Observations on Metal Concentrations in Three Species of Shark (*Deania calcea*, *Centroscymnus crepidater*, and *Centroscymnus owstoni*) from Southeastern Australian Waters

N. J. Turoczy,[†] L. J. B. Laurenson,[†] G. Allinson,*[,†] M. Nishikawa,[‡] D. F. Lambert,[†] C. Smith,[†] J. P. E. Cottier,[†] S. B. Irvine,[†] and Frank Stagnitti[†]

School of Ecology and Environment, Deakin University, Warrnambool, Victoria 3280, Australia, and Regional Environment Division, National Institute for Environmental Studies, 16-2 Onogawa, Tsukuba, Ibaraki 305, Japan

Deep sea dog sharks (*Deania calcea*, *Centroscymnus crepidater*, and *Centroscymnus owstonii*) were captured from the waters off western Victoria, Australia, in April and May 1998. The elements As, Ca, Cd, Cu, Fe, Hg, K, Mg, Mn, Na, Sr, and Zn were detected in the muscle tissue. The concentrations of Al, Ba, Be, Cr, Mo, Ni, Pb, and V were below the detection limits of the instrumental techniques employed (ICP-ES and GF-AAS). However, significant concentration differences between species were detected for As, Cd, Hg, and Zn. *C. owstoni* contained the highest concentrations of each of these elements. The concentrations of Cu, Pb, and Zn in each species were below the maximum levels permitted in food by the Australian Food Standards Code. The maximum permitted concentrations of As and Hg were exceeded in all species, and weekly consumption of 400 g of any of these sharks would result in intake of more than the FAO's provisional tolerable weekly intake. Although *C. crepidator* and *D. calcea* have strong relationships between length and mercury concentration, *C. owstoni* does not. Placing *C. crepidator* and *C. owstoni* in the same genus, therefore, is worth re-examination as the mechanisms for controlling mercury in *C. owstoni* appear to differ from that used by both *C. crepidator* and *D. calcea*.

Keywords: *Shark; metals; bioconcentration; biomagnification; Australia*

INTRODUCTION

The accumulation of mercury by sharks in the Northern Hemisphere is well-known (Fairey et al., 1997; Hornung et al., 1994; U.S. Food and Drug Administration, 1993; Vas and Gordon, 1993, 1988; Marcovecchio et al., 1991; Vas, 1987; Powell et al., 1981; Ramamurthy, 1979; Eustace, 1974; Le Blanc and Jackson, 1973). The same is true for Australasian sharks. For instance, total mercury concentrations exceed 3.0 mg kg^{-1} of wet weight in *Carcharhinus amblyrhynchoides*, *Carcharhinus melanopterus*, *Carcharhinus amboienensis*, and *Sphyrna mokarran* in Northern Australian waters (Lyle, 1984, 1986). To the east, in tropical Queensland waters, *Brachaelurus waddi*, *Carcharhinus amblyrhynchoides*, *Carcharhinus coatesi*, *Carcharhinus spallanzani*, *Carcharias arenarius*, and *Physodon taylori* had maximum mercury concentrations of 1.18, 0.42, 0.71, 2.14, 3.41, and 1.48 mg kg^{-1}, respectively (Denton and Breck, 1981) and in cooler southwestern Australian waters, mean mercury concentrations in the larger specimens of *Carcharhinus dorsalis*, *Orectolobus* sp., *Sphyrnia lewini*, and *Odentaspis taurus* were approximately 1.2, 1.4, 1.3, and 2.7 mg kg^{-1}, respectively (Caputi et al., 1979).

Although a number of studies have investigated relationships between length and mercury concentration in elasmobranchs, the bioaccumulation and bioconcentration of other priority elements in sharks is less well understood. This is especially true for elasmobranchs in Australian waters. There exists the possibility that long-lived sharks such as the golden (or long-nosed) velvet dogfish, *Centroscymnus crepidater*, Owston's dogfish, *Centroscymnus owstonii*, and the brier (or shovelnose) shark, *Deania calcea*, may accumulate high concentrations of other toxicologically significant metals in their flesh, such as aluminum, arsenic, cadmium, lead, or zinc. These three species of deep sea dog shark contribute to the trawl component of the Australian South East Fishery (Figure 1). Although these sharks have only recently been legally fished for human consumption (Office of Legislative Drafting 1998), fillets of the firm, white flesh of these, and other sharks, are often consumed in Victoria, deep-fried as "flake" in popular take-out meals, such as "fish and chips", because of their flavor and lack of bones (Yeow et al., 1986), and may, therefore, pose a human health threat for those who consume large amounts of their flesh.

In this study, we investigated the concentrations of 20 metals in fillets of muscle tissue of *D. calcea*, *C. crepidater*, and *C. owstoni* captured in the Southern Ocean at depths of ~600 m on the shelf slope south of Victoria. Inductively coupled plasma emission spectrophotometry (ICP-ES) was used to assess the concentrations of Al, As, Ba, Be, Ca, Cr, Cu, Fe, K, Mg, Mn, Mo, Na, Ni, Pb, Sr, V, and Zn in the shark flesh after nitric/perchloric acid digestion. However, when used as a screening tool in which excessively high concentrations can be quickly recognized, ICP-ES is not sufficiently

* Corresponding author (telephone +61 3 55 633316; fax +61 3 55 633462; e-mail graemea@deakin.edu.au).
[†] Deakin University.
[‡] National Institute for Environmental Studies.

10.1021/jf000285z CCC: $19.00
Published on Web 08/16/2000

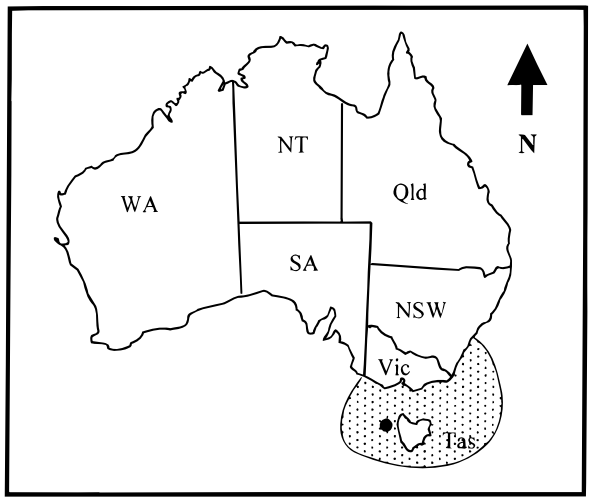

Figure 1. Location of southeastern Australian fishery (shaded area) and approximate location of trawls from which samples were collected (◆).

sensitive to detect cadmium and lead in the digests at toxicologically significant concentrations. Concentrations of these metals were determined using graphite furnace atomic absorption spectrophotometry (GF-AAS). Open vessel nitric acid/perchloric acid digestions are not suitable for the detection of mercury in fish. The concentrations of mercury in the shark flesh were determined using cold vapor atomic absorption spectrophotometry after closed-vessel nitric acid digestion.

Herein we report the results of our study. The data are examined to determine if there are significant differences in metal concentrations between the three species of shark and to determine if metal concentrations vary with the size or sex of sharks. When relevant, the concentrations measured are compared to the maximum values permitted by the Australian Food Standards Code and the human health implications for heavy consumers of this food discussed.

MATERIALS AND METHODS

Reagents. Deionized water having a resistivity of at least 18 MΩ cm^{-1} was produced by passing singly distilled water through a Milli-Q water purification system. Nitric acid (of both AnalaR and Aristar grade), hydrochloric acid (of AnalaR grade), and perchloric acid (of AnalaR grade) and Extran 300 detergent were obtained from BDH Chemicals. All dissection equipment, glassware, and plasticware used for the processing or storage of samples were soaked prior to use for at least 24 h in a 5% solution of Extran 300, followed by three rinses with deionized water. Glassware and plasticware were further soaked for at least 1 week in 10% nitric acid (of AnalaR grade) solution. The equipment was finally rinsed at least three times with deionized water.

Samples. The dog sharks were captured off the coast of Victoria by a commercial otter board trawler during April and May 1998 and were frozen until processing. The sharks were then thawed and identified to species level prior to dissection. A total of 39 sharks were investigated: 18 *D. calcea* (6 male, 12 female), 10 *C. crepidator* (6 male, 4 female), and 11 *C. owstoni* (5 male, 6 female). The sharks were dissected in the following manner: one set of dissecting equipment was used to cut a flap of skin (approximately 3 × 3 cm) immediately below the first dorsal fin above the lateral line. The flap of skin was folded posteriorly before a second set of dissecting equipment was used to remove ~30 g of muscle and transfer the tissue to a polyethylene bottle. In this way, contamination of the sample by extraneous material on the skin was minimized. The muscle samples were then freeze-dried and ground to a fine powder using a plastic spatula.

Sample Preparation. For metals other than mercury, tissue samples were digested using the nitric/perchloric digestion procedure described by Ward et al. (1980), although, as an additional step, the digests were filtered through 0.2 μm cellulose acetate disposable syringe filters (Micro Filtration Systems) to protect the ICP-ES equipment from damage from particulate matter.

Analysis. Digests were analyzed using an IRIS ICP-AES (Thermo Jarrell Ash). The following analytical wavelengths were monitored: Al, 396.1 nm; As, 189.0 nm; Ba, 233.5 nm; Be, 313.1 nm; Ca, 317.9 nm; Cr, 205.5 nm; Cu, 324.7 nm; Fe, 259.9 nm; K, 766.4 nm; Mg, 285.2 nm; Mn, 260.5 nm; Mo, 202.0 nm; Na, 588.9 nm; Ni, 231.6 nm; Sr, 421.5 nm; V, 311.0 nm; Zn, 206.2 nm. The same digests were also analyzed for cadmium and lead using the procedure described by van Loon (1985), in which $NH_4H_2PO_4$ is used as a matrix modifier. Cadium and lead concentrations were determined using a Hitachi 7000 polarized Zeeman atomic absorption spectrophotometer. The wavelengths monitored were Cd, 228.8 nm for Cd and 283.3 nm for Pb.

Mercury concentrations were determined using the closed-vessel digestion and cold vapor generation techniques described by the AOAC (1990) using a GBC 901 hydride generator and a GBC 901 atomic absorption spectrophotometer.

Data Analysis. Concentrations of elements among species and between sexes were compared using multiple analysis of covariance (MANCOVA) (Statview 5.0 for Macintosh). Preliminary analysis having indicated that variances among samples were heterogeneous, data were thus log transformed which, in most cases, resulted in homogeneous variances. Post hoc testing of means was carried out using a Games−Howell (1976) a posteriori test; this test has the added advantage that it does not assume homogeneity among variances. Principle component analysis (PCA) (Statistica 4.1 for Macintosh) was used to estimate the relationships among species and between sexes. For consistency with the MANCOVA analysis, PCA analysis was also carried out with log transformed data.

RESULTS AND DISCUSSION

Dogfish reference material (DORM 2) obtained from the National Research Council Canada (NRCC) was used to assess the performance of the analytical methods. Quantitative agreement with the respective certified values was achieved for As, Cd, Cu, Hg, and Zn [recoveries in the range of 85−110%, Table 1; detection limits indicated are based on mean + 10 standard deviations for the digested blanks (APHA, 1995)]. The elements Al, Cr, Fe, Mn, and Ni were recovered semiquantitatively, with recoveries in the range of 70−85%. The comparatively low recoveries for these elements, in contrast to the quantitative recoveries reported by Ward et al. (1980), may be related to the filtration step introduced into the procedure to protect the instrumentation. The performance of the analytical methods for those elements not certified in DORM 2, or for which the certified concentrations were below the detection limits achieved in this study, were studied by making standard additions of set point standards [Analytical Products Group (trace metals, minerals, and nutrients)] to samples of DORM 2. Quantitative recoveries were obtained for Be, Ca, K, Mg, Mo, Na, Pb, Sr, and V, and semiquantitative recovery was obtained for Ba. Reagent blanks were also processed, with nitric acid quantities adjusted to maintain similar treatment volumes in samples and blanks.

On the basis of the analysis of DORM 2, quantitative results were expected for 13 elements. Ten of these (As, Ca, Cd, Cu, Hg, K, Mg, Na, Sr, and Zn) were detected

Table 1. Summary of Analytical Parameters for Metals under Investigation, Including Limits of Determination (MDL), Recovery after Digestion of Certified Reference Material (Number of Samples, $n = 7$ for All Metals except Mercury, for Which $n = 3$), and Repeatability

element	MDL (mg kg^{-1})	recovery (%)	precision (CV %)[a]	element	MDL (mg kg^{-1})	recovery (%)	precision (CV %)
Al	2.0	71	18	K	40	90	1
As	4.0	101	4	Mg	10	77	3
Ba	2.0	82	NA[b]	Mn	0.1	80	8
Be	0.01	90	NA	Mo	2.0	96	NA
Ca	7.0	110	3	Na	30	98	2
Cd	0.08	110	6	Ni	0.6	70	13
Cr	0.7	72	14	Pb	0.1	92	NA
Cu	0.2	88	9	Sr	0.02	89	3
Fe	7.0	79	8	V	5.0	92	NA
Hg	1.0	109	17	Zn	0.4	92	6

[a] CV % = coefficient of variation. [b] NA = not applicable (recoveries quoted represent the average of two determinations of spiked blanks).

Table 2. Metal Concentrations in Muscle Tissue from Three Species of Dog Shark[a]

	metal concn (mg kg^{-1} of dry wt)					
element	*D. calcea* ($n = 18$)		*C. crepidator* ($n = 10$)		*C. owstonii* ($n = 11$)	
Al	<2		<2		<2	
As	60 ± 8	(26000)	68 ± 13	(30000)	114 ± 20	(50000)
Ba	<2		<2		<2	
Be	<1		<1		<1	
Ca	360 ± 30	(0.9)	420 ± 80	(1.0)	420 ± 80	(1.0)
Cd	0.38 ± 0.017	(760)	0.011 ± 0.007	(220)	0.053 ± 0.04	(1100)
Cr	<7		<7		<7	
Cu	0.23 ± 0.04	(80)	0.26 ± 0.04	(90)	0.29 ± 0.04	(100)
Fe	3.8 ± 1.6	(1300)	3.2 ± 1.8	(1100)	2.4 ± 0.08	(800)
Hg	7.2 ± 2.3	(140000)	4.3 ± 2.4	(90000)	11.9 ± 1.1	(240000)
K	16500 ± 900	(40)	16500 ± 700	(40)	16100 ± 1000	(39)
Mg	1200 ± 40	(0.9)	1160 ± 60	(0.9)	1230 ± 80	(0.9)
Mn	0.47 ± 0.03	(200)	1.0 ± 0.3	(500)	0.74 ± 0.12	(400)
Mo	<2		<2		<2	
Na	5100 ± 600	(0.5)	4300 ± 600	(0.4)	5900 ± 1500	(0.5)
Ni	<0.6		<0.6		<0.6	
Pb	<0.1		<0.1		<0.1	
Sr	1.0 ± 0.2	(0.1)	1.0 ± 0.3	(0.1)	1.6 ± 0.8	(0.2)
V	<5		<5		<5	
Zn	7.8 ± 0.4	(1600)	8.8 ± 0.4	(1800)	9.6 ± 0.5	(1900)

[a] Range gives 95 % confidence interval. Numbers in parentheses represent bioconcentration factors relative to average concentration in seawater.

in the dog shark muscle. Only these elements are considered in the statistical analysis now described. Metal concentrations in the shark tissue are summarized in Table 2. The concentration factor of each element relative to its average concentration in seawater (Riley and Chester, 1971) is also stated.

The major cations in seawater (Na, Mg, Ca, and K) were generally present in the shark flesh at concentrations similar to their concentrations in seawater, although potassium is somewhat enriched. Some other elements were concentrated by factors of many thousands relative to seawater; for instance, cadmium, copper, iron, and zinc were generally concentrated by factors of ~100−1000 compared to seawater. Mercury was concentrated by a factor of ~100000. For some elements, for example, phosphorus, bioconcentration arises because of the physiological role played by the element. Elements without a known biological role may be accumulated if they behave in a chemically analogous way to an essential element, for example, arsenic in the case of phosphorus (Phillips, 1990).

The elemental concentrations of cadmium, copper, iron, manganese, and zinc measured in this study are generally of the same order of magnitude as those reported elsewhere for shark (Table 3). In contrast, the concentrations of nickel and lead are lower than reported for other sharks. The flesh of each of the three species analyzed in this study was found to comply with the Australian Food Standards Code maximum concentrations for copper, lead, and zinc of less than 10.0, 1.0, and 150 mg kg^{-1} of wet weight, respectively (Australia New Zealand Food Authority, 1998). Zinc was the only element that complied with the Australian Food Standards Code and for which statistically significant different concentrations ($p = 0.05$) were found between species. In this case, *C. owstoni* had statistically significantly higher zinc concentrations than *C. crepidater*, which in turn had significantly higher zinc concentrations than *D. calcea* (Table 4).

The concentrations of arsenic in these deep sea dog sharks were higher than have been reported in other sharks (Table 3). Again, *C. owstoni* had statistically significantly higher arsenic concentrations than *C. crepidater* and *D. calcea* (Table 4). Although in general seafood contains <5 mg kg^{-1} of wet weight of arsenic, some bottom-feeding fish, crustaceans, and shellfish have been reported as having arsenic concentrations of several tens of milligrams per kilogram of wet weight (IPCS, 1981). The arsenic concentrations seen in these sharks, therefore, may be a reflection of their feeding habits and diet. For many coastal dwelling Australians, the dietary intake of arsenic will depend on the amount of seafood in their diet. The Australian Food Standards Code specifies that the maximum concentration of inorganic arsenic permitted in fish is 1.0 mg kg^{-1} of wet weight (Australia New Zealand Food Authority, 1998).

Table 3. Comparison of Mean Trace Metal Concentrations Measured in Dog Shark (This Study) with Mean Concentrations Reported in Sharks in Other Studies

		metal concn (mg kg^{-1} of wet wt)									
species	location	As	Cd	Cu	Fe	Hg	Mn	Ni	Pb	Zn	ref
Alopias vulpinus	S.E. Australia					0.64					Walker, 1988
Asymbolus vincenti	S.E. Australia					0.64					Walker, 1988
Brachaelurus waddi	Queensland					1.18					Denton and Breck, 1981
Callorrhyncus milli	S.E. Australia					0.26					Walker, 1988
Carcharodon carcharius	S.E. Australia					1.29					Walker, 1988
Carcharhinus	Queensland					0.42					Denton and Breck, 1981
amblyrhynchoides	N.T.					1.90					Lyle, 1986
	N.T.					2.07					Lyle, 1984
Carcharhinus amboinensiss	N.T.					1.51					Lyle, 1986
Carcharias arenarius	Queensland					3.41					Denton and Breck, 1981
Carcharhinus brachyurus	S.E. Australia										Walker, 1988
Carcharhinus brevipinna	N.T.					0.14					Lyle, 1986
	S.E. Australia					1.48					Walker, 1988
Carcharhinus cautus	N.T.					1.14					Lyle, 1986
Carcharhinus coatesi	Queensland					0.71					Denton and Breck, 1981
Carcharhinus dorsalis	W.A.					0.80					Caputi et al., 1979
Carcharhinus dussumieri	N.T.					0.35					Lyle, 1986
Carcharhinus fitzroyensis	N.T.					0.90					Lyle, 1986
	N.T.					0.90					Lyle, 1984
Carcharhinus limbatus	N.T.					1.50					Lyle, 1986
	N.T.					1.05					Lyle, 1984
	PNG	1.8	0.01	0.31						0.2	Powell et al., 1981
Carcharhinus maclotis	N.T.					0.25					Lyle, 1986
Carcharhinus melanopterus	N.T.					1.59					Lyle, 1986
Carcharhinus sorrah	N.T.					0.33					Lyle, 1986
	N.T.					0.44					Lyle, 1984
Carcharhinus spallanzani	N.T.					2.14					Denton and Breck, 1981
Carcharodon carcharias	S.E. Australia					1.29					Walker, 1988
Centrophorus uyato	S.E. Australia					2.50					Walker, 1988
Centroscyllium fabricii	N.E. Atlantic			0.07			0.24	0.43			Vas and Gordon, 1993
Centroscymnus coelolepis	N.E. Atlantic			0.19			0.03	0.98			Vas and Gordon, 1993
	Tasmania					3.08					Davenport, 1995
Centroscymnus crepidator	N.E. Atlantic			1.91			0.08	<0.02			Vas and Gordon, 1993
	Tasmania					0.81					Davenport, 1995
	Tasmania	17	0.003	0.065	0.8	1.08	0.25	<0.01	<0.03	2.2	present study
Centroscymnus owstonii	Tasmania					2.33					Davenport, 1995
	Tasmania	29	0.013	0.0073	0.6	2.98	0.19	<0.01	<0.03	2.4	present study
Cephaloscyllium laticeps	Tasmania					10.5					Thompson, 1985
	S.E. Australia					1.28					Walker, 1988
Dalatias licha	N.E. Atlantic			3.80			0.13	<0.02			Vas and Gordon, 1993
Deania calcea	N.E. Atlantic			<0.02			<0.02	2.77			Vas and Gordon, 1993
	Tasmania					1.21					Davenport, 1995
	Tasmania	15	0.01	0.058	1.0	1.8	0.12	<0.1	<0.03	2.0	present study
Diplodus sargus	Mediterranean					1.02					Hornung et al., 1994
Etmopterus princeps	N.E. Atlantic			0.13			0.10	0.37			Vas and Gordon, 1993
Etmopterus spinax	N.E. Atlantic			5.31			0.44	1.54			Vas and Gordon, 1993
			0.25				0.86		<0.10		Vas, 1991
Furgaleus macki	S.E. Australia					0.70					Walker, 1988
Galeocerdo cuvieri	N.T.					0.77					Lyle, 1986
Galeorhinus australis	Tasmania		<0.05	0.5			<0.5			5.0	Eustace, 1974
	S.E. Australia	23	0.06	0.6	0.6	2.2				4.2	Glover, 1979
	S. E. Australia					0.90					Walker, 1976
Galeorhinus galeus	U.K.		<0.02	0.44	3.14		0.03	1.79	0.16	2.12	Vas, 1991
	U.K.		0.01	0.50	3.55		0.04	2.01	0.14		Vas, 1987
	S.E. Australia					0.64					Walker, 1988
Galeus melastomas	N.E. Atlantic			0.22			<0.02	1.41			Vas and Gordon, 1993
	N.E. Atlantic			0.22	1.99		<0.02	1.41			Vas and Gordon, 1988
	N.E. Atlantic		0.08	0.22	1.99		<0.02	1.41	0.16		Vas, 1991
Galeus murinus	N.E. Atlantic			<0.02			<0.02	<0.02			Vas and Gordon, 1993
Halaculurus bivius	Argentina		0.15			2.20				26.4	Marcovecchio et al., 1991
Heptranchias perlo	S.E. Australia					1.33					Walker, 1988
Heterodontus portjacksoni	S.E. Australia					0.41					Walker, 1988
Hexanchus griseus	Canada (Pacific)	4.6									Le Blanc and Jackson, 1973
Isurus oxyrinchus	New Zealand	4.4		0.35	12.0	1.58	0.05			4.0	Vlieg et al., 1993
	S.E. Australia					3.20					Walker, 1988
Lamna nasus	New Zealand	2.3		0.40	18.0	0.68	0.06			4.0	Vlieg et al., 1993
	U.K.		0.79		0.60		0.93			7.21	Vas, 1991
Lithognathus mormyrus	Mediterranean					0.29					Hornung et al., 1994
Mustelus antarcticus	Tasmania		<0.05	<0.25			<0.5			4.6	Eustace, 1974
	S.E. Australia					0.41					Walker, 1988
	S.E. Australia					0.37					Walker, 1976
Mustelis henlei	San Francisco	<0.7	<2.3								Fairey et al., 1997
Mustelus schmitti	Argentina		0.14			0.77				16.9	Marcovecchio et al., 1991

Table 3 (Continued)

species	location	metal concn (mg kg^{-1} of wet wt)									ref
		As	Cd	Cu	Fe	Hg	Mn	Ni	Pb	Zn	
Negaprion acutidenss	N.T.					0.50					Lyle, 1986
Notorynchus cepedianus	S.E. Australia					1.39					Walker, 1988
Notorhyncus spp.	Argentina		0.18			2.99				18.3	Marcovecchio et al., 1991
Odentaspis taurus						2.0					Caputi et al., 1979
Orectolobus sp.						1.25					Caputi et al., 1979
Parascyllium ferrugeneum	Tasmania					1.59					Thompson, 1985
	S.E. Australia					0.54					Walker, 1988
Parascyllium variolatum	S.E. Australia					0.25					Walker, 1988
Physodon taylori	Queensland					1.48					Denton and Breck, 1981
Prionace glauca	Tasmania					0.57					Davenport, 1995
	U.K.		0.45	0.24	6.34		1.55	2.58	<0.02		Vas, 1991
	S.E. Autralia					0.41					Walker, 1988
Pristiophorus cirratus	Tasmania					0.35					Thompson, 1985
	S.E. Australia					0.52					Walker, 1988
Pristiophorus nudipinnus	Tasmania					0.61					Thompson, 1985
	S.E. Australia					0.47					Walker, 1988
Rhizoprionodon acutus	PNG	3.4	0.01	0.35					0.1	3.3	Powell et al., 1981
	N.T.					1.01					Lyle, 1986
Rhizoprionodon taylori	N.T.					1.01					Lyle, 1986
Scyliorhinus canicula	U.K.		0.78	0.39	7.89		2.07	1.70	1.88		Vas, 1987
	U.K.		1.08	2.42	6.79		<0.02	<0.02	0.35		Vas, 1991
Scyliorhinus stellaris	U.K.		<0.02	0.56	1.54		<0.02	<0.02	<0.02		Vas, 1991
Scymnorhinus licha	N.E. Atlantic		<0.02				0.24		<0.05		Vas, 1991
Sphyrnia blochii	N.T.					0.58					Lyle, 1986
	N.T.					1.36					Lyle, 1984
Sphyrna lewini	N.T.					0.56					Lyle, 1986
	N.T.					1.15					Lyle, 1984
	PNG	2.6	0.01	0.32					0.2	3.7	Powell et al., 1981
	W.A.					0.8					Caputi et al., 1979
Sphyrnia mokarran	N.T.					1.52					Lyle, 1986
	N.T.					1.59					Lyle, 1984
Sphyryna zygaena	S.E. Australia					0.89					Walker, 1988
Squalus acanthias	S.E. Australia					0.86					Walker, 1988
Squalus blainvellei	S.E. Australia					2.44					Walker, 1988
Squalus megalops	S.E. Australia					1.70					Walker, 1988
Squalis sp.	Tasmania		<0.05	0.7			<0.5			4.9	Eustace, 1974
Squalis mitsukurii	Japan		0.007	0.3	3.3	1.12				2.5	Taguchi et al., 1979
Squatina australis	S.E. Australia					0.13					Walker, 1988
Triakis semifasciata	San Francisco	<0.7	<2.3								Fairey et al., 1997
Urolophus paucimaculatuss	S.E. Australia					0.20					Walker, 1988
Urolophus testaceaus	S. E. Australia					0.48					Walker, 1988

[a] PNG, Papua New Guinea; U.K., United Kingdom. Queensland, W.A. (Western Australia), N.T. (Northern Territory), and Tasmania are states of Australia.

Table 4. Comparison of Metal Concentrations in Deep Sea Dog Shark Using Games−Howell Test for Significance

element	comparison of concentrations	
	$p = 0.05$	$p = 0.10$
As	*C. owstoni* > *C. crepidater* = *D. calcea*	*C. owstoni* > *C. crepidater* = *D. calcea*
Cd	*C. owstoni* = *C. crepidater* = *D. calcea*	*C. owstoni* > *C. crepidater* = *D. calcea*
Hg	*C. owstoni* > *C. crepidater* > *D. calcea*	*C. owstoni* > *C. crepidater* = *D. calcea*
Zn	*C. owstoni* > *C. crepidater* > *D. calcea*	*C. owstoni* > *C. crepidater* > *D. calcea*

Arsenic found in marine organisms is found in the form of fat or water-soluble organoarsenic compounds, such as arsenobetaine (IPCS, 1981), and these are considered to be several orders of magnitude less toxic than inorganic arsenic. The maximum concentration of inorganic arsenic would still be exceeded by each of the three species examined in this study if, taking the example of the U.S. Food and Drug Administration (1993), 90% of the arsenic found in these dogfish is considered to be present in organic forms. A weekly consumption of 200 g of *C. crepidator* (17 mg kg^{-1} of wet weight of As; Table 3) would result in intake of several milligrams of arsenic or several times the FAO's provisional weekly tolerable intake. Seafood arsenic has been found to be readily absorbed by the human gastrointestinal tract. Fortunately, it is also rapidly excreted, and there appears to be no conclusive evidence for long-term toxicity or carcinogenic effects from ingestion of seafood arsenic.

A number of other coastal species of shark form an important component of the southeastern Australian commercial shark catch, and several of these, too, have been reported as having high mercury concentrations (Table 3). For instance, a single sample of *Cephaloscyllium laticeps* caught on the continental shelf of Tasmania was found to contain 10.5 mg kg^{-1} Hg. Samples of *Parascyllium ferrugenium* contained 1.59 mg kg^{-1} Hg (Thompson, 1985). Walker (1976, 1977, 1988) reported mercury concentrations in 23 species of elasmobranchs taken from depths of <75 m on the southeastern Australian continental shelf, and 2 species of shark and 2 species of dogfish taken from the continental slope. Maximum mercury concentrations observed in *Squalus megalops* and *Isurus oxyrinchus* in these two regions

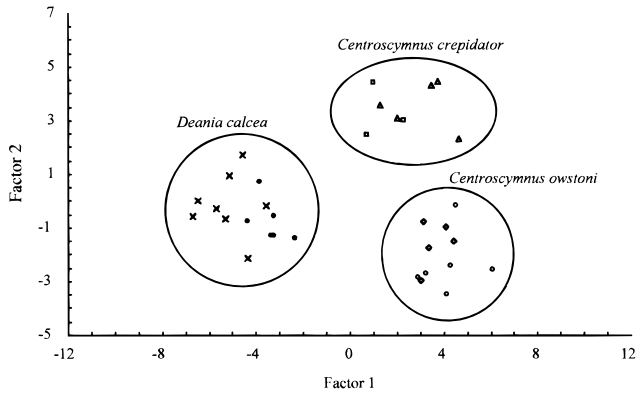

Figure 2. PCA: *D. calcea,* ● = male, × = female; *C. crepidator,* □ = male, △ = female; *C. owstoni,* ◇ = male, ○ = female.

were 1.70 and 3.20 mg kg^{-1}, respectively. The mercury concentrations reported here for *C. owstoni*, *C. crepidater*, and *D. calcea* are similar to those reported by Davenport (1995), with bioconcentration factors similar to those reported for marine fish (IPCS, 1991). Sharks are considered to be long-lived with comparatively slow rates of growth (Lyle, 1984), and their longevity and slow growth rates in conjunction with their high trophic position contribute to their accumulation of mercury. Environmental mercury is often considered to be an indication of anthropogenic pollution. In this case, however, the main source of the mercury is natural, such as degassing of the earth's crust (IPCS, 1991) and natural geological cycling of Hg from offshore cinnabar deposits (Walker, 1988), because the region in which our dogfish were caught is several hundred kilometers from the nearest large city, Melbourne. Indeed, Walker (1988) observed that marine organisms in Port Philip Bay (upon which Melbourne is located) have lower mercury contents than organisms sampled outside the bay in the Southern Ocean. The concentration of mercury in each of the three species was higher than the 1.0 mg kg^{-1} of wet weight permitted by the Australian Food Standards Code (Australia New Zealand Food Authority, 1998). In most foodstuffs, mercury is usually present in the inorganic form. The exception is fish and fish products. Fish retain mercury in their tissues principally as methylmercury, even though most environmental exposure is to inorganic mercury. Methylmercury in fish arises from bacterial methylation of inorganic mercury in bacteria associated with the gills and the gut and is an order of magnitude more toxic than inorganic mercury (IPCS, 1991). In every species studied to date, the nervous system is the primary site of toxicological action of methylmercury. The level of mercury in fish can markedly affect the intake of mercury and, hence, methylmercury. A weekly consumption of 200 g of *C. crepidator* (1 mg kg^{-1} of wet weight Hg; Table 2) would result in intake of 200 μg of mercury, predominantly methylmercury, or 60% of the FAO's provisional tolerable weekly intake. Populations having high fish consumption may attain blood methylmercury levels associated with low neurological damage to adults (IPCS, 1991).

PCA grouping of samples on species and sex and using concentrations of elements as factors shows substantial and clear separation on the basis of species (Figure 2). These results support the data from MANCOVA indicating differences in the concentrations of some of the elements (As, Ca, Cd, Cu, Na, Sr, and Zn) analyzed among species with Games−Howell a posteriori tests identifying significant differences in the concentrations of As, Cd, Hg, and Zn (Table 5). Although the PCA suggests that there are some differences in the relative concentrations of elements between sexes in these species, particularly in *D. calcea* and to a lesser extent in *C. owstoni* (Figure 2), MANCOVA found significant differences in the concentrations of elements between sexes only for copper and mercury. Sex was therefore removed from the analysis (Table 5). Importantly, if both copper and mercury are removed from the PCA, the degree of separation among species and between sexes is not greatly altered (data not presented). The sharks were collected from deep water trawls (at ~600 m), the exact locations of which could not be obtained from fishermen. Trawls in these waters may cover up to nine nautical miles and may cover quite different habitats. Although little is known of the biology of these species, the conjectured causes of their different element signatures, and perhaps even differences in elemental signatures between sexes of some species, are differences in age, maximum size, habitat, and diet. As a general rule in elasmobranchs, older animals are larger animals. In addition, females tend grow to a larger maximum size because greater size equates to greater reproductive potential. Increased size provides increased opportunity to prey on larger and perhaps older prey species with higher metal concentrations and, hence, to ingest greater quantities of metals. The lower temperatures of the deep waters which *C. owstoni*, *C. crepidater*, and *D. calcea* inhabit may also decrease metabolic rates, retard aging, and provide longer periods for bioaccumulation. The MANCOVA also found significant covariance between mercury and length. Two of the three species of dog fish (*C. crepidator* and *D. calcea*) have strong relationships between length and mercury concentration (Figures 4 and 5), suggesting not only that this element is biomagnified but also that rates of bioaccumulation and compartmentalization of mercury by these species exceed those of metabolism and excretion. The length−weight relationship for mercury is consistent with similar findings by Davenport (1995)

Table 5. Summary of MANOVA Testing Incorporating Species, Sex, Length, and Weight

		element									
parameter	MVS	As	Ca	Cd	Cu	Hg	K	Mg	Na	Sr	Zn
species	***	***		**		***			**		***
length	***					***					
weight	***					***					

[a] MVS, multivariate significance; *, significant at $p = 0.1$; **, significant at $p = 0.05$; ***, significant at $p = 0.00$.

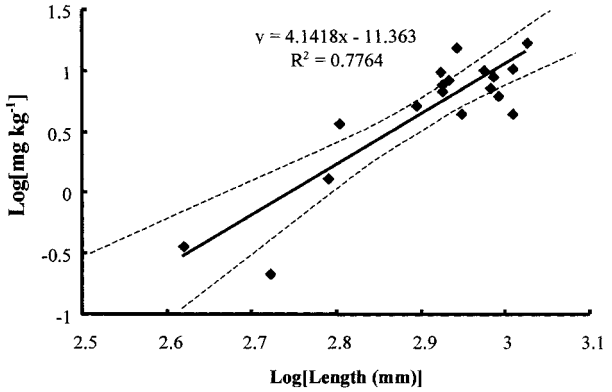

Figure 3. Concentration of mercury in muscle of *C. owstoni*.

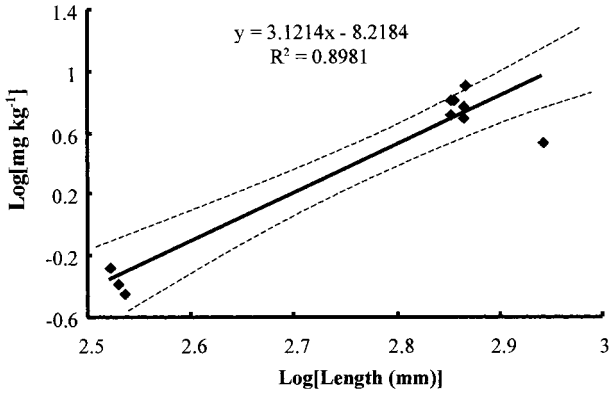

Figure 4. Concentration of mercury in muscle of *C. crepidator*.

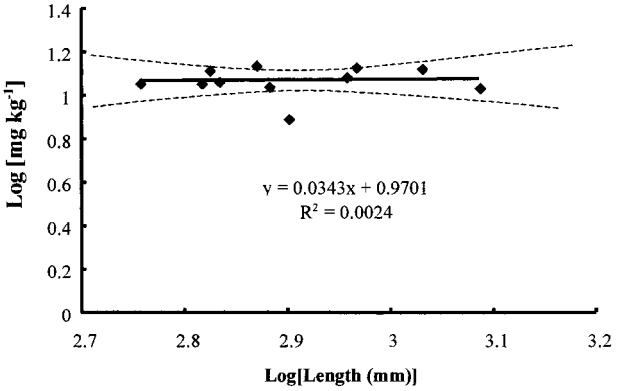

Figure 5. Concentration of mercury in muscle of *D. calcea*.

and supports historical observations that there is a strong exponential relationship between the length of sharks and the concentration of mercury. For instance, Walker (1988) suggested that differences in longevity may be partly responsible for differences in mercury concentrations found in school shark, *Galeorhinus galeus*, and gummy shark, *Mustelus antarcticus*. In contrast, *C. owstoni* does not seem to accumulate mercury in the same manner (Figure 3). Concentrations of mercury plateau just above 1 mg kg^{-1} with no detectable exponential relationship in the data ($R^2 = 0.002$). Walker (1988) also suggested that differences in diet and habitat were responsible for differences in mercury concentrations found in *G. galeus* and *M. antarcticus*. Again, although little is known of the biology of *C. owstoni*, *C. crepidater*, and *D. calcea*, the different species almost certainly utilize different but adjacent habitats and feed on different prey species. Thus, although the different fisheries stocks accumulate characteristically different signatures of trace elements, the analytical chemistry suggests that the mechanisms for controlling mercury in *C. owstoni* may be different from that used by both *C. crepidator* and *D. calcea* and, therefore, that placing *C. crepidator* and *C. owstoni* in the same genus may not be justified.

CONCLUSIONS

Several trace metals exhibit high concentration factors relative to seawater in the muscle tissue of the three dog shark species examined. Significant differences between concentrations in the three species were detected for arsenic, cadmium, mercury, and zinc, with *C. owstoni* containing the highest or equally high concentrations of each element. *C. crepidator* and *D. calcea* have strong relationships between length and mercury concentration; *C. owstoni* does not. The mechanisms for controlling mercury in *C. owstoni* appear to differ from that used by both *C. crepidator* and *D. calcea*. Furthermore, placing *C. crepidator* and *C. owstoni* in the same genus is worth re-examination by modern genetic techniques as this may not be justified. The muscle tissue of these sharks was found to comply with Australian food standards for copper, lead, and zinc but contained higher than permitted concentrations of arsenic and mercury. A weekly consumption of 400 g of any of these sharks would result in intake of more than the FAO's provisional tolerable weekly intake.

LITERATURE CITED

AOAC (Association of Official Analytical Chemists). *Official Methods of Analysis*, 15th ed.; AOAC: Arlington, VA, 1990.

APHA (American Public Health Association). *Standard Methods for the Examination of Water and Wastewater*, 19th ed.; APHA: Washington, DC, 1995.

Australia New Zealand Food Authority. *Food Standards Code*; 1998; Standard A12, Issue 37.

Caputi, N.; Edmonds, J. S.; Heald, D. I. Mercury content of shark from south-western Australian waters. *Mar. Pollut. Bull.* **1979**, *10*, 337−340.

Davenport, S. Mercury in blue sharks and deepwater dogfish from around Tasmania. *Aust. Fish.* **1995**, *54*, 20−22.

Denton, G. R. W.; Breck. W. G. Mercury in tropical marine organisms from North Queensland. *Mar. Pollut. Bull.* **1981**, *12*, 116−121.

Eustace, I. J. Zinc, cadmium, copper and manganese in species of finfish and shellfish caught in the Derwent Estuary, Tasmania. *Aust. J. Mar. Freshwater Res.* **1974**, *25*, 209−220.

Fairey, R.; Taberski, K.; Lamerdin, S.; Johnson, E.; Clark, R. P.; Downing, J. W.; Newman, J.; Petreas, M. Organochlorines and other environmental contaminants in muscle tissues of sportfish collected from San Francisco Bay. *Mar. Pollut. Bull.* **1997**, *34*, 1058−1071.

Games, P. A.; Howell, J. F. Pairwise multiple comparison procedures with unequal n's and/or variances: a Monte Carlo study. *J. Ed. Stat.* **1976**, *1*, 113−125.

Glover, J. W. Concentrations of arsenic, selenium, and 10 heavy metals in school shark, *Galeorhinus australis* (Macleay), and gummy shark, *Mustelus antarcticus* Günther, from south-eastern Australian waters. *Aust. J. Mar. Freshwater Res.* **1979**, *30*, 505−510.

Hornung, H.; Sukeenik, A.; Gabreielides, G. P. Distribution and composition of fatty acids in muscle lipids of inshore fish and deep water sharks from the eastern Meditterranean. *Mar. Poll. Bull.* **1994**, *28*, 448−450.

IPCS. *Environmental Health Criteria 18. Arsenic*; World Health Organization: Geneva, Switzerland, 1981.

IPCS. *Environmental Health Criteria 118. Inorganic Mercury*; World Health Organization: Geneva, Switzerland, 1991.

Le Blanc, P. J.; Jackson, A. L. Arsenic in marine fish and invertebrates. *Mar. Pollut. Bull.* **1973**, *4*, 88−90.

Lyle, J. M. Mercury concentrations in four carcharhinid and three hammerhead sharks from coastal waters of the Northern Territory. *Aust. J. Mar. Freshwater Res.* **1984**, *35*, 441−452.

Lyle, J. M. Mercury and selenium concentrations in sharks from northern Australian waters. *Aust. J. Mar. Freshwater Res.* **1986**, *37*, 309−321.

Marcovecchio, J. E.; Moreno, V. G.; Perez, A. Metal accumulation in tissues of sharks from the Bahia Blanca Estuary, Argentina. *Mar. Environ. Res.* **1991**, *31*, 263−274.

Office of Legislative Drafting. *Fisheries Management Regulations*; Ausinfo: Canberra, Australia, 1998.

Phillips, D. J. H. Arsenic in aquatic organisms: a review, emphasizing chemical speciation. *Aquat. Toxicol.* **1990**, *16*, 151−186.

Powell, J. H.; Powell, R. E.; Fielder, D. R. Trace element concentrations in tropical marine fish at Bougainville Island, Papua New Guinea. *Wat. Air Soil Pollut.* **1981**, *16*, 143−158.

Ramamurthy, V. D. Baseline study of the level of concentration of mercury in the food fishes of the Bay of Bengal, Arabian Sea and Indian Ocean. *Bull. Jpn. Soc. Sci. Fish.* **1979**, *45*, 1405−1407.

Taguchi, M.; Yasuda, K.; Toda, S.; Shimizu, M. Study of metal contents of elasmobranch fishes: Part 1—metal concentration in the muscle tissue of a dogfish, *Squalus mitsukurii*. *Mar. Environ. Res.* **1979**, *2*, 239−249.

Thompson, J. D. Mercury concentrations of the axial muscle tissues of some marine fishes of the continental shelf adjacent to Tasmania. *Aust. J. Mar. Freshwater Res.* **1985**, *36*, 509−517.

U.S. Food and Drug Administration. *Guidance Document for Arsenic in Shellfish*; U.S. GPO: Washington, DC, 1993.

U.S. Food and Drug Administration. *Mercury in Fish: Cause for Concern?* U.S. GPO: Washington, DC, 1994.

Van Loon, J. C. *Selected Methods of Trace Metal Analysis: Biological and Environmental Aamples*; Wiley: New York, 1985.

Vas, P. Observations on trace metal concentrations in a carcharhinid shark, *Galeorhinus galeus*, from Liverpool Bay. *Mar. Pollut. Bull.* **1987**, *18*, 193−194.

Vas, P. Trace metal levels in sharks from British and Atlantic waters. *Mar. Pollut. Bull.* **1991**, *22*, 67−72.

Vas, P.; Gordon, J. D. M. Trace metal concentrations in the scyliorhinid shark *Galeus melastomas* from the Rockall Trough. *Mar. Pollut. Bull.* **1988**, *19*, 396−398.

Vas, P.; Gordon, J. D. M. Trace metals in deep-sea sharks from the Rockall Trough. *Mar. Pollut. Bull.* **1993**, *26*, 400−402.

Vas, P.; Stevens, J. D.; Bonwick, G. A.; Tizini, O. A. Cd, Mn, and Zn concentrations in vertebrae of blue shark and shortfin mako in Australian coastal waters. *Mar. Pollut. Bull.* **1990**, *21*, 203−206.

Vlieg, P.; Murray, T.; Body, D. R. Nutritional data on six oceanic pelagic fish species from New Zealand waters. *J. Food Compos. Anal.* **1993**, *6*, 45−54.

Walker, T. I. Effects of species, sex, length and locality on the mercury content of school shark *Galeorhinus australis* (Macleay) and gummy shark *Mustelis antarcticus* Gunther from south-eastern Australian waters. *Aust. J. Mar. Freshwater Res.* **1976**, *27*, 603−616.

Walker, T. I. Statistical comparison of the results from six analytical chemistry laboratories of the mercury content of muscle tissue of two species of sharks. *Int. J. Environ. Anal. Chem.* **1977**, *5*, 25−33.

Walker, T. I. Mercury concentrations in edible tissues of elasmobranchs, teleosts, crustaceans and molluscs from south-eastern Australian waters. *Aust. J. Mar. Freshwater Res.* **1988**, *39*, 39−49.

Ward, A. F.; Marciello, L. F.; Carrara, L.; Luciano, V. J. Simultaneous determination of major, minor and trace elements in agricultural and biological samples by inductively coupled argon plasma spectrometry. *Spectrosc. Lett.* **1980**, *13*, 803−831.

Yeow, M.; Chambers, A.; Walker, T. Filleted flake—solving an identity problem. *Aust. Fish.* **1986**, Oct, 30−31.

Received for review March 3, 2000. Revised manuscript received June 9, 2000. Accepted June 10, 2000.

JF000285Z