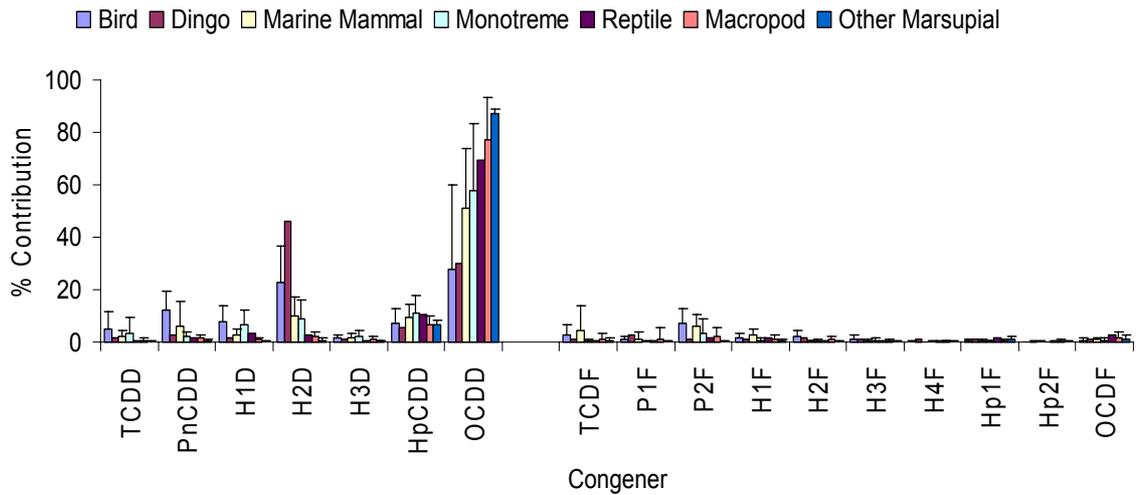
Dioxin-like PCBs contributed only a small fraction of the  $TEQ_{DFP}$  in the dingo and monotremes samples. Conversely, dioxin-like PCBs, in particular mono-ortho PCBs, contributed principally to the sum of TEQs within the marine mammal and the reptile samples. With respect to the other fauna classes (i.e. other marsupials, birds and macropods) non-ortho PCBs were the primary contributors to the  $TEQ_{DFP}$ .



**Figure 3.2 Comparison of the percent contribution of PCDD/PCDFs and dioxin-like PCBs to the sum of the TEQ ( $WHO_{98}-TEQ_{DFP}$ ) across each faunal class<sup>6</sup>**

Octa-chlorinated dibenzo-p-dioxin (OCDD), contributed primarily to the total PCDD/PCDF concentration in most of the fauna classes sampled (Figure 3.3). 1,2,3,4,7,8-Hexa-chlorinated dibenzo-p-dioxin (H2D) contributed 50% to the total PCDD/PCDF concentration in the dingo sampled from Ceduna, SA, despite having contributed notably less to other fauna classes represented in this study. Most of the congeners contributed less than 20% to the concentration of the sum of PCDD/PCDF congeners.

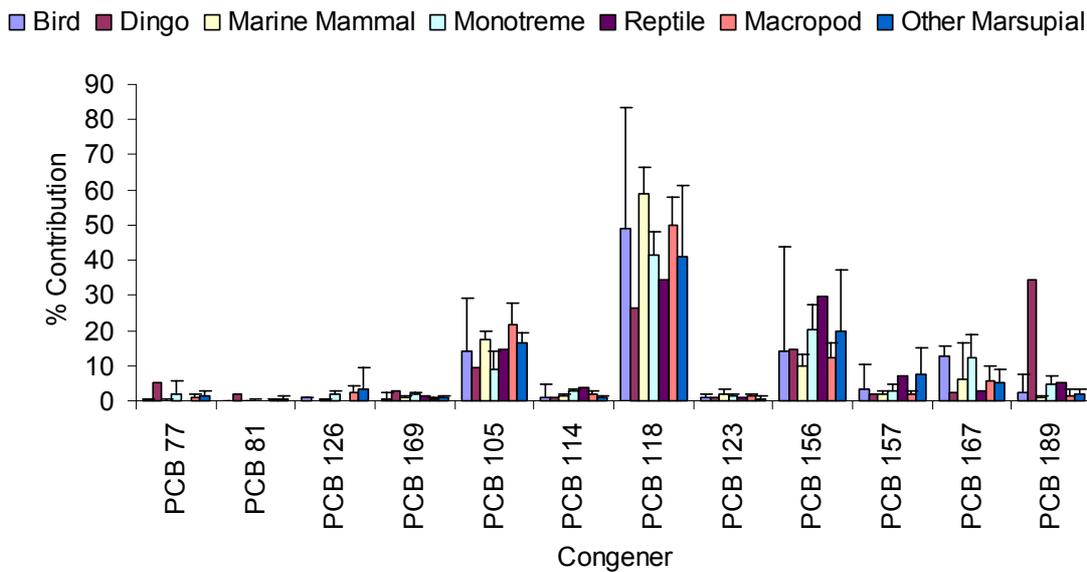
<sup>6</sup> TEQs were calculated with mammalian TEF for dingos, macropods, marine mammals, monotremes and other marsupials. TEQs for reptiles and birds were calculated using avian TEFs.



**Figure 3.3 Percent contribution of PCDD/PCDF congeners to the total PCDD/PCDF concentration in different fauna classes<sup>7</sup>**

Mon-ortho PCB 118 (2,3',4,4',5-pentachlorobiphenyl) contributed most to the sum of mono- and non-ortho PCBs for the majority of the fauna represented in this study (Figure 3.4). 2,3,3',4,4',5,5'-heptachlorobiphenyl (PCB 189), although having contributed a relatively small amount to the sum of the mono-ortho and non-ortho PCB concentration for the majority of the fauna classes, contributed substantially to the sum of dioxin-like PCBs for the dingo from SA. Mono-ortho PCBs 105, 156 and 167 (2,3,3',4,4'-pentachlorobiphenyl, 2,3,3',4,4',5-hexachlorobiphenyl, 2,3',4,4',5,5'-hexachlorobiphenyl) contributed at least 10% to the sum of mono- and non-ortho PCB concentration for the majority of classes while all non-ortho PCBs contributed a relatively small amount to the sum of dioxin-like PCBs.

<sup>7</sup> Error bars on congener profiles are standard deviation for samples with  $n > 2$



**Figure 3.4** Percent contribution of PCB congeners to the total PCB concentration in different fauna classes

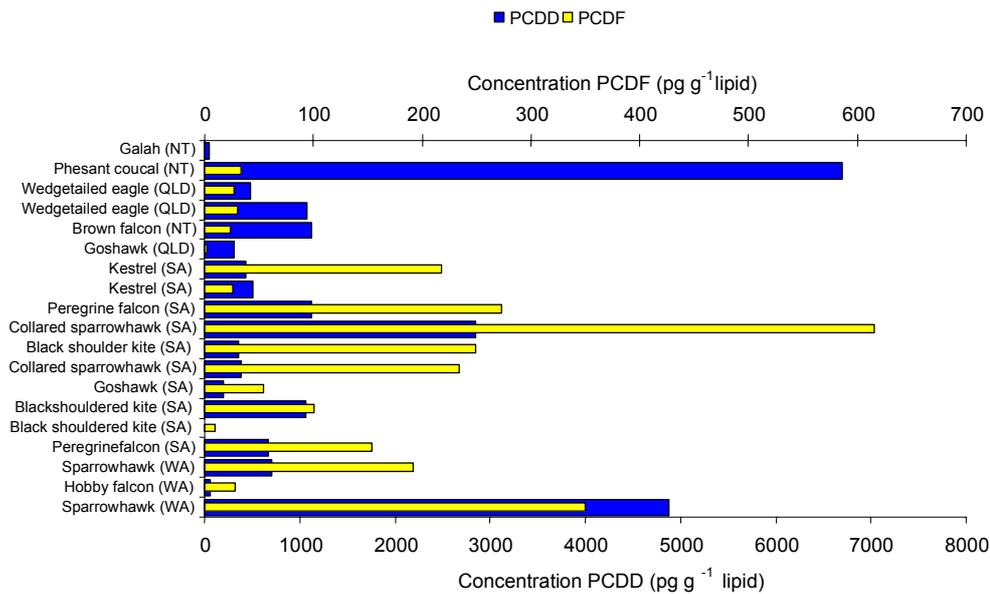
### 3.2 Concentration of PCDD/PCDFs and dioxin-like PCBs within fauna classes

The classification of the fauna into designated classes provided information on the average concentrations and congener profiles for these classes. However, within each of the fauna classes, in particular the birds, a number of different species were grouped together within the one class. To further investigate and provide more detailed assessment of the dioxin concentrations in Australian fauna, samples within each class were considered individually.

#### 3.2.1 Birds

Birds were one of the best represented fauna classes analysed as part of this study, with a total of 19 samples (10 different species) collected across four states, including the north, south-east and south-west regions. When comparing PCDD/PCDFs (Figure 3.5), PCDD concentrations were always greater than those for PCDF. High concentrations of PCDD were displayed for the pheasant coucal from Darwin, Northern Territory and the sparrowhawk from Perth in Western Australia relative to PCDD concentrations for other bird species sampled.

Relatively high concentrations of PCDF were found in one of the sparrowhawk samples from South Australia with at least double the concentration found than in any other of the bird samples. PCDF concentrations in bird species from the northern regions were generally lower than those found in species sampled in the south-east and south-west regions in Australia.

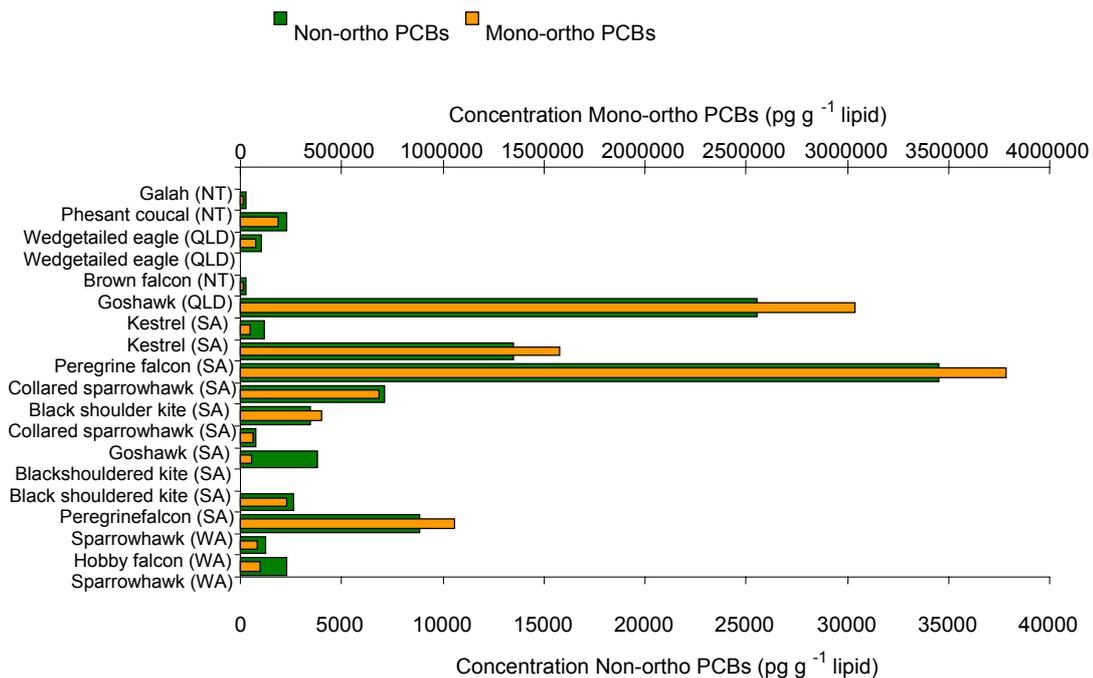


Note that scale for each horizontal axis is different

**Figure 3.5 Concentration of PCDD/PCDFs in the bird species analysed**

Mono-ortho PCB concentrations dominated the sum of PCB concentrations found in all of the bird species, when compared to non-ortho PCBs, by at least a factor of one hundred (Figure 3.6). The concentration of the sum of mono-ortho PCBs ranged from 980 pg g<sup>-1</sup> lipid (galah from Darwin, NT) to 3,800,000 pg g<sup>-1</sup> lipid (collared sparrowhawk from Adelaide, South Australia), whereas the concentration of the non-ortho PCBs ranged from 3 pg g<sup>-1</sup> lipid (galah from Darwin) to 35,000 pg g<sup>-1</sup> lipid (the collared sparrowhawk from Adelaide).

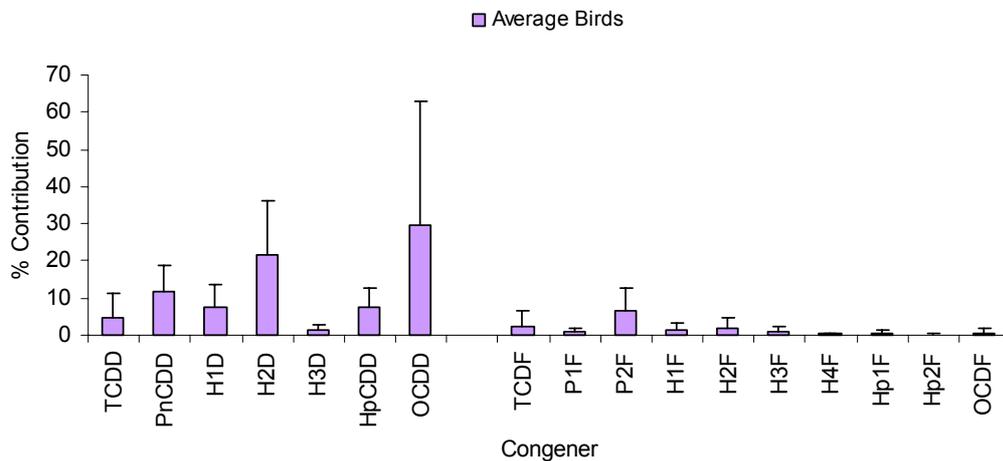
The high PCB values in the collared sparrowhawk from Adelaide, South Australia were accompanied by high levels of PCDDs. PCDFs were also elevated but their concentration was always less than a tenth of that of the corresponding PCDDs.



Note that scale for each horizontal axis is different

**Figure 3.6 Concentration of dioxin-like PCBs in the birds**

Congener profiles of a number of PCDD/PCDFs indicate that the percent contribution of dioxins is greater than the furans (Figure 3.7). Octa-chlorinated dibenzo-p-dioxin dominated the profile, however the standard deviation was considerable, therefore, individual profiles for species from different states were analysed.



**Figure 3.7 Percent contribution of PCDD/PCDF congeners to the total PCDD/PCDF concentration<sup>8</sup>**

<sup>8</sup> Error bars on congener profiles are standard deviation for samples with n > 2

There were similar congener profiles displayed for the two peregrine falcons from two different locations in South Australia (Figure 3.8). The percentage contribution of 1,2,3,4,7,8-hexachlorinated dibenzo-p-dioxin was more than 5% higher for the peregrine falcon from Port Lincoln than the sample from Adelaide, but this is well within the expected range of sampling variation. Dioxins contributed the greatest amount to the sum of PCDD/PCDF concentration, particularly the penta- and two of the hexa-chlorinated dibenzo-p-dioxins (PCDD, H1D and H2D).

The PCDD/PCDF congener profiles for black shouldered kite from South Australia varied markedly between samples from different locations, which could be a result of location, gender or age as well as sampling variation. The sample from Bremer River South Australia was dominated by 1,2,3,6,7,8-HxCDF, but with significant contributions from 2,3,4,7,8-PeCDF and 1,2,3,6,7,8-HxCDF. The sample from Elizabeth also had a high proportion of 1,2,3,6,7,8-HxCDF, but also high levels of TCDD and PeCDD and low contributions from furans. The sample from Penola not only had a high proportion of 1,2,3,6,7,8-HxCDF, but also of HpCDD and OCDD with significant contributions from 2,3,4,7,8-PeCDF and HxCDFs. The proportion of OCDD in the sample from Penola was double that from any of the other samples and should therefore be considered significant.

The goshawks and the sparrow hawks were dominated by dioxins rather than furans with about 30% of the sum coming from 1,2,3,6,7,8-HxCDD, with significant contribution from other PCDDs. The main contributions from the furans came from 2,3,7,8-TCDF and 2,3,4,7,8-PeCDF.

The kestrels were again dominated by dioxins but in that case the main contribution was from OCDD. The difference between the kestrels and other birds is quite marked and may be attributed to selective uptake of dioxins or perhaps some mechanism the kestrel have for coping with the low chlorinated congeners of dioxins or perhaps other factors such as location and diet.

The congener profiles of the birds from Western Australia, Queensland and the Northern Territory are shown in Figure 3.9. The collared sparrowhawk had a similar profile to the sparrowhawk from South Australia, with a large contribution from 1,2,3,6,7,8-HxCDD. The hobby falcon had equal contributions from 1,2,3,6,7,8-HxCDD and HpCDD as well as significant amounts of PeCDD and 2,3,4,7,8-PeCDF.

The samples from the wedge-tail eagle had mainly dioxins rather than furans. About 30% of the dioxins were OCDD, but there were also significant contributions from PeCDD, 1,2,3,4,7,8-HxCDD and 1,2,3,6,7,8-HxCDD and minor contributions from the other dioxin congeners.

The galah, pheasant coucal and brown falcon were all from the Northern Territory. These samples were completely dominated by OCDD with traces of the other dioxin congeners and no detectable furans.

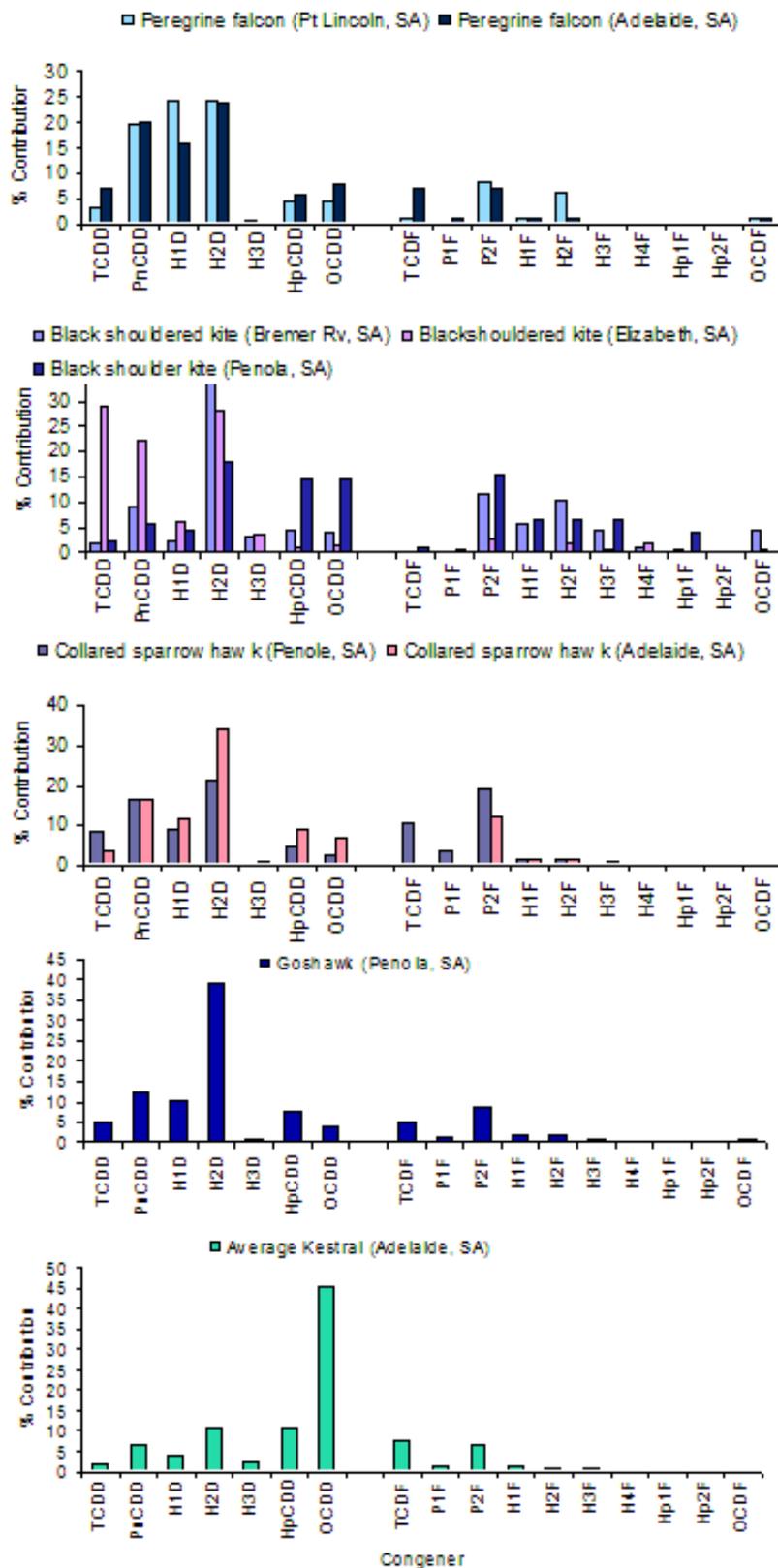
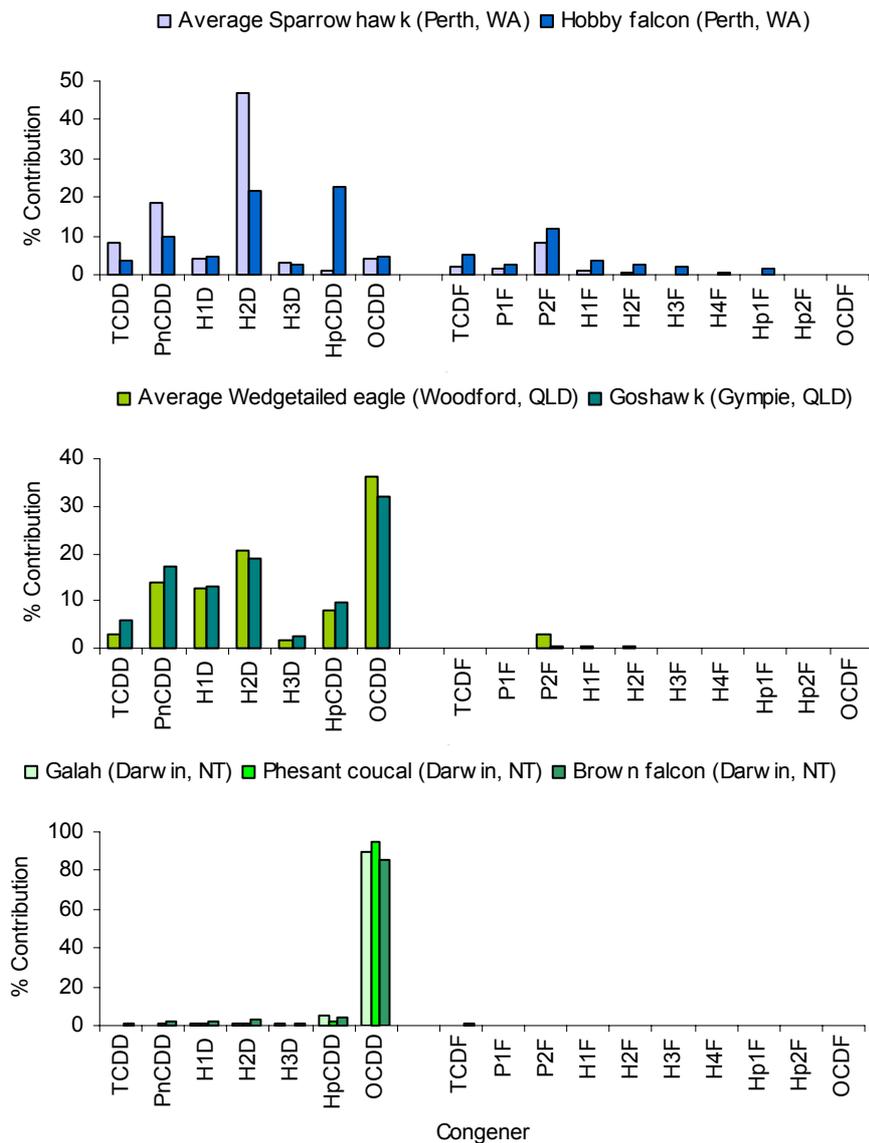
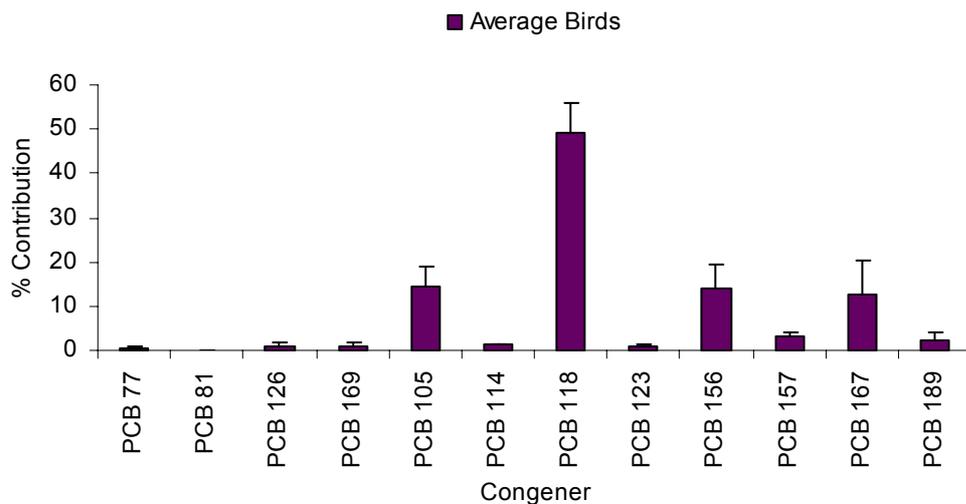


Figure 3.8 Percent contribution of PCDD/PCDF congeners to the total PCDD/PCDF concentration in individual bird species from SA



**Figure 3.9** Percent contribution of PCDD/PCDF congeners to the total PCDD/PCDF concentration in individual bird species from WA, QLD and the NT

The average PCB profile (Figure 3.10) was dominated by PCB 118, which contributed almost 50% of the total load. PCBs 105, 156 and 167 each typically contributed about 15%. Contributions from the other PCBs were minor.



**Figure 3.10** Percent contribution of dioxin-like PCBs to the total PCB concentration in birds

**Table 3.2** Predominant congeners encountered in the samples of birds

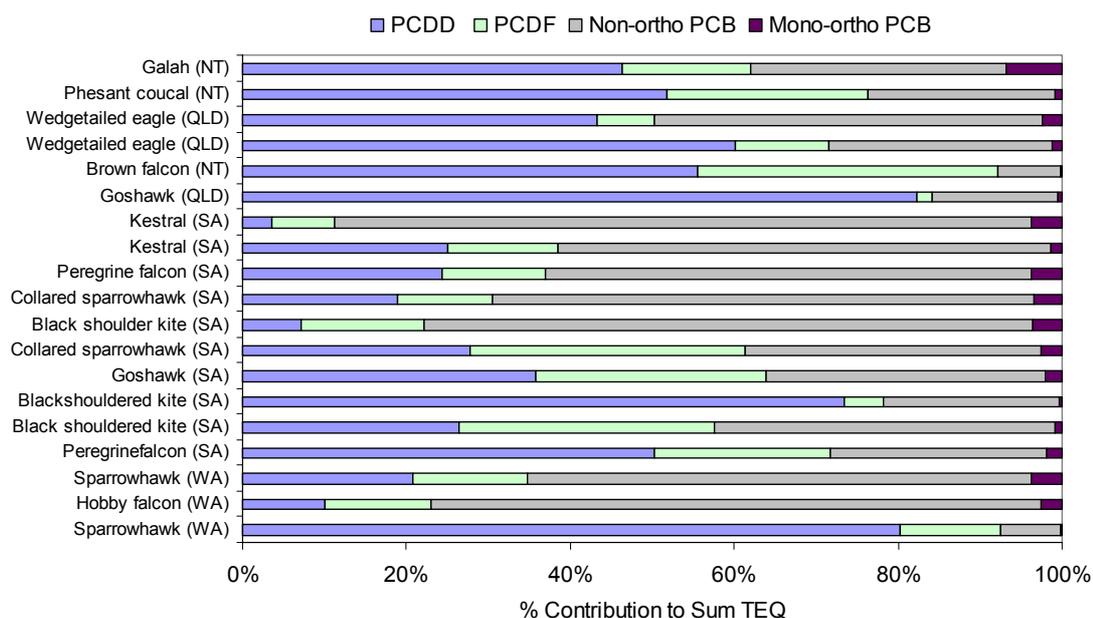
Common name	State	Sample number	2,3,7,8-TCDD	1,2,3,7,8-PeCDD	2,3,7,8-TCDF	2,3,4,7,8-PeCDF	PCB 77	PCB 81	PCB 126	TEQ
Galah	NT	7	0	0	0	0	0	0	0	1
Black shouldered kite	SA	13	0	3	0	3	0	0	5	12
Brown falcon	NT	9	6	20	14	5	2	1	2	52
Goshawk	QLD	4	18	54	0	2	1	1	12	92
Pheasant coucal	NT	3	6	38	0	23	0	0	22	99
Kestrel male	SA	16	5	23	7	9	31	5	39	120
Hobby falcon	WA	3	3	9	5	11	19	8	70	130
Brown goshawk	SA	12	12	32	13	22	8	5	32	130
Eagle-breast	QLD	5	16	110	0	25	7	4	50	220
Eagle-liver	QLD	6	23	86	0	18	14	8	110	280
Peregrine falcon	SA	18	26	160	11	68	22	6	75	400
Collared sparrowhawk	SA	10	52	100	65	120	31	18	150	570
Black shouldered kite	SA	15	15	34	5	94	36	21	490	740
Black shouldered kite	SA	14	340	260	2	33	4	5	168	830
Sparrowhawk	WA	2	58	160	28	120	120	40	510	1100
Peregrine falcon	SA	19	100	280	97	100	150	85	720	1600
Sparrowhawk	WA	1	510	1000	39	190	14	9	120	2000
Kestrel-female	SA	17	17	57	90	72	430	36	1400	2200
Collared sparrowhawk	SA	11	120	570	14	420	250	130	2200	3900

Key to colours: (The units are TEQ avian g<sup>-1</sup> lipid).

<10      10-100      >100

The largest percentage contribution to the sum of the TEQ in birds was typically from the non-ortho PCBs or the PCDDs with a smaller contribution from the PCDFs (see Figure 3.11). The mono-ortho PCBs made little contribution. There were some extreme cases such as one of the kestrels from South Australia had little PCDD and a large contribution from the non-ortho PCBs. One of the black-shouldered kites from South Australia and the hobby falcon from Western Australia also had relatively low contributions of PCDDs. At the other extreme, the goshawk from Queensland and one of the sparrowhawks from Western Australia had over 80% of the TEQ contributed from PCDDs. The contribution to TEQ of PCDFs also varied from an almost negligible amount in Queensland goshawk to over 30% in the brown falcon from Northern Territory, a collared sparrowhawk, the goshawk and a peregrine falcon (all from South Australia).

Details of the absolute contributions to the TEQs of seven congeners that contributed significantly to the TEQ are given in Table 3.2. The largest contributions were by far from PCB 126 and then by 1,2,3,7,8-PeCDD, with about equal contributions from 2,3,7,8-TCDD, 2,3,4,7,8-PeCDF and PCB 77. Other features, such as the relatively low contribution of PCBs to the TEQ in the Queensland goshawk are better studied from Table 3.2.

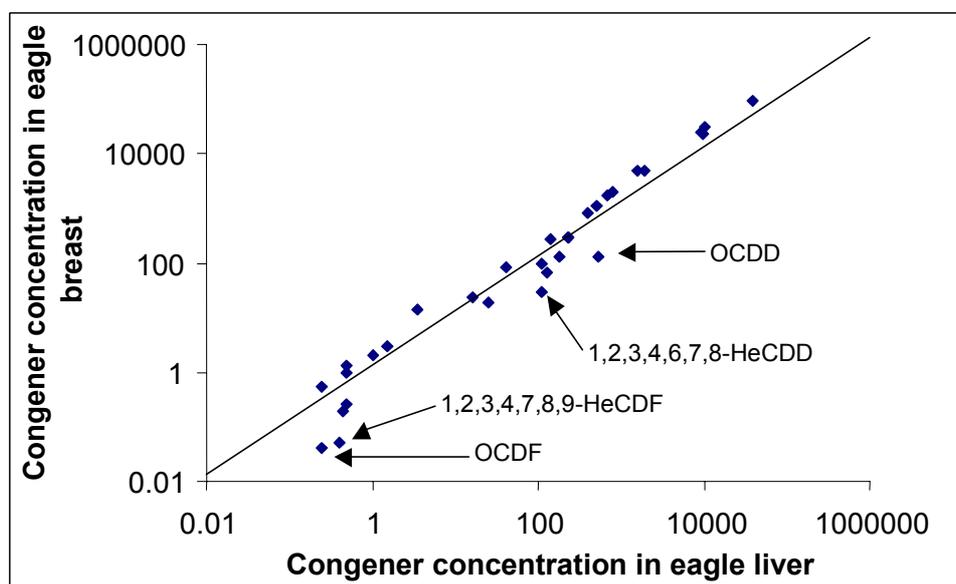


**Figure 3.11 Percent contribution of PCDDs, PCDFs, non-ortho PCBs and mono-ortho PCBs to the total TEQ concentration in birds**

As a part of this study, both liver and breast tissue were collected from a wedge-tail eagle collected from Queensland. The concentration of PCDD/PCDFs and dioxin-like PCBs in the liver was 225 pg TEQ<sub>DFP</sub> g<sup>-1</sup> lipid, with highest contributions to the TEQ<sub>DFP</sub> from the congeners 2,3,7,8-TCDD (16 pg TEQ g<sup>-1</sup> lipid), 2,3,4,7,8-PeCDF (25 pg TEQ g<sup>-1</sup> lipid), 1,2,3,7,8-PeCDD (110 pg TEQ g<sup>-1</sup> lipid) and PCB 126 (50 pg TEQ g<sup>-1</sup> lipid).

The data offer a comparison of the congener concentrations between liver and breast tissue of the eagle. A graphical comparison is given in Figure 3.12. There was a strong relationship between the concentrations in the two tissue types, with the overall concentration being higher in the breast tissue, although the observed difference was within the bounds of laboratory variation. There were some exceptions, all of which were heavily chlorinated congeners, and included OCDD, OCDF, 1,2,3,4,6,7,8-HpCDD and 1,2,3,4,7,8,9-HpCDF.

Overall, the points shown in Figure 3.2 are close to the 1:1 line, indicating that the congener profiles are similar in the two tissues. This result suggests that there are no gross differences between the different tissue types. It gives some support for using entire animals where that was required and taking subsamples in other cases. However, caution should be used in this interpretation as it based on only two tissue types from a single animal.

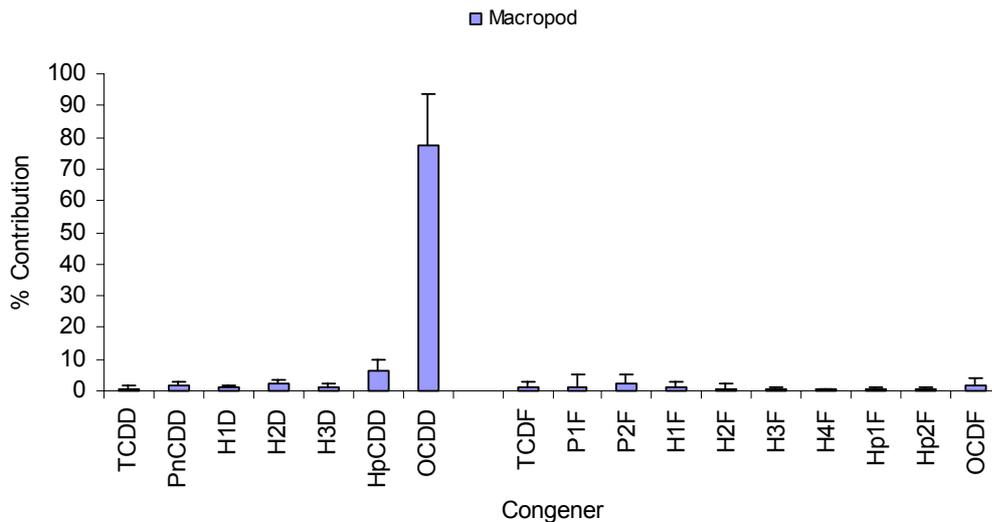


**Figure 3.12 Comparison of congener concentrations ( $\text{pg g}^{-1}$  lipid) in liver and breast tissue of an eagle**

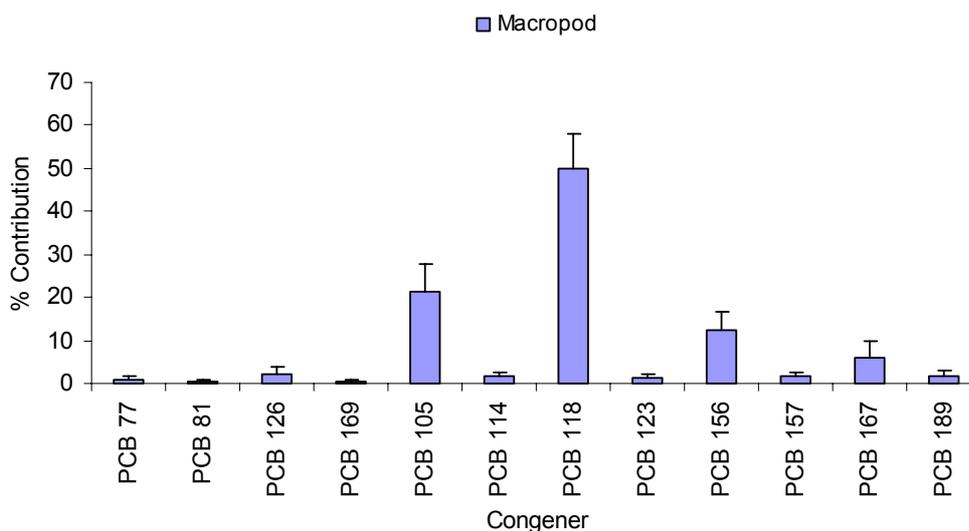
### 3.2.2 Macropods

In total 22 Macropod samples (including wallabies and kangaroos) were analysed as a part of this study and these were collected from 6 states throughout Australia. In general the concentration of all PCDD/PCDF and PCB congeners were relatively low (on a TEQ basis) and in some cases, for example the sample from Tibooburra, NSW congeners were either below the LOD or found in trace amounts (PCB 77, PCB 169, and PCB 189).

The PCDD/PCDF congener profile (expressed as a percentage contribution of individual PCDD/PCDF congeners of the sum of all the 2,3,7,8-substituted PCDD/PCDFs) was similar across the macropod samples. The profile was dominated by PCDDs, in particular OCDD (Figure 3.13). Furthermore, a similar PCB congener profile was also observed across the macropods, with the dominance of PCB 118 (Figure 3.14).



**Figure 3.13** Percent contribution of PCDD/PCDF congeners to the total PCDD/PCDF concentration in macropods<sup>9</sup>



**Figure 3.14** Percent contribution of dioxin-like PCBs to the total PCB concentration in macropods

The most contaminated macropod sample (Sample 37 from Para Wirra) exhibited a different profile compared to the average macropod profile, as well as the other two samples from Para Wirra (Figure 3.15). In particular, the Para Wirra sample (Sample 37) had a lower contribution of OCDD because of a higher contribution of the lower chlorinated PCDFs (TCDF, PeCDF and HxCDF). These results indicated that there might be a specific PCDF contamination within this region. On the other hand, the PCB congener profile (Figure 3.16) was similar for the three pooled samples from Para Wirra, which was also similar to the average macropod PCB congener profile.

<sup>9</sup> Error bars on congener profiles are standard deviation for samples with n > 2

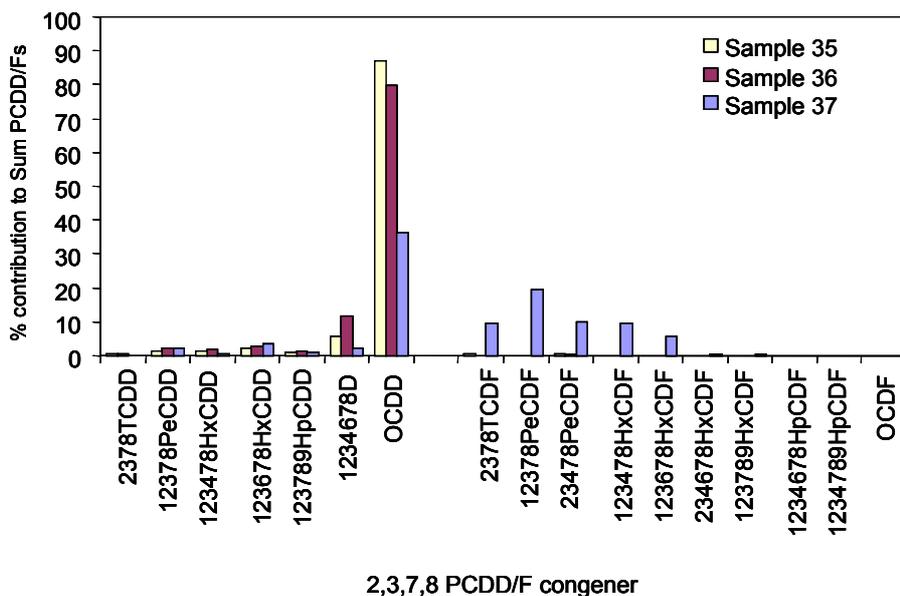


Figure 3.15 PCDD/PCDF congener profile in the 3 macropod samples from Para Wirra

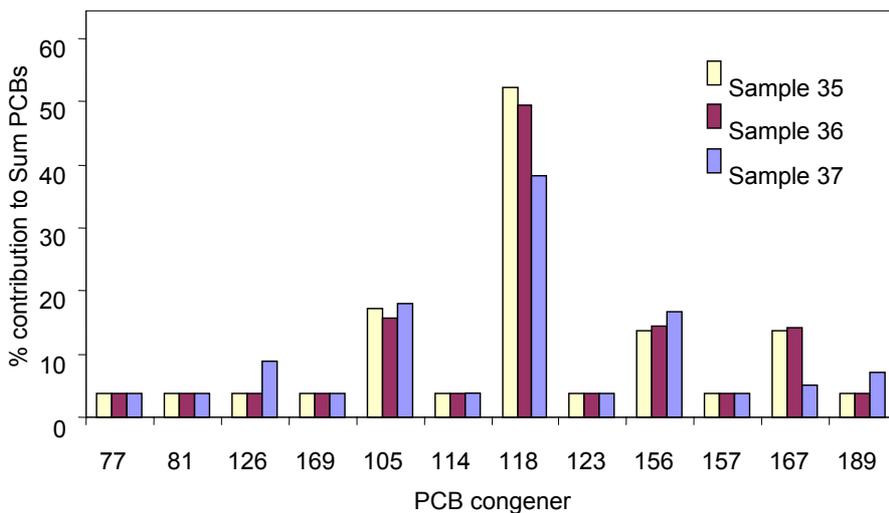
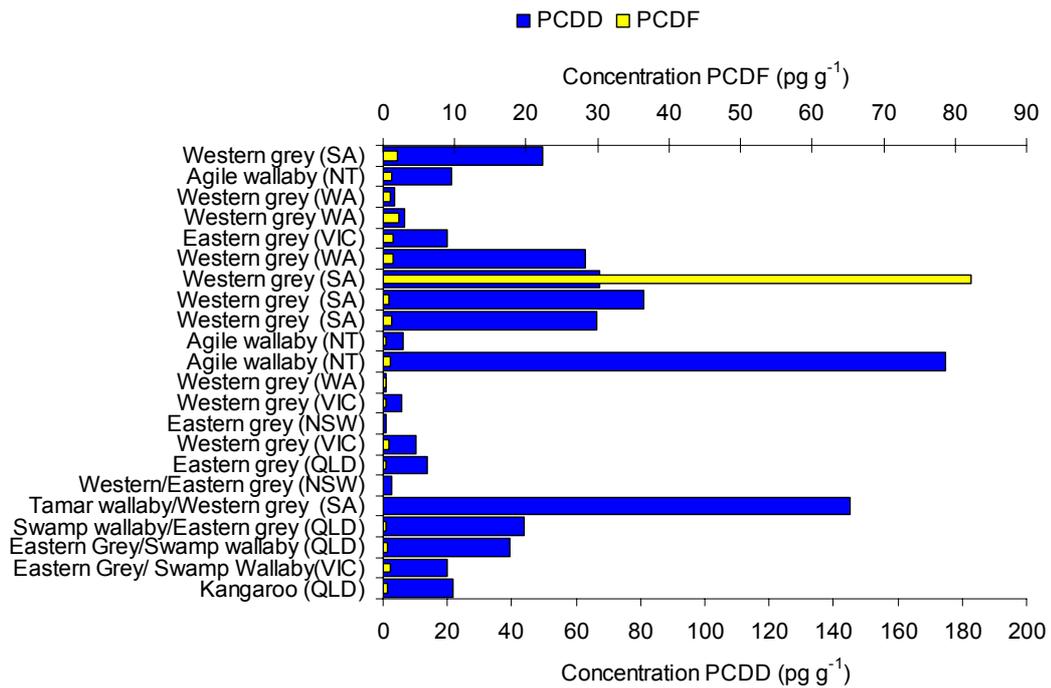
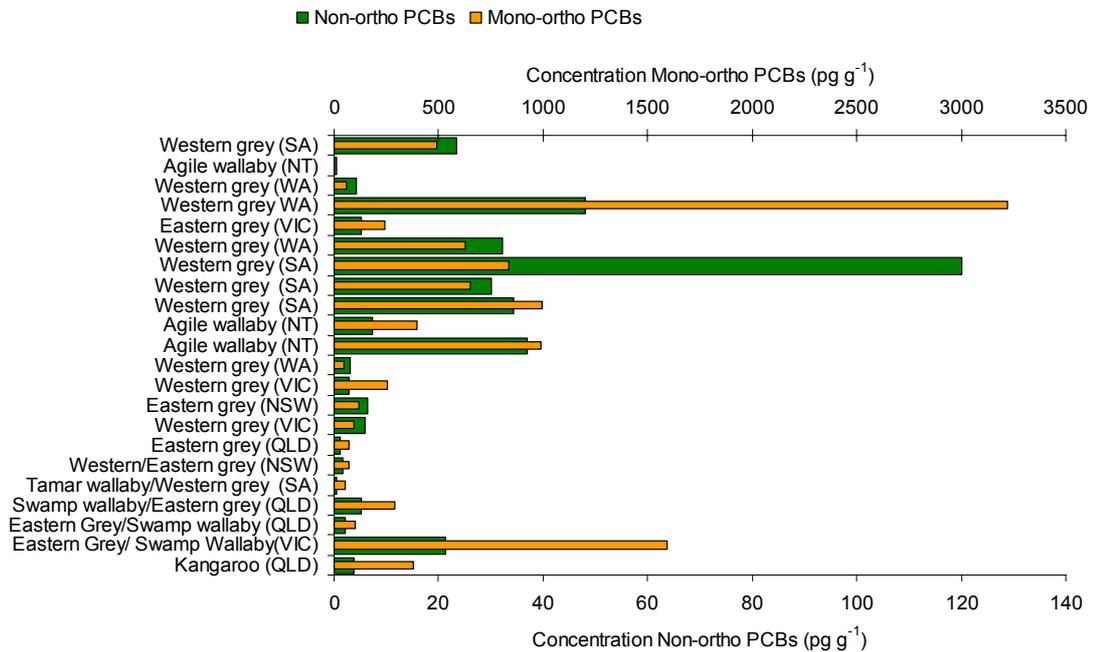


Figure 3.16 PCB congener profile in the 3 macropod samples from Para Wirra

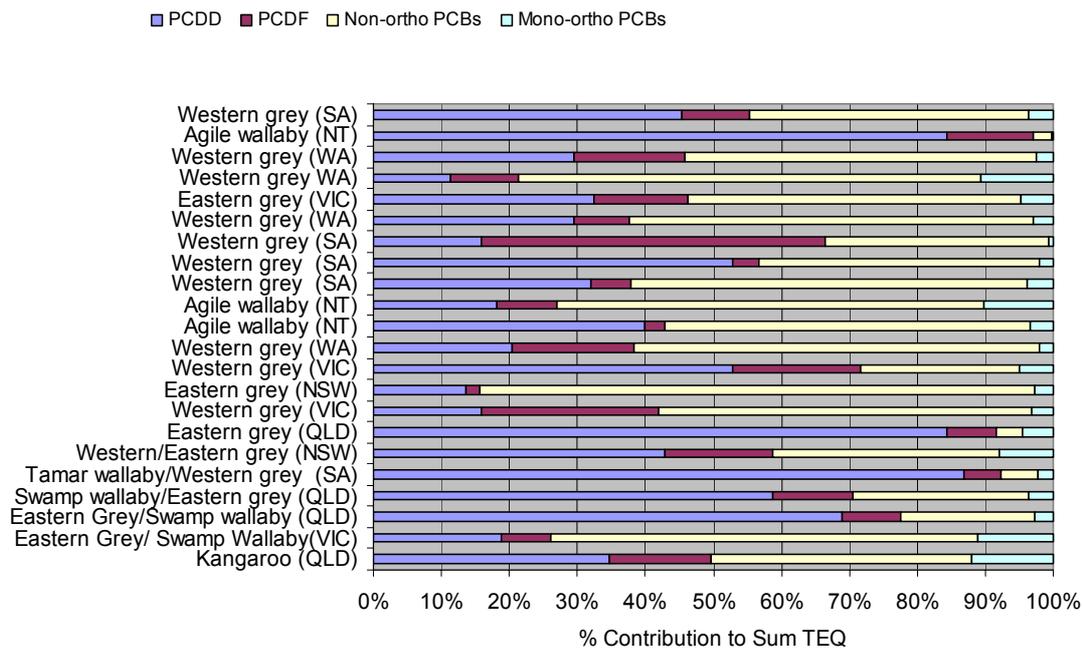
The relatively high concentrations of furans in Sample 37 are also exhibited in Figure 3.17. Further examination of that diagram also indicates that there is an almost similar disparity in PCDDs between the agile wallaby samples from the Northern Territory. This demonstrates the wide range of results from nominally similar samples. The relative concentrations of non-ortho and mono-ortho PCBs are shown in Figure 3.18. One notable feature of that diagram is that the same Para Wirra sample (Sample 37) showed a high level of non-ortho PCBs. Mono-ortho exceeded the non-ortho PCBs in all cases. This may indicate different sources of mono-ortho PCBs although the observed differences are within the range of variation encountered between nominally similar samples. The relative contribution of PCDDs, PCDFs, non-ortho PCBs and mono-ortho PCBs to the total TEQ concentration in macropods is given in Figure 3.19.



**Figure 3.17 Concentration of PCDD/PCDFs in the macropod samples collected from different States across Australia.**



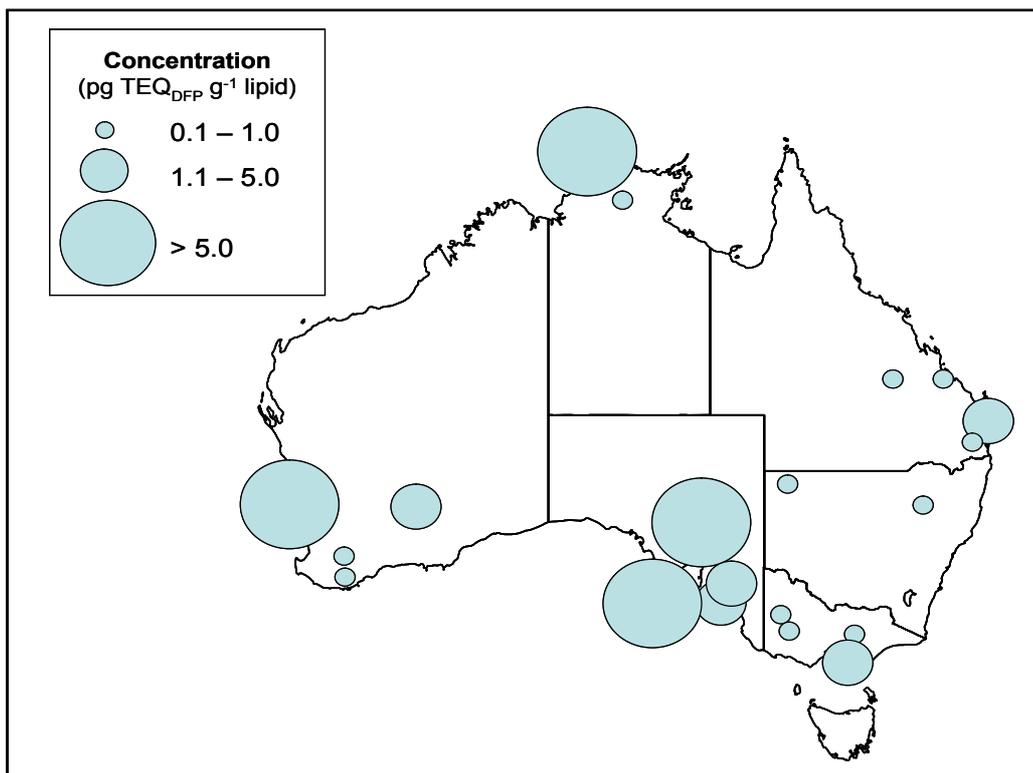
**Figure 3.18 Concentration of dioxin-like PCBs in the macropod samples collected from different States across Australia**



**Figure 3.19 Percent contribution of PCDDs, PCDFs, non-ortho PCBs and mono-ortho PCBs to the total TEQ concentration in birds**

The spatial distribution of concentration of PCDD/PCDFs and dioxin-like PCBs ( $\text{pg TEQ}_{\text{DFP}} \text{g}^{-1}$  lipid) detected in the macropod samples is shown in Figure 3.20.

The highest concentration of PCDD/PCDFs and PCBs ( $25 \text{ pg TEQ}_{\text{DFP}} \text{g}^{-1}$  lipid) was detected in a composite sample (of three macropod tails) from Para Wirra conservation reserve, which is 25 km north north-east of Adelaide (see Appendix H for detailed location). There is no industry in the local area. The lowest concentrations of PCDD/PCDFs and PCBs ( $0.001 \text{ pg TEQ}_{\text{DFP}} \text{g}^{-1}$  lipid) were observed in a macropod sample from Tibooburra.



**Figure 3.20 Concentration of PCDD/PCDFs and PCBs in macropods across Australia.**

In total only nine of the macropod samples analysed had concentrations of PCDD/PCDFs and PCBs that exceeded 1.0 pg TEQ<sub>DFP</sub> g<sup>-1</sup> lipid. The individual congeners (pg TEQ g<sup>-1</sup> lipid) that contributed greatest to the TEQ are shown in Table 3.3. The largest contribution to the TEQ was from PCB 126 followed by 1,2,3,7,8-PeCDD. However, in macropod Sample 37 (from Para Wirra) the PCDF congeners contributed more to the TEQ than did PCDDs.

The overall low TEQ in the macropod samples was influenced by the relatively high concentrations of the PCDD/PCDF congeners with a low TEF value rather than the low concentrations of PCDD/PCDFs and the dioxin-like PCBs. This particularly applied to OCDD. In comparison, when the levels of PCDD/PCDFs and PCBs were expressed as a concentration (i.e. pg g<sup>-1</sup> lipid), the concentration of the 2,3,7,8-substituted PCDD/PCDFs ranged from 0.95-180 pg g<sup>-1</sup> lipid, whereas the concentration of dioxin-like PCBs ranged from 8-3,300 pg g<sup>-1</sup> lipid. The low TEQ in the macropods is therefore due not only to the low levels of dioxin-like compounds but also to the congener profiles.

**Table 3.3 Details of the nine macropod samples displaying the highest TEQ<sub>DFP</sub> concentrations<sup>1</sup>**

State	SA	NT	WA	SA	SA	WA	VIC	SA	QLD
Location	Para Wirra	Jabiru	Perth (Melville)	Para Wirra	Para Wirra	Kalgoorlie	Healesville	Kuipito Forest	Coolum/Noosa
Sample number	37	26	24	36	35	23	42	38	32
Congener									
1,2,3,7,8-PeCDD	3.0	1.3	0.4	1.7	0.9	0.7	0.3	0.5	0.5
1,2,3,6,7,8-HxCDD	0.5	0.2	0.0	0.2	0.1	0.1	0.0	0.2	0.0
2,3,7,8-TCDF	1.4	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
1,2,3,7,8-PeCDF	1.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
2,3,4,7,8-PeCDF	7.5	0.1	0.4	0.2	0.2	0.2	0.1	0.1	0.1
1,2,3,4,7,8-HxCDF	1.4	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
1,2,3,6,7,8-HxCDF	0.8	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
PCB 126	8.0	2.7	3.4	2.1	2.4	2.1	1.4	0.9	0.3
Sum of dominant components	24	4.3	4.3	4.1	3.7	3.2	1.8	1.7	0.9
TEQ	25	5.3	5.1	5.0	4.2	3.6	2.2	2.1	1.1

<sup>1</sup> Units are pg TEQ g<sup>-1</sup> lipid and concentrations below detection limits are treated as 0.5 detection limit

One of the objectives of this study was to investigate the concentration of PCDD/PCDFs and dioxin-like PCBs between different regions (i.e. north, south-east, south-west) and land use categories (i.e. remote, agricultural, urban and industrial). This was not possible for all species, but comparisons are possible using the macropod data.

On a TEQ basis there was no statistically significant difference between different regions or states, even after a covariate adjustment had been made for the local land use. The statistical test had a low power due to the high variation between samples from the same region and also the low number of samples available to the study.

There was a trend for higher concentrations of both PCDD/PCDFs and dioxin-like PCBs in the samples that originated from South Australia (Table 3.3), with all four samples being included in the top nine highest TEQs. Within the other states, the concentrations of both PCDD/PCDFs and PCBs were lower, although one or two samples had elevated concentration, e.g. one sample from Vic and WA had elevated concentrations of PCBs, whereas one of the samples from NT had elevated concentrations of PCDD/PCDFs. However, given the relatively poor representation across the states, it is impossible to draw definitive conclusions concerning the spatial distribution of PCDD/PCDFs and PCBs.

An analysis of covariance was used to test for differences between the local land uses, after removing potential regional effects as covariates. All the PCDD/PCDF and PCB congeners were analysed and four of these showed a statistically significant difference between land use types. The predicted means of these four congeners are shown in Table 3.4. Note that these values were obtained from an analysis of covariance on log-

transformed data. Means are geometric means as no correction for bias to convert them to true arithmetic means was made.

**Table 3.4 Concentration of selected congeners and overall TEQ in macropods across land use types<sup>10</sup>**

Congener	Urban	Agricultural	Remote	P value
2,3,7,8-TCDD	0.28	0.22	0.42	0.024
2,3,4,6,7,8-HxCDF	0.23	0.18	0.31	0.049
PCB 77	1.05	0.86	1.70	0.044
PCB 189	1.57	1.07	2.46	0.033
TEQ	1.09	0.79	1.43	0.068

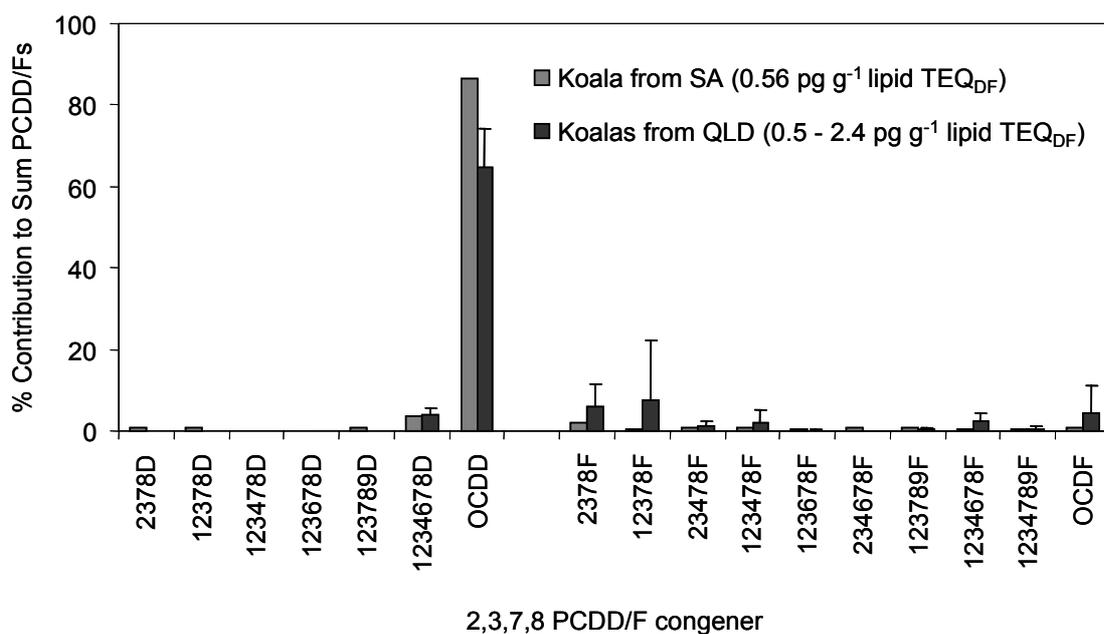
### 3.2.3 Other Marsupials

All the non-macropod marsupials had detectable 1,2,3,7,8-PeCDD levels, with the highest concentrations found in the brown bandicoots (*Isodon macrourus*). The difference is not statistically significant because of the high CV (72%) of the field observations. The possum (*Trichosurus vulpecula*) had higher levels of 1,2,3,4,6,7,8-HpCDD than the koala (*Phascolarctos cinereus*) or the bandicoots. Similarly PCB126 was over ten times higher in the possum than in the koala and the bandicoots. The difference in PCB 126 levels between the two bandicoots from the Northern Territory is indicative of the amount of field variation that is present between the animals.

There are few data available for the levels of PCDD/PCDFs and dioxin-like PCBs for marsupials. In particular many of the species analysed as a part of this study are endemic to Australia (e.g. koala). The concentration of  $\Sigma$ 2,3,7,8-substituted PCDD/PCDFs in the Koala sampled from South Australia was 20 pg g<sup>-1</sup> lipid (0.56 pg g<sup>-1</sup> lipid TEQ<sub>DF</sub>, 0.96 pg g<sup>-1</sup> lipid TEQ<sub>DFP</sub>). In comparison Prange et al. (2003) observed similar or slightly higher concentrations of PCDD/PCDFs in koala fat tissue collected from Queensland (ranging from 30-77 pg g<sup>-1</sup> lipid, 0.5-2.4 pg TEQ<sub>DF</sub> g<sup>-1</sup> lipid) (Figure 3.21).

Prange et al. (2003) calculated that the concentration of PCDD/PCDFs in koalas from Queensland are relatively low, considering their estimated intake of PCDD/PCDFs from the consumption of Eucalyptus leaves. Those authors hypothesised that koalas may have an efficient metabolic capacity for these compounds.

<sup>10</sup> Units are pg g<sup>-1</sup> lipid. The values are predicted values following a covariate adjustment for regional effects and back transforming from a log scale



**Figure 3.21 Comparison of koala 2,3,7,8 PCDD/PCDF profiles between SA and QLD**

The concentrations of the PCDDs and PCDFs in the ‘other marsupials’ are shown in Figure 3.22. That figure shows that the PCDD/PCDF concentrations of the possum were approximately fifty times that of the koala, even though the two species came from similar geographic regions and both graze eucalypts (although the exact diets are not known). This observation is consistent with the hypothesis of Prange et al. (2003) that the koalas have an efficient method for reducing the concentration of PCDD/PCDFs in their system.

Both bandicoot samples came from the NT and had similar levels of PCDDs, but different concentrations of PCDFs.

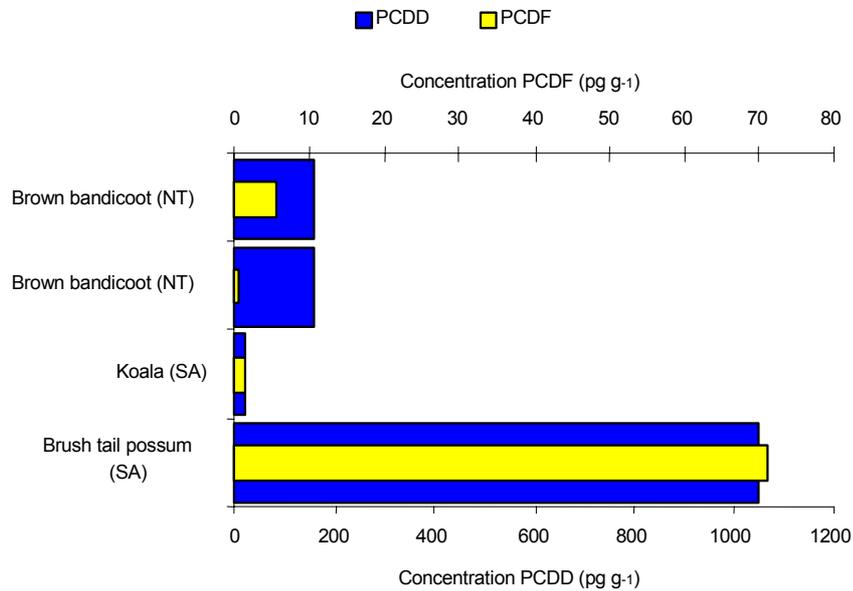
Both the SA and QLD koalas exhibited a congener profile dominated OCDD (Figure 3.21), but there was a suggestion that the OCDD in the SA koalas had a higher relative contribution than the OCDD in the Queensland koalas.

OCDD also dominated the PCDD/PCDF profile of the possum and bandicoots (see Figure 3.24).

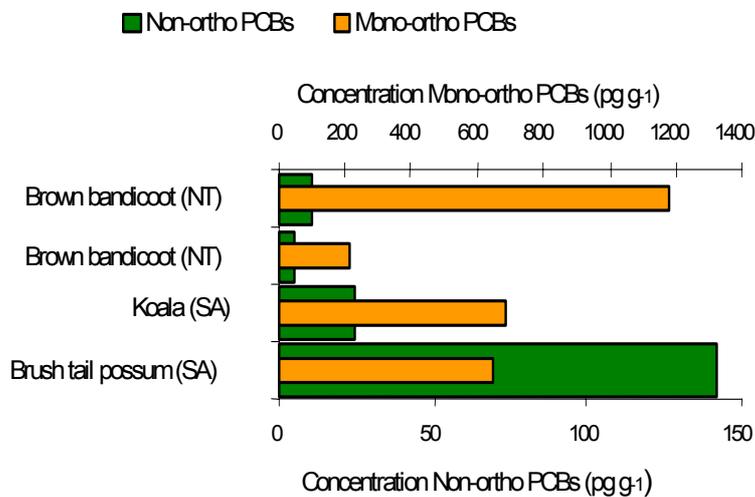
The koala and the possum had similar concentration of mono-ortho PCBs, but quite different levels of non-ortho PCBs (Figure 3.23). The levels of mono-ortho PCBs differed markedly between the two bandicoots, with their range including both the levels found in the koala and the possum.

The PCB profiles (Figure 3.25) differed between the koala, the brush tail possum and the Northern Territory Bandicoots. The main PCB congener in the koala was PCB 118, with small contributions from PCB 105 and PCB 167. The brush tail possum, which was from a similar area to the koala from SA, also had PCB 118 as its dominant component but it also had significant amounts of PCBs 126, 105 and 156. In the Northern Territory bandicoots, the most significant component was PCB 156 but with over 25% coming from PCB 118, with contributions from PCBs 105, 157, 167 and 189. The difference between the koala and the possum may indicate that the koala can break

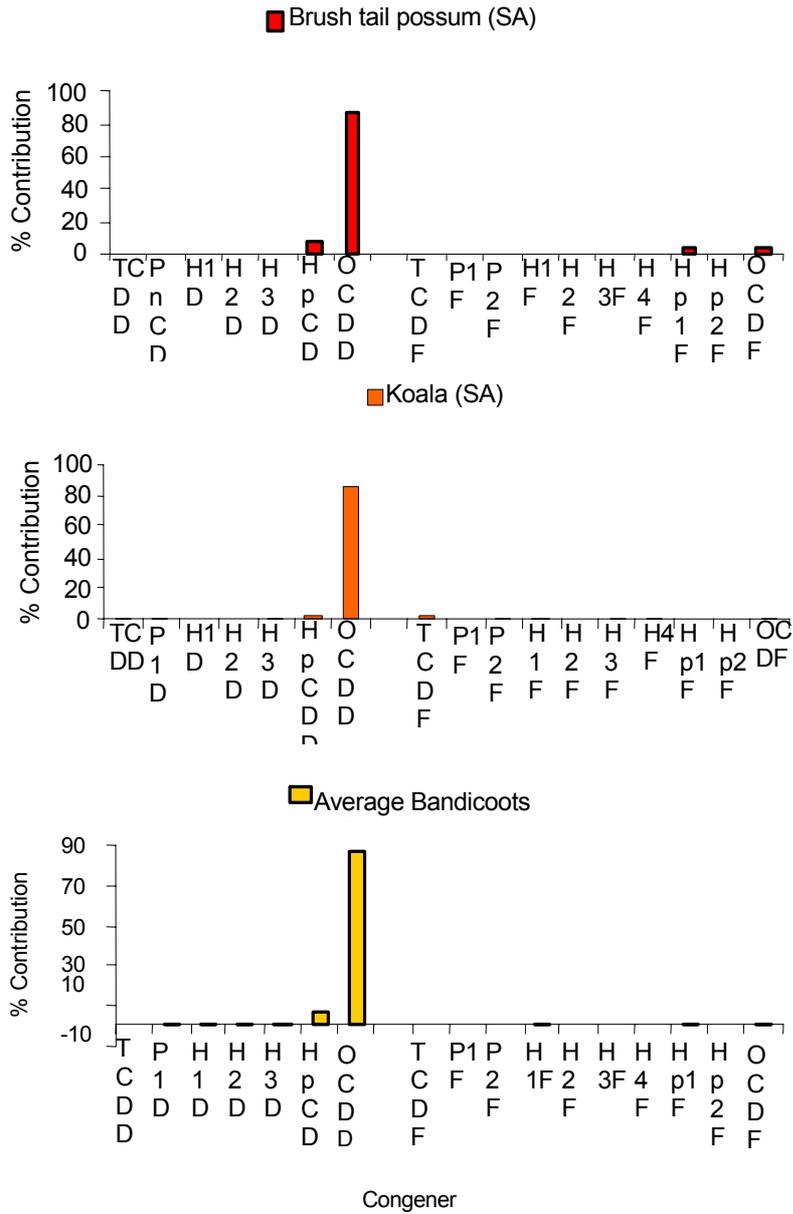
down some PCBs thus leaving a higher relative concentration of PCB 118. Alternatively, the possum may be able to break down the mono-ortho PCBs. Given the high degree of field variability it is difficult to draw any firm conclusions from such a small sample.



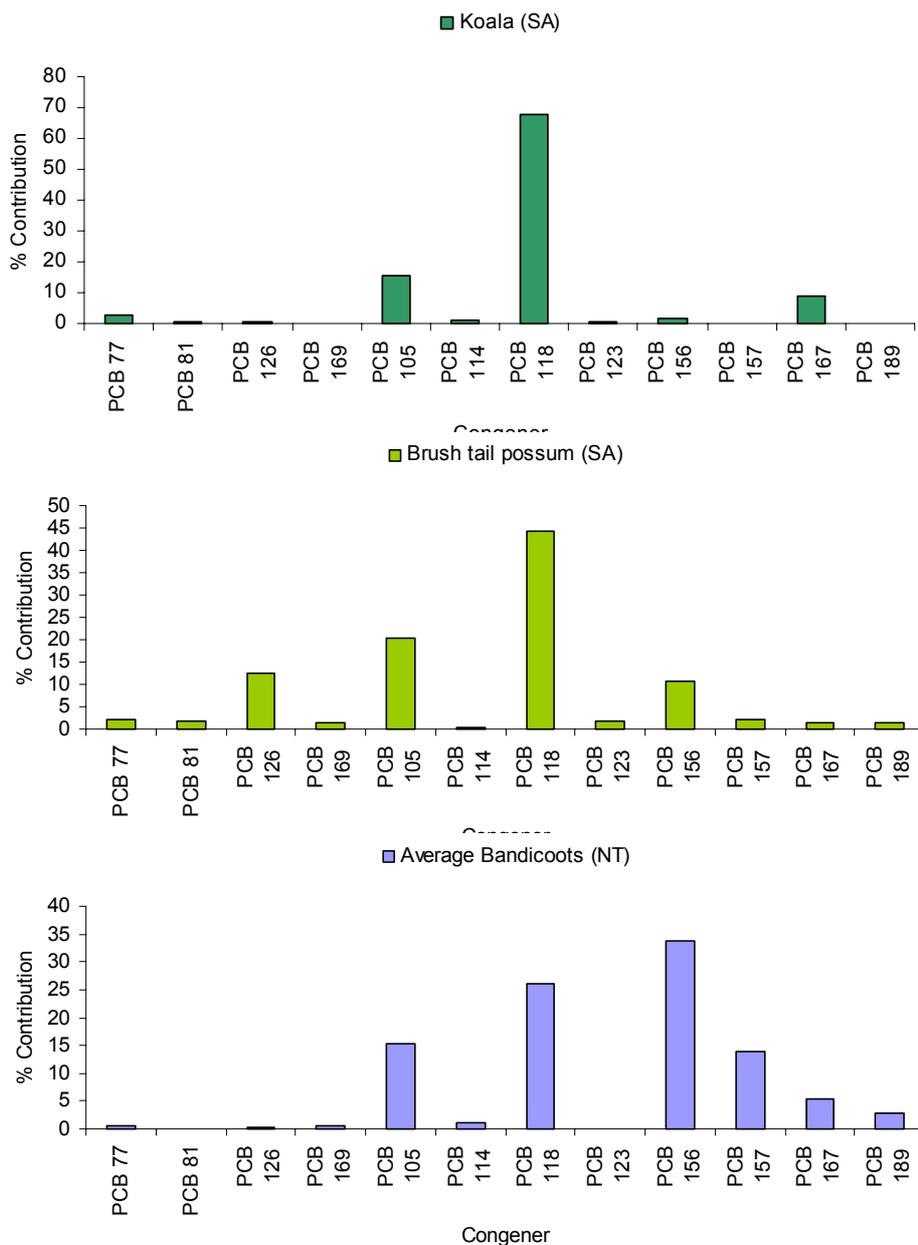
**Figure 3.22** Concentration of PCDD/PCDFs in other marsupial samples collected from different states across Australia.



**Figure 3.23** Concentration of dioxin-like PCBs in other marsupial samples collected from different states across Australia



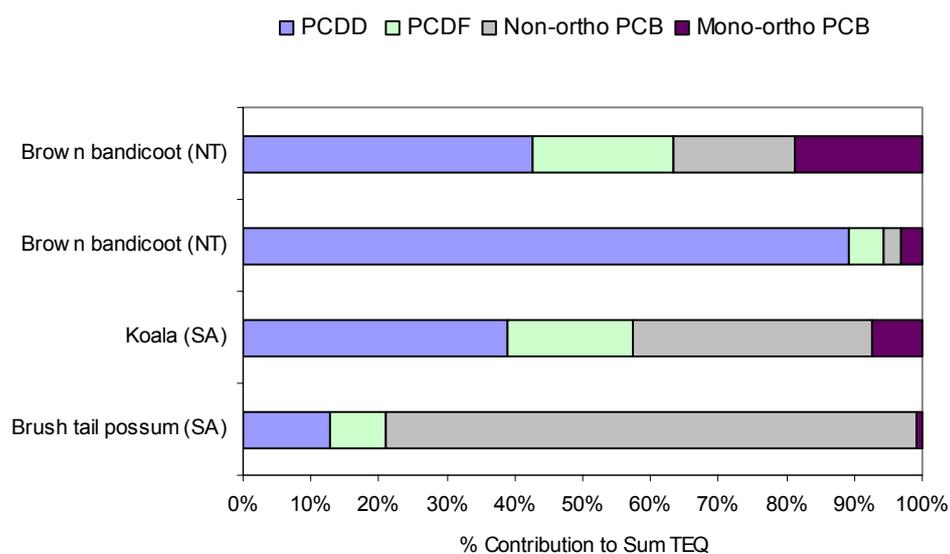
**Figure 3.24** Percent contribution of PCDD/PCDF congeners to the PCDD/PCDF concentrations in other marsupial species



**Figure 3.25 Percent contribution of individual PCB congeners to the total PCB concentration in other marsupial species**

The relative contribution to the TEQ of the PCDDs, PCDFs non-ortho PCBs and mono-ortho PCBs to the total TEQ is shown in Figure 3.26. A striking feature of that figure is the variation between the contributions of the PCDDs to the profiles of two bandicoots from the NT. Apart from that large difference, the ratios of PCDFs, and the non-ortho and mono-ortho PCBs were similar between the two samples.

Another feature of that figure is the relatively large contribution made by the non-ortho PCBs to the overall TEQ in the possum and to a lesser extent in the koala.



**Figure 3.26 Percent contribution of the PCDDs, PCDFs, non-ortho PCBs and mono-ortho PCBs to the total TEQ (TEQ<sub>DFP</sub>) in other marsupial species**

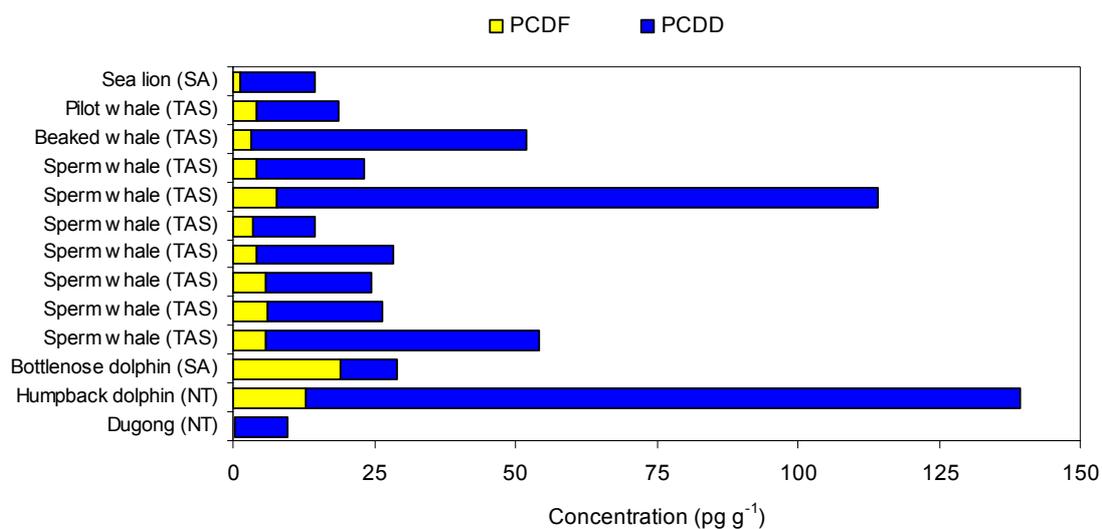
### 3.2.4 Marine Mammals

A total of thirteen marine mammals from three states were analysed for the present study. Among these, the majority of animals (n = 9) represented whale species collected from Tasmania (Sperm whale, [*Physeter catodon*], long fin pilot whale [*Globicephala melas*] and the beaked whale [*Mesoplodon grayi*]), while two animals were collected each from South Australia (bottlenose dolphin [*Tursiops aduncus*] and sea lion [*Neophoca cinerea*]) and the Northern Territory (dugong [*Dugong dugong*] and humpback dolphin [*Sousa chinensis*]).

It has to be noted that only one animal was available for analysis of each species other than sperm whales. The variation in PCDD/PCDF and PCB concentrations within the seven sperm whale samples collected from Tasmania highlight the influences of biological parameters such as age, gender and parity on the levels of persistent organic pollutants in an individual (see Section 2.3). The range of within species variation must be considered when drawing conclusions from the data presented in this report.

Overall, PCDD/PCDF concentrations in marine mammals were relatively low, ranging from 9.8 to 140 pg g<sup>-1</sup> lipid. Among PCDD/PCDFs, the PCDDs dominated the profiles in the majority of marine mammals, the data shown in Figure 3.27 are the sum of the 17 congeners.

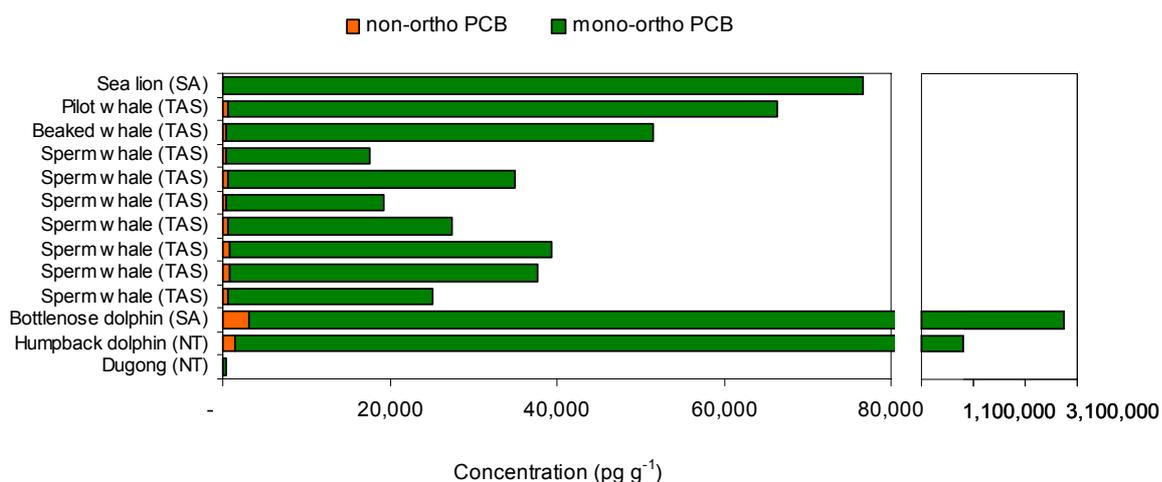
The lowest PCDD/PCDF concentrations within the marine mammal class were present in the dugong from Northern Territory and the sea lion from South Australia as well as one sperm whale from Tasmania. The highest PCDD/PCDF concentrations were found in the humpback dolphin from the Northern Territory and one sperm whale from Tasmania.



**Figure 3.27 PCDD and PCDF concentrations in marine mammals.**

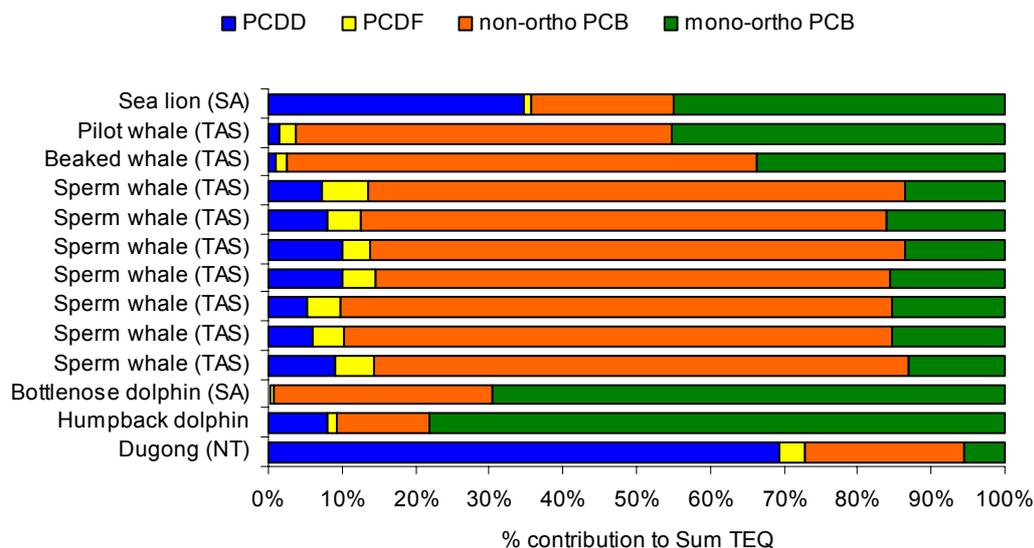
PCB concentrations ranged from 490 to 2,800,000 pg g<sup>-1</sup> lipid with mono-ortho PCBs contributing the highest concentrations in all animals analysed (Figure 3.28).

PCB concentrations were elevated in particular in the humpback and bottlenose dolphins from the Northern Territory and South Australia, respectively. Compared to these two dolphin species, concentrations in the remaining animals were more than a factor of ten lower, and relatively consistent among the seven sperm whale samples, ranging from 18 to 39 ng g<sup>-1</sup> lipid. The sea lion and pilot whale were intermediate between the dolphins and the sperm whales.



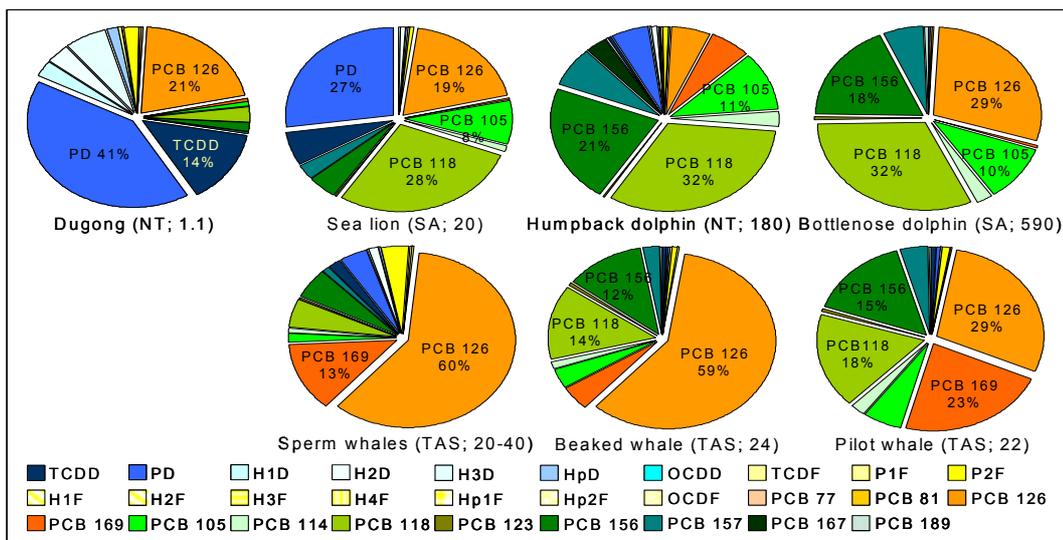
**Figure 3.28 Non-ortho and mono-ortho PCB concentrations in marine mammals**

Total TEQ levels ranged from 1.1 to 586 pg TEQ<sub>DFP(mammal)</sub> g<sup>-1</sup> lipid among all marine mammals with the highest levels present in the two dolphin species from the Northern Territory and South Australia. The lowest TEQ levels were present in the dugong from the Northern Territory, and relatively low TEQ concentrations were also present in the three whale species and the sea lion (range 20-40 pg g<sup>-1</sup> lipid). PCBs contributed the greatest proportion (64-99%) to the total TEQ in all animals except the dugong, which had 27% (see Figure 3.29).



**Figure 3.29 Percent contribution of PCDDs, PCDFs, non-ortho PCBs and mono-ortho PCBs to the total TEQ concentration in marine mammals**

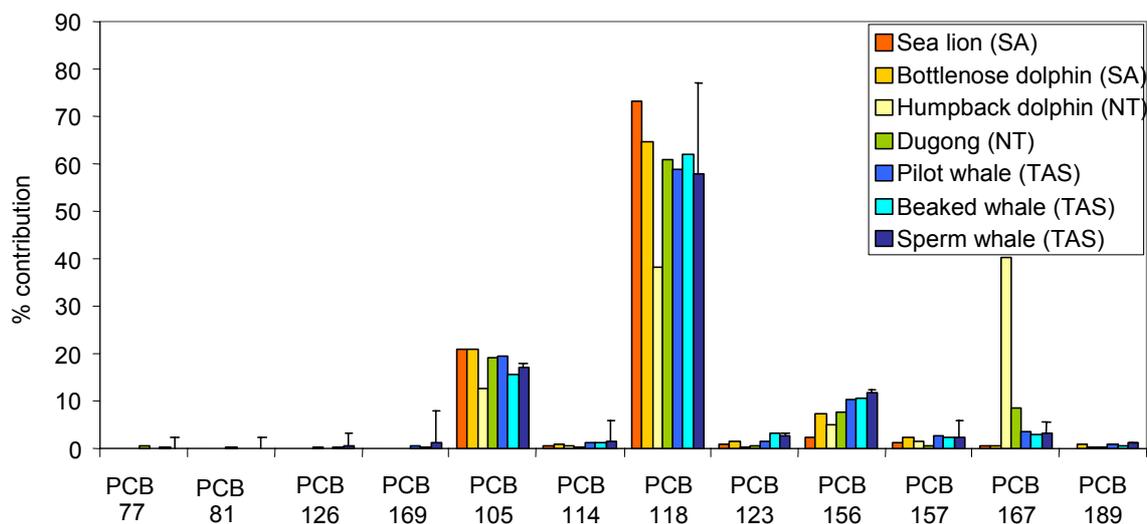
The contribution of individual congeners to the total TEQ varied between marine mammals (see Figure 3.30). The TEQ in all three whale species was dominated by contributions from the two non-ortho PCBs 126 and 169, with additional significant contributions from the mono-ortho PCBs 156 and 118. In contrast, the TEQ in the two dolphin species were dominated by the mono-ortho PCBs 156, 118 and 105. While PCB 126 and 118 contributed major proportions to the total TEQ in the sea lion, PCDD/PCDFs, in particular PCDD and PCDFs contributed significant proportions (33 and 55%) in both the sea lion and the dugong, respectively).



Figures in brackets represent the TEQDFP concentrations in  $\text{pg g}^{-1}$  lipid.

**Figure 3.30 Percent contribution of individual PCDD/PCDF/PCB congeners to the total TEQ in marine mammals.**

PCB congener profiles in the marine mammals analysed for the present study were similar among different species with PCB 118 dominating the profile (59-73%) in all animals except the humpback dolphin (38%) (Figure 3.31). However, with respect to the latter, the low contribution of PCB 118 is due to the elevated contribution of PCB 167. The high contribution is most likely an analytical artefact because it had a high LOD and, because it was reported as below the limit of detection, it was treated as  $0.5 \times \text{LOD}$ . This problem is discussed in Section 2.5. PCB 105 contributed 13-21% to the sum of the 12 PCBs concentrations, followed by PCB 156 (2.2-12%) and PCB 167 (0.6-8.7%), except in the humpback dolphin where, most probably due to an analytical artefact, it appeared to contribute 40%.



Note: the elevated PCB 167 in the humpback dolphin was reported as below the limit of detection, and is most likely an artefact of the treatment of the below detection levels.

**Figure 3.31 Percent contribution of individual PCB congeners to the total PCB concentration in marine mammals**

In contrast to PCB congener distributions, the PCDD/PCDF profiles showed marked differences between some of the marine mammals analysed for this study (Figure 3.32). In particular, the sea lion and the bottlenose dolphin showed unusual high contributions from PeCDD and TCDF, respectively. Despite both samples originating from South Australia and both animals with fish as a predominant food source, the congener profiles were different. The remaining samples showed a congener profile dominated by OCDD with only minor contributions from PCDFs. Among the whale samples from Tasmania, sperm whales showed a higher contribution of lower chlorinated PCDDs and PCDFs compared to both the pilot and beaked whale samples.

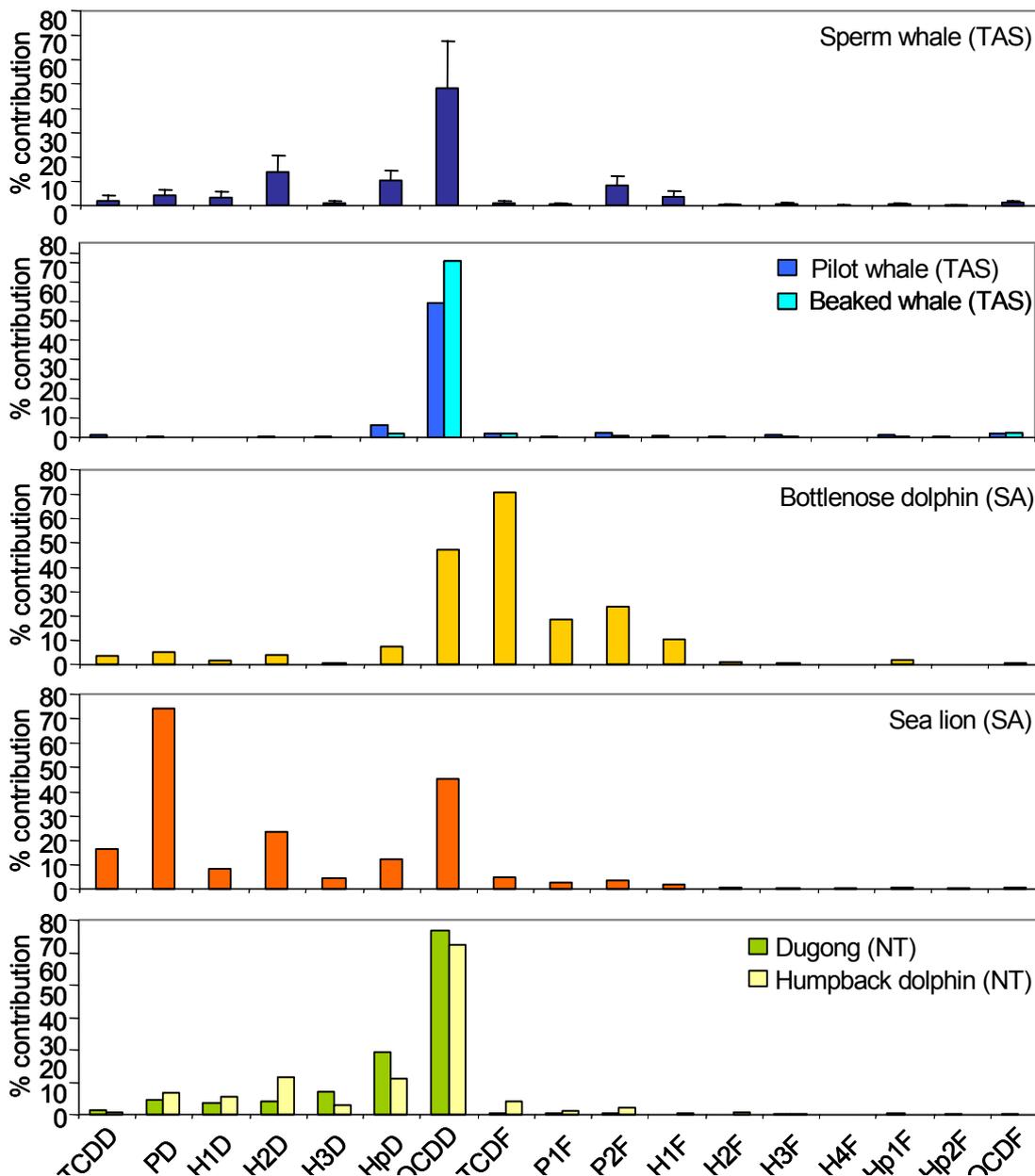
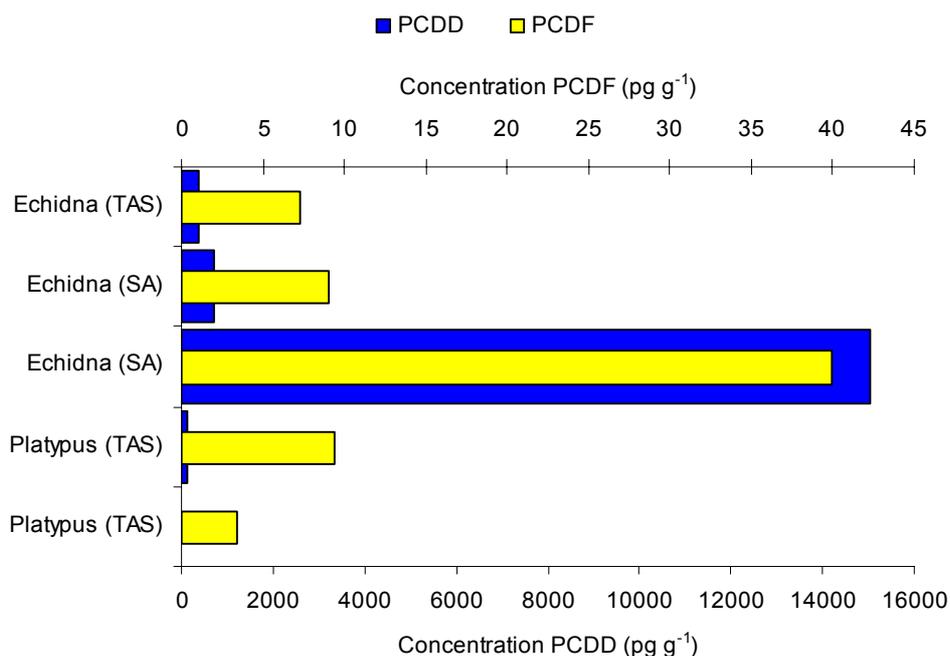


Figure 3.32 Percent contribution of PCDD/PCDF congeners to the total PCDD/PCDF concentration in marine mammals

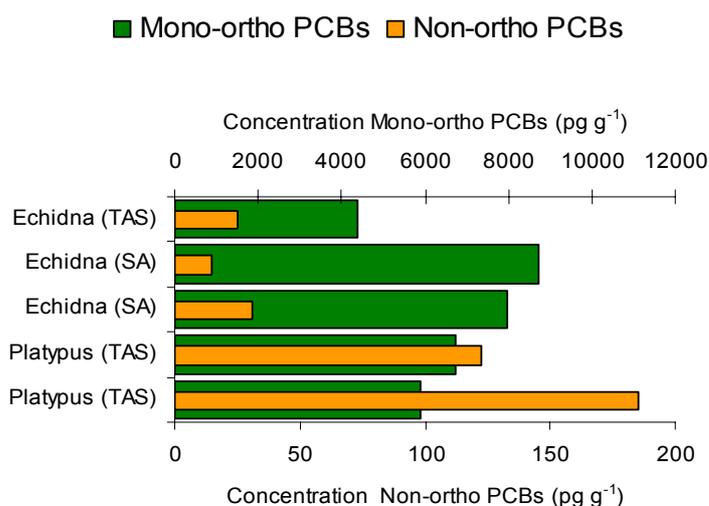
Overall, while the results obtained are indicative of the concentrations of dioxin-like compounds in a marine mammal species from a specific location, caution should be exercised with respect to extrapolating these data to a population without taking into consideration the potential significant variability between individual animals due to biological and other influencing parameters. In addition, it should be noted that the marine mammal data can provide only limited information on regional contamination differences since different species were obtained from each location, there were only a few samples and there persists the problem of the inherent variation.

### 3.2.5 Monotremes

The study considered both groups of monotremes, the echidna (*Tachyglossus aculeatus*) and platypuses (*Ornithorhynchus anatinus*). The monotremes had concentrations of PCDDs ranging from 5 to 14,000  $\text{pg g}^{-1}$  lipid but levels of PCDFs were less than 40  $\text{pg g}^{-1}$  lipid (see Figure 3.33). The outstanding result was an echidna from Pt Elliot in South Australia, which had a total PCDD of over 14,000  $\text{pg g}^{-1}$  lipid. That sample also had the highest concentration of PCDFs with a concentration of almost 40  $\text{pg g}^{-1}$  lipid. The ratio of PCDFs to PCDDs was also much higher in that sample than that observed in the other monotremes.



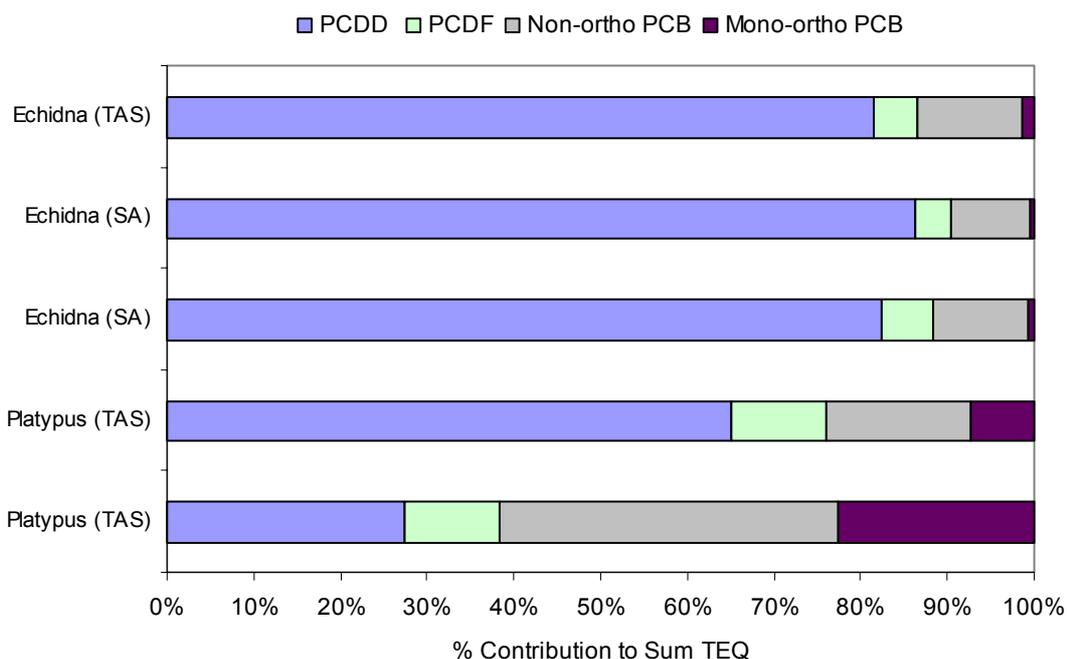
**Figure 3.33 PCDD and PCDF concentrations in monotremes**



**Figure 3.34 Non-ortho and mono-ortho PCB concentrations in monotremes**

The concentration of the non-ortho and mono-ortho PCBs is shown in Figure 3.34. The concentration of non-ortho PCBs was much higher in the platypus samples than in both the Tasmanian and South Australian echidnas. The mono-ortho PCBs varied between the echidna samples, with the Tasmanian sample having only about half the concentration of the South Australian samples, but this difference is within the range of sampling variation encountered between the field replicates. The concentration in the platypus samples was intermediate between that of the echidna from Tasmania and those echidnas from South Australia.

The relative contributions to the TEQs are shown in Figure 3.35. Despite the differences in concentrations of PCDD/PCDFs observed between the three samples of echidna, the proportions of PCDD, PCDF, non-ortho PCBs and mono-ortho PCBs were quite similar. This contrasts with the samples of platypus from Tasmania, which had relatively high proportions of PCBs. An inspection of Table 3.5 indicates that the difference between echidnas and platypuses was due to higher levels of PCDDs in echidnas rather than differing levels of PCBs.



**Figure 3.35 Percent contribution of PCDDs, PCDFs, non-ortho PCBs and mono-ortho PCBs to the total TEQ concentration in monotremes**

The monotremes had a TEQ ranging from 9.3 pg g<sup>-1</sup> lipid in a platypus to 60 pg g<sup>-1</sup> lipid in an echidna as shown in Table 3.5. Much of the contributions were from dioxin congeners, and in particular 1,2,3,7,8-PeCDD. Figure 3.36 indicates the concentrations of the components in each of the monotreme samples. An outstanding feature of that figure is the high level of OCDD in the echidnas, with a maximum level of 14,000 pg g<sup>-1</sup> in the sample from Pt Elliot.

The PCDD/PCDF profiles for the monotremes are shown in Figure 3.37. All the echidnas had similar profiles, with a dominance of OCDD. The contribution of the PCDD congeners generally increased with increasing chlorination levels, with OCDD contributing about 70% of the total PCDD/Fs. There were effectively no PCDFs in the echidnas. The platypus samples had a higher proportion of lower chlorinated congeners in the profile. OCDD was again the dominant component of the PCDD/PCDFs, contributing about 30% of the sum. There was, however, a greater relative contribution from the tetra- and penta-PCDDs than in the echidnas. The platypuses had significant amounts of PCDFs, particularly of the 2,3,4,7,8-PeCDF congener.

The PCB profiles are given in Figure 3.38. These profiles were similar across the samples and the differences could be attributed to sampling variation.

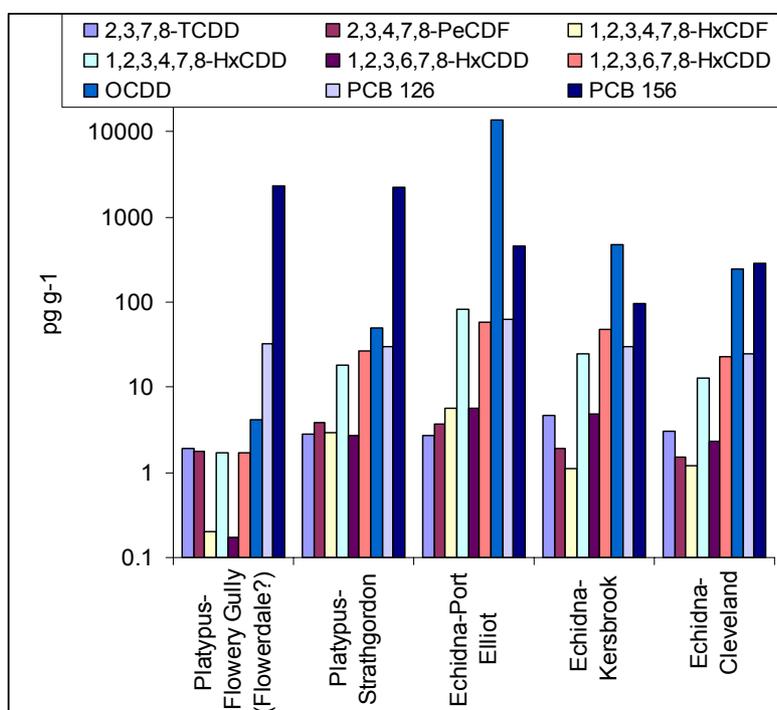
**Table 3.5 Dominant components of the TEQ for five samples of monotremes**

Common name	Platypus Flowery Gully Tas	Platypus Strathgordon Tas	Echidna Port Elliot SA	Echidna Kersbrook SA	Echidna Cleveland SA
2,3,7,8-TCDD	1.9	2.8	2.7	4.6	3.1
1,2,3,7,8-PeCDD	0.3	6.5	20	16	11
1,2,3,4,7,8-HxCDD	0.2	1.8	8.1	2.5	1.3
1,2,3,6,7,8-HxCDD	0.2	2.7	5.7	4.9	2.3
1,2,3,4,6,7,8-HpCDD	0	0.2	8.7	1.3	0.6
OCDD	0	0	1.4	0	0
2,3,4,7,8-PeCDF	0.9	2	1.9	1	0.8
PCB 126	3.2	3	6.2	3.1	2.5
PCB 156	1.2	1.1	0.2	0	0.1
TEQ	9.3	23	60	36	23

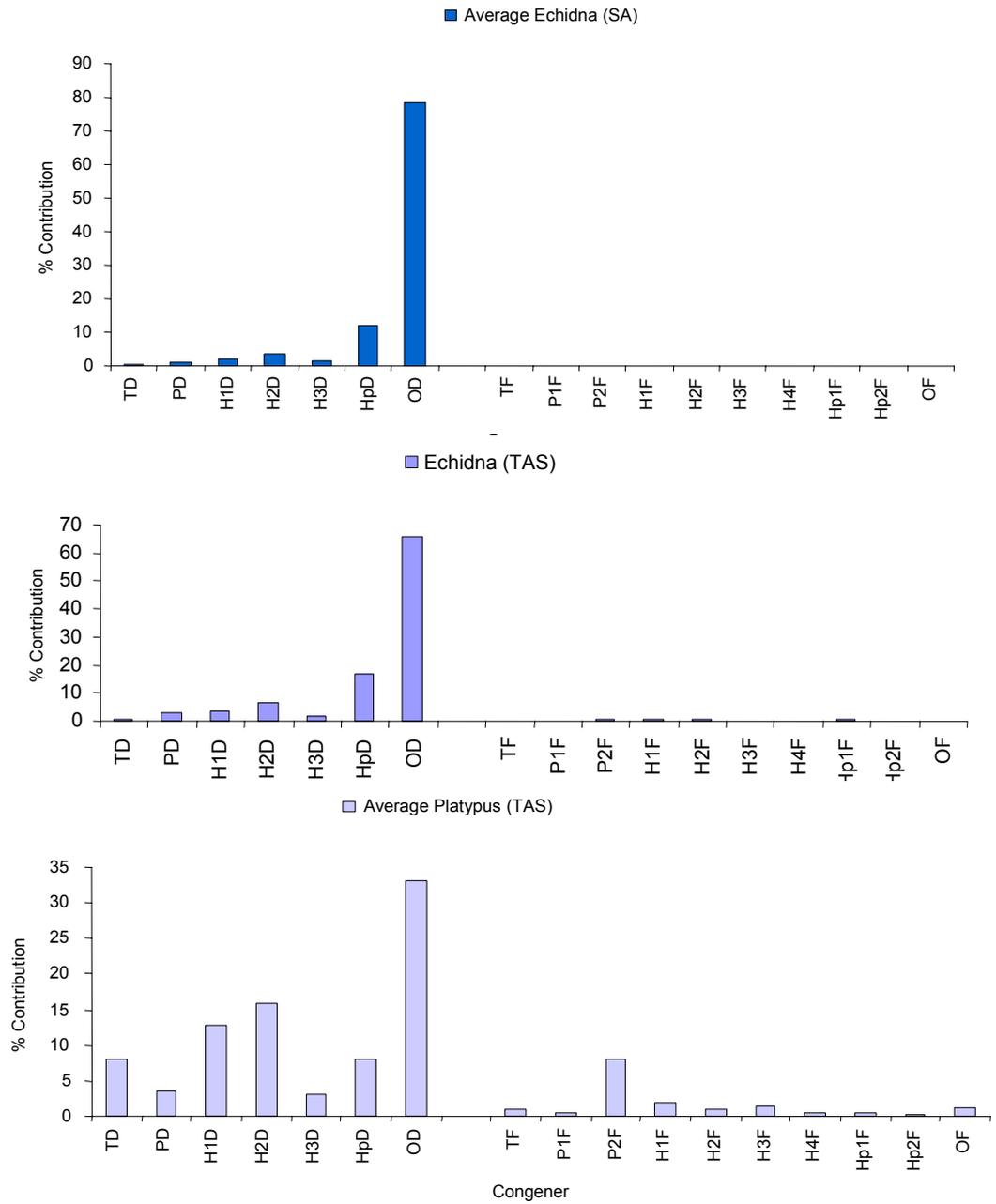
Units are pg TEQ g<sup>-1</sup> lipid. TEQ calculated included 0.5 LOD when concentration was < LOD

Key to colours BLUE 1-10, GREEN 10-100

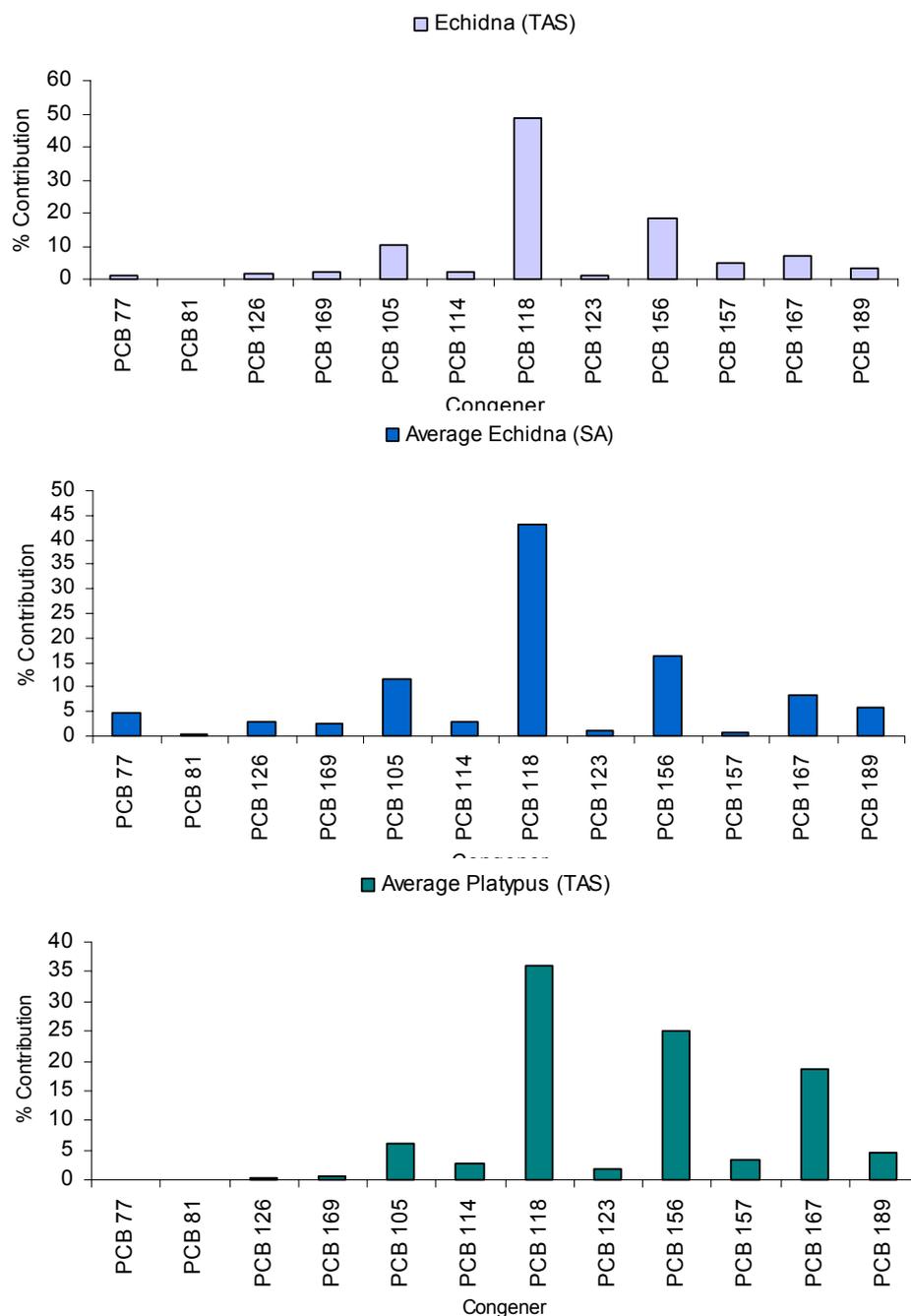
The high level of OCDD in the echidnas may be in part attributed to their consuming considerable amounts of dirt in their diet (for some details see Services of Tasmania <http://www.dpiwe.tas.gov.au/inter.nsf/WebPages/BHAN-5357K5?open>). Soil is known to contain a high proportion of OCDD in general (Tyskling et al. 1993; Koester and Hites, 1992) and OCDD is often elevated in soil samples from Australia (Prange et Müller 2001; Müller et al. NDP soil study in press).



**Figure 3.36 Concentration of 9 congeners from five samples of monotremes**



**Figure 3.37 Percent contribution of PCDD/PCDF congeners to the total PCDD/PCDF concentration in monotremes**



**Figure 3.38** Percent contribution of individual PCB congeners to the total PCB concentration in monotremes

### 3.2.6 Dingo

The dingo (*Canis familiaris dingo*) samples were from remote pastoral country (see Figure 2.1). Both samples had very low concentrations of dioxin-like congeners with TEQ levels of 2.3 and 1.7 pg g<sup>-1</sup> lipid. Much of the TEQ came from two dioxin congeners (1,2,3,7,8-PeCDD and 1,2,3,6,7,8-HxCDD), as shown in Table 3.6. As shown in Figure 3.4, PCB 189 (2,3,3',4,4',5,5'-heptachlorobiphenyl), although having contributed a relatively small amount to the sum of the mono- and non-ortho PCB

concentration for the majority of the fauna classes, contributed substantially to the sum of dioxin-like PCBs for the dingo from South Australia.

The low concentration of congeners was unexpected as dingos are carnivores, because some biomagnification of the dioxin-like compounds was anticipated. The low concentrations in the dingos therefore suggest that the concentrations of dioxin-like compounds in the local primary feeders must be very low.

**Table 3.6 Concentration and contribution to TEQ of two congeners from dingo samples**

Congener	Concentration		TEQ contribution	
	Adult	Immature	Adult	Immature
1,2,3,7,8-PeCDD	0.5	0.5	0.5	0.5
1,2,3,6,7,8-HxCDD	9	7.3	0.9	0.73
Total TEQ			2.3	1.7

Units are  $\text{pg g}^{-1}$  lipid for concentration and for the TEQ. Total TEQ calculated included 0.5 LOD when concentration was  $<$  LOD.

### 3.2.7 Reptile

The only reptile included in the study was a heath goanna from Penneshaw on Kangaroo Island. The TEQ for that animal was only  $0.65 \text{ pg g}^{-1}$  lipid. The main components were from 1,2,3,7,8-PeCDD, and PCBs 126, 156 and 169. As in the case of the dingo, the reptile is not a primary feeder, indicating that the primary feeders must have very low levels of dioxin-like compounds.

## 3.3 Multivariate analyses

The data presented above are essentially multivariate, so it would seem to be appropriate to use multivariate techniques.

There are many ways this can be approached, and these data offer many opportunities for multivariate analyses. The purpose of the analyses should be considered before choosing a particular technique. For example, a principal component analysis would be appropriate for showing the overall structure of the data, whereas differences between fauna classes (or regions) would be better demonstrated by using a canonical variate approach.

A principal component analysis based on the sums of squares matrix of the log concentrations is discussed in Appendix K and is summarised in Table 3.7

**Table 3.7 Principal component analysis.**

Component	Interpretation	% variance explained
1	Overall sum of dioxin-like compounds	79
2	Contrast between PCDD/PCDFs and PCBs	9

The analysis showed that the most important component is how much of the dioxin-like compounds was present (regardless of their type). The second component contrasted those samples dominated by PCDD/PCDFs against those dominated by PCBs.

## 4 Trophic levels in terrestrial fauna

The fauna analysed as part of this study were classified according to their dominant food source as surrogate of the trophic level. Details of the classification used are shown in Table 4.1

**Table 4.1 Classification of terrestrial species into groups based on food source**

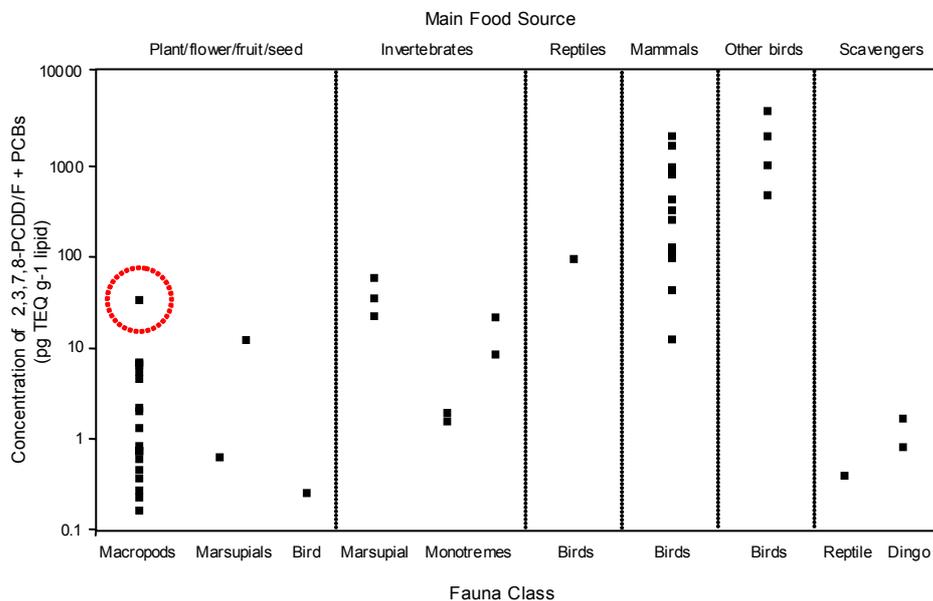
Main food source	Species name	Common name	No of samples
Plant/flower/ fruit/seeds	<i>Macropus agilis</i>	Agile wallaby	3
	<i>Macropus fuliginosus</i>	Western grey kangaroo	10
	<i>Macropus giganteus</i>	Eastern grey kangaroo	4
	<i>Macropus spp.</i>	Wallaby/kangaroo	5
	<i>Phascolarctos cinereus</i>	Koala	1
	<i>Trichosurus vulpecula</i>	Brush tail possum	1
	<i>Cacatua roseicapilla</i>	Galah	1
Invertebrates	<i>Isoodon macrourus</i>	Bandicoot	2
	<i>Ornithorhynchus anatinus</i>	Platypus	2
	<i>Tachyglossus aculeatus</i>	Echidna	3
Reptiles	<i>Centropus phasianinus</i>	Pheasant Coucal	1
Mammals	<i>Accipiter fasciatus</i>	Brown Goshawk	2
	<i>Aquila audax</i>	Wedgetail Eagle	1
	<i>Elanus axillaris</i>	Black Kite	3
	<i>Falco berigora</i>	Brown Falcon	1
	<i>Falco cenchriodes</i>	Kestrel male	2
	<i>Falco longipennis</i>	Hob.Falcon	1
	<i>Falco peregrinus</i>	Peregrine Falcon	2
Other birds	<i>Accipiter cirrhocephalus</i>	Sparrowhawk	4
Carrion	<i>Canis familiaris dingo</i>	Dingo	2
	<i>Varanus rosenbergi</i>	Goanna	1

These classifications are obviously not exclusive, in particular for the higher trophic fauna classes such as the birds. The lower invertebrate feeders refer to those fauna species consuming invertebrates (including aquatic invertebrates) such as insects, worms, beetles, termites and ants. One bird species, the pheasant coucal (*Centropus phasianinus*), was classified as a reptile consumer, eating mainly small reptiles such as lizards and snakes but this species also consumes frogs, aquatic insects, eggs and sometimes the young of other birds. The fauna species that consumed mammals as their dominant food source exclusively belonged to the bird class. These birds consume small mammals such as mice, rabbit, and macropods but also eat other organisms such as lizards, insects, snakes and occasionally other birds. One species of bird, the sparrowhawk (*Accipiter cirrhocephalus*), was classified as a consumer of other birds. This species also predated some insects and small reptiles such as lizards and snakes. The highest trophic consumers were considered those that are mainly scavengers, i.e. the dingo (*Canis familiaris dingo*) and a heath goanna (*Varanus rosenbergi*). The dingo

is mainly a carnivorous mammal, scavenging deceased animals but they may also predate on young or injured animals. The heath goanna is an opportunistic feeder, scavenging other deceased animals as well as consuming mice, bird eggs and other animals. Because it consumes bird eggs it was assigned a higher trophic level than the birds.

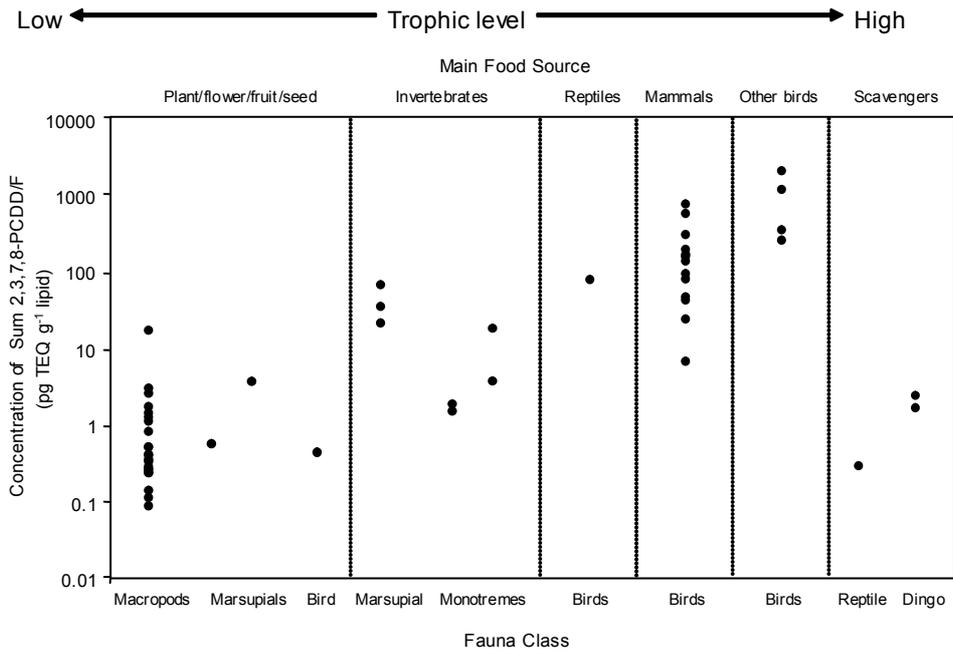
Figure 4.1 indicates a trend for increasing TEQ with increasing trophic level. This is consistent with bio-magnification up the food chain. However, the scavengers (dingo and goanna) did not show this trend. This may indicate that they consume mainly primary feeders (e.g. rabbits) or the samples that came from particularly remote areas.

Not all the variation can be attributed to trophic levels. An example that is circled in the diagram is a macropod sample from Para Wirra (SA), which had a TEQ of 22 pg g<sup>-1</sup> lipid.

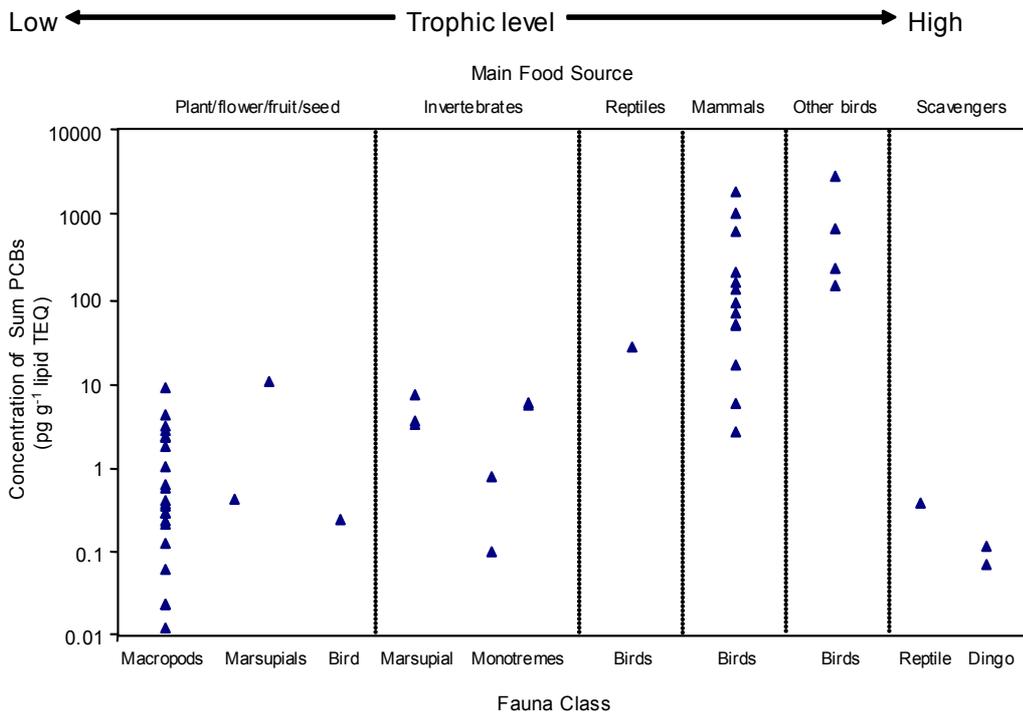


**Figure 4.1 PCDD/PCDF/PCB concentration across fauna class for individual samples.**

Figure 4.2 and Figure 4.3 indicate the 2,3,7,8-PCDD/PCDF and PCB components. There is a similar trend between the two types of compounds, but while the invertebrate consumers had an elevated TEQ, this difference was not apparent in the PCB component. In Figure 4.2 and Figure 4.3 there was a general trend of an increase in the TEQ with increasing trophic level. It is well established that the 2,3,7,8-PCDD/PCDFs and PCBs bioaccumulate in the food chain, thus, the higher trophic levels were expected to have a higher concentration of PCDD/PCDFs and PCBs.



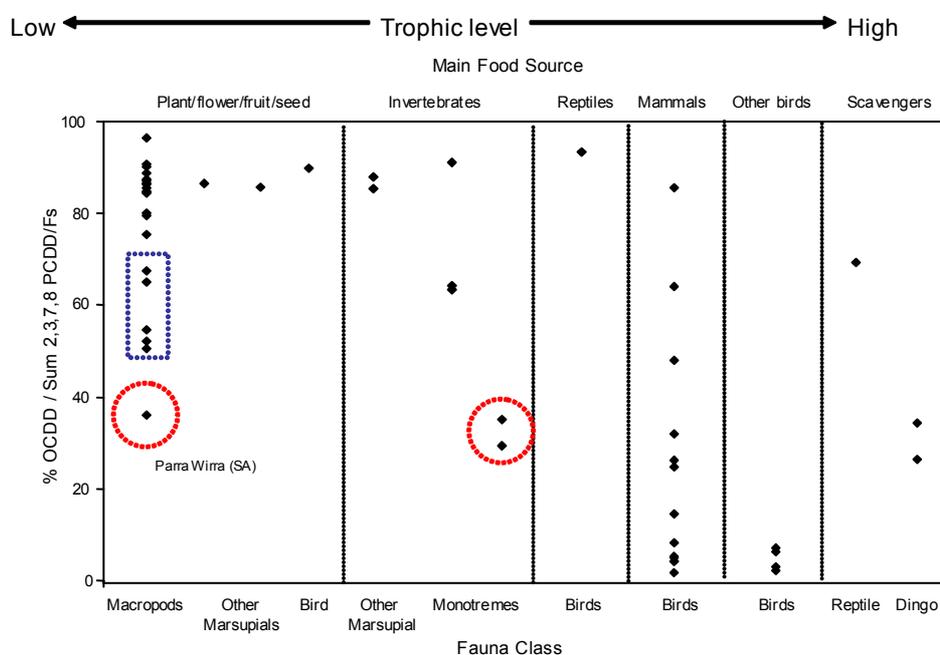
**Figure 4.2 PCDD/PCDF concentration across fauna class for individual samples**



**Figure 4.3 PCB concentration across fauna class for individual samples**

The lower chlorinated congeners are known to be more efficiently bioaccumulated than the higher chlorinated compounds (Broman et al. 1992). Therefore, it would be anticipated that there would be a shift to the lower chlorinated PCDD/PCDFs with increasing trophic level. A good indication of this is the fraction of OCDD in the PCDD/PCDF profile. The percentage contribution of OCDD to the sum of the PCDD/PCDF was calculated for each terrestrial fauna sample. These values were then plotted against the trophic level (food source) as shown in Figure 4.4. Across each of the fauna classes there is a general (but not statistically significant) decrease in the percentage OCDD with increasing trophic level (with respect to the predominant food source).

The proportion of OCDD in the Para Wirra macropod sample (circled in the diagram) was low – this was in part at least to the relatively high contribution of PCDFs in that sample. The two platypus samples (also circled) also had a low proportion of OCDD, which was in part due to some PCDF in the sample but also to contributions from other PCDDs (see Figure 3.37 and Figure 4.4).



**Figure 4.4** Percent OCDD as a percentage of the total PCDD/PCDFs across fauna class and food source for individual samples

In general the low trophic level had a high percentage OCDD contribution, and the higher levels a lower contribution from OCDD. There were many exceptions. Some of the macropods had lower than expected percentage OCDD, and in particular the sample from Para Wirra. The lowest value in the macropods is the Para Wirra sample. Part of the reason for this was the presence of a high percentage contribution from furans (this may be a description and a reason why the calculation worked out as it did – it is not an explanation of what was going on).

## 4.1 Trophic levels in marine mammals

Marine mammals were classified into trophic levels according to standardised diet compositions derived from published accounts of stomach contents and morphological, behavioural and other information (summarised in Pauly et al. 1998). Unlike most terrestrial fauna, however, there is only limited information available on diet composition of many marine mammals and trophic level estimates vary accordingly in the literature. Among the marine mammals analysed for this study, the dugong as a primary consumer (predominant food source: seagrass) represents the lowest trophic level. Among the remaining animals, the humpback dolphin (predominant food source: small pelagic fish [50%] and miscellaneous fish [40%]) and sea lion (predominant food source: miscellaneous fish (55%) and small squid, benthic invertebrates, small pelagic fish represented the lowest trophic level, followed by the beaked whale (since the species was unknown, an average trophic level reported for several beaked whales was assumed for this sample) and the sperm and long-finned pilot whales (predominant food source: small and large squid (70 and 75%, respectively) at the highest trophic level.

An initial comparison of TEQ<sub>DFP</sub> concentrations obtained for all marine mammals analysed versus their estimated trophic level did not reveal any correlations. Since most of these animals were obtained from different locations throughout Australian waters, the influence of regional contamination in the animals' habitat would be expected to play a considerable role in the exposure levels and resulting TEQ concentrations. This comparison was hampered by the small sample size.

Different trophic levels from one location were available only for the dugong and humpback dolphin from Darwin in the Northern Territory. Approximately 14 fold higher TEQ concentrations were present in the higher trophic humpback dolphin compared to the low trophic dugong. A comparison of the congener profiles between these two animals shows similar trends suggesting that both animals were exposed to a similar source (Figure 3.32). However, higher contributions to the total PCDD/PCDF concentration by congeners with no chlorines in the 1,4 and/or 1,9 positions were present in the humpback dolphin (27%) compared to the dugong (12%). These congeners have been demonstrated to biomagnify through food webs (Broman et al. 1992) and include the congeners with the highest Toxic Equivalent Factors.

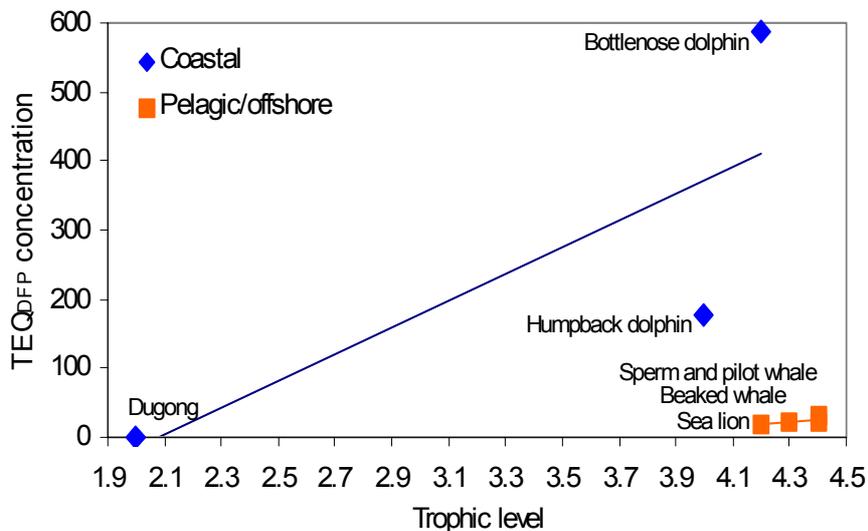
The sea lion and bottlenose dolphin, although both obtained from South Australia, originated from different habitats with the dolphin obtained from Port Adelaide and the sea lion obtained from the Pacific facing site near Kangaroo Island, a habitat with considerably fewer urban and industrial influences. Both animals are on similar trophic levels, however the TEQ in the bottlenose dolphin is approximately 30 fold higher compared to the sea lion. While the PCB congener profiles are similar in both animals, an unusually high contribution of PCDFs has been observed in the bottlenose dolphin, possibly as the result of the elevated exposure to local PCBs contamination in Port Adelaide. In contrast, the PCDD/PCDF congener profile of the sea lion was dominated by PCDD. This highlights that local influences of contamination sources can have significant impacts on the TEQ concentration of wildlife, obscuring the effect of trophic positions. The inclusion of local conditions must therefore be considered rather than relying simply on trophic position alone.

In this respect it has been suggested that in particular the habitat location of an animal in Australian coastal or offshore regions can have significant influences on PCDD/PCDF

contaminant levels in marine wildlife, due to the predominant accumulation of these compounds in the near shore environment from terrestrial runoff (Gaus et al. 2001; Gaus 2002).

The habitat of dugongs and humpback dolphins are predominantly shallow embayments and estuarine environments or occasionally riverine locations near the coastline. The habitat of bottlenose dolphins includes coastal, estuarine, pelagic and oceanic locations, with some animals occurring within a few hundred meters of the coastline. Since the specimen obtained for the present study was obtained from Port Adelaide, it is assumed that its predominant habitat was estuarine/coastal. In contrast to these coastal species, the three whale species are pelagic animals, preferring mainly deep, offshore waters. Similarly, the sea lion was obtained from a location approximately 20 km off the coastline from Adelaide, and on-shelf, less than 200 m deep waters of low productivity represents the typical habitat of an Australian sea lion.

A separation of the marine mammals into “coastal” and pelagic/offshore” according to their predominant habitat, did show a trend of increasing TEQ concentrations with increasing trophic level (Figure 4.5), with more than 10 fold higher levels present in the coastal animals compared to the pelagic/offshore specimens (Pauly et al. 19998). However, the limited number of animals collected overall, and their distribution over three states requires caution in the interpretation of the data with respect to biomagnification processes or ratios. Testing of this hypothesis would require having samples at different levels in the food web from the same or close by sites.



Trend lines are based on insufficient data points to be considered statistically significant.

**Figure 4.5** TEQ<sub>DFP</sub> concentrations versus trophic level in marine mammals from coastal and pelagic/offshore habitats.

## 5 Comparisons with other studies of dioxins

Dioxins have been described as being ubiquitous contaminants in the environment (Richardson and Waid, 1983; Gaus et al. 2001). However, relatively few studies have been conducted to investigate the extent of the occurrence of dioxins in Australian biota. In particular, published literature provides very little data regarding the occurrence of PCDDs and PCDFs while PCB levels in biota have been studied more extensively. Current evidence suggests that relative hotspots of dioxin concentration occur in the vicinity of urban and industrial areas, particularly in the aquatic environment, but further research is needed to establish the extent. Overall, available data indicate that the Australian fauna have relatively low concentrations on a global scale.

### 5.1 Australian Fauna

There are very little data available on dioxin-like chemicals and particularly PCDD/PCDFs in the Australian terrestrial biota. For PCDD/PCDFs Bremner et al. (1990) reported analytical results from subcutaneous fat samples collected from cattle in Victoria. Specifically they compared samples collected from cattle that grazed on land treated with liquid wastes fed by a manufacturing company with samples collected from cattle that grazed in untreated pastures. The authors found concentrations ranging from <0.5 to 0.7 ppt (pg I-TEQ g<sup>-1</sup> lipid) and concluded that the concentrations were very low.

Organochlorine residue testing in Australian meats from May 1987 to May 1989 reported detecting PCBs over the maximum residue limits for export in only nine samples out of a total of 813,330 tested (Corrigan and Sveneiratna, 1990).

Remaining with dioxin-like chemicals in cattle, a study on the occurrence of PCDD/PCDFs in Australian butter as a surrogate measure for dioxin contamination in dairy cattle found that dioxin-like chemicals in Australian butter are relatively low (0.09-0.37 I-TEQ pg g<sup>-1</sup> lipid) (Müller et al. 2001). Tinned butter that had been produced before 1945 (using a sample from the Australian War Memorial) was also analysed by Müller et al. (2002) and found to have a toxicant load of 0.57 pg TEQ g<sup>-1</sup> lipid. The levels of PCDD/PCDF and particularly OCDD were higher in the historic butter (0.57 pg TEQ g<sup>-1</sup> lipid) compared to the then current samples (Müller et al. 2002). Following this relatively surprising result the authors obtained a further set of historic Australian butter samples found on Antarctic tip sites covering samples from the 1950s, 1960s and 1980s and found highest levels of dioxin-like chemicals in the samples from the 1940s to 1960s and a lower concentration in the samples from the 1980s (Müller et al. 2003). Similarly Kalantzi et al. 2001 analysed butter from 23 countries for PCBs and found that the concentration of PCBs in Australian butter are internationally among the lowest.

With respect to endemic species, Prange et al. (2003) reported the concentration of PCDD/PCDFs in koala from Queensland with the  $\Sigma$ PCDD/PCDFs ranging from 30-77 pg g<sup>-1</sup> lipid and 0.5 to 2.4 pg g<sup>-1</sup> lipid TEQ<sub>DF</sub>. These results are comparable to those of the koala analysed in this study, which had 0.56 TEQ<sub>DF</sub> pg g<sup>-1</sup> lipid.

Background levels of PCBs (tetra chlorinated congeners only) in the testicular tissue of brushtail possums from two geographically separate areas in Victoria, one urban and one remote, were found to be similar (range 30-320 ng g<sup>-1</sup> lipid) (Bolton and Ahokas, 1997). In the present study, the concentration of dioxin-like PCBs was 780 pg g<sup>-1</sup> lipid

in a possum from South Australia indicating that the concentrations are low in the Southern part of Australia.

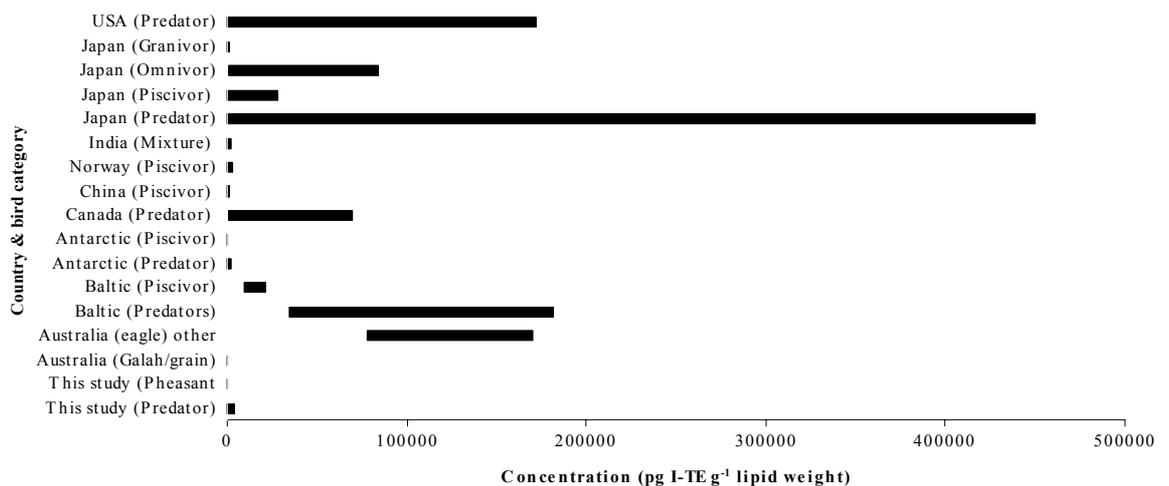
In Australia there had only been one bird analysed for PCDD/PCDFs. This bird species (an eagle) came from a known contaminated region in Sydney (Home Bush Bay) and contained concentrations of PCDD/PCDFs of 78,300 pg TEQ<sub>DF</sub> g<sup>-1</sup> lipid (unpublished data). In comparison, in the present study the maximum concentration of PCDD/PCDFs (1,100 pg TEQ<sub>DF</sub> g<sup>-1</sup> lipid) was detected in a sparrowhawk from South Australia.

## 5.2 International Fauna

Globally there are limited data available for terrestrial fauna (with the exception of birds), in particular there is no data available for the majority species analysed as a part of this study. Nevertheless, the concentration of PCDD/PCDFs and dioxin-like PCBs in polar bears (predator) from the Arctic range from 70-190 pg TEQ g<sup>-1</sup> lipid (Kumar et al. 2002). The concentration of PCDD/PCDFs and non-ortho PCBs (plus PCB 105 and 118) in caribou (a herbivore) from Canadian Arctic ranged from 0.77-6.4 pg TEQ<sub>(NATO)</sub> g<sup>-1</sup> lipid (Herbert et al. 1996; Braune et al. 1999).

Several fauna species were analysed from Japan by Ueda et al. (1999) for PCDD/PCDFs and dioxin-like PCBs, including the Japanese woodmouse (12-2,900 pg TEQ<sub>DFP</sub> g<sup>-1</sup> lipid), the Japanese macaque (0.82-190 pg TEQ<sub>DFP</sub> g<sup>-1</sup> lipid), racoon dogs (15-620 pg TEQ<sub>DFP</sub> g<sup>-1</sup> lipid), Sika deer (3.2-330 pg TEQ<sub>DFP</sub> g<sup>-1</sup> lipid) and bears (0.12-3.3 pg TEQ<sub>DFP</sub> g<sup>-1</sup> lipid). A study in India analysed chicken, lamb and goat meat for PCDD/PCDFs and dioxin-like PCBs and the concentrations ranged from 1.4-5.3 pg TEQ<sub>DFP</sub> g<sup>-1</sup> lipid (Kumar et al. 2001).

Further comparisons could be made using components of these data where they are available. For example, as indicated by the multivariate analysis shown in Appendix , the PCDD/PCDF component could be separated from the PCB component.



**Figure 5.1 Comparison TEQ<sub>DFP</sub> of an international sample of birds compared with the current study**

In contrast numerous studies have investigated the PCDD/PCDF and PCB contamination in birds from around the world. For example, studies in North America have shown that bald eagle populations are declining due to the reproductive effects associated with exposure to persistent organic pollutants (e.g. Elliot et al. 1996). A study by Kumar et al. (2001) found 46-1,800 pg TEQ<sub>DFP</sub> g<sup>-1</sup> lipid in predatory birds from India. In Japan, elevated concentrations of PCDD/PCDFs and dioxin-like PCBs were found in the liver of granivore, piscivore, omnivore and predator bird species. The concentrations ranging from (53-480 pg TEQ<sub>DFP</sub> g<sup>-1</sup> lipid (granivore), 520-28,000 pg TEQ<sub>DFP</sub> g<sup>-1</sup> lipid (piscivore), 560-83,000 pg TEQ<sub>DFP</sub> g<sup>-1</sup> lipid (omnivore) and 430-450,000 pg TEQ<sub>DFP</sub> g<sup>-1</sup> lipid (predator) with the highest concentration detected in a mountain hawk eagle (Kumar et al. 2002).

Further studies in Japan by the Ministry for the Environment (1999) found 66-15,000 pg TEQ<sub>DFP</sub> g<sup>-1</sup> lipid in the muscle from kites. In contrast in low industrial and low urban regions in Japan the concentration of PCDD/PCDFs and dioxin-like PCBs in the fat tissue of black-tailed gull ranged from 263-1,050 pg TEQ<sub>DFP</sub> g<sup>-1</sup> lipid.

Blood analysis of turkey and black vultures from the USA found 57-650 pg TEQ<sub>DFP</sub> g<sup>-1</sup> lipid (Kumar 2003). Further studies by Kumar et al. (2002) found between 260-17,700 pg TEQ<sub>DFP</sub> g<sup>-1</sup> lipid (in several different tissue components) in bald eagles. Elliot et al. (1996) analysed the liver of bald eagles from Canada and found 1,325-68,500 pg TEQ<sub>DFP</sub> g<sup>-1</sup> lipid (assuming a 4% lipid content). Insectivorous tree swallows from Canada, which were nesting along rivers in contaminated areas, were analysed for PCDD/PCDFs and dioxin-like PCBs with the concentrations ranging from approx. 18-145 pg TEQ g<sup>-1</sup> lipid.

In Norway, Grey Herons were analysed for PCBs only with the concentrations ranging from 390-2,510 pg TEQ<sub>(Nordic)</sub> g<sup>-1</sup> lipid. Piscivorous and predator birds were analysed from the Baltic and the concentrations ranged from 10,100-11,500 pg TEQ<sub>(Nordic)</sub> g<sup>-1</sup> lipid in the piscivores and 34,800-147,000 pg TEQ<sub>(Nordic)</sub> g<sup>-1</sup> lipid in the predator birds (Koistinen et al. 1995). A summary of these studies is given in Figure 5.1.

### **5.2.1 Dioxin-like data in marine mammals from Australia; previous studies**

Limited information exists to date on the levels of dioxin-like compounds in marine mammals from Australia. Among the few studies reported, the majority have investigated PCB concentrations. Analysis for PCDD/PCDFs are restricted to marine mammals from Queensland, however, some data is available for fish as well as invertebrates from Victoria (summarised in Appendix A).

TEQ levels in marine mammals analysed previously from Australia and compared to the results of the present study are presented in Figure 5.2. A general trend from this data is the presence of relatively low TEQ<sub>DFP</sub> levels in all whale species, whereas the highest TEQ<sub>DFP</sub> levels are present in dolphins from South Australia and the Northern Territory. With the exception of the dugong, the TEQs of all marine mammals in the current study are dominated by PCBs.

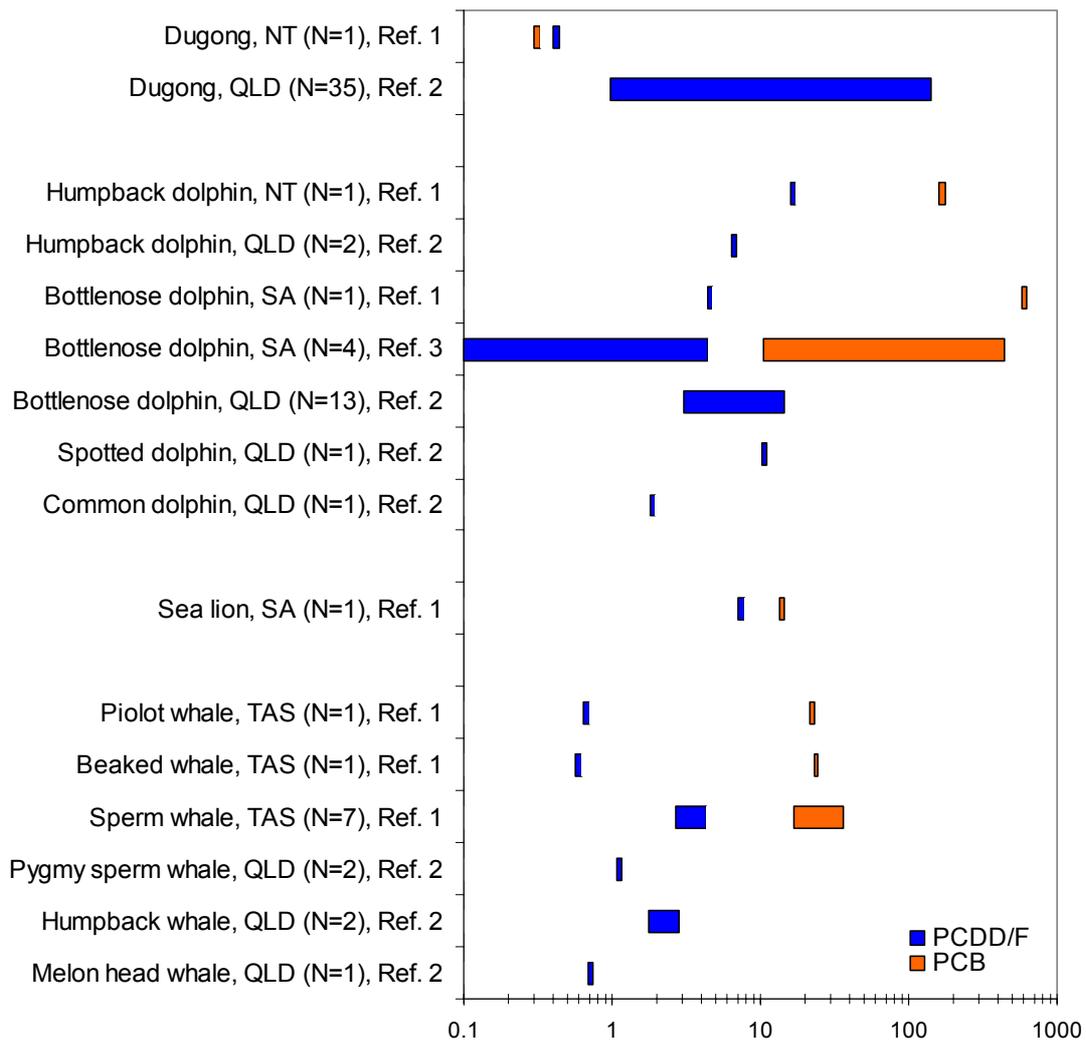
Among whales, TEQ<sub>P</sub> levels have not been reported previously from Australia, however, TEQ<sub>DF</sub> levels observed in the present study are comparable with concentrations reported in other species analysed previously from Queensland (Gaus 2002), Gaus (unpublished data). These TEQ<sub>DFP</sub> concentrations are relatively low

compared to those reported for whales from elsewhere (see Jones et al. (1999) and Symons et al. (2003)).

Among the dolphin species, the  $TEQ_{DF}$  levels observed in the present study are comparable to previous reports from dolphins in Queensland (Gaus et al. 2001; Gaus 2002) and South Australia (Ruchel 2001), and similar to the relatively low  $TEQ_{DF}$  levels observed in whales. In contrast to  $TEQ_{DF}$ ,  $TEQ_P$  levels were elevated in the dolphins from Port Adelaide and Darwin (580 and 160  $pg\ g^{-1}$  lipid, respectively). The only previous report on  $TEQ_P$  levels in dolphins included 4 bottlenose dolphins, 2 of which originated from Port Adelaide (Ruchel 2001). These latter contained similarly elevated  $TEQ_P$  levels (280 and 440  $pg\ g^{-1}$  lipid) compared to the animal obtained from Port Adelaide for the present study (580  $pg\ g^{-1}$  lipid). Interestingly, two bottlenose dolphins obtained from the nearby Spencer Gulf (Ruchel 2001) as well as the sea lion obtained from Kangaroo Island (this study), contained an order of magnitude lower  $TEQ_P$  levels (dolphins: 10 and 37  $pg\ g^{-1}$  lipid; sea lion: 5.7  $pg\ g^{-1}$  lipid) compared to the animals originating from the adjacent Port Adelaide, indicating the presence of a significant PCB point source into the bay of Port Adelaide.

On an international scale, the  $TEQ_{DFP}$  levels in bottlenose dolphins from Port Adelaide are comparable to those reported in cetaceans from areas considered relatively polluted such as the Mediterranean (Risso's and bottlenose dolphins, average 300  $pg\ g^{-1}$  lipid;  $n=8$  (Jimenez et al. 2000)) or British Columbia (killer whale average 660  $pg\ g^{-1}$  lipid (Ross 2000)) and higher compared to cetaceans from Japan (average 120  $pg\ g^{-1}$  lipid;  $n=22$ ).

The dugong analysed for the present study contained the lowest  $TEQ_{DFP}$  levels among all animals analysed, which is in accord with their low trophic position as herbivorous mammals. Similar  $TEQ_{DF}$  levels have been reported in dugongs from the Torres Strait; however, dugongs in six regions along the Queensland coastline have been reported with  $TEQ_{DF}$  levels a factor of 5-170 higher ( $N=35$ ; (Gaus et al. 2001; Gaus 2002) and  $N=3$  (Haynes et al. 1999)). In particular, one previously analysed specimen from Darwin contained more than 30 times higher  $TEQ_{DF}$  concentrations (33  $pg\ g^{-1}$  lipid; (Gaus 2002)) compared to the animal from Darwin analysed for this study. Such high variability within a region may be attributed to biological parameters or local point source influences, however, the PCDD/PCDF congener profiles are similar in all dugongs analysed to date and  $TEQ_{DF}$  covariance in Queensland animals from the same area have been found to be within 12-80%. The generally elevated  $TEQ_{DF}$  levels in dugongs from Queensland have been suggested to be the result of elevated riverine PCDD/PCDF inputs to the nearshore dugong habitats in combination with a low metabolism of toxicologically relevant congeners (Gaus 2002). The differences in  $TEQ$  concentrations observed between the samples analysed for this study and those analysed during previous studies highlight that analysis of a single sample from one region may lead to an incorrect conclusion.



Ref 1 – this study; Ref 2 – (Haynes et al. 1999; Gaus et al. 2001; Gaus 2002, including Gaus, unpublished data); Ref 3 – Ruchel (2001)

**Figure 5.2 TEQ<sub>DF</sub> and TEQ<sub>P</sub> concentrations in marine mammals analysed compared to those reported previously in Australia.**

## 6 Guidelines and recommendations

Some macropod species that have been subject to analysis in this study are also used as a potential food source. A provisional evaluation of these samples is presented with respect to the concentration of the dioxin-like chemicals in the Australian fauna samples was therefore made.

Food guidelines that have been proposed by different countries for different foods of animal origin are presented in Table 6.1 and Table 6.2. The European guideline for dioxin-like chemicals in fats (initially limited to PCDD/PCDFs only, but with the intention to extend it to include dioxin-like PCBs) suggest that levels in meat products should be less than 3 pg TEQ<sub>DF</sub> g<sup>-1</sup> lipid with highest acceptable levels in meat from ruminants. It should be noted that preparation of the guidelines considered the levels that have been found in the testing programs in the specific food types and cannot easily applied to samples collected from macropods. This discussion is only included to compare the values found in samples from this study. The guidelines include a maximum level for farmed game of 2 pg TEQ<sub>DF</sub> g<sup>-1</sup> lipid.

**Table 6.1 Maximum permissible levels for dioxins and dioxin-like substances for EU**

<b>Product</b>	<b>Maximum level<sup>(1)</sup></b>
Meat and meat products originating from:	
• ruminants (bovine animals, sheep)	3 pg TEQDF g <sup>-1</sup> lipid
• poultry and farmed game	2 pg TEQDF g <sup>-1</sup> lipid
• pigs	1 pg TEQDF g <sup>-1</sup> lipid
Liver and derived products	6 pg TEQDF g <sup>-1</sup> lipid
Muscle meat of fish and fishery products and products thereof	4 pg TEQDF g <sup>-1</sup>
Milk and milk products, including butter	3 pg TEQDF g <sup>-1</sup> lipid
Oils and fats:	
Animal fat from:	
• ruminants	3 pg TEQDF g <sup>-1</sup> lipid
• poultry and farmed game	2 pg TEQDF g <sup>-1</sup> lipid
• pigs	1 pg TEQDF g <sup>-1</sup> lipid
• mixed animal fat	2 pg TEQDF g <sup>-1</sup> lipid
Vegetable oil	0.75 pg TEQDF g <sup>-1</sup> lipid
Fish oil intended for human consumption	2 pg TEQDF g <sup>-1</sup> lipid

<sup>(1)</sup>Upper bound concentrations: upper bound concentrations are calculated assuming that all values of the different congeners less than the limit of determination are equal to the limit of determination.

Source: Maximum levels in food in the EU which will apply as of July 1st 2002. "Position paper on dioxins and dioxin-like PCBs, including methods of analysis for dioxins and dioxin-like PCBs", Codex Committee on Food Additives and Contaminants, Thirty-fourth Session, Rotterdam, The Netherlands, 11–15 March 2002

**Table 6.2 (Provisional) legal limits or action limits for dioxins in foods in various Countries**

<b>Country</b>	<b>Foodstuffs of animal origin</b>
Austria	Provisional limits: Pork 2, milk 3, poultry and eggs 5 and beef 6 pg WHO-TEQD g <sup>-1</sup> lipid
Belgium	Milk, bovine, poultry, animal fats and oils, eggs and derived products, if >2% fat: 5 pg WHO-TEQDF g <sup>-1</sup> lipid Pork and derived products, if >2% fat: 3 pg WHO-TEQDF g <sup>-1</sup> lipid
France	Milk and dairy products: 5 pg g <sup>-1</sup> fat
Germany	Recommendations for milk and dairy products in pg I-TEQ g <sup>-1</sup> milk lipid: < 0.9 (desirable target) >3.0 (identification of sources; measures to reduce input recommendations for land use; recommendation to stop direct supply of milk products to consumers) 5.0 (ban on trade of contaminated milk products)
Luxembourg	Recommended: pork 2, beef 6, poultry 5, milk 3 and eggs 5 pg (dioxins) g <sup>-1</sup> lipid
Spain	Levels > 5 pg (dioxins) g <sup>-1</sup> lipid are considered as non-acceptable in dairy products
The Netherlands	Milk, bovine (excluding kidney and liver), poultry (excluding kidney and liver), animal fats and oils, eggs and derived products, if >2% fat: 5 pg WHO-TEQDF g <sup>-1</sup> lipid Pork and derived products, if >2% fat: 3 pg WHO-TEQDF g <sup>-1</sup> lipid. Eel: 8 pg WHO-TEQDF g <sup>-1</sup> eel Milk and derived products with < 2% fat: 0.120 pg TEQDF kg <sup>-1</sup> foodstuff
United Kingdom	Guideline for cows' milk: 0.66 g WHO-TEQ g <sup>-1</sup> whole milk (16.6 pg WHO-TEQ g <sup>-1</sup> lipid) (NB: for dioxins and dioxin-like PCBs together)
Republic of Korea	Beef, pork, chicken meats and eggs: 5 pg WHO-TEQDF g <sup>-1</sup> lipid. (NB: Levels are applied on a temporary basis until reliable scientific evidence is obtained)

<sup>(1)</sup>Upper bound concentrations: upper bound concentrations are calculated assuming that all values of the different congeners less than the limit of determination are equal to the limit of determination.

Source: "Position paper on dioxins and dioxin-like PCBs, including methods of analysis for dioxins and dioxin-like PCBs", Codex Committee on Food Additives and Contaminants, Thirty-fourth Session, Rotterdam, The Netherlands, 11–15 March 2002.

Twenty two kangaroo and wallaby samples were included in the study. Interestingly, 6 of the 22 macropod samples (all of which were grey kangaroos) were found to exceed 3 pg TEQ g<sup>-1</sup> fat and thus the maximum level for meat products. It is, however, noteworthy that the fat content of kangaroo meat is generally low and hence consumption of kangaroo meat is unlikely to contribute substantially to the overall PCDD/PCDF body burden.

For completeness a list of the maximum limits in feed is given in Table 6.3.

**Table 6.3 Maximum levels in feed in the EU**

<b>Product</b>	<b>Maximum level <sup>1</sup></b>
All feed materials of plant origin including vegetable oils and by-products.	0.75 pg WHO-TEQDF g <sup>-1</sup>
Minerals.	1.0 pg WHO-TEQDF g <sup>-1</sup>
Animal fat, including milk fat and egg fat.	2.0 pg WHO-TEQDF g <sup>-1</sup>
Other land animal products including milk and milk products and eggs and egg products.	0.75 pg WHO-TEQDF g <sup>-1</sup>
Fish oil.	6 pg WHO-TEQDF g <sup>-1</sup>
Fish, other aquatic animals, their products and by-products with the exception of fish oil.	1.25 pg WHO-TEQDF g <sup>-1</sup>
Compound feedstuffs, with the exception of feedstuffs for fur animals and feedstuff for fish.	0.75 pg WHO-TEQDF g <sup>-1</sup>
Feedstuffs for fish.	2.25 pg WHO-TEQDF g <sup>-1</sup>

<sup>1</sup> Upper bound concentrations: upper bound concentrations are calculated assuming that all values of the different congeners less than the limit of determination are equal to the limit of determination.

## **7 Summary of findings**

### **7.1 Sampling bias**

The material used in this study was opportunistic. No animals were killed to obtain samples for this study. In some cases, for example the macropod samples from Para Wirra in South Australia, samples were obtained from animal culling. Such studies do have a potential for bias towards the slow and sick animals. This may in turn have led to a bias in the measurement of the concentrations in the animal samples.

The potential bias was recognised, but it was considered that to obtain random samples would be unacceptable ethically. It would be almost impossible operationally to have defined a sampling frame from which to select samples.

In most cases the samples were obtained following the accidental deaths of the animals. The animals were, thus, active at the time of death, suggesting that they were at least in reasonable health. There are, therefore, grounds to assume that any bias from the selection procedure would have been small.

Of greater concern was the effect of the drought that occurred in 2002. The drought was severe and affected much of southern Australia. In seven cases, samples were collected and found to contain insufficient lipid to enable testing for dioxins (see Appendix G). In other cases there was a minimal quantity present. It is postulated that the animals would have been in poor condition due to the drought. It is likely that the animals would have used stored body fat during the drought. Furthermore, it is likely that the lipid would have been removed preferentially to the dioxin-like compounds, so those compounds would have been concentrated in the fatty tissue of the animal.

The effects of the drought would not have been confined to primary feeders such as kangaroo, but would have been perpetrated throughout the food chain.

Both the bias of selection of mammals and the bias due to the drought would be positive, so the results presented in this study would if anything be an overestimate of the general levels in Australia.

### **7.2 Spatial effects**

The initial design of this survey was to obtain a measure of the dioxin levels in macropods across Australia. A spatial analysis of those data was then envisaged. However, as data became available, it became apparent that the concentration of dioxin-like compounds in macropods was generally very low. In fact some cases most of the compounds were at concentrations less than the LOD.

There was no apparent trend with latitude in those data.

However, as shown in Table 3.4, there was a suggestion of an effect of land use. In particular the remote sites had higher levels of 2,3,7,8-TCDD, 2,3,4,6,7,8-HxCDF, PCB 77 and PCB 189. This was a surprising result, as the remote areas would be expected to have little influence from industry and agriculture. Other sources, perhaps wild fires, must be considered to explain this pattern.

Although the above suggestion is based on limited data, it would suggest that the urban/industrial and agricultural areas represented in this survey do not have a large problem from dioxin-like compounds.

### **7.3 Local anomalies**

The detection of outliers in multivariate data is a well-known statistical problem (see for example Barnett and Lewis 1994). However, there are some cases where there is strong evidence of outliers. These outliers are indicative of some local anomaly.

One example is the high level of OCDD in the echidna sample from Port Elliot. This anomaly is supported by the high levels of HxCDD in the same sample (see Table 3.5 and Figure 3.33).

Perhaps the most interesting local anomaly is from the third sample of macropods from Para Wirra. That sample had a high TEQ  $\text{g}^{-1}$  lipid.

Another anomaly is the high level of 1,2,3,6,7,8-HxCDD in the dingos. These samples were from 30 km north of Ceduna on the west coast of South Australia, well away from industry and urban environments. The levels were consistent across the two samples, reinforcing the significance of the anomaly. The local source of the dioxins is unknown, but presumably is in the food chain.

These anomalies are not the only ones present in the data. A multivariate search for outliers may prove to be a fruitful project in its own right.

### **7.4 Bio-magnification**

Different trophic levels had very different concentrations of the analytes. The increase in concentration with the higher trophic levels is consistent with bio-magnification. The high levels of some of the birds of prey (as shown in Table 3.2) indicate potential bio-magnification of three orders of magnitude. These effects were examined in more detail.

This did not occur consistently, as evidenced by the relatively low levels that were found in the black-shouldered kite.

There were only low levels of the analytes in the dingos. This may be an indication that they do not bio-magnify as efficiently as the birds of prey, or perhaps their prey has very low levels of the analytes.

The monotremes also showed elevated levels not only of PCBs (as discussed by Munday et al. 2002), but the PCDD/PCDFs showed even higher levels relative to other species. The data presented in this study are the first analyses of PCDD/PCDFs performed on monotremes tissue. The levels in this fauna class are second only to the birds of prey. The effect of elevated levels of PCDD/PCDFs on monotremes is, therefore, unknown.

### **7.5 Marine mammals**

The whale data included in this survey has been examined by Symons et al. (2003). The dioxin-like PCBs accounted for 85-98% of the combined PCDD/PCDFs and dioxin-like PCB TEQ. All the whales investigated had much lower concentrations of the analytes than Northern Hemisphere whales.

The identical rankings of the top five components of the TEQ across all the marine mammals, despite their species, trophic and geographic diversity, suggests that there is a common source (or at least pathway) for much of the Australian coast.

Symons et al. (2003) noted that 1,2,3,7,8-PeCDD was the largest congener contribution to the TEQ. This congener was a significant contributor to all the marine mammals, but its contribution is almost unrelated to the contribution of the PCB component. The source of the high levels found in the sea lion, which came from a colony on Kangaroo Island is not known.

The highest level of 1,2,3,7,8-PeCDD was found in the humpbacked dolphin from Darwin, although that level does not differ significantly from that in the sea lion. Nevertheless, it is interesting to note that the highest ratio of the concentration of that congener relative to the PCBs was in the dugong that also came from Darwin. Furthermore, the highest contribution to the most contaminated macropod from Jabiru (see Table 3.3) was also from 1,2,3,7,8-PeCDD.

## 7.6 Monotremes

The very high levels of PCB 156 and to a lesser extent PCB 126, in the platypus samples are consistent with the findings of Munday et al. (2002). PCB 156 also had the largest contribution to the echidnas. The echidnas also had larger contributions from PCDD/PCDFs.

The echidnas also had significant concentrations of OCDD. There was a very high level of 14 000 pg g<sup>-1</sup> lipid in the sample from Port Elliot. That sample also had 870 pg g<sup>-1</sup> lipid of 1,2,3,4,6,7,8-HpCDD. The source of this is unknown. Presumably this high concentration has resulted from biomagnification from ants or termites. A possible source may be from insects that have encountered treated timber.

The data presented in this study are the first analyses for PCDD/PCDFs performed on monotremes tissue. The levels in this fauna class were second only to the birds of prey.

## 7.7 Conclusions

The levels of dioxins in the Australian fauna are very variable, especially across fauna classes. Even within a species a CV exceeded 80%.

The overall level of PCDD/PCDFs and dioxin-like PCBs in Australia are generally low by world standards. However, the TEQ of six of the 22 macropod samples exceed the 3 pg TEQ g<sup>-1</sup> lipid, but it is noted that the lipid content of macropod muscle is very low.

There was no evidence from this survey of higher concentrations of dioxin-like chemicals in fauna in the industrial/urban or agricultural areas, the higher levels found were in 'remote levels'.

There was significant evidence of bio-magnification in birds of prey, but the amount of bio-magnification was very variable, ranging from 10-1,000. The levels found in the survey were lower than those found in some other international studies.

The PCB profile of the marine mammals was similar across all species examined, suggesting that these have a common origin or pathway.

There was evidence of some local sources of dioxin-like chemicals.

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## 9 Appendices

### Appendix A Toxic equivalents of congeners for fauna

Congener		WHO <sub>98</sub> -TEF (mammal)	WHO <sub>98</sub> -TEF (avian)
<b>Dioxins</b>	2,3,7,8-TCDD	1	1
	1,2,3,7,8-PeCDD	1	1
	1,2,3,4,7,8-HxCDD	0.1	0.05
	1,2,3,6,7,8-HxCDD	0.1	0.01
	1,2,3,7,8,9-HxCDD	0.1	0.1
	1,2,3,4,6,7,8-HpCDD	0.01	0.01
	OCDD	0.0001	0.0001
<b>Furans</b>	2,3,7,8-TCDF	0.1	1
	1,2,3,7,8-PeCDF	0.05	0.1
	2,3,4,7,8-PeCDF	0.5	1
	1,2,3,4,7,8-HxCDF	0.1	0.1
	1,2,3,6,7,8-HxCDF	0.1	0.1
	2,3,4,6,7,8-HxCDF	0.1	0.1
	1,2,3,7,8,9-HxCDF	0.1	0.1
	1,2,3,4,6,7,8-HpCDF	0.01	0.01
	1,2,3,4,7,8,9-HpCDF	0.01	0.01
	OCDF	0.0001	0.0001
<b>Non-Ortho PCBs</b>	PCB 77	0.0001	0.05
	PCB 81	0.0001	0.1
	PCB 126	0.1	0.1
	PCB 169	0.01	0.001
<b>Mono-Ortho PCBs</b>	PCB 105	0.0001	0.0001
	PCB 114	0.0005	0.0001
	PCB 118	0.0001	0.00001
	PCB 123	0.0001	0.00001
	PCB 156	0.0005	0.0001
	PCB 157	0.0005	0.0001
	PCB 167	0.00001	0.00001
	PCB 189	0.0001	0.00001

## **Appendix B      Details of PCDD/PCDF and dioxin-like PCB analyses**

### **Materials**

The following standards were all purchased from Wellington Laboratories (Ontario, Canada) and were used for calibration, quantification and determination of recovery of PCDD/PCDFs and dioxin-like PCBs.

#### **PCDD/PCDFs**

- EPA-1613-CVS calibration and verification solutions (CS-1 to CS-5)
- EPA-1613-LCS labelled compound surrogate solution
- EPA-1613-ISS-ST internal standard solution

#### **Dioxin-like PCBs**

- WP-CVS calibration and verification solutions (CS-1 to CS-7),
- WP-LCS labelled surrogate spiking solution
- WP-ISS internal standard solution

Acetone, dichloromethane, hexane, and toluene were all OmniSolv® grade sourced from Merck KgaA (Darmstadt, Germany). Ethyl acetate and anhydrous sodium sulfate (granular) were both AR grade sourced from Mallinckrodt (Kentucky, USA). AnalaR® sulfuric acid S.G. was sourced from Merck (Victoria, Australia).

All chromatographic columns were purchased from Fluid Management Systems. (Waltham, MA, USA) and were used without any further treatment. They comprised multi-layer (basic/neutral/acidic) silica, basic alumina and PX-21 carbon dispersed on celite, which are packed in individual teflon columns and vacuum sealed in aluminium foil packages.

### **Sample preparation**

Lipid extraction was performed by sample digestion using concentrated hydrochloric acid with dichloromethane solvent extraction on most of the biological samples. In some cases, accelerated solvent extraction was performed on samples that had been mixed with hydromatrix using a ASE 100 (Dionex, Utah, USA) with ethanol:toluene (68:32) as the extracting solvent and a temperature and pressure of 150°C and 1500 psi, respectively. Approximately 5-10 g of the extracted lipid was accurately weighed and spiked with a known amount of the respective PCDD/PCDFs and dioxin-like PCB isotopically labelled  $^{13}\text{C}_{12}$  surrogate spiking solutions. Lipid was dissolved in hexane and subsequently cleaned up using multiple extractions with concentrated sulfuric acid until the acid layer remained colourless. The hexane extracts were washed several times with water and dried through cleaned anhydrous sodium sulfate. The extracts were then concentrated prior to clean-up on the Power-Prep™ system. Elution through the different columns was computer controlled and required applying the hexane extract first onto the multi-layer silica and using hexane at a flow rate of 10 mL min<sup>-1</sup> onto the alumina column. Dichloromethane:hexane (2:98) at 10 mL min<sup>-1</sup> was used initially. The solvent strength was then modified to dichloromethane:hexane (50:50) and transferred to the carbon column, which was eluted with ethyl acetate:toluene (50:50) in

the forward direction at 10 mL min<sup>-1</sup>. The flow was then reversed and the carbon column was eluted with toluene at 5 mL min<sup>-1</sup>.

Two fractions were collected. The first fraction was collected from the alumina column during elution using dichloromethane:hexane (50:50) and contained the mono-ortho and di-ortho PCBs. The second fraction containing PCDD/PCDFs and non-ortho PCBs were eluted from the carbon column during the reverse elution with toluene. The two fractions were concentrated separately under vacuum and the respective recovery standards (EPA-1613-ISS-ST and WP-ISS) were added and then further concentrated using clean dry nitrogen to a final volume of 10 µL prior to HRGC/HRMS analysis.

### **High-resolution gas chromatography high resolution mass spectrometric (HRGC-HRMS) analysis**

All experiments were conducted on a MAT95XL HRMS (ThermoFinnigan MAT GmbH, Bremen, Germany) coupled to an Agilent 6890 GC (Palo Alto, CA, USA) equipped with a CTC A200S autosampler. A DB-5 (J & W Scientific, Folsom, CA, USA) capillary column (60 m x 0.25 mm internal diameter, film thickness 0.25 µm) was used as the primary analytical column with ultra-high purity helium as the carrier gas. A flow rate of 1.0 mL min<sup>-1</sup> was maintained throughout the chromatographic run. The temperature program was from 100 °C (isothermal for 1 min) then ramp 1 to 200 °C at 40 °C/min, ramp 2 to 235 °C (isothermal for 10 min) at 3 °C min<sup>-1</sup> and then ramp 3 to 310 °C (isothermal 9 min) at 5 °C min<sup>-1</sup>. A 1 µL splitless injection with an injector temperature of 290 °C was employed for all standards and sample extracts. The mass spectrometer operating conditions were: ion source and transfer line temperatures, 240 °C and 280 °C, respectively; ionisation energy 45 eV, filament current 0.7 mA and electron multiplier voltage set to produce a gain of 106. Resolution was maintained at 10,000 (10% valley definition) throughout the sample sequence. Multiple ion detection (MID) experiments were performed in the electron impact mode with monitoring of the exact masses of either M<sup>+</sup>, [M+2]<sup>+</sup> or [M+4]<sup>+</sup> ions for native and labelled compounds. Individual congeners were identified using the GC retention time and ion abundance ratios with reference to internal standards. A DB-dioxin (J & W Scientific, Folsom, CA, USA) capillary column (60 m x 0.25 mm i.d., film thickness 0.15 µm) was used for confirmation analysis when necessary.

## Appendix C Background levels of dioxin-like compounds in Australian terrestrial biota

Mean values are presented with the range in parentheses; n = number; ND = not detected; wwt = wet weight; a = PCDD/PCDFs only; b = Aroclor 1254; c = concentration of 2,3,7,8 TCDD; d = Tri-CBP; e = Tetra-CBP; f = average concentration of PCB corresponding to Aroclor 1260; g = Aroclor 1262 detected in one sample only.

Medium	Compounds			n	Notes	Reference
	Dioxins		Total PCBs			
	$\Sigma 17$ PCDD/PCDF (pg g <sup>-1</sup> lipid)	TEQs g <sup>-1</sup> lipid	WHO-TEQ g <sup>-1</sup> lipid			
Butter	34.3	0.52	0.57	1	QLD, <1945	Müller et al. 2002
	1.3	0.09-0.2	0.11a	2	QLD, 2000	Müller et al. 2001
		0.06-0.12	0.15-0.26 <sup>a</sup>	2	NSW, 2000	"
		0.2-0.37	0.26-0.46 <sup>a</sup>	2	Vic, 2000	"
		0.13	0.15 <sup>a</sup>	1	Tas, 2000	"
		0.1	0.13 <sup>a</sup>	1	SA, 2000	"
		0.1	0.11 <sup>a</sup>	1	WA, 2000	"
			5	AUS	Kalantzi et al. 2001	
Dairy Products (butter, cheese and cream cheese)			4.1 (1.2-8.2) wwt	3-4/ city, pooled	Sydney, Perth, Hobart and Atherton	Kannan et al. 1994
Yoghurt			5 (2-12) <sup>b</sup>	7	Australia	McMahon 1975 in Richardson et al. 1986
Cattle Fat	(<0.4-0.7) <sup>c</sup> (0.3-<0.5) <sup>c</sup>			5 3	Werribee Treat-ment Compl., Vic	Bremner et al. 1990 "
Brush-tail Possum Testicles ( <i>Trichosurus vulpecular</i> )			241 (73-324) <sup>d</sup> 157 (50.3-247.9) <sup>e</sup> 108 (102-121) <sup>d</sup> 66.7 (30.5-107.9) <sup>e</sup>	5 5	Non-urban Vic  Melbourne (urban), Vic	Bolton and Ahokas 1997 "
Platypus ( <i>Ornitho-rhynchus anatinus</i> )			237 (40-570)	9	Tas	Munday et al. 1998

## Appendix C (continued)

Medium	Compounds			n	Notes	Reference
	Dioxins		Total PCBs			
	$\sum 17$ PCDD/PCDF (pg g <sup>-1</sup> lipid)	TEQs g <sup>-1</sup> lipid	WHO-TEQ g <sup>-1</sup> lipid			
Python ( <i>Morelia spilota</i> )	Analysed data not currently available		Majority of con-geners <LOD PCB 153: 160 PCB 138: 140 PCB 180: 50 PCB: 170: 30	1	Northern QLD	Vetter et al. 2001
Pelican ( <i>Pelecanus conspicillatus</i> )			170 000f	6	Brisbane River, QLD	Shaw and Connell 1982
Silver Gull ( <i>Larus novahollandiae</i> )			58 000f	3	Brisbane River, QLD	"
Pacific Black Duck			(ND-400) wwtg	1/8	South-eastern AUS	Olsen et al. 1980
Peregrine Falcon Eggs			(100-4300) wwtg	32	Vic	Pruett-Jones et al. 1981
Beef			160	3 – 4/ city, pooled	Sydney, Perth, Hobart and Townsville	Kannan et al. 1994
Lamb			95	"	"	"
Pork			67	"	"	"
Chicken			34	"	"	"
Chicken Egg			9 (7-10)	?	NSW	McMahon 1975 in Richardson et al. 1986

## Appendix D Background levels of dioxin-like compounds in Australian aquatic biota.

Mean values are presented with the range in parenthesis

ND = not detected; ww = wet weight; sd = standard deviation; a = Mean calculated from 3 individuals as sum PCBs for other 4 individuals were not calculated (see b); b = Not calculated due to many congeners being below the detection limit so that a comparison with other sum PCB levels is not justified; c = Mean calculated from published data.

Medium	Compounds			n	Notes	Reference
	Dioxins		Total PCBs			
	•17 PCDD/PCDF (pg g <sup>-1</sup> lipid)	TEQs g <sup>-1</sup> lipid	WHO-TEQ g <sup>-1</sup> lipid			
<i>Dugong (Dugong dugong)</i>	320 (260 – 390)	17.3 (13 – 22)	ND	3	Northern QLD	Haynes et al. 1999
	370 (22 – 2000)	34	ND	35	QLD	Gaus 2002
			131.3 <sup>a</sup> (not calc <sup>b</sup> – 209)	7	QLD	Vetter et al. 2001
<i>Bottlenose dolphin (Tursiops truncatus)</i>	132 (96 – 162)	2.0 (1.1 – 4.3)	ND	3	QLD	Gaus et al. 2001
			8860.25 <sup>c</sup> (794, 25 524)	4	Northern QLD	Vetter et al. 2001
	30 (15 – 57)	1.3 (0 – 4.4)	60 wwt 9800 (976-23500) TEQ: 190 (10-440)	6	AUS SA	Kemper et al. 1994 Ruchel 2001
<i>Common Dolphin (Delphinus delphis)</i>			627	1	Northern QLD	Vetter et al. 2001
			180 wwt	1	Australia	Kemper et al. 1994

Medium	Compounds			Total PCBs (ng g <sup>-1</sup> lipid)	n	Notes	Reference
	Dioxins		WHO-TEQ g <sup>-1</sup> lipid				
	•17 PCDD/PCDF (pg g <sup>-1</sup> lipid)	TEQs g <sup>-1</sup> lipid					
Dolphin ( <i>Stenella attenuata</i> )				820 wwt	1	Australia	“
( <i>Mesoplodon spp.</i> )				390 wwt	2	Australia	“
Pinniped ( <i>Hydrurga leptonyx</i> )				1130 wwt	1	Australia	“
Australian Sea Lion milk ( <i>Neophoca cinera</i> )				57.16 <sup>d</sup>	5	Australia	Bacon et al. 1992
Sperm Whale ( <i>Physeter macrocephalus</i> )				160 wwt	1	WA	Canella and Kitchener 1992 Kemper et al. 1994
Whales (various species)	ND		1.3	ND	5	South east QLD	Gaus 2002
Green Turtle ( <i>Chelonia mydas</i> )	2000 (170 – 5700)		4.5 (4.7 – 140)	ND	4	South east QLD	Gaus 2002
Fur Seal Blubber ( <i>Arctocephalus pusillus doriferus</i> )				696.7 <sup>c</sup> (53.4 – 3876.8) wwt	11	Seals Rocks, Vic	Smillie and Waid 1987
Shark Muscle (Grey Reef and Bronze Whaler)				35.8 (6.5 – 85.8) wwt	?	Great Barrier Reef, QLD	Smillie and Waid 1984
Sea Mullet ( <i>Mugil cephalus</i> )				16 000 <sup>f</sup>  (64 – 100) wwt 1881 (sd 2214)	? 15 3 11	Port Phillip Bay, Vic Brisbane River, QLD Brisbane, QLD Sydney Harbour, NSW	EPA 1991 Shaw and Connell 1982 Kannan et al. 1994 Roach and Runcie 1998

**Appendix D (continued)**

Medium	Compounds			Total PCBs (ng g <sup>-1</sup> lipid)	n	Notes	Reference
	Dioxins		WHO-TEQ g <sup>-1</sup> lipid				
	•17 PCDD/PCDF (pg g <sup>-1</sup> lipid)	TEQs g <sup>-1</sup> lipid					
Oyster Blennie ( <i>Petroscirtes anolius</i> )				15 000 <sup>f</sup>	5	“	Shaw and Connell 1982
Bony Bream ( <i>Nematolosa come</i> )				27 000 <sup>f</sup>	3	“	“
Polychaete ( <i>Capitella capitata</i> )				10 000 <sup>f</sup>	20	“	“
Sleepy Crab ( <i>Sesarma erythrodractyla</i> )				5 000 <sup>f</sup>	19	“	“
Fiddler Crab ( <i>Uca spp.</i> )				46 <sup>f</sup>	2	“	“
Striped Butterfish ( <i>Selenotica multifasciata</i> )				4 <sup>f</sup>	1	“	“
Mud Crab ( <i>Heliograpsus haswellianus</i> )				38 <sup>f</sup>	1	“	“
Whiting ( <i>Sillago ciliata</i> )				25 <sup>f</sup>	2	“	“
Stilt ( <i>Himantopus himantopus</i> )				9 <sup>f</sup>	1	“	“
Catfish ( <i>Neoarius australis</i> )				16 <sup>f</sup>	1	“	“

<sup>f</sup> = Average concentration of PCB corresponding to Araclor 1260.

**Appendix D (continued)**

Medium	Compounds			Total PCBs (ng g <sup>-1</sup> lipid)	n	Notes	Reference
	Dioxins		WHO-TEQ g <sup>-1</sup> lipid				
	•17 PCDD/PCDF (pg g <sup>-1</sup> lipid)	TEQs g <sup>-1</sup> lipid					
Flathead ( <i>Platycephalus sp.</i> )		1.7 <sup>e</sup> 147		41 <sup>f</sup>  <0.64 1511.6 <sup>c</sup> (667 – 4899)	1 ? ? 5 10	“ Port Phillip Bay, Vic Bass Strait, Vic Coles Bay, Tas Deceitful Cove, Tas	“ EPA 1991 Mosse and Haynes 1993 Mondon et al. 2001 “
Whiting ( <i>Sillago bassensis</i> )		47.8			?	Bass Strait, Victoria	Mosse and Haynes 1993
Snapper ( <i>Chrysophrus auratus</i> )				(22 – 34)	3	Sydney	Kannan et al. 1994
Rubberlip Morwong ( <i>Nemadactylus douglasii</i> )				(99 – 100)	2	“	“
Blue Groper ( <i>Achoerodus viridis</i> )				720	1	“	“
Shovelnose Ray ( <i>Aptchotrema rostrata</i> )				5.8 – 160	4	“	“
Estuarine Crabs ( <i>Australoplax spp.</i> and <i>Scylla spp.</i> )				ND		QLD	Mortimer 2000
King Crab		0.269			1	South of Port Fairy, Vic	Nelson 1994

ND = not detected; ww = wet weight; sd = standard deviation; c = Mean calculated from published data; e = Value represents a conservative rounding up of toxic equivalency data; f = Average concentration of PCB corresponding to Aroclor 1260.

Medium	Compounds			n	Notes	Reference
	Dioxins		Total PCBs			
	•17 PCDD/PCDF (pg g <sup>-1</sup> lipid)	TEQs g <sup>-1</sup> lipid	WHO-TEQ g <sup>-1</sup> lipid			
Spider Crab		0.442		1	Southern Port Phillip Bay, Vic	“
		11		1	Northern Port Phillip Bay, Vic	“
Mussels ( <i>Mytilus coruscus</i> )			23 <sup>f</sup>	2	Brisbane River, QLD	Shaw and Connell 1982
Mussels ( <i>Mytilus edulis</i> )	<0.1 <sup>e</sup>	0.365		?	Port Phillip Bay, Vic	EPA 1991
	(0.298 – 0.709) wwt				Port Phillip Bay, Vic	Haynes and Toohey 1995
			(18-656)		Port Phillip Bay, Vic	Richardson and Waid 1983
			(15-589)	?	Corio Bay, Vic	“
			<298g		Port Phillip Bay,	Philips and Rainbows 1993
			(<10-879)	39	Inshore	
			<103 <sup>g</sup>	21	Offshore	
			(<10-257)			
			348 (20-930)	18	Corio Bay, Inshore	
			185 (110-275)	9	Offshore	“
			140-656 <sup>h</sup>	>400	Corio Bay, Victoria	Prest et al. 1995
			ND	>600	Western Australia	Burt and Ebell 1995

ND = not detected; ww = wet weight; sd = standard deviation; e = Value represents a conservative rounding up of toxic equivalency data; f = Average concentration of PCB corresponding to Aroclor 1260; g = Calculated using the limit of detection (10 µg kg<sup>-1</sup>); h = Range of means presented for each of seven sites.

**Appendix D (continued)**

Medium	Compounds			Total PCBs (ng g <sup>-1</sup> lipid)	n	Notes	Reference
	Dioxins		WHO-TEQ g <sup>-1</sup> lipid				
	•17 PCDD/PCDF (pg g <sup>-1</sup> lipid)	TEQs g <sup>-1</sup> lipid					
Oysters ( <i>Crassostrea gigas</i> )				5139.6c (2644-8836)	5	Deviot, Tasmania	Mondon et al. 2001 "
				5524.2c (1499-8034)	5	Deceitful Cove, Tasmania	
Corals ( <i>Fungi sp.</i> and <i>Acropora sp.</i> )				ND	40	Great Barrier Reef, Queensland	Olafson 1978

ND = not detected; ww = wet weight; sd = standard deviation; a = Mean calculated from 3 individuals as sum PCBs for other 4 individuals were not calculated (see b); b = Not calculated due to many congeners being below the detection limit so that a comparison with other sum PCB levels is not justified; c = Mean calculated from published data; d = Sum of PCB congeners 44, 99, 118, 153, 138, 187, 180 from values presented in paper; e = Value represents a conservative rounding up of toxic equivalency data; f = Average concentration of PCB corresponding to Aroclor 1260; g = Calculated using the limit of detection (10 µg kg<sup>-1</sup>); h = Range of means presented for each of seven sites.

# Appendix E Specimen Collection Form

Sheet to be completed in duplicate. Please send one copy to AGAL with sample and one to David Ellis.

**National Dioxins Program – FAUNA ASSESSMENT:**

SPECIMEN COLLECTION SHEET

**Collector details:** Name:.....  
 Address:.....  
 Phone: ( ).....  
 Animal of origin: .....  
 Species (if known): .....  
 Approximate age (juvenile/mature/old).....  
 Tissue type: .....  
 Date of collection: (dd/mm/yy).....  
 Collection location: State:.....  
 Nearest town:.....

**Site details:**

**Surrounding landuse(s) circle one: Agricultural - if agriculture, please circle dominant form:**

- Cereals – cotton – sugar – grazing – forestry
- Urban (residential)
- Urban (industrial)
- Remote (Note landscape type below)
- Site characteristics: If “Remote”, please circle: Savannah
- Semi-desert savannah
- Woodland
- Open forest

Collection history:

**If sample was collected / excised from archived material, please provide all relevant archive sample information. If sample was collected from a carcass located opportunistically, please indicate where found (e.g. roadside, in park) and also provide an estimate of probable cause of death.....**

Estimated time of death: (as hours / days before collection): .....

Sampling / sub sampling methods: Please describe the process employed for obtaining, handling, packaging and forwarding sample.

**Sampling / sub sampling methods:** Please describe the process employed for obtaining, handling, packaging and forwarding sample.

**For office use only**  
 AGAL sample code.....  
 Sample received AGAL Date:.....  
 Person received AGAL Name:.....  
 AGAL forwarding to: Treatment / location.....

## Appendix F List of collaborators who collected samples

Collector	Organisation	Location(s)	Specimen(s) provided
Niels Andersen	University of Tasmania	Cleveland TAS	Echidna (fat tissue)
Jared Archibald	NT Museum	Darwin NT	Dugong (fat tissue) Humpback dolphin (fat tissue)
Sarah Betts	Emerald Council	Springsure QLD	Eastern grey kangaroo (tail tissue)
Alan Braithwaite	Parks Victoria	Dimboola VIC	Western grey kangaroo (tail tissue)
Kerry Catford	Yarra Park Meat Processors	Gunnedah NSW	Eastern grey kangaroo (tail tissue)
Ray Correll	CSIRO Urrbrae SA	Crafers SA	Koala
David Ellis	CSIRO Urrbrae SA	Hanson Bay (Kangaroo Island SA) Hanson Bay (Kangaroo Island SA) Penneshaw KI SA Penneshaw KI SA Angaston SA Para Wirra Recreation Park SA	Tamar wallaby (tail tissue) 2 Western grey kangaroos (tail tissue) Heath goanna Possum Eastern grey kangaroo (tail tissue) 9 Western grey kangaroos (tail tissue)
Chris Gairns	Moonshadow Raptor Rehabilitation	Jane Brook Perth WA Beldon Perth WA Balga Perth WA	Collared Sparrowhawk Collared Sparrowhawk Hobby falcon
James Gilbert	Kowanyama Natural Resource Management Council	Kowanyama QLD	Agile wallaby (tail tissue)
G. Gondie	C/o Department of Conservation and Land Management WA	Melville (Perth) WA	Western grey kangaroo (tail tissue)
John Gras	CSIRO Atmospheric Research	Mansfield VIC	Eastern grey kangaroo (tail tissue)
Richard Harris	National Parks and Wildlife NSW	Bulga NSW	Eastern grey kangaroo (tail tissue)
John Harris	Parks Victoria	Halls Gap VIC	Western grey kangaroo (tail tissue)
J. Heal	C/o Department of Conservation and Land Management WA	Mt Barker WA	Western grey kangaroo (tail tissue)
Bryan Heywood	Department of Environment and Heritage SA	Mount Gambier SA	Brown goshawk Collared sparrowhawk Black-shouldered kite
Ben Hoffman	CSIRO Sustainable Ecosystems	Winellie NT	Pheasant coucal 2 Brown falcons 2 Brown bandicoots Galah

<b>Collector</b>	<b>Organisation</b>	<b>Location(s)</b>	<b>Specimen(s) provided</b>
Philippa Horton	SA Museum	Adelaide SA	Black shouldered kite
Dan Hough	National Parks and Wildlife Service NSW	Tibooburra NSW Tibooburra NSW	Eastern grey kangaroo (tail tissue) 2 Western grey kangaroos (tail tissue)
Kath Kemper	SA Museum	Kersbrook forest SA Port Elliot SA Port Adelaide SA	Echidna (fat tissue) Echidna (fat tissue) Indo Pacific bottlenose Dolphin (body fat)
Ms Chris Kilpatrick	Fauna carers	Gladstone QLD Gladstone QLD	Swamp Wallaby (tail tissue) 2 Eastern Grey kangaroos (tail tissue)
A. Larwood	SA Museum	Seal Bay Conservation Park SA	Australian sea lion (body fat)
Ian Mason	Australian National Wildlife Collection	Canberra ACT	2 Eastern grey kangaroos (tail tissue)
Peter Mawson	Department of Conservation and Land Management WA	Collie WA	Western grey kangaroo (tail tissue)
Barry Munday	University of Tasmania	Flowery Gully / Glenorchy TAS	2 Platypuses (tail fat)
National Parks and Wildlife Service SA	Monarto Zoological Park SA	Elizabeth SA	Black shouldered kite Peregrine falcon 2 Australian kestrels Collared sparrow hawk
Trevor Patterson	National Parks and Wildlife	Cotton Tree QLD Noosa QLD Gympie QLD Calandra QLD Woodford QLD	Swamp Wallaby (tail tissue) Eastern Grey kangaroo (tail tissue) Brown Goshawk Boobook Owl Wedge-tail eagle
Leonie Perry	Parks and Wildlife Commission NT	Humpty Doo, NT	Agile Wallaby (tail tissue)
Rod Kennett	Environment Australia NT	Jabiru - Kakadu National Park NT	2 Agile wallabies (tail tissue)
Warrick Roe	Department of Conservation and Land Management WA	Kalgoorlie WA Katanning WA	Western grey kangaroo (tail tissue) Western grey kangaroo (tail tissue)
Peter Swinkels	Museum Victoria	Melbourne VIC	Eastern grey kangaroo (tail fat veins) Swamp Wallaby (tail tissue) Rufous Wallaby (tail tissue)
Lindsay Wilson	Forestry Tasmania	Geeveston TAS	
Mr Jim Wood	Forestry SA	Kuitpo forest SA	Western grey kangaroo (tail tissue)
Anthony Yendell	Dog Fence Board SA	Ceduna SA	2 Dingos

## **Appendix G      Samples found to contain insufficient lipid for analysis**

<b>State</b>	<b>Animal type</b>	<b>Laboratory reference</b>
NT	Wallaby	N02/036849
TAS	Wallaby	N02/035204
ACT	Kangaroo	N02/036786
ACT	Kangaroo	N02/036787
QLD	Boobook Owl	N02/035375
QLD	Wallaby	N03/009189
SA	Bearded Dragon	N03/014156

## Appendix H Location of samples and other field details

Sample	Region	State	Town	Collection date	Latitude	Longitude	Use	Common name	Class	Species
1	SW	WA	Jane Brook, Perth	Jan-03	-31.86	116.06	ur	Sparrowhawk	Bi	<i>Accipiter cirrhocephalus</i>
2	SW	WA	Beldon, Perth	Feb-03	-31.77	115.76	ur	Sparrowhawk	Bi	<i>Accipiter cirrhocephalus</i>
3	SW	WA	Balga, Perth	Jan-03	-31.85	115.83	ur	Hobby Falcon	Bi	<i>Falco longipennis</i>
4	N	QLD	Gympie	Oct-02	-26.17	152.58	ag	Goshawk	Bi	<i>Accipiter fasciatus</i>
5	N	QLD	Woodford	Nov-02	-26.28	152.78	ag	Eagle-liver	Bi	<i>Aquila audax</i>
6	N	QLD	Woodford	Nov-02	-26.28	152.78	ag	Eagle-breast	Bi	<i>Aquila audax</i>
7	N	NT	Darwin	?2002	-12.38	130.73	ur	Galah	Bi	<i>Cacatua roseicapilla</i>
8	N	NT	Darwin	Oct-02	-12.38	130.73	ur	Pheasant coucal	Bi	<i>Centropus phasianinus</i>
9	N	NT	Darwin	Oct-02	-12.38	130.73	ur	Brown falcon	Bi	<i>Falco berigora</i>
10	SE	SA	Penola	Jun-05	-37.38	140.17	ag	Collared Sparrowhawk	Bi	<i>Accipiter cirrhocephalus</i>
11	SE	SA	Adelaide	unknown	-34.27	138.60	ur	Collared sparrowhawk	Bi	<i>Accipiter cirrhocephalus</i>
12	SE	SA	Penola	Jun-05	-37.38	140.17	ag	Brown Goshawk	Bi	<i>Accipiter fasciatus</i>
13	SE	SA	Bremer River	2001, Jan	-35.50	138.67	re	Black shouldered kite	Bi	<i>Elanus axillaris</i>
14	SE	SA	Elizabeth	?2002	-34.50	138.65	ur	Black Shouldered Kite	Bi	<i>Elanus axillaris</i>
15	SE	SA	Penola	Jun-05	-37.38	140.17	ag	Black shouldered Kite	Bi	<i>Elanus axillaris</i>
16	SE	SA	Adelaide	unknown	-34.27	138.60	ur	Kestral male	Bi	<i>Falco cenchriodes</i>
17	SE	SA	Adelaide	unknown	-34.27	138.60	ur	Kestrel-female	Bi	<i>Falco cenchriodes</i>
18	SE	SA	Pt Lincoln	2001/2	-34.70	135.85	ag	Peregrine Falcon	Bi	<i>Falco peregrinus</i>
19	SE	SA	Adelaide	unknown	-34.27	138.60	ur	Peregrine Falcon	Bi	<i>Falco peregrinus</i>
20	SE	SA	Ceduna	Jun-03	-31.25	133.68	re	Dingo	Di	<i>Canis familiaris dingo</i>
21	SE	SA	Ceduna	Jun-03	-31.25	133.68	re	Dingo	Di	<i>Canis familiaris dingo</i>
22	SW	WA	Mt Barker	Nov-02	-34.50	117.63	ag	Western grey kangaroo	Ma	<i>Macropus fuliginosus</i>
23	SW	WA	Kalgoorie	Dec-02	-30.76	121.47	ag	Western grey kangaroo	Ma	<i>Macropus fuliginosus</i>
24	SW	WA	Melville, Perth	Dec-02	-33.01	115.70	ur	Western grey kangaroo	Ma	<i>Macropus fuliginosus</i>
25	SW	WA	Katanning	Nov-02	-33.68	117.66	ag	Western grey kangaroo	Ma	<i>Macropus fuliginosus</i>
26	N	NT	Jabiru	Mar-03	-12.67	132.22	re	Agile wallaby	Ma	<i>Macropus agilis</i>
27	N	NT	Jabiru	Apr-03	-12.67	132.22	re	Agile wallaby	Ma	<i>Macropus agilis</i>
28	N	NT	Jabiru	May-02	-13.00	133.00	re	Agile wallaby	Ma	<i>Macropus agilis</i>
29	N	QLD	Springsure	Sep-02	-24.15	148.07	ag	Eastern grey kangaroo	Ma	<i>Macropus giganteus</i>
30	N	QLD	Amberly		-27.36	153.97	ag	Kangaroo	Ma	<i>Macropus sp.</i>
31	N	QLD	Gladstone	Sep-02	-23.20	151.27	ur	Wallaby/kangaroo	Ma	<i>Macropus spp.</i>
32	N	QLD	Coolum/ Noosa	Oct-02	-26.38	153.12	ag	Wallaby/kangaroo	Ma	<i>Macropus spp.</i>
33	SE	VIC	Dimboola	Jan-03	-36.45	142.03	ag	Western grey kangaroo	Ma	<i>Macropus fuliginosus</i>
34	SE	VIC	Halls Creek	Mar-03	-37.13	142.52	ur	Western grey kangaroo	Ma	<i>Macropus fuliginosus</i>
35	SE	SA	Para Wirra	Apr-03	-34.72	138.17	re	Western grey kangaroo	Ma	<i>Macropus fuliginosus</i>
36	SE	SA	Para Wirra	Apr-03	-34.72	138.17	re	Western grey kangaroo	Ma	<i>Macropus fuliginosus</i>
37	SE	SA	Para Wirra	Apr-03	-34.72	138.17	re	Western grey kangaroo	Ma	<i>Macropus fuliginosus</i>
38	SE	SA	Kuipito Forest	f.n.t	-35.18	138.75	re	Western grey kangaroo	Ma	<i>Macropus fuliginosus</i>

Sample	Region	State	Town	Collection date	Latitude	Longitude	Use	Common name	Class	Species
39	SE	NSW	Tibooburra	Nov-02	-29.47	142.07	re	Eastern grey kangaroo	Ma	<i>Macropus giganteus</i>
40	SE	NSW	Gunnedah	Mar-03	-30.32	150.25	ag	Eastern grey kangaroo	Ma	<i>Macropus giganteus</i>
41	SE	VIC	Mansfield	Oct-02	-37.07	146.07	ag	Eastern grey kangaroo	Ma	<i>Macropus giganteus</i>
42	SE	VIC	Healsville	Mar-99	-37.67	145.52	ur	Wallaby/kangaroo	Ma	<i>Macropus spp.</i>
43	SE	SA	Parndana	Nov-02	-35.73	137.23	ag	Wallaby/kangaroo	Ma	<i>Macropus spp.</i>
44	N	NT	Darwin	Jun-92	-12.33	130.73	ur	Dugong	Mm	<i>Dugong dugon</i>
45	N	NT	Darwin	Oct-00	-12.33	130.73	ur	Humpback dolphin	Mm	<i>Sousa chinensis</i>
46	SE	TAS	Sisters Beach	Sep-02	-40.27	145.58	re	Long fin pilot whale	Mm	<i>Globicephala melas</i>
47	SE	TAS	Cloudy Bay	Dec-02	-43.47	147.22	re	Beaked whale	Mm	<i>Mesoplodon grayi</i>
48	SE	SA	Seal Bay	Nov-99	-36	137.33	re	Australian sea lion	Mm	<i>Neophoca cinerea</i>
49	SE	TAS	Waterhouse	Nov-02	-40.27	147.65	re	Sperm Whale	Mm	<i>Physeter catodon</i>
50	SE	TAS	Waterhouse	Nov-02	-40.27	147.65	re	Sperm Whale	Mm	<i>Physeter catodon</i>
51	SE	TAS	Waterhouse	Nov-02	-40.27	147.65	re	Sperm Whale	Mm	<i>Physeter catodon</i>
52	SE	TAS	Waterhouse	Nov-02	-40.27	147.65	re	Sperm Whale	Mm	<i>Physeter catodon</i>
53	SE	TAS	Waterhouse	Nov-02	-40.27	147.65	re	Sperm Whale	Mm	<i>Physeter catodon</i>
54	SE	TAS	Waterhouse	Nov-02	-40.27	147.65	re	Sperm Whale	Mm	<i>Physeter catodon</i>
55	SE	TAS	West Coast	Nov-02	-40.27	144.62	re	Sperm whale	Mm	<i>Physeter catodon</i>
56	SE	SA	Port Adelaide	Aug-98	-34.75	138.50	ur	Bottlenose dolphin-blubber	Mm	<i>Tursiops aduncus</i>
57	SE	TAS	Glengarry	2001, 1997	-40.30	145.18	ag	Platypus	Mo	<i>Ornithorhynchus anatinus</i>
58	SE	TAS	Strathgordon	2001, 1997	-42.77	146.05	re	Platypus	Mo	<i>Ornithorhynchus anatinus</i>
59	SE	SA	Port Elliot	Mar-01	-35.50	138.68	ag	Echidna	Mo	<i>Tachyglossus aculeatus</i>
60	SE	SA	Kersbrook	?2000	-34.78	138.18	ag	Echidna	Mo	<i>Tachyglossus aculeatus</i>
61	SE	TAS	Cleveland	Jan-03	-41.80	147.40	re	Echidna	Mo	<i>Tachyglossus aculeatus</i>
62	N	NT	Darwin	Oct-02	-12.38	130.73	ur	Northern Brown bandicoot	Ot	<i>Isodon macrourus</i>
63	N	NT	Darwin	Dec-02	-12.38	130.73	ur	Northern Brown bandicoot	Ot	<i>Isodon macrourus</i>
64	SE	SA	Crafers	Mar-03	-35.00	138.72	re	Koala	Ot	<i>Phascolarctos cinereus</i>
65	SE	SA	Hanson Bay	?2000	-35.73	137.23	ag	Brush tail possum	Ot	<i>Trichosurus vulpecula</i>
66	SE	SA	Penneshaw	Nov-02	-35.72	137.27	ag	Heath Goanna	Re	<i>Varanus rosenbergi</i>
5	N	QLD	Woodford	Nov-02	-26.28	152.78	ag	Wedgetail eagle-breast	Bi	<i>Aquila audax</i>
27	N	NT	Jabiru	Mar-03	-12.67	132.22	re	Agile wallaby	Ma	<i>Macropus agilis</i>
28	N	NT	Jabiru	Apr-03	-12.67	132.22	re	Agile wallaby	Ma	<i>Macropus agilis</i>
41	SE	SA	Para Wirra	Apr-03	-34.72	138.17	re	Western grey kangaroo	Ma	<i>Macropus fuliginosus</i>
42	SE	SA	Para Wirra	Apr-03	-34.72	138.17	re	Western grey kangaroo	Ma	<i>Macropus fuliginosus</i>
45	N	NT	Darwin	Oct-00	-12.33	130.73	ur	Humpback dolphin	Mm	<i>Sousa chinensis</i>
56	SE	SA	Port Adelaide	Aug-98	-34.75	138.50	ur	Bottlenose dolphin-blubber	Mm	<i>Tursiops aduncus</i>
60	SE	SA	Kersbrook	?2000	-34.78	138.18	ag	Echidna	Mo	<i>Tachyglossus aculeatus</i>

## **Appendix I      Concentrations of PCDD/PCDFs and PCBs in Australian fauna**

The following tables indicate concentrations of PCDD/PCDFs and PCBs in the fauna samples. Field details of the samples are given in Appendix F.

The units are  $\text{pg g}^{-1}$  lipid for concentration or  $\text{TEQ g}^{-1}$  lipid.

The sample number gives a link to Appendix H, where the field data have been recorded. An 'r' following the number indicates a repeat analysis.

The laboratory reference is the internal code used by AGAL

The fauna class is defined in Appendix H.

Appendix I (Continued)

Sample	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19
State	WA	WA	WA	QLD	QLD	QLD	NT	NT	NT	SA	SA	SA	SA	SA	SA	SA	SA	SA	SA
Class	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi
2,3,7,8-TCDD	510	58	3.3	18	16	23	<0.08	5.7	6	52	120	12	0.43	340	15	5.1	17	26	100
Total TCDD isomers	510	59	3.9	18	16	23	0.084	5.7	8.6	59	160	14	0.45	340	16	7.5	18	26	110
1,2,3,7,8-PeCDD	1000	160	8.9	54	110	86	<0.3	38	20	100	570	32	2.5	260	34	23	57	160	280
Total PeCDD isomers	1000	160	8.9	54	110	86	<2	38	0.9	100	620	32	2.7	260	34	24	57	160	290
1,2,3,4,7,8-HxCDD	78	57	4.5	41	130	69	0.47	49	22	53	410	26	0.66	69	27	15	28	200	220
1,2,3,6,7,8-HxCDD	2900	350	20	59	180	120	0.53	51	35	130	1180	99	10	330	110	40	89	200	330
1,2,3,7,8,9-HxCDD	270	12	2.4	8	<7	12	0.33	22	9.2	<1	31	1.4	0.87	43	<2	12	11	<9	<5
Total HxCDD isomers	3200	420	27	110	310	200	1.5	120	61	190	1630	130	12	440	270	70	130	410	560
1,2,3,4,6,7,8-HpCDD	19	19	21	31	110	27	2.7	140	46	29	300	20	1.2	12	90	67	60	36	80
Total HpCDD isomers	19	23	22	31	110	27	2.7	140	59	30	310	20	1.5	12	90	72	66	36	90
OCDD	97	55	<9	100	530	120	43	6400	990	17	240	10	<2	16	88	340	170	38	110
2,3,7,8-TCDF	39	28	4.7	<0.1	<1	<0.5	<0.05	<0.2	14	65	14	13	<0.1	1.9	5.4	6.9	90	11	97
Total TCDF isomers	41	46	8	<0.8	1.3	<5	<0.5	0.38	47	120	65	18	<1	28	13	20	100	11	120
1,2,3,7,8-PeCDF	69	15	2.5	<0.2	<1	1.5	<0.06	<0.3	3.5	24	<20	3.9	0.14	2.5	5.2	2.4	14	2.7	20
2,3,4,7,8-PeCDF	190	120	11	1.5	25	18	<0.1	23	4.6	120	420	22	3.1	33	94	9.1	72	68	100
Total PeCDF isomers	260	140	17	2.3	25	20	<1	23	12	170	470	30	3.3	41	100	15	100	71	130
1,2,3,4,7,8-HxCDF	15	17	3.4	<0.6	<2	0.84	<0.1	1.5	<0.6	<20	<100	<10	1.5	5.3	40	<0.6	16	6.9	17
1,2,3,6,7,8-HxCDF	23	8.7	2.6	<0.4	<3	2.8	<0.1	5.1	0.68	7.6	68	5.6	2.7	20	39	2.6	8.6	51	12
2,3,4,6,7,8-HxCDF	<2	<4	1.9	<0.08	<1	0.97	<0.06	1.3	<0.2	3.4	34	2.6	1.1	9.9	38	1.5	8.9	1.7	5.3
1,2,3,7,8,9-HxCDF	11	<0.9	0.26	<0.2	<0.5	<0.5	<0.06	2	0.3	0.15	<1	<0.1	0.3	19	<1	0.66	<2	<0.3	0.61
Total HxCDF isomers	49	37	8.2	8.1	<10	7.4	<0.7	10	22	<30	<300	<20	6.2	64	120	6	46	63	51
1,2,3,4,6,7,8-HpCDF	<0.6	<0.5	1.3	<0.1	<0.9	<0.4	<0.2	<1	<0.2	<1	8.7	0.82	<0.3	2.4	24	0.7	<8	<0.9	3.6
1,2,3,4,7,8,9-HpCDF	<0.7	<0.7	<0.2	<0.1	<0.8	<0.2	<0.2	<0.2	<0.1	<0.3	0.74	0.13	0.1	0.73	<3	0.14	<3	<0.4	0.64
Total HpCDF isomers	<2	5.6	1.3	3	<2	<0.6	<0.8	<2	<0.4	<2	87	<4	<0.8	4.1	30	<2	<20	2.9	4.2
OCDF	<2	<0.6	<0.1	<0.2	<0.5	<0.08	<0.3	<0.4	<0.6	3.1	9.7	1.3	1.1	6.5	<4	2	<3	12	17
Total PCDD/PCDF's (exc) <sup>1</sup>	5200	950	96	330	1100	480	47	6700	1200	690	3600	255	27	1210	760	560	690	830	1480
WHO98-TEQ <sub>DF</sub> (inc) <sup>2</sup>	1900	330	22	84	170	140	0.4	70	37	240	1100	71	6.2	670	120	41	140	270	500
WHO98-TEQ <sub>DF</sub> (exc)	1900	330	22	84	170	140	0.16	70	37	240	1100	70	6.2	670	120	41	140	270	500

<sup>1</sup> = excluding LOD values

<sup>2</sup> = including half LOD values

Appendix I (Continued)

Sample	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19
Region	SW	SW	SW	N	N	N	N	N	N	SE									
State	WA	WA	WA	QLD	QLD	QLD	NT	NT	NT	SA									
Class	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi	Bi
Laboratory reference	N03/014163	N03/014165	N03/014164	N02/035374	N03/008891	N03/008891/1	N03/003568	N03/003564	N03/003565	N03/021808	N03/021810	N03/021807	N03/014157	N03/021806	N03/021809	N03/021812	N03/021813	N03/013899	N03/021811
PCB 77	280	2400	380	28	140	280	<3	<5	30	610	4920	150	3.4	76	720	620	8540	430	3060
PCB 81	67	400	79	6.1	41	82	<0.3	2.3	5.3	180	1280	48	1.8	49	210	45	359	64	852
PCB 126	1200	5100	700	120	500	1100	<2	220	20	1540	21700	320	46	1680	4910	390	13900	750	7200
PCB 169	730	990	61	110	390	800	0.35	75	28	1130	6610	240	37	1990	1260	150	2760	1380	2360
PCB 105	18000	180000	14000	1300	9700	26000	270	1900	130	49000	321000	9640	290	12200	120000	7640	239000	18600	209000
PCB 114	1000	9300	1300	100	780	2000	11	240	16	4070	46900	990	20	440	4020	740	29100	3380	26900
PCB 118	50000	610000	49000	4600	39000	94000	590	5800	470	211000	2E+06	35100	850	23900	289000	28600	1E+06	119000	765000
PCB 123	1100	11000	880	110	690	1800	<20	<60	<20	3140	43100	990	23	420	5800	600	11900	2070	13500
PCB 156	11000	110000	11000	2200	9100	26000	<100	4200	220	56400	583000	9570	390	8850	78300	4360	239000	33300	219000
PCB 157	2800	34000	2100	590	1900	4600	22	800	51	10600	103000	1950	95	2830	18200	1080	48100	4960	38400
PCB 167	8200	92000	6500	2500	10000	29000	<30	<600	180	62200	713000	<2000	230	3070	147000	5100	896000	43700	282000
PCB 189	2400	12000	750	340	1600	4700	8.8	510	32	7110	53000	1220	150	1930	23300	650	117000	3800	22600
Sum of PCBs (exc) <sup>1</sup>	97000	1E+06	87000	12000	74000	190000	900	14000	1200	410000	4E+06	60000	2100	57000	690000	50000	3E+06	230000	2E+06
WHO98-TEQ <sub>p</sub> (inc) <sup>2</sup>	140	680	84	15	65	150	0.23	26	2.5	230	2800	45	5.4	200	600	47	1800	120	990
WHO98-TEQ <sub>p</sub> (exc) <sup>1</sup>	140	680	84	15	65	150	0.11	26	2.5	230	2800	45	5.4	200	600	47	1800	120	990

<sup>1</sup> = excluding LOD values

<sup>2</sup> = including half LOD values

Appendix I (Continued)

Sample	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35	36	37	38
State	SA	SA	WA	WA	WA	WA	NT	NT	NT	QLD	QLD	QLD	QLD	VIC	VIC	SA	SA	SA	SA
Class	Di	Di	Ma	Ma	Ma														
2,3,7,8-TCDD	0.47	<0.2	<0.03	<0.2	0.16	<0.06	0.47	<0.03	0.088	<0.04	<0.04	<0.09	<0.1	<0.1	0.12	<0.2	0.45	<0.5	<0.2
Total TCDD isomers	0.96	<1	0.036	0.55	0.17	<0.4	0.58	0.04	0.09	<0.5	<1	<1	<0.3	<0.7	0.14	<1	0.51	0.65	<1
1,2,3,7,8-PeCDD	<1	<1	<0.1	0.73	0.35	<0.2	1.3	<0.05	0.18	<0.2	<0.2	<0.03	0.47	<0.03	0.2	0.9	1.9	3	<0.9
Total PeCDD isomers	<5	<6	<0.8	0.75	0.36	<1	1.4	<0.4	0.18	<0.7	<1	<1	0.23	<0.2	0.21	<2	2	3	<6
1,2,3,4,7,8-HxCDD	0.29	<0.5	<0.02	0.61	0.12	<0.09	1.2	0.036	<0.1	0.31	<0.2	0.5	0.36	<0.1	0.15	0.89	1.4	<1	0.99
1,2,3,6,7,8-HxCDD	9	7.3	<0.08	1	0.41	<0.08	1.9	0.39	0.28	<0.4	<0.3	0.5	0.42	<0.1	0.24	1.4	2.3	5.3	2.1
1,2,3,7,8,9-HxCDD	0.21	<0.2	<0.03	<0.5	<0.08	<0.1	0.96	<0.2	0.17	0.44	<0.3	0.3	<0.3	<0.3	<0.1	0.7	1.4	1.1	<0.8
Total HxCDD isomers	10	8	0.14	4	0.62	<0.4	4.8	<0.7	0.69	<1	<1	<1	<1	<1	0.52	3	5.6	7.8	6.4
1,2,3,4,6,7,8-HpCDD	0.92	1	<0.1	3.2	0.76	0.26	10	0.43	1.6	<1	2.3	2.8	2.5	0.5	0.83	3.8	10	3.3	6.6
Total HpCDD isomers	0.92	<1	<0.2	3.2	0.76	0.4	13	0.43	1.9	<0.2	<1	<1	<0.8	<0.6	1.1	4.5	11	4.1	8.4
OCDD	4.8	5.8	<1	57	4.8	2.9	150	3.6	19	12	19	35	40	9.4	4	58	72	54	39
2,3,7,8-TCDF	<0.3	<0.3	<0.06	<0.3	0.21	<0.1	<0.1	<0.01	<0.2	<0.04	<0.06	<0.6	<0.03	<0.3	<0.02	0.19	<0.2	14	<0.3
Total TCDF isomers	<3	<3	<0.5	0.29	0.33	<0.8	<0.8	0.061	<5	<0.6	<1	<1	<0.7	<2	<0.5	0.3	0.35	14	<3
1,2,3,7,8-PeCDF	0.74	<0.4	<0.08	0.34	0.1	<0.06	0.052	<0.01	0.067	<0.04	<0.06	<0.05	<0.04	<0.07	<0.02	<0.09	<0.2	29	<0.1
2,3,4,7,8-PeCDF	0.37	<0.2	<0.2	0.46	0.86	<0.2	0.38	<0.3	<0.08	<0.02	<0.2	<0.1	0.23	0.25	<0.5	0.39	0.39	15	0.28
Total PeCDF isomers	1.3	<4	<1	0.98	1	<1	<0.6	<0.7	0.16	<0.6	<1	<3	0.47	0.25	<1	0.61	<3	49	0.89
1,2,3,4,7,8-HxCDF	<0.2	<0.5	<0.06	<0.05	<0.2	<0.1	<0.1	<0.06	0.069	<0.05	<0.05	<0.1	<0.06	<0.05	<0.03	<0.2	<0.1	14	<0.2
1,2,3,6,7,8-HxCDF	0.26	<0.5	<0.05	0.19	0.19	0.13	<0.1	0.038	0.065	<0.06	<0.05	<0.06	<0.05	<0.05	<0.03	<0.2	<0.1	8.3	0.23
2,3,4,6,7,8-HxCDF	<0.2	<0.5	<0.03	<0.07	0.2	<0.09	0.094	<0.05	<0.03	<0.03	<0.05	<0.04	<0.05	<0.05	<0.02	<0.5	<0.05	0.77	<0.2
1,2,3,7,8,9-HxCDF	<0.2	<0.4	<0.03	<0.06	<0.07	<0.03	<0.03	<0.5	<0.04	<0.07	<0.3	<0.07	<0.03	<0.09	<0.01	<0.2	<0.08	0.7	0.1
Total HxCDF isomers	<2	<4	<0.6	0.3	0.7	<0.6	0.33	<2	0.3	<0.5	<1	<3	0.78	<0.4	<0.1	<2	<1	26	1.6
1,2,3,4,6,7,8-HpCDF	<0.3	<0.4	<0.02	<0.04	0.16	<0.1	0.25	0.059	<0.09	<0.1	<0.2	<0.2	<0.03	<0.1	<0.01	0.11	<0.06	<0.5	0.32
1,2,3,4,7,8,9-HpCDF	<0.1	<0.3	<0.05	<0.07	0.049	<0.03	0.037	<0.02	<0.07	<0.05	<0.1	<0.1	<0.02	<0.07	<0.02	<0.05	<0.06	<0.2	<0.2
Total HpCDF isomers	<0.8	<1	<0.2	<0.2	0.33	<0.2	0.63	<0.1	<0.2	<0.2	<4	<4	2.5	<0.2	<0.08	<0.3	<0.2	<2	0.76
OCDF	<0.4	<0.4	<0.04	<0.08	<0.5	0.55	<0.7	<0.03	0.65	<0.2	<0.1	<0.3	<0.1	<0.3	<0.02	<0.09	<0.07	<0.3	0.64
Total PCDD/PCDF's (exc) <sup>1</sup>	18	14	0.18	67	9	4	171	4	23	12	19	35	44	10	6	66	91	159	58
WHO98-TEQ <sub>DF</sub> (inc) <sup>2</sup>	2.2	1.5	0.14	1.3	1.1	0.23	2.5	0.21	0.39	0.24	0.26	0.29	0.77	0.25	0.5	1.6	3.2	17	1.2
WHO98-TEQ <sub>DF</sub> (exc) <sup>1</sup>	1.7	0.74	140	1.2	1.1	0.016	2.5	0.052	0.35	0.076	0.025	0.16	0.69	0.13	0.37	1.5	3.2	16	0.56

<sup>1</sup> = excluding LOD values

<sup>2</sup> = including half LOD values

Appendix I (Continued)

Sample	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35	36	37	38
Region	SE	SE	SW	SW	SW	SW	N	N	N	N	N	N	N	SE	SE	SE	SE	SE	SE
State	SA	SA	WA	WA	WA	WA	NT	NT	NT	QLD	QLD	QLD	QLD	VIC	VIC	SA	SA	SA	SA
Class	Di	Di	Ma	Ma	Ma	Ma	Ma	Ma	Ma	Ma	Ma	Ma	Ma	Ma	Ma	Ma	Ma	Ma	Ma
Laboratory reference	N03/021815	N03/021816	N03/014308	N03/014305 +306	N03/014307	N03/014309	N03/013926	N03/013927	N03/020664 /5	N02/041220	Daisy	N02/036533-4 /N02/034688	N02/035372 +N02/035373	N03/006136	N03/013082	N03/014296	N03/014299	N03/014302	N03/021814
PCB 77	4.1	15	<0.9	6.5	5.8	<1	5.3	<0.8	<0.6	0.94	<2	0.71	1.3	1.6	<0.5	5.1	5.7	5.1	12
PCB 81	3	1.3	0.34	2.2	4.3	0.46	2.5	0.23	<0.2	<0.3	<1	<0.3	0.7	0.88	0.44	2.4	2.7	8.9	1.7
PCB 126	<0.4	<0.4	2.1	21	34	2.5	27	4.6	<0.2	<0.2	<4	<2	2.8	3.2	1.6	23	23	80	8.7
PCB 169	3.4	5.7	<0.4	2.4	3.9	0.78	2.2	1.4	<0.09	<0.05	0.4	<0.4	<0.6	<0.5	0.52	2.9	<2	26	0.99
PCB 105	10	22	<20	82	620	<20	150	49	<3	<40	150	26	69	<40	66	170	110	160	76
PCB 114	<1	2.5	<0.6	16	58	2	19	6.2	<0.2	<3	12	<1	4.1	<3	4.9	19	19	34	12
PCB 118	28	61	<50	300	1860	<50	450	150	<9	<70	150	60	160	<80	160	480	350	340	270
PCB 123	<2	<2	<2	14	45	<2	19	4	<0.3	<3	<7	<2	4.1	<5	3.3	14	15	19	<9
PCB 156	17	31	<10	100	430	12	150	57	<1	<20	50	9	21	22	17	130	99	150	64
PCB 157	2	3.9	<2	16	81	2.3	26	9.9	<0.4	<0.6	<10	<2	5	<3	<3	22	20	<50	<8
PCB 167	<6	<8	<4	89	90	4.1	86	<9	0.34	<6	<9	<4	26	<10	<6	120	94	<90	51
PCB 189	25	110	2.2	9.7	37	2.3	12	7.3	<0.4	<1	<3	<0.8	<2	<4	1.6	13	9.1	63	7.2
Sum of PCBs (exc) <sup>1</sup>	93	250	4.6	660	3300	26	950	290	0.34	0.94	360	96	290	28	260	1000	750	890	500
WHO98-TEQ <sub>p</sub> (inc) <sup>2</sup>	0.071	0.12	0.22	2.2	4	0.27	2.9	0.53	0.012	0.022	0.27	0.12	0.32	0.34	0.2	2.5	2.4	8.4	0.96
WHO98-TEQ <sub>p</sub> (exc) <sup>1</sup>	0.051	0.097	0.21	2.2	4	0.27	2.9	0.53	3E-06	9E-05	0.065	0.013	0.32	0.33	0.2	2.5	2.4	8.4	0.96

<sup>1</sup> = excluding LOD values

<sup>2</sup> = including half LOD values

Appendix I (Continued)

Sample	39	40	41	42	43	44	45	46	47	48	49	50	51	52	53	54	55	56	57
State	NSW	NSW	VIC	VIC	SA	NT	NT	TAS	TAS	SA	TAS	TAS	TAS	TAS	TAS	TAS	TAS	SA	TAS
Class	Ma	Ma	Ma	Ma	Ma	Mm	Mm	Mm	Mm	Mm	Mm	Mm	Mm	Mm	Mm	Mm	Mm	Mm	Mo
2,3,7,8-TCDD	<0.06	<0.1	0.049	<0.02	<0.03	<0.3	1	<0.4	<0.2	1.2	<0.6	<0.6	<0.3	<1	<2	<2	<1	0.58	1.9
Total TCDD isomers	<0.7	<0.7	0.051	<5	<0.3	<2	6.6			1.2								<3	<3
1,2,3,7,8-PeCDD	<0.04	<0.06	0.12	<0.5	<0.2	<0.9	8.9	<0.2	<0.2	5.4	1.8	<2	<3	1.8	<2	<2	<1	0.69	<0.6
Total PeCDD isomers	<0.6	<0.4	0.13	<7	<1	<6	9.1			5.4								<2	<4
1,2,3,4,7,8-HxCDD	<0.06	<0.05	0.092	<0.4	<0.2	<0.7	7.6	<0.07	<0.2	0.61	<2	2.3	<2	<1	<1	<2	<1	0.28	1.7
1,2,3,6,7,8-HxCDD	<0.08	<0.1	0.34	<0.6	0.65	<0.8	15	<0.2	<0.2	1.7	4.5	5.7	<5	4.2	3.4	6.1	3.3	0.58	1.7
1,2,3,7,8,9-HxCDD	<0.03	<0.09	<0.1	<0.3	<0.1	0.68	4.2	<0.1	<0.2	0.32	<0.7	0.76	<0.7	<0.6	<0.3	<0.4	<0.3	<0.09	<0.07
Total HxCDD isomers	<0.4	<0.6	0.7	<1	<2	<3	26			2.7								<2	3.8
1,2,3,4,6,7,8-HpCDD	<0.2	<0.2	1.1	<2	4.2	1.9	15	1.2	<2	0.88	5.5	4.2	3.5	<2	<2	9.1	2.9	1	<1
Total HpCDD isomers	<0.2	<0.3	1.4	<1	<10	1.9	16			0.88								1	<2
OCDD	<5	<1	18	18	140	5.6	72	13	47	3.3	35	6.2	9.9	16	4.1	88	11	7.2	4.1
2,3,7,8-TCDF	<0.09	<0.05	<0.1	<0.03	<0.02	<0.1	5.6	<0.7	<2	0.35	<0.4	<0.4	<1	<0.4	<0.4	<0.9	<1	11	<0.5
Total TCDF isomers	<0.8	<0.5	0.51	<3	<0.3	<0.8	5.7			0.35								28	<5
1,2,3,7,8-PeCDF	<0.01	<0.008	0.11	<0.05	<0.02	<0.1	1.5	<0.1	<0.09	0.19	<0.4	<0.4	<0.7	<0.4	<0.2	<0.3	<0.4	2.7	<0.2
2,3,4,7,8-PeCDF	<0.04	<0.03	<0.2	<0.5	<0.03	<0.1	2.9	<0.9	<0.7	<0.5	2.7	2.7	3.3	2.3	<3	2.7	2.2	3.6	1.8
Total PeCDF isomers	<0.7	<0.1	0.66	<6	<0.3	<0.7	4			0.58								11	<3
1,2,3,4,7,8-HxCDF	<0.05	<0.01	0.21	<0.08	<0.06	<0.02	<0.3	<0.3	<0.2	<0.3	1.4	1.9	<2	<1	1	<1	<0.9	<2	<0.4
1,2,3,6,7,8-HxCDF	<0.007	<0.03	0.096	<0.2	<0.02	<0.02	0.51	<0.08	<0.1	<0.1	<0.3	<0.3	<0.2	<0.1	<0.1	<0.3	<0.2	<0.4	<0.3
2,3,4,6,7,8-HxCDF	<0.03	<0.02	<0.03	<0.1	<0.03	<0.03	<0.4	<0.5	<0.5	<0.05	<0.4	<0.5	<0.4	<0.3	<0.4	<0.6	<0.5	<0.2	0.34
1,2,3,7,8,9-HxCDF	<0.05	<0.03	<0.04	<0.1	<0.02	<0.02	<0.04	<0.05	<0.09	<0.03	<0.3	<0.05	<0.04	<0.06	<0.04	<0.07	<0.1	<0.04	<0.2
Total HxCDF isomers	<0.6	<0.2	0.39	<4	<0.4	<0.2	0.7			<0.6								13	<2
1,2,3,4,6,7,8-HpCDF	<0.04	<0.05	0.24	<0.2	<0.06	<0.1	<0.2	<0.4	<0.4	<0.09	<0.5	<0.5	<0.3	<0.3	<0.3	<1	<0.4	0.3	<0.2
1,2,3,4,7,8,9-HpCDF	<0.04	<0.04	<0.06	<0.2	<0.03	<0.05	<0.1	<0.1	<0.2	<0.04	0.15	<0.2	<0.2	<0.2	<0.09	<0.1	<0.1	<0.05	<0.1
Total HpCDF isomers	<0.2	<0.2	0.3	<4	<0.1	<0.3	1	0	0	<0.2	0	0	0	0	0	0	0	<0.8	<0.4
OCDF	<0.08	<0.05	0.62	<0.3	<0.2	<0.07	<0.07	<0.7	1.2	<0.1	<1	<0.7	<0.1	<1	<0.3	2.8	<0.5	<0.1	0.34
Total PCDD/PCDF's (exc) <sup>1</sup>	0	0	23	18	140	8	141			14								60	8
WHO98-TEQ <sub>DF</sub> (inc) <sup>2</sup>	0.082	0.11	0.33	0.49	0.27	0.8	15	0.87	0.57	7.1	4.3	3.8	3.9	4.1	3.3	4.3	2.7	4.5	3.6
WHO98-TEQ <sub>DF</sub> (exc) <sup>1</sup>	0.5	1.6	0.26	0.0018	0.12	0.088	15	0.013	0.0048	6.9	3.8	2.5	1.7	3.4	0.44	2.1	1.5	4.4	3.2

<sup>1</sup> = excluding LOD values

<sup>2</sup> = including half LOD values

Appendix I (Continued)

Sample	39	40	41	42	43	44	45	46	47	48	49	50	51	52	53	54	55	56	57
Region	SE	SE	SE	SE	SE	N	N	SE	SE	SE	SE	SE	SE	SE	SE	SE	SE	SE	SE
State	NSW	NSW	VIC	VIC	SA	NT	NT	TAS	TAS	SA	TAS	SA	TAS						
Class	Ma	Ma	Ma	Ma	Ma	Mm	Mm	Mm	Mm	Mm	Mm	Mm	Mm	Mm	Mm	Mm	Mm	Mm	Mo
Laboratory reference	N02/041392-94	N03/010034	N03/010252	N02/031844-5	N02/040955-57	N03/007082	N03/007083	W9	W7	N03/014154	W1	W2	W3	W4	W5	W6	W8	N03/014155	N03/007023
PCB 77	0.95	<0.8	<0.7	2	<0.5	2.9	270	48	98	11	6.6	8.6	6.6	11	6.1	5.9	14	540	14
PCB 81	<0.3	<0.4	0.63	2.4	<0.04	0.67	120	34	47	8.1	8.4	11	11	7.2	7.8	9.4	14	430	8.3
PCB 126	<0.9	5.6	3.4	14	<0.3	2.3	100	64	140	38	180	230	250	160	140	200	120	1700	32
PCB 169	0.14	<0.4	0.75	3	<0.1	<1	1000	500	110	2.8	410	490	480	360	330	480	230	430	44
PCB 105	<40	39	54	420	13	95	240000	13000	8100	16000	4300	6000	6700	4600	3100	6300	3400	610000	850
PCB 114	<0.2	2	4.2	<30	0.82	<4	12000	930	740	480	390	540	610	440	320	500	280	29000	360
PCB 118	<70	<100	150	900	31	<600	630000	39000	32000	56000	14000	23000	23000	16000	11000	20000	10000	1800000	4500
PCB 123	<2	<3	<3	<20	<1	<7	6100	<2000	1600	720	960	1200	1200	<800	<600	1000	570	47000	230
PCB 156	<20	<30	20	190	<6	38	94000	6800	5600	1700	3100	4200	4400	3400	2700	4300	1800	210000	2300
PCB 157	<0.6	<3	<4	35	<1	<6	27000	1800	1200	1100	560	640	840	620	500	790	400	74000	280
PCB 167	<8	<20	8.1	<40	<4	43	<40000	<5000	<3000	<900	<2000	<2000	<3000	<2000	<1000	<2000	<1000	<70000	2100
PCB 189	1.6	<3	2	<10	<0.3	2.6	5000	610	400	56	290	340	440	350	260	410	200	28000	480
Sum of PCBs (exc) <sup>1</sup>	2.7	47	240	1600	45	180	1000000	63000	50000	76000	24000	37000	38000	26000	18000	34000	17000	2800000	11000
WHO98-TEQ <sub>p</sub> (inc) <sup>2</sup>	0.058	0.58	0.38	1.7	0.022	0.3	170	22	23	13	26	34	36	24	21	30	17	580	5.7
WHO98-TEQ <sub>p</sub> (exc) <sup>1</sup>	0.0017	0.56	0.38	1.7	0.0048	0.26	170	21	23	13	26	34	36	24	20	30	17	580	5.7

<sup>1</sup> = excluding LOD values

<sup>2</sup> = including half LOD values

Appendix I (Continued)

Sample	58	59	60	61	62	63	64	65	66	67r	68r	69r	70r	71r	72r	73r	74r
State	TAS	SA	SA	TAS	NT	NT	SA	SA	SA	QLD	NT	NT	SA	SA	NT	SA	SA
Class	Mo	Mo	Mo	Mo	Ot	Ot	Ot	Ot	Re	Bi	Ma	Ma	Ma	Ma	Ma	Mm	Mo
2,3,7,8-TCDD	2.8	2.7	5	3.1	<0.2	<0.09	0.14	<0.2	<0.09	25	<0.4	<0.07	0.15	0.44	1.2	0.43	4.2
Total TCDD isomers	2.8	4	6.3	3.3	<1	0.11	0.3	<1	<0.7	25	0.58	<0.2	0.22	<3	11	0.49	5.1
1,2,3,7,8-PeCDD	6.5	20	18	11	1	0.42	<0.4	<0.5	<0.2	100	1.2	<0.2	0.88	1.4	10	0.81	14
Total PeCDD isomers	6.5	20	18	11	1.1	0.48	<2	<4	<1	100	1.3	<0.4	0.93	<5	10	1	14
1,2,3,4,7,8-HxCDD	18	81	30	13	0.74	0.58	<0.05	0.61	0.19	67	1.3	<0.2	<0.6	1.1	7.6	0.2	20
1,2,3,6,7,8-HxCDD	27	57	56	23	2.1	2.1	<0.07	2.5	<0.3	140	2	<0.3	1.5	1.8	17	0.52	41
1,2,3,7,8,9-HxCDD	8	29	24	7.3	1	<0.5	0.14	0.75	<0.07	17	0.96	<0.1	0.64	1	3.9	<0.2	20
Total HxCDD isomers	51	170	110	43	6.6	<4	0.17	28	<1	224	4.7	<1	2.9	4.3	30	<1	81
1,2,3,4,6,7,8-HpCDD	17	870	150	61	11	12	0.75	84	0.6	32	9.7	<0.2	3.8	8.7	16	1.1	110
Total HpCDD isomers	17	920	150	66	17	18	0	190	1.1	32	12	<0.8	4.4	9.4	17	1.1	110
OCDD	50	14000	550	240	140	140	18	960	<8	130	170	7.5	60	58	74	6.5	380
2,3,7,8-TCDF	<0.1	<0.2	0.14	0.13	<0.05	<0.1	0.42	<0.3	<0.05	<1	<0.1	<0.07	0.22	0.11	5.6	9.5	<0.2
Total TCDF isomers	<0.8	0.9	1.8	0.44	0.19	0.22	0.72	5.1	<0.5	7.1	<0.8	<0.2	0.32	<1	6	14	<2
1,2,3,7,8-PeCDF	<0.1	<0.5	0.16	<0.05	0.097	<0.1	<0.2	0.47	<0.05	1.2	<0.06	<0.07	0.11	<0.07	1.9	2.7	<0.07
2,3,4,7,8-PeCDF	3.9	3.7	2.2	1.5	0.14	0.39	<0.3	0.61	<0.2	20	<0.2	<0.09	0.34	0.27	3.3	3.3	1.6
Total PeCDF isomers	4	5.1	2.6	1.5	0.32	3.3	<3	7.3	<1	26	<0.7	<0.3	0.45	<0.6	7.5	12	1.6
1,2,3,4,7,8-HxCDF	2.9	5.6	1.4	1.2	<0.1	1.5	0.14	<2	<0.2	3.3	<0.09	<0.08	<0.07	<0.05	0.91	2	0.84
1,2,3,6,7,8-HxCDF	<2	4.9	3.2	2.1	<0.07	0.37	0.088	1.5	<0.07	3.1	<0.08	<0.09	<0.2	<0.07	1.6	<0.09	2.4
2,3,4,6,7,8-HxCDF	0.78	4.6	1.5	0.86	<0.06	0.18	0.13	1.4	<0.07	1	<0.1	<0.06	0.078	0.05	0.46	<0.1	0.93
1,2,3,7,8,9-HxCDF	<0.4	<0.7	0.19	<0.05	<0.08	<0.2	0.15	0.15	<0.09	0.6	<0.1	<0.1	<0.06	<0.07	<0.08	<0.04	<0.07
Total HxCDF isomers	6.9	17	7.3	4.3	<0.8	2.6	<1	22	<1	12	<0.4	<0.33	<1	<1	5.8	<3	5
1,2,3,4,6,7,8-HpCDF	<0.9	5	1.4	1.2	0.037	1.9	0.069	28	<0.2	<0.6	<0.05	<0.02	<0.1	<0.06	<0.2	0.26	1
1,2,3,4,7,8,9-HpCDF	<0.1	<0.9	0.053	0.11	<0.1	0.14	0.084	0.93	<0.07	<0.1	<0.2	<0.2	<0.05	<0.08	<0.2	<0.04	<0.06
Total HpCDF isomers	<2	6.3	2.6	1.3	<2	2.7	1.3	48	0.4	<1	<0.4	0.56	0.12	<0.3	1.2	0.34	2.1
OCDF	<0.1	<30	0.45	<0.2	<0.08	0.83	<0.3	37	<0.3	<0.2	<0.2	<0.2	<0.08	<0.1	<0.1	<0.2	<0.8
Total PCDD/PCDF's (exc) <sup>1</sup>	138	15100	850	370	165	168	20	1300	1.5	560	189	8	69	72	163	35	600
WHO98-TEQ <sub>DF</sub> (inc) <sup>2</sup>	17	53	37	20	1.7	1.3	0.54	2.7	0.27	160	2	0.21	1.5	2.5	17	4.3	29
WHO98-TEQ <sub>DF</sub> (exc) <sup>1</sup>	17	53	37	20	1.6	1.2	0.26	2.2	0.025	160	1.7	0.0008	1.5	2.5	17	4.3	29

<sup>1</sup> = excluding LOD values

<sup>2</sup> = including half LOD values

Appendix I (Continued)

Sample	58	59	60	61	62	63	64	65	66	67r	68r	69r	70r	71r	72r	73r	74r
Region	SE	SE	SE	SE	N	N	SE	SE	SE	N	N	N	SE	SE	N	SE	SE
State	TAS	SA	SA	TAS	NT	NT	SA	SA	SA	QLD	NT	NT	SA	SA	NT	SA	SA
Class	Mo	Mo	Mo	Mo	Ot	Ot	Ot	Ot	Re	Bi	Ma	Ma	Ma	Ma	Ma	Mm	Mo
Laboratory reference	N03/007024	N03/014158	N03/014159	N03/025830	N03/003566	N03/003567	N03/014152	N03/014153	N03/014160	N03/008891	N03/013926 /1	N03/013927 /1	N03/014296	N03/014299	N03/007083	N03/014155	N03/014159
PCB 77	4	26	99	14	2.6	2.4	19	17	<0.7	280	5.1	2.4	5.1	6.2	290	556	67
PCB 81	1.2	2.1	5.3	1.1	0.26	<0.1	2.5	13	0.16	84	2.6	0.29	2.8	2.4	120	440	3.8
PCB 126	30	62	36	25	<0.5	3.2	3.3	100	1.1	1140	27	4.3	25	18	110	1690	25
PCB 169	77	43	32	33	2.1	5.4	<0.05	12	9.8	800	2	1.2	2.4	1.3	1110	390	22
PCB 105	360	120	220	160	38	160	110	160	99	20400	150	86	160	91	1E+05	6E+05	130
PCB 114	<300	43	27	36	2.9	9.8	9.7	<6	25	2040	21	<0.9	20	15	8550	29100	<30
PCB 118	2400	780	590	780	69	250	480	350	230	90700	510	300	520	280	5E+05	2E+06	370
PCB 123	140	14	14	15	<0.5	<2	<10	14	<10	1770	20	<8	12	11	7470	40700	9.7
PCB 156	2200	460	110	290	58	490	10	85	200	23000	180	83	130	85	56800	2E+05	84
PCB 157	330	22	<3	77	24	200	1.8	16	48	5510	30	14	24	16	23500	71000	3.5
PCB 167	1400	260	<40	110	17	34	63	<20	<40	31200	140	<20	140	85	<80000	<10000	55
PCB 189	370	170	39	55	6.8	33	<3	10	34	4980	15	<8	11	<5	5510	26100	26
Sum of PCBs (exc) <sup>1</sup>	7300	2000	1200	1600	220	1200	700	780	650	2E+05	1100	490	1100	610	8E+05	3E+06	800
WHO98-TEQ <sub>P</sub> (inc) <sup>2</sup>	5.5	7	4.1	3.1	0.1	0.77	0.4	10	0.38	150	2.9	0.53	2.7	1.9	130	580	2.8
WHO98-TEQ <sub>P</sub> (exc) <sup>1</sup>	5.4	7	4.1	3.1	0.075	0.77	0.4	10	0.38	150	2.9	0.53	2.7	1.9	130	580	2.8

<sup>1</sup> = excluding LOD values

<sup>2</sup> = including half LOD values

## Appendix J Comparison of laboratory duplicates

### Selection of samples for repeat analysis

A series of 8 laboratory duplicates was re-analysed to give a measure of the precision of the results (Table J.1). The samples were selected to include both high and low levels of the dioxin-like compounds, however, the choice was limited by the availability of material, as in many cases the entire lipid was used for the initial analysis.

The choice of material for the repeat analyses was made in conjunction with AGAL staff and used material in their custody. It was not feasible to treat the repeats as 'blinds'. All the agreed analyses were undertaken and are reported.

The majority of the individual dioxin and dioxin-like congeners were detected in various concentrations in the samples selected for repeat analysis. The samples are, therefore, representative of all the samples considered in this study. Accordingly, the estimates of precision obtained from the repeats can be extrapolated with some degree of confidence to the entire sample set.

There was bias in the selection process against samples in which the entire animal was analysed, because these samples were small and typically had only enough lipid for a single analysis.

**Table J.1 Repeat samples used to measure the precision of the laboratory analyses**

Common name	Fauna class	Sample number	Report number
Eagle (breast)	Bird	N03/008891	DAU03_187
Agile wallaby	Macropod	N03/013926/1	DAU03_187
Agile wallaby	Macropod	N03/013927/1	DAU03_187
Western grey kangaroo	Macropod	N03/014296	DAU03_187
Western grey kangaroo	Macropod	N03/014299	DAU03_187
Bottlenose dolphin (blubber)	Marine mammal	N03/014155	DAU03_132
Humpback dolphin	Marine mammal	N03/007083	DAU03_068
Echidna	Monotreme	N03/014159	DAU03_187

### Estimation of mean from duplicate samples

When both measures were above the limit of detection (LOD), the mean was calculated as the average of the two measures. However, this method could not be used when one or both values were below the LOD. These cases are discussed below, with examples given in Table J.2.

Case 1 is the average of the two measures (no concentration below LOD).

In case 2, the LOD of one sample (e.g. <6) is higher than the measured concentration of the other sample (e.g. 4). As the most likely estimate for the non-detectable concentration is 4, the average of the two values is therefore taken as 4.

In case 3, the LOD of measure 2 is lower than measure 1. Thus, the most likely value for measure 2 is the upper limit of its range. Therefore, the true value is taken as the LOD and the average is calculated between the two concentrations.

Case 4 has both measures below the LOD. The average in that case is the range below the lower of the both LODs.

**Table J.2 Estimation of mean and relative error for samples where duplicate data were available**

Case	Measure 1	Measure 2	Estimate of Mean	Relative error
1	4	6	5	2/5 = 0.4
2	4	<6	4	NA
3	6	<4	5	2/5 = 0.4
4	<6	<4	<4	NA

### Estimation of precision

One measure of precision that is commonly used (for example Hall 2000) is the relative error (RE).

$$RE = \frac{\text{mod}(m_1 - m_2)}{(m_1 + m_2)/2}$$

In the equation above,  $m_1$  and  $m_2$  represent the initial and repeat measurements, respectively, and  $\text{mod}(m_1 - m_2)$  is the absolute value of the difference of the two measurement.

For example, with the eagle breast tissue, the initial value for 1,2,3,4,7,8-HxCDF was 0.84 and 3.3 for the repeat sample (refer to Table J.3). In that case the relative error was

$$RE = \frac{\text{mod}(m_1 - m_2)}{(m_1 + m_2)/2} = \frac{\text{mod}(0.84 - 3.3)}{(0.84 + 3.3)/2} = \frac{2.46}{2.07} = 1.19$$

Examples of the calculation for four cases are shown in Table J.2.

There was generally close agreement between the laboratory duplicates, but there were also some differences. An example of a large difference is that from 1,2,3,4,7,8-HxCDF from the eagle breast tissue that was shown above. There were also large relative errors between the two analyses of the macropod tissue. For example the results for PCB 77 were <0.8 and 2.4, which gave a relative error of 1.00. An even more extreme difference was obtain for PCB 114, with values of 6.2 and <0.9 which gave a relative error of 1.49. Examples for laboratory duplicates are given in Table J.3.

**Table J.3 Comparison of analyses for breast tissue from a wedge-tail eagle and an agile wallaby.**

Congener	Eagle breast		Macropod (N03/013927)	
	Initial	Repeat	Initial	Repeat
2,3,7,8-TCDD	23	25	<0.03	<0.07
1,2,3,7,8-PeCDD	86	100	<0.05	<0.2
1,2,3,4,7,8-HxCDD	69	67	0.036	<0.2
1,2,3,6,7,8-HxCDD	120	140	0.39	<0.3
1,2,3,7,8,9-HxCDD	12	17	<0.2	<0.1
1,2,3,4,6,7,8-HpCDD	27	32	0.43	<0.2
OCDD	120	130	3.6	7.5
<hr/>				
2,3,7,8-TCDF	<0.5	<1	<0.01	<0.1
1,2,3,7,8-PeCDF	1.5	1.2	<0.01	<0.07
2,3,4,7,8-PeCDF	18	20	<0.3	<0.09
1,2,3,4,7,8-HxCDF	0.84	3.3	<0.06	<0.08
1,2,3,6,7,8-HxCDF	2.8	3.1	0.038	<0.09
2,3,4,6,7,8-HxCDF	0.97	1	<0.05	<0.06
1,2,3,7,8,9-HxCDF	<0.5	0.6	<0.5	<0.1
1,2,3,4,6,7,8-HpCDF	<0.4	<0.6	0.059	<0.02
1,2,3,4,7,8,9-HpCDF	<0.2	<0.1	<0.02	<0.2
OCDF	<0.08	<0.2	<0.03	<0.2
<hr/>				
PCB 77	280	280	<0.8	2.4
PCB 81	82	84	0.23	0.29
PCB 126	1100	1140	4.6	4.3
PCB 169	800	800	1.4	1.2
<hr/>				
PCB 105	26000	20400	49	86
PCB 114	2000	2040	6.2	<0.9
PCB 118	94000	90700	150	300
PCB 123	1800	1770	4	<8
PCB 156	26000	23000	57	83
PCB 157	4600	5310	9.9	14
PCB 167	29000	31200	<9	<20
PCB 189	4700	4980	7.3	<8

Units are pg g<sup>-1</sup> lipid

The relative error is a measure of the coefficient of variation  $\times \sqrt{2}$ , but it is based on only a single degree of freedom. A more stable measure of the between duplicate variation can be obtained by averaging across the pairs of samples, which leads to the coefficient of variation (CV). The formula used was

$$CV(\%) = \sqrt{\frac{\sum_{i=1}^{i=n} RE_i^2}{2n}} \times 100$$

where  $n$  was the number of samples that were used in the estimate of the CV (up to eight, the number of duplicates used as described in Table J.1). This formula is an

approximation because not only is the standard deviation subject to error but the mean is as well. The approximation is satisfactory because the mean has a much a lower variance than the standard deviation.

**Table J.4 Estimates of coefficient of variation for each congener based on laboratory duplicates**

Congener	Number of pairs	CV*
2,3,7,8-TCDD	6	12%
1,2,3,7,8-PeCDD	7	13%
1,2,3,4,7,8-HxCDD	7	19%
1,2,3,6,7,8-HxCDD	8	13%
1,2,3,7,8,9-HxCDD	6	15%
1,2,3,4,6,7,8-HpCDD	8	21%
OCDD	8	21%
2,3,7,8-TCDF	3	8%
1,2,3,7,8-PeCDF	5	28%
2,3,4,7,8-PeCDF	7	22%
1,2,3,4,7,8-HxCDF	4	58%
1,2,3,6,7,8-HxCDF	3	44%
2,3,4,6,7,8-HxCDF	4	17%
1,2,3,7,8,9-HxCDF	2	47%
1,2,3,4,6,7,8-HpCDF	5	54%
1,2,3,4,7,8,9-HpCDF	0	
OCDF	0	
PCB 77	8	27%
PCB 81	8	11%
PCB 126	8	11%
PCB 169	7	13%
PCB 105	8	24%
PCB 114	7	41%
PCB 118	8	22%
PCB 123	7	15%
PCB 156	8	18%
PCB 157	8	13%
PCB 167	5	19%
PCB 189	7	21%
<b>Average</b>	<b>5.9</b>	<b>21%</b>

\* The average CV was obtained by using weights proportional to the number of pairs contributing to its estimation

No estimate of the CV was available for 1,2,3,4,7,8,9-HpCDF and OCDF due to the number of observations that were below the LOD. In other cases there were only a few pairs that could be used for estimating the CV. Table J.4 shows that the average CV was 21%. Most congeners had a CV less than 21%, the average being increased by a

few congeners that had higher coefficients of variation and this was linked to their low concentrations.

### Estimation of field variability

An estimate of the field variability was obtained by considering samples of the same species that were collected from a similar location. An example of the method used in the estimation of field variability (for 2,3,7,8-TCDF) is given in Table J.5.

**Table J.5 Method used in the estimation of average field variability.**

Location	Common name	No. of replicates	Degrees of freedom	CV	Weighted sum of squares
Jabiru	Agile wallaby	3	2	55%	0.61
Ceduna	Dingo	2	1	0%	0.00
Adelaide	Kestrel	2	1	121%	1.47
Darwin	Northern brown bandicoot	2	1	47%	0.22
Waterhouse	Whale	6	5	49%	1.20
Para Wirra	Western grey kangaroo	3	2	167%	5.61
Overall		18	12	87%	9.11

The analyte used in this example was 2,3,7,8-TCDF

Where pairs were available, a similar approach could be used to that for the laboratory duplicates. In particular, a CV based on a single degree of freedom could be estimated for each pair. As shown above, CV is closely related to the relative error approach used by Hall (2000).

The CV was also derived where triplicates or multiple samples were available (as shown in Table J.6). An average CV was obtained by first multiplying the square of CV by its degrees of freedom to obtain a sum of squares, totalling the sums of squares across each animal type, dividing by the total degrees of freedom, and finally taking the square root to obtain the average CV. The formula for obtaining the average CV is shown below.

$$CV_{\text{average}} = \sqrt{\frac{\sum df \times CV^2}{\sum df}}$$

An example of the calculation in Table J.6 is given here. The first step was to calculate the degrees of freedom, which was one less than the number of observations. For the agile wallaby, the sum of squares was calculated as  $2 \times 0.55 \times 0.55 = 0.61$ . The sums of squares were totalled to give 9.11 and subsequently divided by the total number of degrees of freedom (12) to give a mean square of 0.755. The square root of this number yielded the average CV of 87%.

**Table J.6 Estimated coefficients of variation between field replicates**

Location	Jabiru	Ceduna	Adelaide	Darwin Northern brown bandicoot	Waterhouse Whale	Para Wirra Western grey kangaroo	Overall
Common name	Agile wallaby	Dingo	Kestrel				
Number of replicates	3	2	2	2	6	3	18
2,3,7,8-TCDD	117%	92%	76%	54%	49%	53%	72%
1,2,3,7,8-PeCDD	126%	0%	60%	58%	30%	58%	65%
1,2,3,4,7,8-HxCDD	156%	10%	43%	17%	63%	46%	79%
1,2,3,6,7,8-HxCDD	110%	15%	54%	0%	31%	71%	59%
1,2,3,7,8,9-HxCDD	126%	50%	6%	85%	61%	28%	72%
1,2,3,4,6,7,8-HpCDD	132%	6%	8%	6%	75%	61%	77%
OCDD	139%	13%	47%	0%	121%	9%	98%
2,3,7,8-TCDF	55%	0%	121%	47%	49%	167%	87%
1,2,3,7,8-PeCDF	31%	81%	100%	45%	42%	172%	86%
2,3,4,7,8-PeCDF	114%	81%	110%	67%	24%	162%	93%
1,2,3,4,7,8-HxCDF	30%	61%	136%	132%	51%	172%	97%
1,2,3,6,7,8-HxCDF	32%	3%	76%	117%	45%	169%	86%
2,3,4,6,7,8-HxCDF	55%	61%	101%	101%	24%	136%	76%
1,2,3,7,8,9-HxCDF	43%	47%	29%	61%	109%	151%	98%
1,2,3,4,6,7,8-HpCDF	80%	20%	99%	136%	56%	87%	78%
1,2,3,4,7,8,9-HpCDF	64%	71%	117%	67%	43%	81%	67%
OCDF	112%	0%	20%	128%	142%	76%	113%
PCB 77	107%	81%	122%	6%	27%	9%	63%
PCB 81	141%	56%	110%	96%	18%	78%	81%
PCB 126	137%	0%	134%	121%	22%	80%	84%
PCB 169	90%	36%	127%	62%	16%	139%	80%
PCB 105	102%	53%	133%	87%	27%	25%	67%
PCB 114	135%	94%	134%	77%	23%	39%	79%
PCB 118	101%	52%	136%	80%	28%	26%	67%
PCB 123	130%	0%	128%	85%	47%	23%	76%
PCB 156	105%	41%	136%	111%	19%	24%	69%
PCB 157	104%	46%	135%	111%	20%	16%	69%
PCB 167	152%	20%	140%	47%	32%	48%	81%
PCB 189	95%	89%	140%	93%	20%	113%	83%

## **Discussion of sources of variation**

The large variability observed between replicate field samples was a consistent feature across the six species shown in Table J.6. The whale had a variability that was typical of the other species, so it had little effect on the estimate of the average CV. However, the inclusion of the data yielded a much more stable estimate of the CV because it was based on 12 degrees of freedom.

The observed field variability is a function of the true field variability (including sampling the lipid from the animal) and the analytic uncertainty. The variances of these components are additive, so the true field variability can be derived from the observed and analytic variances. These estimates, which are given in Table J.7, show that the field component was in most cases much larger than the laboratory component. There is therefore potential for reducing the field variability by using composite samples that include several animals. The increased precision was bought at the expense of losing information on between animal variation. This policy was followed where material was available.

A point of interest here is that the variability between the Para Wirra samples contributed a large component to the large between sample variability. These three samples were in fact each composites of three kangaroo tails. However, there were other large outliers as well, one whale sample (W6) had approximately 5 times as much OCDD as the average of the other whale samples (W1 to W5) from that location.

The field variability is of interest in its own right. This prompts questions as to what other factors are contributing to this variability. These factors may include age, breeding status or variability in the food chain.

Finally, this study of the variability of the samples from similar environments indicates that caution is required in the interpretation of the results presented in this study. From Table J.7 it can be seen that the average CV is 79%. The CV for TEQ (including LODs as 0.5 the LOD) was 85%. This large CV means that a doubling of concentration between samples could well be attributable to sampling variation.

**Table J.7 Estimation of true field variability**

Congener	Laboratory variability (%)	True field variability (%)	Observed field variability (%)
2,3,7,8-TCDD	12	71	72
1,2,3,7,8-PeCDD	13	63	65
1,2,3,4,7,8-HxCDD	19	77	79
1,2,3,6,7,8-HxCDD	13	58	59
1,2,3,7,8,9-HxCDD	15	70	72
1,2,3,4,6,7,8-HpCDD	21	74	77
OCDD	21	95	98
2,3,7,8-TCDF	8	87	87
1,2,3,7,8-PeCDF	28	81	86
2,3,4,7,8-PeCDF	22	91	93
1,2,3,4,7,8-HxCDF	58	78	97
1,2,3,6,7,8-HxCDF	44	74	86
2,3,4,6,7,8-HxCDF	17	74	76
1,2,3,7,8,9-HxCDF	47	86	98
1,2,3,4,6,7,8-HpCDF	54	56	78
1,2,3,4,7,8,9-HpCDF	NA	NA	67
OCDF	NA	NA	113
PCB 77	27	57	63
PCB 81	11	80	81
PCB 126	11	84	84
PCB 169	13	79	80
PCB 105	24	62	67
PCB 114	41	68	79
PCB 118	22	63	67
PCB 123	15	75	76
PCB 156	18	67	69
PCB 157	13	68	69
PCB 167	19	78	81
PCB 189	21	80	83
<b>Average</b>	<b>23</b>	<b>74</b>	<b>79</b>

## **Appendix K      Multivariate analysis**

### **Methodology**

The profiles of the dioxin-like analytes can be further described using multivariate analyses. There are many ways this can be approached, and these data offer many opportunities for multivariate analyses. The purpose of the analyses should be considered before choosing a particular technique. For example, a principal component analysis would be appropriate for showing the overall structure of the data, whereas differences between fauna classes (or regions) would be better demonstrated by using a canonical variate approach.

Even within the principal components, a decision must be made as to whether the analysis is looking at the overall contributions to the variance, the contributions of components after standardising for their relative variability, or whether those components with the highest coefficient of variation should be emphasised. These analyses require operations on the sums of squares and products matrix, the correlation matrix, or the sums and squares of products after a logarithmic transformation. Each form of the analysis highlights different aspects of the data.

There are many other alternatives that could be used with these data. These include using subsets of the data (e.g. only macropods), using subsets of the analytes, comparing ordinations from one set of analytes with other sets (perhaps contrasting the dioxins and furans with the PCBs).

Another approach is to weight the data by their toxicity, so the input variables are then the contributions to the TEQ. Weighting in this manner would have no effect if the principal components were based on the correlation matrix. A useful approach would be to base the weighted analysis on the sums of squares and products matrix of the logarithmically transformed data.

The above methods give information about the samples based on the analytes. It would also be feasible to do the analysis of the analytes based on their concentrations in different samples.

### **Analysis based on all samples**

A principal component analysis was performed on all the single analyte data, where the components were extracted from the sums of squares and products matrix of the logarithmically transformed data. The first component of this analysis contained 79% of the information, and the first and second together included 88%. The loadings of these two components are shown in Table K. The loadings of the first component were all the same sign, suggesting that that component was a measure of the overall degree of contamination. The second component was a contrast between the PCDD/PCDFs and the PCBs.

The analysis indicates that for the samples considered, almost 80% of the variation was due to how much of the dioxin-like compounds was present. The next component (or almost half of the remainder) was whether these compounds were PCDD/PCDFs or PCBs. Together these components accounted for 88% of the variation.

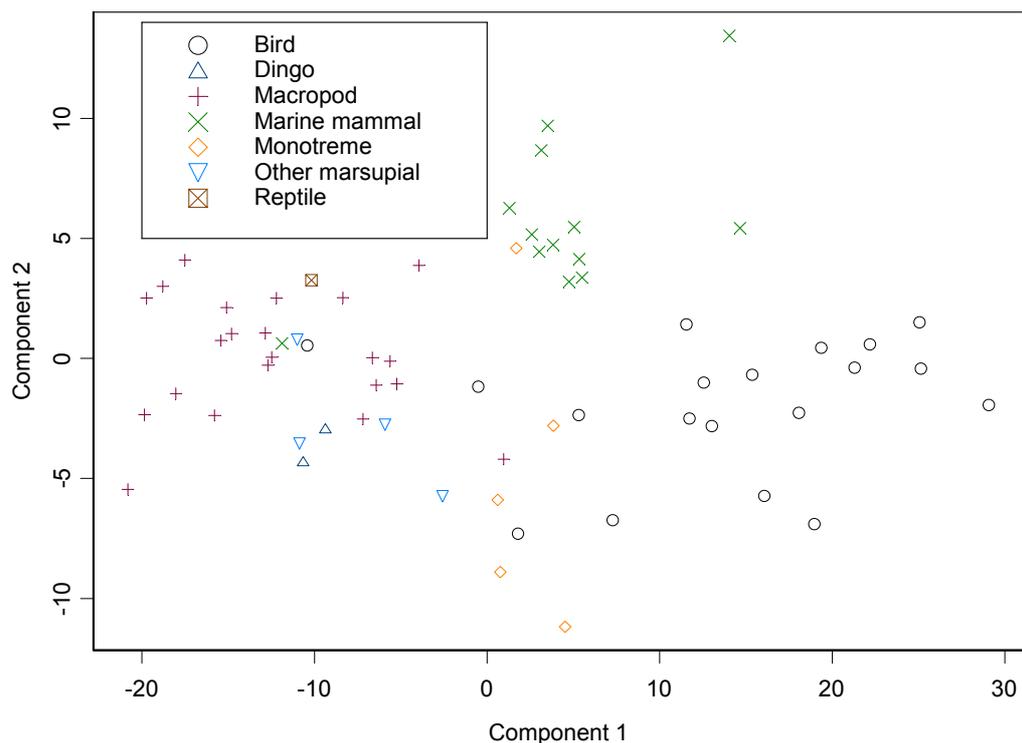
**Table K.1 Principal component loadings for overall analyses.**

Congener	Component1	Component 2
2,3,7,8-TCDD	0.186	-0.183
1,2,3,7,8-PeCDD	0.191	-0.243
1,2,3,4,7,8-HxCDD	0.176	-0.283
1,2,3,6,7,8-HxCDD	0.186	-0.297
1,2,3,7,8,9-HxCDD	0.117	-0.299
1,2,3,4,6,7,8-HpCDD	0.111	-0.27
OCDD		-0.273
2,3,7,8-TCDF	0.157	
1,2,3,7,8-PeCDF	0.149	
2,3,4,7,8-PeCDF	0.193	-0.101
1,2,3,4,7,8-HxCDF	0.157	-0.113
1,2,3,6,7,8-HxCDF	0.155	-0.258
2,3,4,6,7,8-HxCDF	0.14	-0.115
1,2,3,7,8,9-HxCDF		-0.18
1,2,3,4,6,7,8-HpCDF		-0.125
1,2,3,4,7,8,9-HpCDF		
OCDF		-0.126
PCB 77		0.188
PCB 81	0.183	
PCB 126	0.223	
PCB 169	0.247	
PCB 105	0.225	0.231
PCB 114	0.238	0.198
PCB 118	0.241	0.227
PCB 123	0.237	0.258
PCB 156	0.243	0.168
PCB 157	0.258	0.195
PCB 167	0.262	0.116
PCB 189	0.248	0.101

The analyses were based on the sums of squares and products of the logarithmically transformed data. Loadings with very small contributions have been excluded.

A plot of the components is given in Figure K.1. In general the classes of animals are in their own clusters. The macropods are grouped together, but with one high value on Component 1, which was a macropod sample from Para Wirra. That sample was also relatively high in PCDD/PCDFs compared to PCBs.

The birds generally high values for Component 1 and low values for Component 2, the galah was an exception having a low value for Component 1.



**Figure K.1 Principal component analysis of all the data.**

The marine mammals had several outliers, with the dugong having a low value for Component 1 and also for Component 2 compared to the other marine mammals. The dolphin from the Port River (Adelaide) was high both in the Component 1 (overall load) and in Component 2 (proportion of PCBs), whereas the humpback dolphin from Darwin had a similar score for Component 1 but much lower value of Component 2 (proportion of PCBs).

The monotremes all had similar values for Component 1 but very mixed values for Component 2. The two platypus samples had the higher values of Component 2, indicating that they have relatively higher levels of PCBs.

The dingo, goanna and other marsupial samples had similar scores to those of the macropods.

### **Discussion of principal component analysis**

The principal component analysis presented above was very interpretable and distinguished the fauna classes, and to a lesser extent the trophic levels in a very clear manner. The application of other multivariate techniques may also offer useful insights into this data set. Such analyses should be arranged so that they focus on questions of interest to the environmental chemist, rather than using the multivariate techniques in their own right.

A simple extension of the above analysis would be to use subsets of the data (e.g. marine mammals). However, with the resultant smaller sample size, the estimates of the loadings would become less reliable and hence more difficult to interpret.