



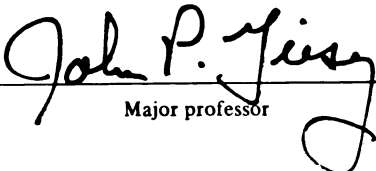
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Heidi Joan Auman

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**PLASTIC INGESTION, BIOMARKERS OF HEALTH, PCBS AND DDE  
IN TWO SPECIES OF ALBATROSSES ON SAND ISLAND, MIDWAY ATOLL**

**By**

**Heidi Joan Auman**

**A THESIS**

**Submitted to  
Michigan State University  
in partial fulfillment of the requirements  
for the degree of**

**MASTER OF SCIENCE**

**Department of Fisheries and Wildlife**

**1994**

## ABSTRACT

### PLASTIC INGESTION, BIOMARKERS OF HEALTH, PCBS and DDE IN TWO SPECIES OF ALBATROSSES ON SAND ISLAND, MIDWAY ATOLL

By

Heidi Joan Auman

Possible impacts of pollution in a remote area of the Pacific Ocean on Laysan (*Diomedea immutabilis*) and black-footed albatrosses (*Diomedea nigripes*) were assessed. Dead Laysan albatross chicks had significantly greater weights of plastic in their proventriculi and gizzards and had significantly lighter body weights than injured but otherwise healthy chicks. Ingested plastic is probably not a significant direct cause of death in Laysan albatross chicks, but likely cause varying degrees of physiological distress. Concentrations of PCBs, DDE and biomarkers of exposure were measured in the plasma of chicks and adults of both species. Concentrations of retinol, retinyl palmitate, total vitamin A, total and free triiodothyronine (T3) and thyroxine (T4) in plasma were not correlated to total PCB concentrations. Significant differences in total PCB concentrations were detected between species, between chicks and adults, and among nesting periods. DDE concentrations were also significantly different between species.

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**This thesis is dedicated to my Mother, who made all things possible for me.**

## **ACKNOWLEDGMENTS**

**I would like to express my appreciation to some of the people instrumental to my research. C.P.O. Gregg E. Diefenderfer of the U. S. Navy spent hundreds of donated hours assisting me in blood sampling, banding, necropsying and monitoring chicks during my visits to Midway. His effort, courage and stamina, along with his knowledge of computers, have earned him a place in this thesis and in my heart.**

**Dr. James Ludwig, my friend, mentor and boss, gently showed me the inspiration and dedication needed to achieve any goal, and provided me with the opportunity to have a role in such an exotic and fulfilling project. Dr. John Giesy, my advisor, honed my scientific skills and helped me focus on my goals and objectives. I would also like to thank my committee members, Dr. Jim Sikarskie, Dr. Scott Winterstein, and Dr. Steve Bursian, for their valuable input, encouragement, and friendship. Dave Verbrugge put in many hours of overtime to make sure the PCB and DDE data got to me in time. Jill Dedoes was my tireless technician and always available to assist me in a time crunch. Cheryl Summer was a reliable and organized fellow field-worker and consistently willing to put in the extra effort to get a job done. Bill Bowerman and John Besser patiently led me**



through the agonies of the SAS program, and Jody Kubitz assisted me with photographs and slides. The residents of Midway were very supportive of my research, and I thank Peter Paris and Florante DeLeon for use of equipment in the Medical Center, and "Burma" Prematilaka of Pest Control for the daily carcass delivery. I am also indebted to the U. S. Environmental Protection Agency and the World Wildlife Fund for funding, and to the U. S. Navy and U. S. Fish and Wildlife Service for permission to conduct my research on Midway.

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## **GENERAL INTRODUCTION**

The incidence of man-made, persistent, synthetic compounds dispersed by atmospheric and aquatic transport and the adverse effects they cause in wildlife have been documented in birds, including the fish-eating birds of the North American Great Lakes. The contamination of the northcentral Pacific Ocean and the related bioeffects in seabirds has not been as well documented as the birds of the Great Lakes. Accumulation of organohalogens such as DDE, the ubiquitous PCBs and ocean-borne plastic materials is an increasing cause for concern (Fisher, 1973; Tanabe et al., 1987; Day and Shaw, 1987; Sileo et al., 1990). The north Pacific Ocean is a remote area and hypothesized to be less affected by contamination than more industrialized and urbanized areas. This research was conducted to determine the status and trends of contaminants and biomarkers in a remote area in order to assess the extent of global contamination. Midway Atoll is located in the north central Pacific Ocean (28° 11' N, 177° 22' W), about 1200 miles northwest of Honolulu, almost equidistant between California and Japan. Midway was a United States Naval Air Facility from 1941 to 1993, a National Wildlife Refuge, and the breeding area for several million seabirds.

Laysan (*Diomedea immutabilis*) and black-footed (*Diomedea nigripes*) albatrosses were studied because both species are at the top of the food chain and feed over a wide area of the north Pacific Ocean. The diet of the Laysan albatross consists primarily of eight families of squid while that of the black-footed albatross consists primarily of flying fish eggs (family Exocoetidae) (Harrison, 1990). Therefore, while these two species are classified as being at the same trophic level, they fill different ecological niches and were thought to be exposed to different types or amounts of contaminants (Fisher, 1973). Both species of albatrosses are long-lived, and representatives of all age classes can be identified in the banded populations on Midway Atoll. The estimated annual adult mortality of 8.6% insures that many banded birds return each year for long-term studies (Rice and Kenyon, 1962). Albatrosses have little fear of humans, and the sheer numbers and densities make an adequate sample size easy to attain. The strong nest site tenacity exhibited by these birds also facilitates long-term studies. Finally, the metabolism of adipose tissue and concomitant weight loss in chicks and adults through the nesting season should result in a dynamic flux of organochlorine contaminants, which are released when lipids are utilized under starvation conditions (Fisher, 1973).

This thesis is divided into two chapters which address two different categories of pollution found in albatrosses of the north Pacific Ocean. The first chapter presents information on the incidence and ingestion of plastic

by Laysan albatross chicks, and compares the masses of plastic in dead and injured chicks. The second chapter presents information on concentrations of chlorinated hydrocarbons, vitamin A and thyroid hormone concentrations in both black-footed and Laysan albatrosses as a function of age and season.



**CHAPTER 1:**  
**PLASTIC INGESTION IN LAYSAN ALBATROSS CHICKS**

## **INTRODUCTION**

The 13 species of albatrosses are members of the class Procellariiformes or "tube noses", family Diomedidae, genus *Diomedea*, and are among the world's largest flying birds. Approximately 2.5 million Laysan albatrosses (*Diomedea immutabilis*) and 200,000 black-footed albatrosses (*Diomedea nigripes*) are found worldwide. Breeding and non-breeding pairs total 380,000 Laysan and 50,000 black-footed albatrosses in the Hawaiian Islands, with 53% and 15% of these two species breeding on Midway Atoll, respectively (McDermond and Morgan, 1993).

Laysan and black-footed albatrosses have different diets and feeding strategies. All albatrosses feed by surface-seizing, characterized by settling on the surface of the water and grabbing prey items. Laysan albatrosses feed mainly on several squid species while black-footed albatrosses feed mainly on flying fish (family Exocoetidae) eggs that stick to floating objects in the water, but also commonly scavenge refuse from ships. Both albatross species are associated with cold (40-65° F) current upwellings of greater salinity, located in the most productive, nutrient- and plankton-rich areas of the ocean (Fisher and Fisher, 1972). Black-footed albatrosses feed mainly

during daylight, while Laysan albatrosses feed during twilight and in darkness. This difference in time of feeding is possibly due to greater amounts of rhodopsin in Laysan retinas than the retinas of black-footed albatrosses (Harrison, 1990).

Because seabirds have a tendency to ingest plastic materials while feeding, plastic pollution on the surface of the Pacific Ocean may have adverse effects on these birds that feed on the surface and thus is a cause for concern. Laysan albatrosses are known to carry assorted ingested plastic materials in their proventriculi, the thin-walled, glandular section of the stomach, and the gizzard, the muscular grinding section. This species is reported to have a greater incidence, a wider variety, and larger volume of ingested plastic than any other seabird (Kenyon and Kridler, 1969; Sileo and Fefer, 1987; Sileo et al., 1989). Fledging chicks normally egest a bolus or casting of squid beaks, fish bones, and other indigestible matter, but large pieces of plastic may complicate or prohibit this process (Petit et al., 1981).

While the populations of both Laysan and black-footed albatrosses are expanding on the islands of Midway Atoll, chicks on Sand, Eastern and Spit Islands of Midway Atoll die by the thousands during the summer months. Some chicks die when they are abandoned by their parents or the parents cannot bring a sufficient amount of food, and others die due to accidents. Some of these chicks die of starvation or dehydration even when they are not abandoned and the adults seem to be providing adequate food. Thus, a

study was conducted to determine if currently ingested quantities of plastic are causing adverse effects on Laysan albatross chicks on Midway Atoll.

The objective of this research was to determine if the population of Laysan albatross chicks found dead have statistically greater weights of plastic in their gastrointestinal tracts than Laysan albatross chicks found accidentally injured, but assumed otherwise healthy. A true cause-effect relationship could not be established between greater plastic loads and increased death rates, but the purpose was to determine if the two populations differ. The following null hypothesis was tested:

$H_0$ : The mass of plastic in the gastrointestinal tracts of Laysan albatross chicks found dead was not significantly different from the mass of plastic found in chicks which had been injured in accidents.

## **METHODS**

Dead or injured Laysan albatross chicks were collected on Sand Island, Midway Atoll in the northcentral Pacific Ocean (28° 11' N, 177° 22' W). Collecting data on dead animals alone did not give an accurate appraisal of whether the plastic loads were involved in the death of the animal. Living, healthy birds were also considered, but killing healthy chicks was not allowed under the permits; therefore, injured chicks were sacrificed. The assumption was made that any plastics that the chicks ingested from parental feeding did not contribute to their injuries resulting from incidents such as vehicular accidents. The birds found dead and injured birds represent a random population within the areas of the island populated by humans, since birds in uninhabited areas were rarely found. Six days per week from 7 June to 8 July 1994, 10 fresh Laysan albatross chick carcasses were randomly collected each day. From these birds, a visual assessment of the glossiness of the eyes determined which carcasses were selected for necropsy. In this manner, only the most recently dead birds were necropsied, and the issues of desiccation and decomposed organs and tissues was minimized. No birds with maggots visible inside or outside were

selected for this study. Once the carcass was opened, a visual inspection of the internal organs was used to determine the decomposition state of the birds that were recovered after death. In several cases, carcasses that passed the first inspection proved too decomposed to assess critically: the proventriculus was thin, peeling and transparent, and the contents were easily visible. In a pilot study on Sand Island (May - June 1993), it was noted that some carcasses had impacted proventriculi in the later stages of decomposition with lacerated linings. Therefore, it could not be concluded that the plastics had caused the tearing.

The injured Laysan albatross chicks were collected 25 May - 21 July 1994. The majority of these birds had compound fractures in their wings and/or legs from being run over by vehicles. Although no regurgitated boluses were observed near the site of injury, the assumption was made that no chicks regurgitated stomach contents as a reaction to their injury. "Droopwing" chicks, thought to be caused by lead poisoning, were not taken as part of the study since they were not considered healthy, although plastic load has not been correlated with lead poisoning or droopwing (Sileo et al., 1990). Injured chicks were processed within 24 h of collection, but it was not possible in many cases to determine exactly how long the bird had been injured. These birds were restrained and about 1 cc of blood was drawn from the brachial vein. A hematocrit and a blood smear were made, and glucose was measured. After sampling blood, the bird was decapitated

and a necropsy performed within 10 min.

The carcasses of chicks found dead and injured chicks were necropsied in the following manner. First, the body was weighed to the nearest 50 g using a spring scale. The percentage of down covering the body was estimated, and length of the first primary feather to the nearest mm was measured to indicate age. Beak length was measured to the nearest mm. A general external survey was also performed to note physical damage or abnormalities.

The carcasses were opened by a ventral incision from cloaca to furcula, with care being taken not to cut into the visceral organs, to expose the entire digestive tract. By visual inspection of the visceral and subcutaneous fat reserves, a fat index was assigned, with 0 = no fat, 1 = low fat, 2 = good fat, and 3 = high fat. The proventriculus and gizzard were removed by cutting the conspicuous junction between the gizzard and small intestine, and the poorly defined area between the esophagus and proventriculus. Since the esophagus gradually increases in width to form the proventriculus with no characteristic demarcation, difficulties arose in making consistently equivalent cuts between carcasses. Therefore, a point was chosen to make each cut just ventral to the caudal tip of the heart, approximately halfway between the taper of the esophagus and the proventriculus. The entire proventriculus and gizzard were removed from the body cavity and the fat was peeled off both organs with gloved fingers. It

was determined during the pilot study that cutting with scissors could damage the proventricular lining, creating punctures or tears. The proventriculus and gizzard were measured together on an Ohaus electronic balance to the nearest 0.1 g. Length, width, and height of the proventriculus were measured to the nearest mm at the midpoints with calipers. The proventriculus and gizzard were opened by making an incision down their entire length with scissors.

Contents of the proventriculus and gizzard were noted and categorized as food, plastic and other man-made materials, and rock (mainly pumice). Plastic and other man-made materials were removed, washed in fresh water, air dried, and weighed to the nearest 0.1 g. Twenty-eight plastic bolus samples from individual birds were saved in chemically clean I-Chem jars and sent back to the Michigan State University Pesticide Research Center for instrumental analyses. The lining of the emptied proventriculus was examined for lacerations, ulcerations, and punctures. Finally, the lining condition was categorized as excellent (well ridged, high surface area), good (somewhat ridged, moderate surface area), fair (poorly ridged) or poor (flat surface). Observations were also recorded on distention of the proventriculus (none, slightly distended, highly distended) (Table 1).

The appropriate number of dead chicks to examine was determined in the following manner. Thirty birds were necropsied, and using the raw data, the variances of the mass of plastic in the proventriculus and gizzard were



calculated. Additional birds were necropsied 5 - 10 at a time, until the estimate of the parametric variance reached a fairly steady line when plotted against the number of necropsies completed.

## **RESULTS**

**Ninety-five Laysan albatross chicks found dead and 39 injured chicks were necropsied and the mass of plastic in their proventriculus and gizzard measured. Of 134 chicks, only one (0.7%) did not contain any plastic. Several observations on categories were recorded, including chips and shards of unidentified plastic, bottle caps, Styrofoam, O-rings, beads, fishing line, rope, buttons, checkers, disposable lighters, toys, PVC pipe, golf tees, magic markers and a light bulb (Figure 1). In addition to plastics, natural objects such as ironwood pine cones, peach pits, walnuts, twigs, wood chips and pumice were found. Indigestible food items such as squid beaks were seen in almost all chicks. Objects that do not float, such as glass shards and copper wire, were also observed in the proventriculus, inferring that chicks and/or adults also pick up detritus from the ground and ingest it.**

**No evidence of punctures or tears in the lining of the proventriculi aside from two separate cases was observed. In both of these chicks found dead, two small, neat punctures were found in the ventral surface of the proventriculus. These two birds also had leaking gall bladders and puncture wounds in the livers and breast tissue, probably caused by a pitchfork,**

which is often used by the Pest Control to pick up carcasses. The lining categories and degree of distention of the proventriculus were categorized in chicks found dead and in injured chicks (Table 1).



Figure 1. Plastic materials in the boluses of Laysan albatross chicks.

Table 1. Proventricular lining categories in necropsied Laysan albatrosses.

	Dead (N = 95)	Dead %	Injured (N = 39)	Injured %
Excellent	11	11.6	26	66.7
Good	26	27.4	13	33.3
Fair	34	35.8	0	0
Poor	24	25.3	0	0
Very Distended	57	60.0	4	10.3
Slightly Distended	22	23.1	8	20.5

Table 2. Variables measured in dead and injured Laysan albatross chicks.

	Mean				Range			STD		P
	Dead	N	Injured	N	Dead	Injured		Dead	Injured	
Body Weight (g)	1938	95	2231	39	1300-2900	1600-2900		277	337	0.0001
% Down on Body	12	95	12	39	0-50	0-60		10	12	
First Primary Length (cm)	27.4	95	29.3	39	11.9-34.6	12.5-36.1		5.6	6.9	
Beak Length (cm)	10.5	95	10.7	39	8.9-11.7	9.5-11.9		0.5	0.5	
Provent. Length (cm)	13.3	93	12.3	38	8.8-21.0	8.5-23.0		2.0	2.8	0.0011
Provent. Width (cm)	5.9	93	3.8	38	2.2-9.4	1.6-7.8		1.8	1.8	0.0001
Provent. Height (cm)	4.7	93	2.9	38	0.8-8.3	0.6-6.9		1.8	1.9	0.0001
Provent. Weight (g)	140.4	93	85.1	38	19.4-393.7	18.5-313.9		81.9	76.0	
Plastic Mass (g)	23.8	95	11.3	39	0.5-136.3	0.0-122.7		26.7	21.3	0.0001
Largest Piece (g)	3.7	95	1.6	39	0.05-34.9	0.0-12.5		6.1	2.7	0.0099
Fat Index	2.0	95	2.5	39	1-3	1-3		0.7	0.6	0.0001
Pumice Weight (g)	30.0	95	10.0	39	0.0-194.8	0.0-121.1		41.6	23.9	0.0001

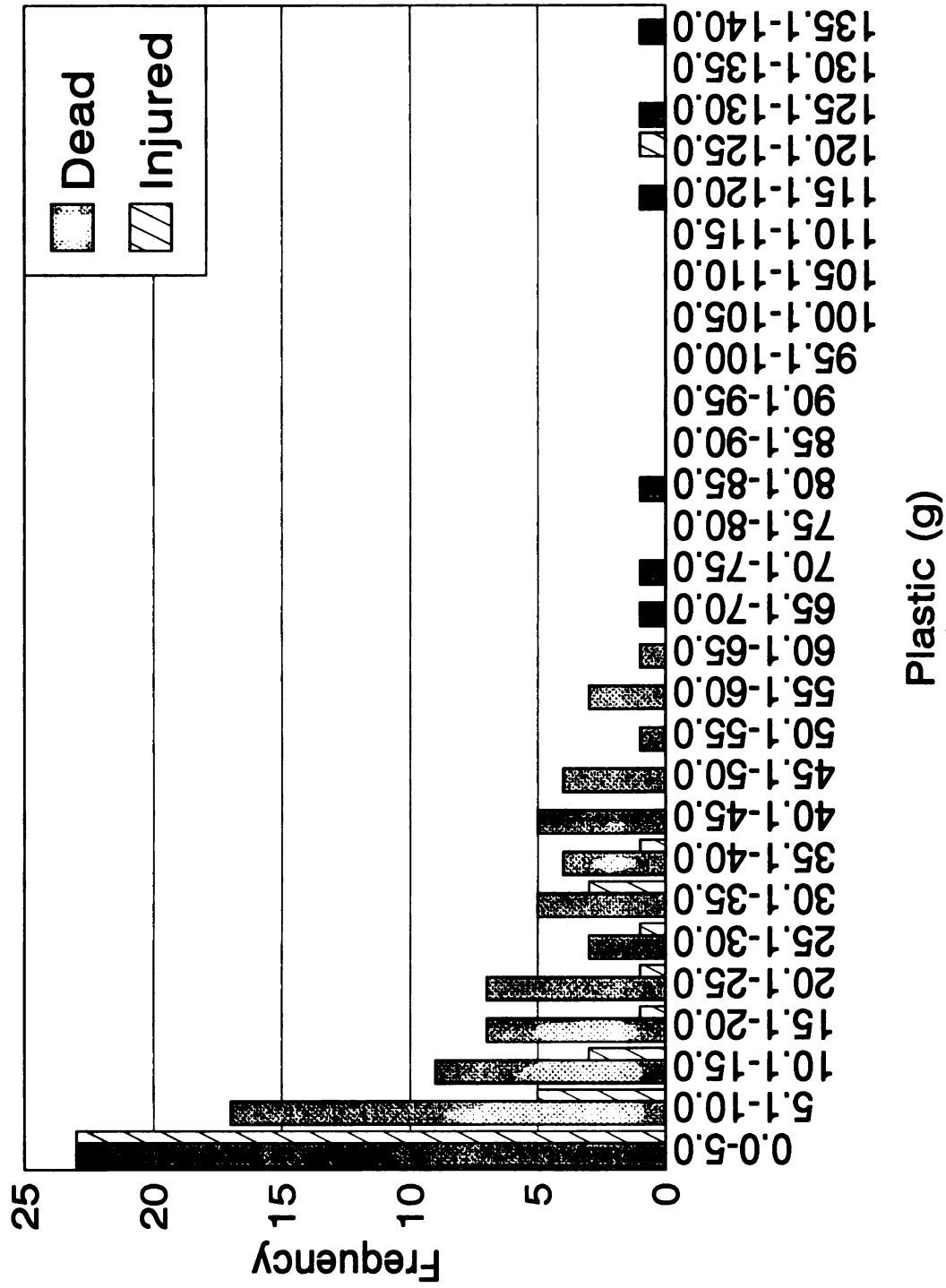


Figure 2. Frequency of occurrence of plastic masses in 95 dead and 39 injured Laysan albatross chicks.

The mean mass of plastic was significantly greater in the proventriculi and gizzards of birds found dead than that found in injured birds ( $p = 0.0001$ ) (Wilcoxon 2-way sample test, NPAR1WAY procedure, SAS/STAT 6.03, SAS Institute, Inc. 1991). Also, the largest pieces of plastic in the dead chicks tended to be heavier than those in injured chicks ( $p = 0.0099$ ). Dead chicks contained an average of 23.8 g of plastic and injured chicks contained an average of 11.3 g of plastic (Table 2). It should be noted that within the sample of injured chicks, only one bolus of plastic greater than 40.0 g was found. If this 122.7 g sample is removed as an outlier, the mean plastic mass drops to 8.4 g. The mass of plastic in neither injured nor dead Laysan albatross chicks followed a normal distribution (Figure 2); therefore, non-parametric statistics were used.

Measures of the health status of the dead or injured chicks at the time of necropsy were compared using a Wilcoxon 2-way sample test. The dead chicks were significantly lighter, with greater proventricular lengths, widths and heights, lesser fat indices, and greater weights of proventricular pumice (Table 2). There was no significant difference in the frequency of males and females between the two groups ( $p = 0.50$ ). Additionally, there were no significant differences in weight ( $p = 0.11$ ), proventricular width ( $p = 0.65$ ), proventricular height ( $p = 0.21$ ), mass of plastic bolus ( $p = 0.83$ ), heaviest piece ( $p = 0.87$ ), or fat index ( $p = 0.36$ ) between male and female chicks. Males had slightly longer proventriculi ( $p = 0.03$ ) and heavier loads



of proventricular pumice ( $p = 0.08$ ) than females.

The weight of the plastic bolus in the proventriculus was correlated with some of the measures of health. Body weight was positively correlated with the fat index, primary feather length and beak length, and negatively correlated with the percent down. The fat index was positively correlated with primary feather length and beak length and negatively correlated with percent down and mass of plastic. Percent down on the body was negatively correlated with primary feather length, beak length and weakly negatively correlated with plastic load (Table 3).

The dimensions of the proventriculus were correlated with the weight of plastic bolus and pumice. Proventricular length was positively correlated with proventricular width, height and weight, largest piece, and weights of plastic and pumice. Proventricular width was positively correlated with proventricular height and weight, largest piece of plastic, plastic and pumice. Proventricular height was positively correlated with proventricular weight, largest piece of plastic, and weights of plastic and pumice. The proventricular weight was positively correlated with the largest piece of plastic, and weights of plastic and pumice. The largest piece of plastic was positively correlated with plastic and pumice weights, and plastic weight was positively correlated with pumice weight (Table 4).

Table 3. Correlations among measurements of health status and with mass of plastic in necropsied Laysan albatross chicks.

	Weight	Fat index	Primary feather length	Beak length	Percent Down	Plastic mass
Weight	$r = 1.00$ $p = 0.00$	$r = 0.55$ $p = 0.0001$	$r = 0.18$ $p = 0.04$	$r = 0.40$ $p = 0.0001$	$r = -0.19$ $p = 0.03$	$r = 0.04$ $p = 0.60$
Fat index		$r = 1.00$ $p = 0.00$	$r = 0.21$ $p = 0.02$	$r = 0.23$ $p = 0.008$	$r = -0.19$ $p = 0.03$	$r = -0.22$ $p = 0.01$
Primary feather length			$r = 1.00$ $p = 0.00$	$r = 0.57$ $p = 0.0001$	$r = -0.75$ $p = 0.0001$	$r = 0.01$ $p = 0.98$
Beak length				$r = 1.00$ $p = 0.00$	$r = -0.41$ $p = 0.0001$	$r = -0.02$ $p = 0.81$
Percent down					$r = 1.00$ $p = 0.00$	$r = -0.15$ $p = 0.09$
Plastic mass						$r = 1.00$ $p = 0.00$

Table 4. Correlations among proventricular dimensions and plastic mass and pumice in Laysan albatross chicks.

	Provent. Length	Provent. Width	Provent. Height	Provent. Weight	Largest Piece	Plastic Mass	Pumice Mass
Provent. Length	$r = 1.00$ $p = 0.00$	$r = 0.53$ $p = 0.0001$	$r = 0.50$ $p = 0.0001$	$r = 0.59$ $p = 0.0001$	$r = 0.31$ $p = 0.0004$	$r = 0.50$ $p = 0.0001$	$r = 0.45$ $p = 0.0001$
Provent. Width		$r = 1.00$ $p = 0.00$	$r = 0.93$ $p = 0.0001$	$r = 0.85$ $p = 0.0001$	$r = 0.48$ $p = 0.0001$	$r = 0.66$ $p = 0.0001$	$r = 0.57$ $p = 0.0001$
Provent. Height			$r = 1.00$ $p = 0.00$	$r = 0.86$ $p = 0.0001$	$r = 0.47$ $p = 0.0001$	$r = 0.66$ $p = 0.0001$	$r = 0.61$ $p = 0.0001$
Provent. Weight				$r = 1.00$ $p = 0.00$	$r = 0.48$ $p = 0.0001$	$r = 0.71$ $p = 0.0001$	$r = 0.70$ $p = 0.0001$
Largest Piece					$r = 1.00$ $p = 0.00$	$r = 0.72$ $p = 0.0001$	$r = 0.21$ $p = 0.01$
Plastic Mass						$r = 1.00$ $p = 0.00$	$r = 0.44$ $p = 0.0001$
Pumice Mass							$r = 1.00$ $p = 0.00$

## **DISCUSSION**

The occurrence of plastic in seabirds and its potential hazard has been documented (Kenyon and Kridler, 1969; Rothstein, 1973; Day, 1980; Petit et al., 1981; Bourne and Imber, 1982; Connors and Smith, 1982; Furness, 1985; van Franeker, 1985; Fry et al., 1987; Ryan, 1987b; van Franeker and Bell, 1988; Sileo et al., 1990 ). The accidental ingestion of plastic refuse is weakly correlated with survival and health. The possible hazards posed by ingestion of plastics include starvation (Dickerman and Goelet, 1987), suppressed appetite and reduced growth in controlled studies with chickens (Ryan, 1988), lower fledging weights in Laysan albatrosses (Sievert and Sileo, 1993), decreased fat deposition (Connors and Smith, 1982), PCB and other organochlorine assimilation (Ryan et al., 1988; Carpenter, 1972; Colton et al., 1974) and obstruction in the gut (Fry et al., 1987). Other research has not demonstrated significant deleterious physiological effects of plastic ingestion. No significant differences were distinguished between white-chinned petrels fed plastic and controls in assimilation efficiency and rate of plastic mass loss, suggesting that plastic does not hamper digestive efficiency (Ryan and Jackson, 1987). In a study on Sand Island, Midway

Atoll in 1987, no Laysan albatross chick deaths, impactions or ulcerations in proventricular linings were attributed to ingested plastic (Sileo et al., 1990). No conclusive evidence of negative effects from plastic consumption by several seabird species was reported in two studies, but suggestions were made that Procellariiformes may be more vulnerable than other seabirds due to their surface-seizing feeding methods, inferior egestion behavior, and smaller gizzards (Day, 1980; Furness, 1985).

Significant correlations among measures of proventricular size, plastic, pumice, body weight, beak, and feather measurements, and fat indices were observed. Some of these correlations were expected, but the significant difference in plastic loads between dead and injured chicks was remarkable. However, the results should be interpreted cautiously, as no cause and effect linkages can be confirmed. While it is possible that the death of healthy chicks may have been the eventual result of ingested plastic, it is feasible that unhealthy chicks eat greater amounts of plastic as a result of their poorer condition. Factors such as satiation and reduced resistance to disease may eventually contribute to death. Without controlled experiments, it is not possible to discern the role of plastic ingestion in the health status of seabirds (Ryan, 1987a).

Satiation, or appetite suppression, can be caused by distention of the proventriculus, gizzard and intestines, dehydration, warm temperatures, and physical activity (Day, 1980). Albatross chicks with greater volumes of

plastic in their proventriculi experience all of these factors in the summer months prior to fledging. In fact, dehydration was considered to be the most common cause of death in Laysan albatross chicks of Midway Atoll in 1987 (Sileo et al., 1990). The displacement of food matter by the plastic bolus may be enough to starve or dehydrate chicks that are already in inferior physical condition. The displacement of food by plastic over time could lead to lighter birds with less fat reserves, which the growing chick relies upon during the periods when the parents are absent just prior to fledging. The uninjured dead chicks in this study were significantly lighter than the injured chicks, caused to some degree by desiccation in the sun. However, only the most recently dead chicks were used and processed in the mornings, and therefore a mean difference in body weight of close to 300 g cannot be explained by desiccation alone. The mean fat indices for dead and injured chicks were 2.0 and 2.5, respectively. This may account for some of the weight difference, since the fat index was positively correlated with body weight ( $p = 0.0001$ ).

Mechanical blockages of the proventriculus or gizzard are both a possible threat from plastic ingestion (Fry et al., 1987). Three (3.2%) of the chicks found dead contained a solid piece of plastic that completely blocked the junction of the esophagus and proventriculus. Even in those cases it could not be determined postmortem if or to what extent the blockage had played a role in the death of the bird. The lifespan of plastic particles in the

digestive system of seabirds has been conservatively estimated to be about two years (Ryan and Jackson, 1987), although radio transmitters inadvertently fed by Laysan albatross adults to chicks stayed in the digestive tract for over 40 d (Petit et al., 1981). Only the smallest plastic pieces ( $< 0.1$  g) can enter the narrow gizzard opening. No plastic has been observed in seabird excretions (Day, 1980; Petit et al., 1981), yet plastic in the intestine was noted in 39% of Laysan albatrosses in 1986 and 1987 (Sileo et al., 1990).

Polychlorinated biphenyls (PCBs) accumulate on the surfaces of plastic in the ocean and have been detected at concentrations as high as 5 parts per million in polystyrene spherules (Carpenter et al., 1972). Additionally, the colorants, softeners, and antioxidants used in the conversion to user-friendly plastic could be harmful toxicants (van Franeker, 1985). It is possible that large masses of plastic ingested by albatrosses may increase their PCB concentrations in addition to PCBs derived from dietary sources (Day, 1980; Bourne and Imber, 1982; van Franeker, 1985). Positive correlations were reported between PCBs and masses of plastic in Great Shearwaters (Ryan et al., 1988). Total PCB concentrations in plasma of albatrosses will be discussed elsewhere in this thesis.

Plastic ingestion in Laysan albatross chicks appears to vary among years, but the incidence, weight and volume is generally increasing. Of 91 Laysan albatrosses studied in 1966 at Pearl and Hermes Reefs in the north-

west Hawaiian Islands, 74% contained plastic material, with 8 pieces being the greatest number found in any individual (Kenyon and Kridler, 1969). A study on Sand Island in 1983 reported a mean of 35.7 g (39.3 cc) plastic in live Laysan albatross chicks and 76.7 g (85.0 cc) plastic in dead chicks (Fry et al., 1987). An average of 46 cc plastic was collected in 1986 and 5 cc in 1987. However, these latter two volumes are likely to be underestimated due to the 50% average yield of the stomach pumping method used (Sileo et al., 1990).

Plastic production continues to increase yearly, and the majority of plastic refuse is generated from three possible sources: municipal solid waste disposal at sea, coastal landfills, and disposal from sea-going vessels (Colton et al., 1974). Currents, winds, and the geographic point sources determine the abundance and distribution of plastic. The warm Kurishio current, which flows north and east around the Japanese Islands, and the North Pacific current, the eastward extension flowing across the northcentral Pacific Ocean, move through the main feeding areas of north Pacific albatrosses and probably transport most plastic material by this route (Fry et al., 1987). Although a study on Midway Atoll in 1979-1980 reported 108 of 109 items found in Laysan albatrosses' egested boluses and carcasses to be of Japanese origin (Petit et al., 1981), the distribution and abundance of plastic has been studied in the Atlantic Ocean as well (Shaw and Mapes, 1979; Day and Shaw, 1987; Colton et al., 1974; Carpenter et al., 1972).



## **CONCLUSIONS**

- 1) Dead Laysan albatross chicks had significantly greater masses of plastic in their proventriculi and gizzards and had significantly lighter body weights than injured chicks.**
- 2) Ingested plastic is probably not a significant direct cause of death in Laysan albatross chicks, but is likely to cause varying degrees of physiological distress.**
- 3) Controlled studies are needed to verify the possible effects of plastics on satiation, PCB uptake, and mechanical blockages. The plastic material used should reflect what is found on the ocean, and seabirds should be included as experimental animals, based on their unique diets and digestive systems.**

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## CHAPTER 2:

### PCBS, DDE AND BIOMARKERS OF HEALTH IN LAYSAN (*Diomedea immutabilis*) AND BLACK-FOOTED (*Diomedea nigripes*) ALBATROSSES OF MIDWAY ATOLL

## **INTRODUCTION**

**As a result of human activities, the global environment has become contaminated with synthetic hydrocarbons in complex mixtures, but the toxic effects resulting from exposure to these polyhalogenated aromatic hydrocarbons (PHAHs) on wildlife have been difficult to establish. The accumulation of PHAHs by birds has been studied in a number of locations. These studies have been primarily in areas such as the North American Great Lakes that are known to be contaminated to a degree significant enough to cause adverse effects (1-2). Accumulation of halogenated hydrocarbons has not been studied extensively in albatrosses of the north Pacific Ocean. This chapter reports the concentrations of PHAHs in albatrosses of the northcentral Pacific Ocean. Although albatross populations appear to be healthy, the health of individual birds exposed to PHAHs was unknown before this study.**

**Two synthetic, widely distributed PHAHs that cause adverse effects in wildlife are PCBs (polychlorinated biphenyls) and DDE [1,1-dichloro-2,2'-bis (p-chlorophenyl)-ethane]. PCBs form a major group of persistent, bioaccumulating, environmental contaminants that have been detected**



ubiquitously in the environment (3). PCBs, used for a variety of purposes such as transformer fluids, coolants and paints, were manufactured as Aroclors. There are 209 theoretically possible PCB congeners, and many can cause several effects in multiple organs and physiological processes. PCBs have been demonstrated to influence fecundity, embryology, metabolism, immunocompetence, endocrine function, and cell growth. PCB toxicity may be assessed by biochemical or physiological biomarkers, which are sensitive measures of impairment and sublethal endpoints from which changes may be measured to make qualitative or quantitative predictions. The chlorinated insecticide DDT [1,1,1-trichloro-2,2'-bis (4-chlorophenyl)-ethane], which is metabolized to the persistent DDE, was banned in the United States by the U. S. Environmental Protection Agency in 1972. DDT is still manufactured and used in some developing countries, and is transported through the atmosphere into distant ecosystems. Egg shell thinning is the most significant bioaccumulative effect of DDE in avian predators (4).

Vitamin A has been found to be a sensitive biomarker of wildlife exposure to certain PHAHs (5-7). One advantage of using vitamin A as a biomarker is that it can be measured repeatedly in plasma by collecting a relatively small amount of blood. Vitamin A is present in three main forms: retinol, found mainly in the serum and liver, retinyl palmitate, the main storage form in the liver, and retinoic acid (Figure 3). Retinoids are

metabolically important in mammals and birds. Retinoids cannot be synthesized *de novo* by vertebrates, thus their precursors must be taken in through the diet (8). Vitamin A is necessary for vision, reproduction, growth and development, nerve and epithelial function, and disease resistance (9). Serum vitamin A is homeostatically controlled, and is released from the liver as retinol only on the demands of the extrahepatic tissue; consequently, an elevated concentration of serum retinol could indicate metabolic alteration and abnormal homeostasis due to toxicity from exposure to certain PHAHs, such as PCBs, polychlorinated dibenzodioxins (PCDDs), polychlorinated dibenzofurans (PCDFs) and compounds which are structurally similar (6, 8, 10). Transthyretin (TTR; prealbumin) is the main transport protein of thyroid hormone, and forms a complex with retinol binding protein (RBP) which transports vitamin A. The TTR-RBP complex is necessary to transport both retinol and thyroid hormone to their target organs. Exposure to certain synthetic halogenated hydrocarbons may alter the usual regulatory response in the liver or a positive feedback signal to release retinol-RBP-TTR. Peripheral tissue effects of PHAH-exposed animals are minimal if vitamin A stores are adequate, but several effects are seen in individuals with depleted stores (8). Vitamin A supplementation is also thought to partially protect against the effects of contaminant exposure (6).

The exact mechanism of altered vitamin A homeostasis in response to PHAHs is not completely understood. It is currently hypothesized that

PHAHs cause mis-regulation of several enzymes in the vitamin A pathway through the Ah receptor (5, 8, 12). One Ah-linked mechanism is the P-450 regulated production of PCB metabolites that decrease plasma retinol concentrations through interference of the vitamin A transport system. Induction of uridine diphosphate glucuronyltransferase (UDPGT) is another enzyme-mediated mechanism that increases plasma retinol concentrations by activating hepatic vitamin A mobilization (13).

Thyroid hormone is a critical hormone in physiological processes, being vital in both fetal development and adult metabolism. The follicular cells of the thyroid gland secrete T4 (thyroxine) and small amounts of T3 (triiodothyronine) ( Figure 4). Once in the target organs, T4 is converted to T3, which is the more biologically active form. Thyroid hormone is necessary for nervous system development, maturation of reproductive and skeletal systems, and regulation of metabolic rates. Thyroid hormone involvement in the normal functioning of organs such as the liver, heart and kidney, as well as its roles in influencing protein, carbohydrate and lipid metabolism, make thyroid hormone a critical endocrine hormone (14).

Thyroid hormone concentrations are also influenced by exposure to some PHAHs. Exposure to chlorinated or brominated biphenyls, PCDDs, or PCDFs produces a number of physiological and metabolic alterations (6-7, 15-17). PCB congener TCB-77 dosed eider ducklings in a semi-field experiment showed decreases in plasma T4 and T3 (18). In field experiments, PCB

concentrations were correlated with thyroid hormone concentrations in common terns (*Sterna hirundo*) (13) and in cormorants (*Phalacrocorax carbo*) (19). Marine environments are iodide-rich and may contribute to healthy thyroid concentrations. The effects from exposure to PHAHs in reference to iodine is exemplified by an experiment where TCB was fed to ring doves (*Streptopelia risoria*) on a reduced iodine diet (15). Large-colloid goiters, lower body temperature, and decreased serum T4 and T3 concentrations were observed.

The mechanisms of thyroid hormone reduction after exposure to PCBs, PBBs (polybrominated biphenyls), PCDDs and PCDFs are similar. Hydroxylated metabolites (Figure 4) of these compounds interfere with thyroxine transport and induce T4 glucuronidation, which solubilizes and removes thyroid hormone from the liver (18).

The hypothesis that PCB concentrations in the plasma of Laysan or black-footed albatrosses were correlated with concentrations of vitamin A and thyroid hormone was tested. Possible relationships between PCBs, the p,p'-DDE isomer and biomarkers were also tested in albatross chicks or known-aged adults, between species, and as a function of various periods within the nesting season.

The following null hypotheses were tested:

H<sub>0</sub>1a: PHAHs such as PCBs and DDE are not detectable in plasma of Laysan albatrosses.

**H<sub>0</sub>1b: PHAHs such as PCBs and DDE are not detectable in plasma of black-footed albatrosses.**

**H<sub>0</sub>2: No relationship exists between PCB concentrations and forms of vitamin A in albatross plasma.**

**H<sub>0</sub>3: No relationship exists between PCB concentrations and forms of thyroid hormone in albatross plasma.**

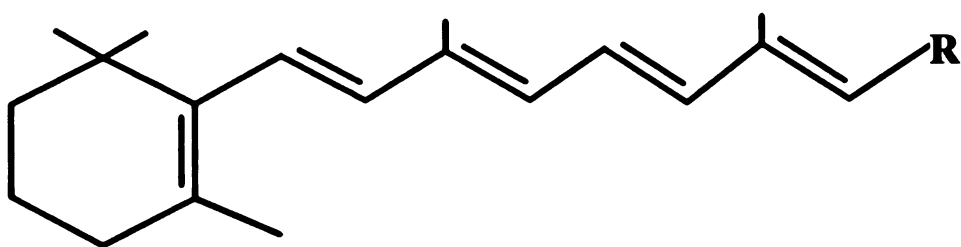


Figure 3. Vitamin A -- Retinol:  $R = \text{CH}_2\text{OH}$ , Retinal:  $R = \text{CHO}$ , Retinoic acid:  $R = \text{COOH}$ , Retinyl palmitate  $R = \text{CH}_3(\text{CH}_2)_{14}\text{COOH}$ .

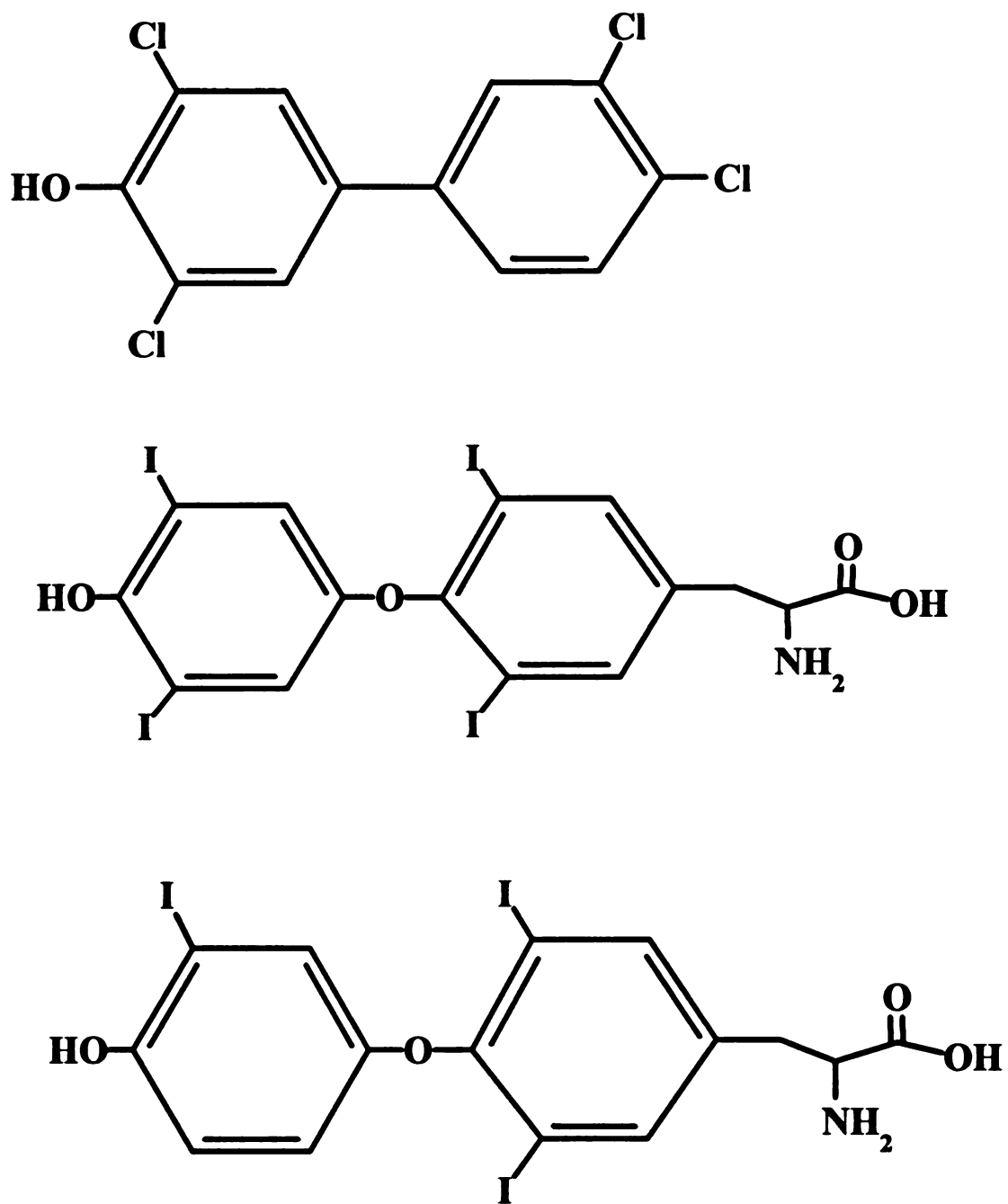


Figure 4. A hydroxylated PCB congener (4-hydroxy-3,3',4',5-tetrachlorobiphenyl), thyroxine (T4) and triiodothyronine (T3).

## **MATERIALS AND METHODS**

Tissue samples were collected from known-aged adults and chicks of both Laysan and black-footed albatrosses on Sand Island, Midway Atoll in the northcentral Pacific Ocean (28° 11' N, 177° 22' W) during three time periods over the nesting season. The two fall sampling periods, November through December, 1992 and 1993, occurred during courtship, nest building and egg laying. In the winter sampling period, January through February 1993, the parent albatrosses were incubating eggs or newly hatched chicks. The spring sampling period, April through May 1993, was during the chick rearing phase. Blood was collected from both chicks and adults during the spring.

Seven ml of blood were drawn from the brachial vein of each bird using 22-gauge needles and vacutainers containing 0.015 ml EDTA anticoagulant. Care was taken not to expose the blood to sunlight, which can affect vitamin A. Blood was centrifuged for 10 min at 3000 rpm within 3 h of sampling. Approximately 4 to 5 ml of plasma were obtained from whole blood and divided in the following manner. Plasma was pipetted into three 0.6 ml polypropylene centrifuge tubes for analysis of vitamin A,



thyroid hormone, and an archived sample. Approximately 2.2 ml remaining plasma were used for instrumental quantification of PHAHs. Samples were stored at -20° C until analyses could be performed.

Vitamin A analyses were performed at the Animal Health Diagnostic Laboratory at Michigan State University. High performance liquid chromatography (HPLC) techniques were used to identify the different forms of retinoids as total vitamin A, retinyl palmitate and retinol (20).

The thyroid hormone analyses consisted of two separate procedures for T3 and T4. The Animal Health Diagnostic Laboratory at Michigan State University analyzed the plasma samples for free T3, T3, free T4 and T4. The quantitative determination of T4 used a Corning MAGIC radioimmunoassay (Corning Magnetic Immunochemistries), a competitive assay using <sup>125</sup> I competing with sample plasma for rabbit anti-T4 antibodies. A similar in-house procedure was implemented for T3 determination.

The methods for extracting and analyzing PCBs and chlorinated insecticides in the plasma have been described previously (21). One ml of albatross plasma was denatured with methanol, extracted with hexane-diethyl ether and cleaned up on Florisil<sup>®</sup> and Silica gel columns. The fractions were then analyzed by gas chromatography and electron capture detection (GC-ECD) at Michigan State University Pesticide Research Center Aquatic Toxicology Laboratory. The COMSTAR linear regression program

was used to calculate total PCB and DDE concentrations.

Statistical methods were performed by SAS (NPAR1WAY, regression, ranks and Tukey's procedures, SAS/STAT 6.03, SAS Institute, Inc. 1991).

In the five albatrosses sampled twice in different sample periods, the mean values of biomarkers and PCBs of each pair were used in ANOVA calculations.

## **RESULTS AND DISCUSSION**

### ***Biomarkers***

Concentrations of vitamin A and thyroid hormone in plasma were compared between Laysan and black-footed albatrosses (Table 9). There were no differences in concentrations of either vitamin A as total vitamin A, retinol or retinyl palmitate or in thyroid hormone as total and free T4, and total and free T3 between the two species, which indicates the physiological similarity between Laysan and black-footed albatrosses. Adult age and sample periods were not related to the biomarkers, except for total T4, which differed with age of adult albatrosses. There were no correlations between concentrations of PCBs and vitamin A or thyroid hormone in plasma (Table 10). The lack of relationships could be explained by the low toxicological influence and narrow range of PCB concentrations measured, coupled with adequate vitamin A and iodine stores.

## ***PHAHs***

PCB and DDE were detected in all plasma samples, with a range of 10 to 1037 ppb and 66 to 702 ppb, respectively (Table 5). Statistically weak differences in total PCB concentrations were detected between all (adults and chicks) Laysan and black-footed albatrosses. However, when categorized into chicks or adults, the difference between Laysan and black-footed albatross chicks was not significant, while the difference in concentrations of total PCBs and DDE between adults of the two species became strongly significant. The concentrations of total PCBs and DDE were more than twice as great in plasma of adult black-footed albatrosses as they were in that of adult Laysan albatrosses (Table 5). This difference between species could be in part due to diet and feeding strategy differences between the two species, which occupy different ecological niches. Black-footed albatrosses commonly scavenge human garbage, a habit the Laysan albatross rarely adopts. Additionally, the fish eggs common in the black-footed albatross diet also represent residue sources from a higher trophic level than taken in the diet of Laysan albatrosses. PCB concentrations in plasma of Laysan albatross chicks were significantly less than in Laysan albatross adults. Similarly, PCB concentrations in black-footed albatross chicks were significantly less than those in black-footed albatross adults. Significant differences in DDE concentrations were

observed between adult Laysan and black-footed albatrosses. Total concentrations of both PCBs and DDE compared with age of adults were not statistically significant, indicating the PHAH accumulation is not age-related in adults (Table 6).

Concentrations of ranked total PCBs and DDE were compared among nesting periods in both species of adult albatrosses using a Tukey's test of honestly significant differences. Among adults, significant differences in PCB concentrations were observed among certain sampling periods (Table 7a; 7b) but not in DDE concentrations (Table 8a; 8b). Total concentrations of PCBs were greatest in the spring of 1993, which were significantly greater than other sample periods, while the fall 1992 sample period had significantly less total PCBs than the other periods. This may be due to mobilization of fat reserves at an increased rate due to extended periods of egg incubation without feeding and to foraging great distances for food for the chicks (22).

Total concentrations of PCBs and DDE were measured in one Laysan and four black-footed albatrosses sampled during two different nesting periods. Although the sample size is too small to assess with great statistical power, PCBs and DDE appear relatively comparable among sampling periods within individual birds, with the exception of total PCB concentrations in one black-foot, 887-13544 (Table 9). It is possible that the former albatrosses were non-breeding, and therefore were not mobilizing

contaminants in adipose tissue on a seasonal basis.

Concentrations of PHAHs in the adipose tissue were compared to those in Laysan and black-footed albatrosses accidentally killed in December 1969, even though protocols used at that time showed interference and poor separation (23). The concentrations of PCBs and DDE in visceral fat of Laysan albatrosses were 22.3 ppm PCBs and 13.7 ppm DDE wet weight, respectively in 1969, while black-footed albatrosses contained a mean of 15.3 ppm PCBs and 15.3 ppm DDE, respectively, in visceral fat. The birds were killed in the early part of the breeding cycle when fat reserves were greatest; therefore, sampling later in the season could have shown greater PCB concentrations in the blood due to metabolized fat reserves.

Concentrations of total PCBs in adipose tissue of Laysan albatrosses in 1993 were 2.1 to 2.8  $\mu\text{g/g}$  wet weight (ppm) (24), which is an order of magnitude less than those observed in 1969 (23).

### ***Comparisons to Great Lakes fish-eating birds***

Concentrations of total PCBs and p,p'-DDE in albatross plasma and eggs were compared to concentrations of plasma and eggs in Great Lakes fish-eating birds. Mean concentrations of total PCB in the plasma of Great Lakes-influenced bald eagle chicks, defined as living within 8 km of Great

Lakes shorelines (25), were less than the mean total PCB concentrations in blood of adult black-footed albatrosses, but greater than that of adult Laysan albatrosses. Bald eagle chicks from interior regions in the Great Lakes basin had similar mean total concentrations of PCBs in plasma as both Laysan and black-footed albatross chicks (Figure 5). Mean total PCB concentrations in Laysan and black-footed albatross eggs were less than PCB concentrations of eggs from Great Lakes-influenced bald eagles and bald eagles from the interior of Michigan (26), Caspian terns and double-crested cormorants (*Phalacrocorax auritus*) (27) (Figure 6). Mean DDE concentrations in plasma of adult Laysan and black-footed albatrosses were greater than both Great Lakes-influenced and interior bald eagle chicks (25). Adult Caspian terns (21) had mean concentrations of DDE in the plasma that were greater than DDE concentrations in Laysan but less than black-footed albatrosses (Figure 7). Mean concentrations of DDE in eggs of both species of albatrosses were less than those found in Great Lakes-influenced and interior bald eagles (26), Caspian terns and cormorants (27) (Figure 8).

### ***Hazard assessments***

Assessments of the hazard presented by current concentrations of total PCBs and DDE in Laysan and black-footed albatrosses were conducted

by calculating the hazard quotient (HQ), which is the quotient of the average current concentrations of PCB or DDE divided by the NOAEL (No Observable Adverse Effect Level) (Table 11). NOAEL numbers were generated from field studies using herring gulls (*Larus argentatus*), bald eagles, Caspian terns and double-crested cormorants, and laboratory studies with white leghorn chickens (*Gallus domesticus*). A HQ of 1.0 would indicate that the NOAEL had been achieved. The ratio between the NOAEL and Lowest Observed Adverse Effect Level (LOAEL) is approximately 10 (2). Assuming that both albatross species have similar sensitivities to these PHAHs, and that their sensitivities are similar to other wild fish-eating birds, the ratios indicate that concentrations of both PCBs and DDE in Laysan albatrosses are at the lower end of the range of possible effects, with current concentrations of neither total PCBs nor DDE expected to cause adverse effects in the Laysan albatross population. The black-footed albatross hazard ratio is close to 10 for current total PCB concentrations, predicting that adverse population effects would begin to be observed. The hazard quotients of DDE concentrations were much less than the threshold for adverse population level effects.

Based on these hazard assessments, predictions were made on effects seen in the populations of the two species. Historical productivity data are scarce, but indicate that the egg crushing rates and hatching rates from 1962 to 1964 (28) were comparable to both Laysan and black-footed



albatross species from 1993 and 1994 (Table 12). However, the recent rate of cracked eggs in black-footed albatrosses is over 5%, twice the rate of cracked eggs found in Laysan albatrosses (J.P. Ludwig, personal communication). This was not reflected in the hazard assessment using DDE concentrations, which indicated a ratio close to the NOAEL, where no adverse effects, specifically egg shell thinning, would be predicted. In fact, no decreases in eggshell weight or increase in egg breakage were reported between 1910 and 1969 in either albatross species, when DDE concentrations were much greater than present times (23).

**Table 5. Age, biomarkers, DDE and total PCB concentrations in the plasma of two albatross species.**

<b>Variable</b>	<b>LAAL Mean</b>	<b>N</b>	<b>BFAL Mean</b>	<b>N</b>	<b>Range</b>	<b>STD</b>
Adult age (years)	12.6	34	19.1	22	0-38	11.8
Retinyl palmitate (ng/ml)	415.3	17	507.8	11	155-1639	319.6
Retinol (ng/ml)	1386.0	17	1462.3	11	617-2196	340.4
Total vitamin A (ng/ml)	1801.3	17	1969.6	11	946-2665	416.5
Total T4 (nmol/L)	51.1	17	43.4	11	21-84	15.6
Total T3 (nmol/L)	2.8	17	3.2	11	0.8-6.6	1.2
Free T4 (nmol/L)	12.6	10	8.8	5	5-30	6.6
Free T3 (nmol/L)	9.5	10	9.7	5	3.9-20.1	4.6
PCBs (ppb)	100.2	35	231.2	31	10-1037	202.6
DDE (ppb)	100.7	9	296.9	15	66-702	163.1

**Table 6. Analysis of variance of variables PCB and DDE classified by species, age, and sampling period.**

	PCBs		DDE	
	N	P	N	P
LAAL (chicks v. adults)	35	0.0011	-	-
BFAL (chicks v. adults)	31	0.0001	-	-
Adults (LAAL v. BFAL)	78	0.009	24	0.0016
Chicks (LAAL v. BFAL)	25	0.23	-	-
LAAL v. BFAL	66	0.08	-	-
Adult age (yrs)	39	0.94	24	0.65
Sample period	38	0.004	22	0.24

LAAL = Laysan albatross

BFAL = black-footed albatross

**Table 7a. Analysis of variance of total PCB concentrations among sampling periods. \***

	N	Mean	Variance	STD	CV
Fall 1992	19	148	35,790	189	128
Winter 1993	9	413	37,858	194	47
Spring 1993	3	454	25,954	161	35
Fall 1993	6	321	25,543	160	50

$\alpha = 0.05$ ,  $df = 33$ ,  $P = 0.003$

**Table 7b. Tukey's test of honestly significant differences in ranked total PCB concentrations among sample periods. \***

Tukey Grouping		Mean	N	Sample Period
B B  B B  B	A A A A	454	3	Spring 1993
		412	9	Winter 1993
	A	321	6	Fall 1993
		148	19	Fall 1992

\* Means not sharing the same letter are significantly different

**Table 8a. Analysis of variance of DDE concentrations among sampling periods.**

	N	Mean	Variance	STD	CV
Fall 1992	4	341	24,552	157	46
Winter 1993	9	258	41,792	204	79
Spring 1993	3	238	18,430	136	57
Fall 1993	6	127	2930	54	42

$\alpha = 0.05$ ,  $df = 18$ ,  $P = 0.24$

**Table 8b. Tukey's test of honestly significant differences in ranked total DDE concentrations among sample periods.\***

Tukey Grouping	Mean	N	Sample Period
A A A A A A A	341	4	Fall 1992
	258	9	Winter 1993
	238	3	Spring 1993
	127	6	Fall 1993

\* Means not sharing the same letter are significantly different

Table 9. Biomarkers, PCBs and DDE in five adult albatrosses sampled twice.

Species	BFAL 1* 887-29394		BFAL 2 887-13544		BFAL 3 767-92977		BFAL 4 887-13810		LAAL 1* 1117-82529	
	1**	4**	1	4	1	4	2**	4	2	4
Sample period	20	21	25	26	26	27	26	26	28	28
Age (yrs)	433	323	359	1831	709	1347	-	448	-	703
Retinyl palmitate (ng/ml)	887	1525	1399	1469	1135	1673	-	1018	-	1606
Retinol (ng/ml)	1320	1848	1758	3300	1844	3020	-	1466	-	2309
Total vit A (ng/ml)	38	39	53	35	50	44	-	45	-	55
Total T4 (nmol/L)	2.6	2.4	2.5	2.2	5.2	1.9	-	2.8	-	2.3
Total T3 (nmol/L)	6	-	11	-	7	-	-	-	-	-
Free T4 (nmol/L)	5.5	-	4.4	-	13.6	-	-	-	-	-
Free T3 (nmol/L)	223	206	258	1037	412	791	482	226	290	278
PCBs (ppb)	257	155	266	54	106	345	68	54	36	39
DDE (ppb)										

\*\*Sample period 1 = November-December, 1992

Sample period 2 = January-February, 1993

Sample period 4 = November-December, 1993

**Table 10. Regression of total PCB concentrations with biomarkers and DDE concentrations.**

	<b>PCBs (ppb)</b>	
	<b>N</b>	<b>R<sup>2</sup></b>
<b>Retinyl palmitate (ng/ml)</b>	<b>28</b>	<b>0.28</b>
<b>Retinol (ng/ml)</b>	<b>28</b>	<b>-0.01</b>
<b>Total vitamin A (ng/ml)</b>	<b>28</b>	<b>0.04</b>
<b>Total T4 (nmol/L)</b>	<b>28</b>	<b>-0.03</b>
<b>Total T3 (nmol/L)</b>	<b>28</b>	<b>0.05</b>
<b>Free T4 (nmol/L)</b>	<b>15</b>	<b>-0.01</b>
<b>Free T3 (nmol/L)</b>	<b>15</b>	<b>-0.08</b>
<b>DDE (ppb)</b>	<b>24</b>	<b>0.53</b>

**Figure 5. Total PCB concentrations in Laysan and black-footed albatross adults and chicks compared to bald eagle chicks in the interior and Great Lakes-influenced areas of the Great Lakes. Data from Bowerman et al., 1990.**



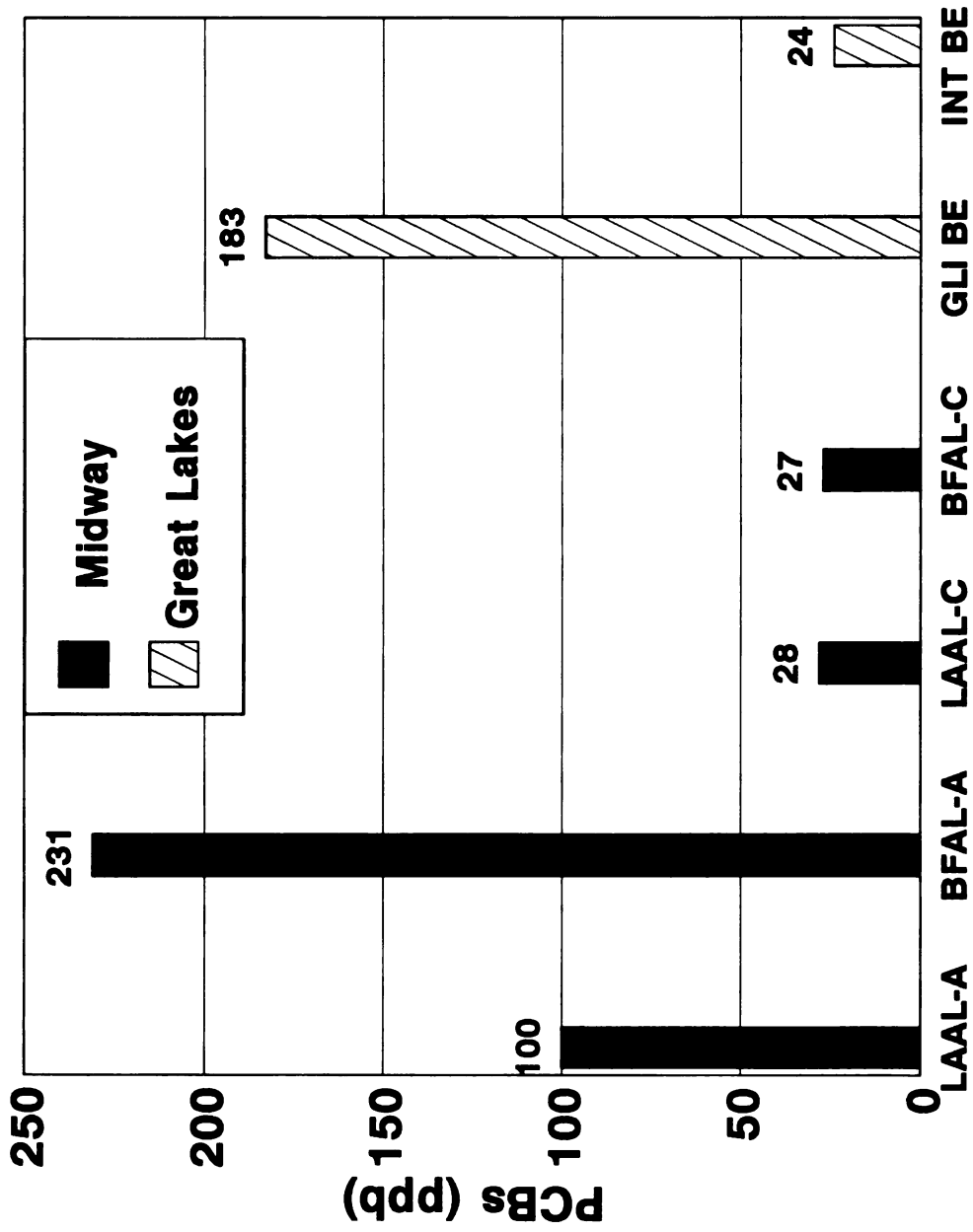


Figure 5.

**Figure 6. DDE in the plasma of Laysan and black-footed albatross adults compared to Caspian tern adults and bald eagle chicks in interior and Great Lakes-influenced areas of the Great Lakes. Data from Mora et al., 1993 and Bowerman et al., 1990.**

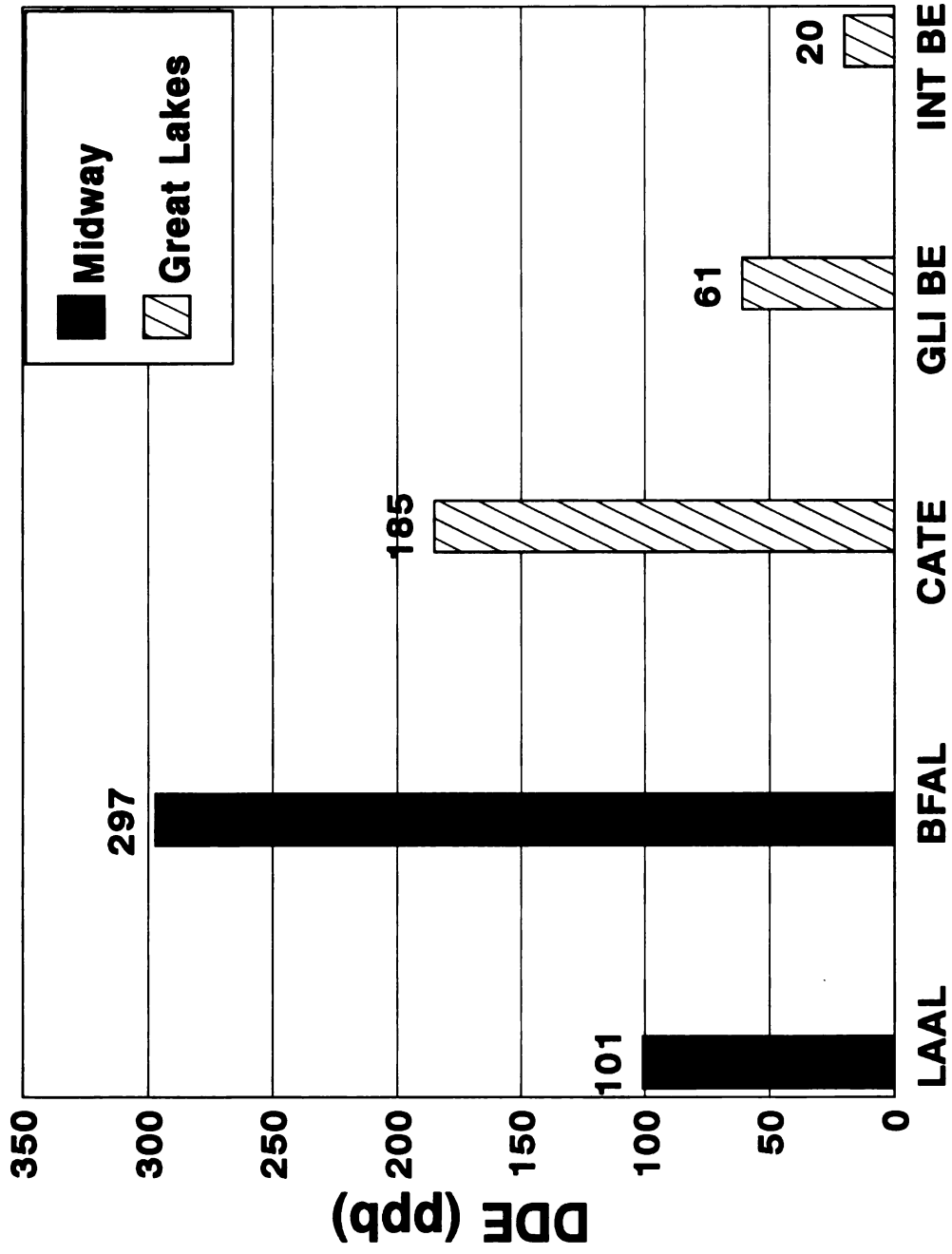


Figure 6.

**Figure 7. Total PCB concentrations in the eggs of Laysan and black-footed albatrosses compared to eggs of Great Lakes-influenced and interior bald eagles, Caspian terns and double-crested cormorants of the Great Lakes. Data from Bowerman, 1991 and Yamashita, 1992.**

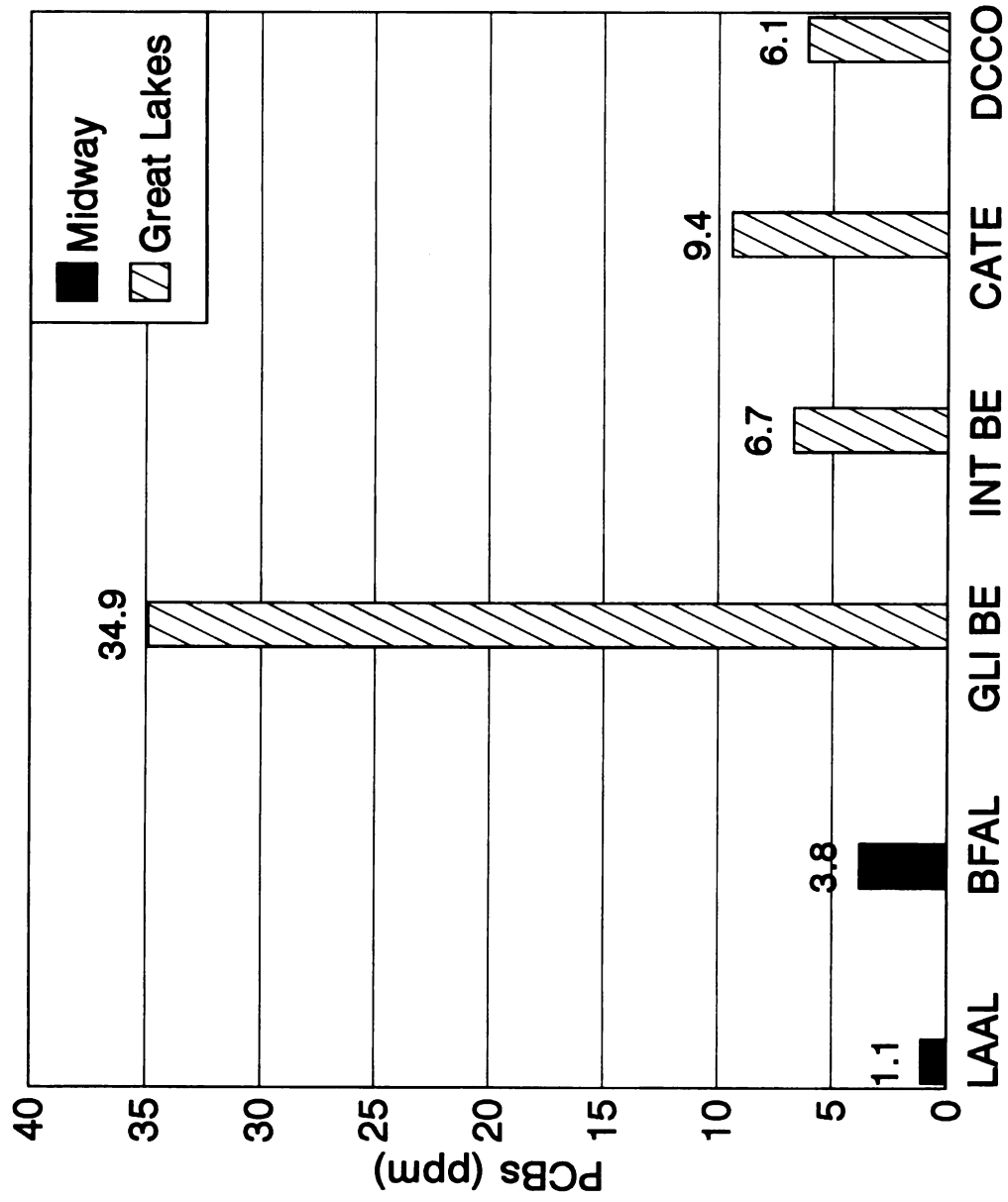


Figure 7.

**Figure 8.** DDE concentrations in the eggs of Laysan and black-footed albatrosses compared to eggs of Great Lakes-influenced and interior bald eagles, Caspian terns and double-crested cormorants. Data from Bowerman, 1993; Mora et al., 1993 and Yamashita et al., 1992.

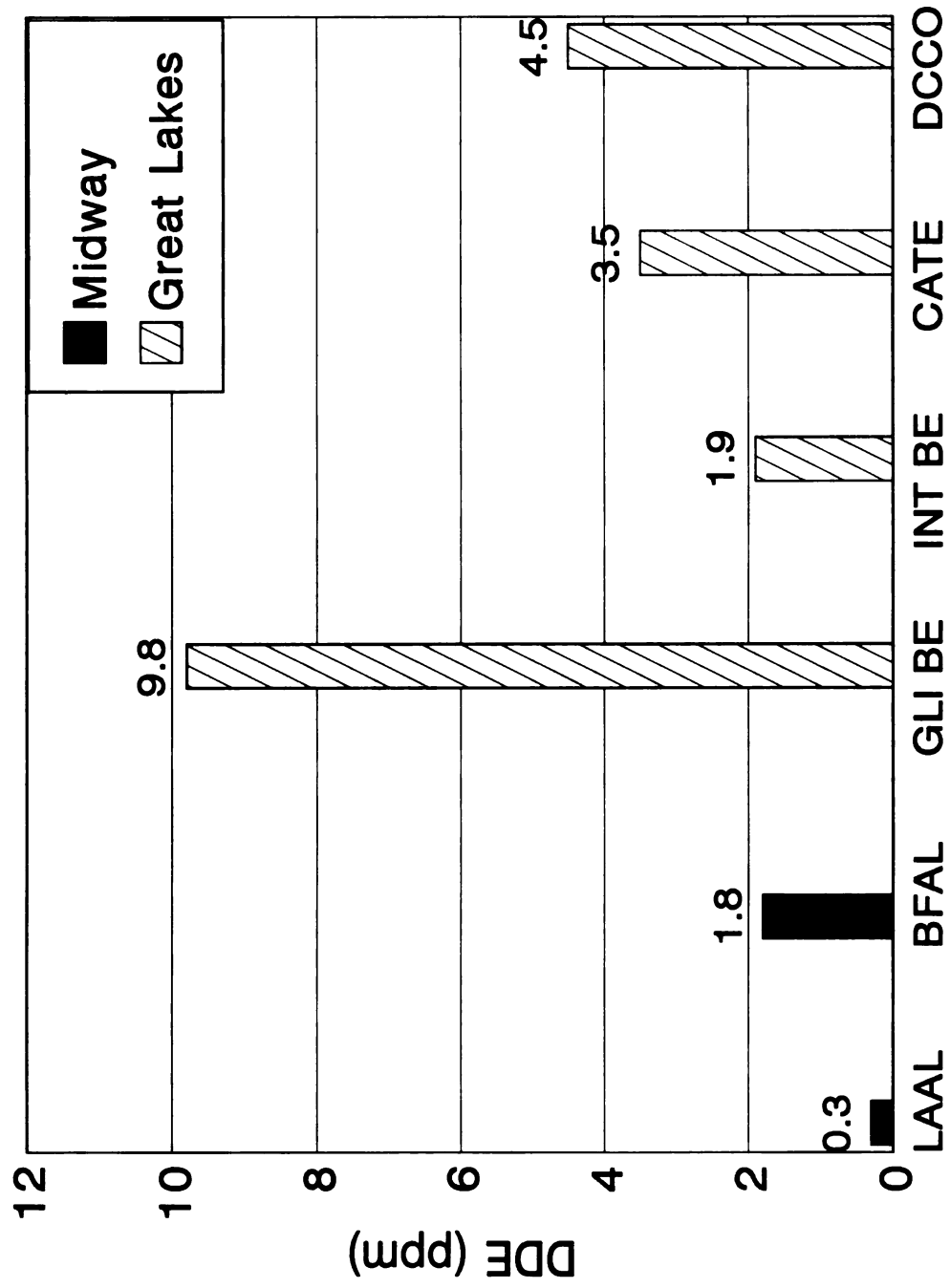


Figure 8.

Table 11. Hazard quotients for Laysan and black-footed albatrosses.

	PCBs	DDE
LAAL	2.6	0.09
BFAL	9.7	0.51

Hazard quotient = egg concentration of PCB or DDE/NOAEL  
 LOAEL = NOAEL x 10

Table 12. Mean egg crushing, egg cracking, and hatching rates for Laysan and black-footed albatrosses on Midway Atoll.

	% Eggs Crushed	% Eggs Cracked	% Eggs Hatched
BFAL 1993-1994	2.62	5.34	71.53
LAAL 1993-1994	2.21	2.84	74.45
LAAL 1962-1964	1-3	-	71-73

Data from Ludwig et al. (unpublished data) and Fisher (1971a).



## **CONCLUSIONS**

- 1. No significant differences were detected in the vitamin A or thyroid hormone concentrations, indicating physiological similarities between species. There were no correlations between concentrations of PCBs and vitamin A or thyroid hormone in plasma.**
- 2. PCBs were detected in all albatrosses, with significant differences seen between species, between chicks and adults, and among certain sample periods within the nesting season.**
- 3. DDE was detected in all adult albatrosses, with significant differences seen between species.**
- 4. The hazard assessment of black-footed albatrosses using PCB concentrations approached the LOAEL, which predicts that adverse effects will begin to be seen at the population level.**

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## LIST OF REFERENCES

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