Trace Element and Stable Isotope Analyses of Deep Sea Fish from the Sulu Sea, Philippines

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Abstract

Thirty-five deep sea fishes belonging to 22 species and one unidentified specimen obtained from the Sulu Sea, located in the southwestern area of the Philippines were analyzed in the late 2002, for 23 trace elements using ICP-MS, HG-AAS and CV-AAS. Predominant accumulation of strontium (Sr) was observed in all the samples. This stems from the fact that the whole body of fish was homogenized since Sr is known to accumulate in bones and hard tissues. Mercury concentrations in all the 36 samples were below the detection limit. Cadmium concentrations were generally below 1 µg/g dry weight (dw) except in *Pterygotrigla* spp. (4.29 µg/g dw) and *Sternoptyx pseudodiaphana* (2.89 µg/g dw). Concentrations of Pb were predominantly low with about 90% of the specimens having less than 1 µg/g dw. In general, concentrations of Sr, Zn, Cu, Se and Cd appeared to increase with increasing depth of occurrence of the species. Manganese, Tl, Pb, Bi, In, Cs and As showed significant positive correlation ($p < 0.05$) with $\delta^{15}N$, suggesting that these elements were biomagnified. To our knowledge, this is the first study reporting Tl biomagnification in fish. Rubidium and Cs showed significant positive correlation with $\delta^{13}C$, implying that Rb and Cs would originate from offshore waters as oceanic plankton has high $\delta^{13}C$. Comparing results from this study to the dietary standards and guidelines for Hg, Pb, Cu and Zn in fish and shellfish of the Ministry of Agriculture, Fisheries and Food of the United Kingdom, these levels were not high to warrant concern if they were to be consumed by humans. However, 16.7% of the fish samples had high Cr levels when compared with the Hong Kong's safe limit of 4 µg/g dw for Cr in sea food. This constitutes a health risk to humans, as Cr is potentially toxic.

Introduction

The deep sea is a vast area which remains largely unexplored, especially in terms of its biota (Ruhl & Smith, 2004). Collectively the deep sea and its ecosystems are considered to be the sink and final reservoir for contaminants (Tatsukawa & Tanabe, 1984). Pollution of marine ecosystems by trace elements is of global environmental concern. Elements in trace concentrations are normal constituents of marine organisms. At high levels they are potentially toxic and may disrupt biological activities of aquatic ecosystems. The ability of trace elements to be concentrated in the organs of marine organisms accounts for their toxicity and also poses a direct threat to both the aquatic biota and man (Watling, 1983).

Many deep sea species are long lived, with slow growth rates and are likely to reach maturity at a much more advanced age than commercial species from continental shelf areas (Gordon *et al.,* 1995). They tend to feed at higher trophic levels than their shallow-water counterparts and, thus, could be exposed to higher levels of elements for longer periods hence the accumulation of trace elements could be greater (Gordon *et al.,* 1995). A study by Mormede & Davies (2001) revealed that trace elements such as cadmium, mercury, lead, copper and zinc were found in relatively high concentrations in some deep sea fish species (*Nezumia aequalis*, *Lepidon eques* and *Raja fyllae*) from the Rockall Trough, west of Scotland.

In contrast to the enormous amount of knowledge on trace element concentrations in near shore organisms, little is known about accumulation levels of trace elements in deep sea organisms (Cronin *et al.,* 1998; Vas *et al.,* 1993; Windom *et al.,* 1987) and their species-specific accumulation profile. There is a paucity of study of these organisms to elucidate their accumulation levels. There is increasing interest in studying the degree of contamination of the deep sea environment. However, many studies do not typically examine the total body burden of trace elements in fish but only concentrations in muscle tissue were investigated, as from a human health perspective. This is, therefore, one of the very few studies that have dealt with the analyses of trace elements in whole fish from deep sea.

Stable isotope analysis has emerged as a powerful tool to trace diet as isotope ratios of a consumer are related to those of their preys (DeNiro & Epstein, 1978, 1981). Carbon and nitrogen isotope values ($\delta^{13}C$ and $\delta^{15}N$) differ between organisms and their diets because of a slight selective retention of the heavier isotope and excretion of the lighter one. As a result, organisms have a higher ′ value than their diet. Changes in ratios of stable isotopes of carbon and nitrogen have been used to elucidate trophic relationships within marine food webs (Hobson *et al*., 1997). For nitrogen, enrichment in ^{15}N typically shows a stepwise increase with trophic level within a food chain with a trophic enrichment value of about 3‰ (Hobson & Welch, 1992). Thus, carbon-13 value, rather than being a reliable indicator of the trophic level, is preferentially used to indicate relative contributions to the diet of different potential primary sources in a trophic network, indicating the aquatic *vs*. terrestrial, inshore *vs*. offshore, or pelagic *vs*. benthic contribution to food intake (Hobson *et al*., 1995; Dauby *et al*., 1998).

Trace elements are merely transferred through diet. Indeed, trace element levels found in marine organisms depend not only on the contamination of the environment but also on several other ecological or physiological factors among which the diet and trophic position are determining elements (Das *et al*., 2003). By using a combination of stable isotope and trace element analyses, one could compare the diet and position in the trophic web of predators.

Asia is the world's leading fish producer and accounts for over 63% of the total fish production (Briones *et al*., 2004). Fish forms an important part of Asian diets. In the Philippines, marine fisheries provide various economic and social benefits (Luna *et al*., 2004). In 2003, the sector produced 2.03 million tonnes of fish and invertebrates (BAS, 2004). Most people fish for personal consumption, and fish consumption is estimated at 30 kg per capita. There is no information on the contamination or baseline status of trace elements in water and fish from the Sulu Sea, despite that fish is an important component of the diet of Filipinos. It was deemed necessary to focus on trace element levels in deep sea fish from the Sulu Sea in an attempt to establish baseline levels and whether they meet the statutory levels recognized as safe for human consumption.

The aim of the study was to investigate and provide data on the concentrations of trace elements in fish species collected from deep water in the Sulu Sea in order to elucidate the accumulation characteristics. Stable carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) isotopes analyses were used to evaluate the relationship between accumulation of trace elements and trophic levels in deep sea ecosystem.

Materials and methods

The Sulu Sea is located in the southwestern area of the Philippines (Fig. 1). It is separated from the South China Sea in the northeast by the Palawan Island and from the Celebes Sea in the southeast by a chain of islands known as Sulu Archipelago. The Sulu Sea, together with the Celebes Sea, constitutes the Sulu-Celebes Sea Large Marine Ecosystem (LME). It has an area of about 900,000 km^2 and much of the LME has a depth greater than 3000 m.

Some deep sea fishes were collected from the Sulu Sea (Fig. 1) and surrounding areas during November-December 2002, with either a mid-water trawl, a plankton net, or a beam trawl (for data collection see Nishida & Gamo, 2004). Thirty-five specimens belonging to 22 species and one unidentified specimen were analyzed for trace elements. All the samples were kept in an ice box and transported to the environmental specimen bank (es-bank) at Ehime University, Japan and kept at – 20 °C until chemical analysis.

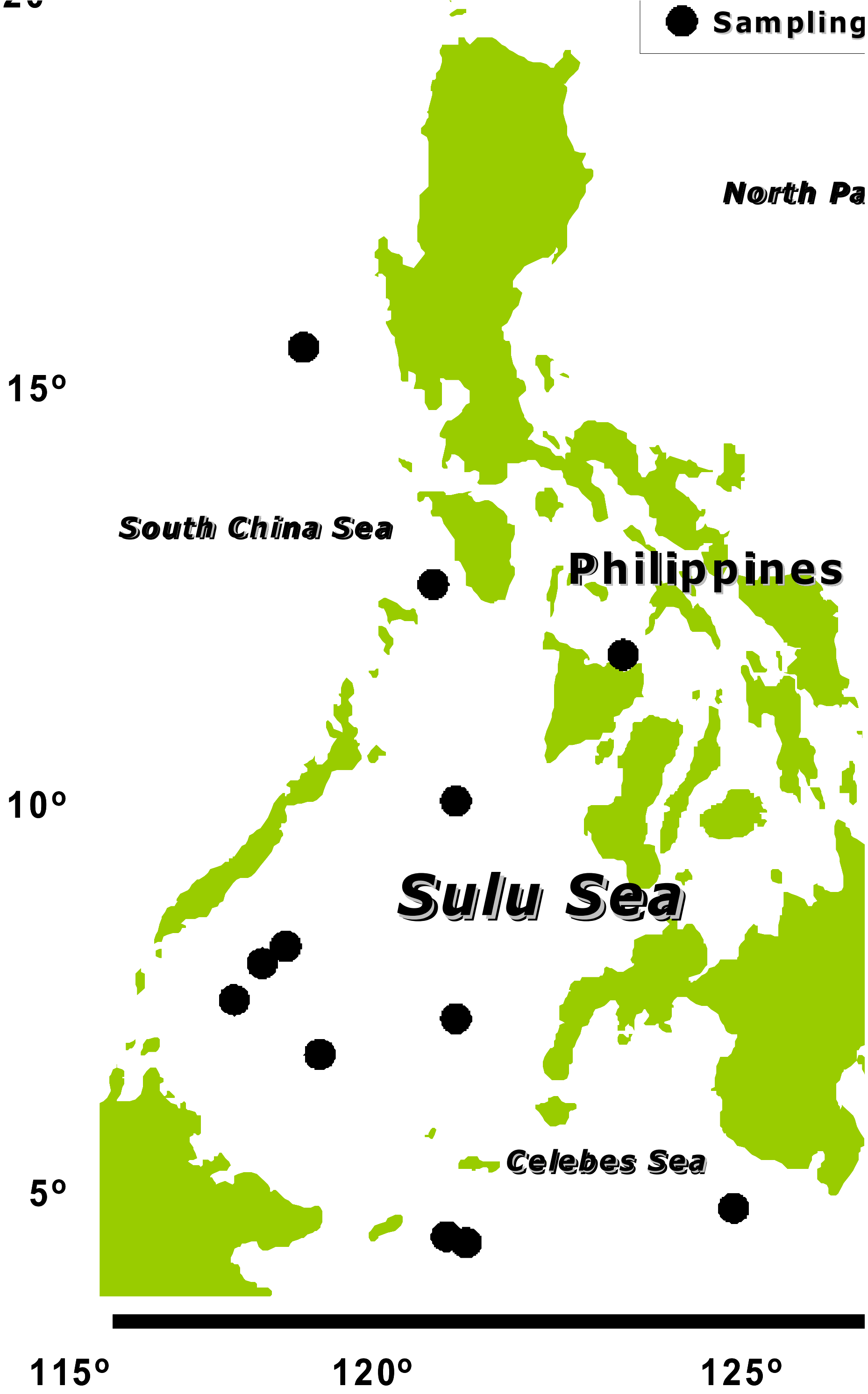

Fig. 1. Map of the Sulu Sea showing sampling locations

The whole bodies of the specimens of the same species were pooled and homogenized to prepare a composite sample. Where possible, the size ranges of fish have been provided (Table 1). The samples were dried at 80 °C for 12 h and about 0.2 g dry weight digested by microwave in the presence of concentrated nitric acid in Teflon vials. Concentrations of trace elements (V, Cr, Mn, Co, Cu, Zn, Ga, Rb, Sr, Mo, Ag, Cd, In, Sn, Sb, Cs, Ba, Tl, Pb and Bi) were measured by an inductively-coupled plasma mass spectrometer (ICP-MS, HP-4500, Hewlett-Packard, Avondale, PA, USA). Matrix effects and instrumental drift in the ICP-MS measurements were corrected by the internal standard method with indium as the internal standard. Concentrations of Hg and Se were determined by cold vapour-atomic absorption spectrometry (CV-AAS, model HG-3000, Sanso, Tsukubu, Japan) and hydride generation-atomic absorption spectrometry (HG-AAS, HVG-1 hydride system, Shimadzu, Kyoto, Japan), respectively. For As determination, about 0.05 g dry weight of the samples were treated with acid mixture (HNO_3:$HClO_4$: H_2SO_4 = 1:2:1) and digested by heating until the perchloric acid was removed. After cooling, they were diluted with Milli-Q water and arsenic concentrations were determined by HG-AAS using a Shimadzu HVG-1 hydride generation system coupled to a Shimadzu AA680 atomic absorption spectrometer (Shimadzu, Kyoto, Japan).

Table 1

Trace element concentrations (µg/g dry wt.) in whole body of deep-sea fish from the Sulu Sea (November - December 2002)

Sample ID	*species*	*Length*	*Weight*	*Depth (m)*	*'^{15}N*	*'^{13}C)*	*V*	*Cr*
Mn	*CoCu*	*Zn*						
		(mm)	*(g) ww*		*(‰)*	*(‰*		
	Acropomatidae							
S1-B	Malakichthys elegans	100.1	10.8	292–296	10.30	-17.31	0.22	13
5.06	0.18	21.5	56.1					
	Alepocephalidae							
7-B	*Rouleina* spp. (1)	173.2	35.1	688–693	12.45	-18.10	1.9	2.7
261	0.42	29.0	76.7					
8-A	*Rouleina* spp. (2)	NA	33.5	796–804	12.36	-17.65	0.74	1.5
99.3	0.18	3.61	48.8					
8-B	*Rouleina* spp. (3)	197	47.8	1012–1015	11.95	-17.30	0.54	1.0
43.3	0.13	11.9	49.5					
	Gonostomatidae							
4	*Cyclothone acclinidens* (1)	29.1–46.6	0.2	NA	11.92	-17.76	3.6	0.37
3.59	0.061	10.6	47.1					
6	*Cyclothone acclinidens* (2)	31.2–45	0.3	NA	11.83	-17.50	2.9	1.3
3.53	0.066	9.48	56.4					
E4	*Cyclothone pallida* (1)	37.8–57	0.6	NA	12.09	-18.28	1.4	1.1
3.81	0.051	10.0	63.3					
17	*Cyclothone pallida* (2)	36.6–52.5	0.6	NA	10.24	-19.30	1.6	1.9
3.23	0.059	7.28	57.7					
2	*Gonostoma elongatum* (1)	141	9.4	NA	10.18	-17.69	0.25	0.37
2.99	0.046	7.51	32.7					
3	*Gonostoma elongatum* (2)	196	20.5	1299	11.57	-16.74	0.14	0.68
5.68	0.043	5.63	35.7					
	Macrouridae							
7-B	*Bathygadus* spp. (1)	223.3	25.4	688–693	13.19	-17.47	0.53	5.2
28.7	0.22	50.3	62.2					
8-A	*Bathygadus* spp. (2)	228	85.4	796–804	12.51	-16.60	0.13	2.2
8.93	0.052	3.33	13.9					
S1-A	*Caelorinchus kamoharai*	119.6	4.9	214–216	10.31	-17.66	1.8	4.0
33.3	0.43	6.19	96.0					
7-A	*Hymenocephalus striatissimus* (1)	159.5	4.5	514–516	10.11	-17.57	0.53	0.81
12.3	0.15	7.80	106					
S1-C	*Hymenocephalus striatissimus* (2)	125.8	6.6	367–368	11.39	-18.07	0.54	0.73
9.66	0.11	3.71	143					
	Macrurocyttidae							

7-A	*Zenion hololepis*	106.2	23.7	514 - 516	11.65	-17.91	0.13	2.5
4.48	0.068	2.87	54.6					
	Myctophidae							
E3	*Ceratoscopelus warmingii* (1)	44–65	1.1	200	12.22	-14.37	3.0	8.4
6.02	0.13	3.80	39.0					
E6	*Ceratoscopelus warmingii* (2)	62–81.5	2.8	200	8.60	-17.84	0.47	3.1
6.59	0.13	5.26	47.0					
E8	*Ceratoscopelus warmingii* (3)	32–79	1.2	200	10.41	-16.10	0.44	3.0
5.67	0.13	5.06	41.0					
2	*Diaphus problematicus*	89.2	6.8	NA	9.38	-17.80	0.15	0.23
3.39	0.081	3.78	39.9					
E8	*Diaphus regani*	60.3 - 71.8	2.9	200	9.53	-17.81	0.59	1.2
6.92	0.11	5.55	36.1					
	Neoscopelidae							
14	*Solivomer arenidens*	118.6	11.1	1482–1488	12.20	-17.71	0.66	2.3
60.9	0.13	4.11	32.4					
	Ophidiidae							
S1-C	*Glyptophidium japonicum*	154.8	14	367–368	11.68	-17.87	0.37	2.6
9.57	0.12	6.45	115					
8-B	*Lamprogrammus niger*	316	143	1012–1015	11.65	-17.79	0.83	1.4
112	0.19	5.98	28.3					
	Serrivomeridae							
3	*Stemonidium hypomelas*	540	48	NA	11.94	-17.36	0.11	0.71
2.51	0.039	6.00	47.8					
	Setarchidae							
3	*Ectreposebastes imus*	122	50.2	NA	11.94	-17.71	0.10	0.51
2.30	0.076	3.76	55.9					
S1-C	*Lioscorpius longiceps*	116.6	14.5	367–368	10.33	-16.13	0.21	7.6
9.47	0.13	17.2	75.6					
	Sternoptychidae							
2	*Sternoptyx pseudodiaphana* (1)	29.7 - 49.6	1.7	NA	7.17	-17.40	0.25	0.36
4.92	0.070	12.4	56.5					
3	*Sternoptyx pseudodiaphana* (2)	10 - 46.8	0.8	NA	8.63	-18.24	0.13	1.3
4.14	0.084	11.0	58.6					
	Stomiidae							
2	*Chauliodus sloani* (1)	151–193	13.1	NA	10.92	-17.48	0.075	0.32
1.86	0.041	6.61	30.2					
3	*Chauliodus sloani* (2)	117–134	6.1	NA	9.50	-17.91	0.092	0.30
1.83	0.039	6.80	29.8					
4	*Chauliodus sloani* (3)	132–190	11.5	NA	10.04	-18.22	0.10	0.80
2.77	0.037	7.46	31.4					
E12	*Chauliodus sloani* (4)	141	6	200	9.08	-18.64	0.13	1.0
3.40	0.050	7.06	34.9					
3	*Idiacanthus fasciola*	256	9.1	NA	10.46	-17.81	0.25	0.47
9.73	0.047	10.4	45.9					
	Triglidae							
S1-B	*Pterygotrigla spp.*	63.9	3	292–296	9.64	-18.13	1.2	1.5
7.73	0.11	21.6	109					
8-B	Unidentified	423	116.5	1012–1015	11.56	-17.25	0.17	4.4
16.0	0.11	13.6	36.6					

TABLE 1 (cont.)

Sample ID	*species*	*Ga*	*As*	*Se*	*Rb*	*Sr*	*Mo*	*Ag*	*Cd*	*In*	*Sn*	*Sb*	*Cs*
Ba	*Hg*	*Tl*		*Pb*	*Bi*								
	Acropomatidae												
S1-B	*Malakichthys elegans*	0.094	124	7.3	1.85	371	0.094	0.009	0.130	0.002	0.155	0.01	0.06
1.7	<0.05	0.001	1.00	0.001									
	Alepocephalidae												
7-B	*Rouleina* spp. (1)	0.806	149	2.3	2.29	249	0.120	0.032	0.549	0.003	0.215	0.05	0.08
	<0.05	0.013	1.58	0.007									
8-A	*Rouleina* spp. (2)	0.198	322	4.6	2.74	381	0.267	0.19	0.848	0.005	0.080	0.03	0.07
3.0	<0.05	0.006	0.210	0.004									
8-B	*Rouleina* spp. (3)	0.164	128	2.8	2.47	205	0.090	0.039	0.479	0.007	0.144	0.03	0.07

2.8	<0.05	0.008	0.599	0.006										
	Gonostomatidae													
4	*Cyclothone acclinidens* (1)	0.069	55.0	2.9	1.88	329	0.105	0.064	0.525	0.007	0.218	0.02	0.04	
1.4	<0.05	0.007	0.314	0.008										
6	*Cyclothone acclinidens* (2)	0.058	40.9	2.6	1.96	312	0.093	0.034	0.578	0.002	0.224	0.02	0.03	
1.2	<0.05	0.002	0.202	0.003										
E4	*Cyclothone pallida* (1)	0.155	63.6	2.4	1.50	269	0.134	0.071	0.475	0.005	0.562	0.04	0.02	
3.5	<0.05	0.005	0.560	0.004										
17	*Cyclothone pallida* (2)	0.088	27.0	2.4	1.93	249	0.128	0.094	0.861	0.006	0.268	0.03	0.04	
1.7	<0.05	0.007	0.461	0.005										
2	*Gonostoma elongatum* (1)	0.030	48.7	2.6	2.16	169	0.143	0.020	0.346	0.006	0.322	0.02	0.03	
0.53	<0.05	0.005	0.100	0.006										
3	*Gonostoma elongatum* (2)	0.042	34.3	3.2	2.26	259	0.060	0.017	0.280	0.001	0.166	0.01	0.04	
0.86	<0.05	0.002	0.184	0.002										
	Macrouridae													
7-B	*Bathygadus* spp. (1)	0.137	146	4.7	2.29	320	0.092	0.12	0.280	0.007	0.309	0.02	0.06	
2.2	<0.05	0.006	2.67	0.007										
8-A	*Bathygadus* spp. (2)	0.044	135	2.1	3.04	24.5	0.038	0.006	0.031	0.007	0.078	0.02	0.08	
0.39	<0.05	0.008	0.246	0.007										
S1-A	*Caelorinchus kamoharai*	0.388	63.3	2.5	2.60	661	0.128	0.031	0.132	0.002	0.062	0.01	0.13	
4.8	<0.05	0.021	0.595	0.005										
7-A	*Hymenocephalus striatissimus* (1)	0.109	24.4	4.2	2.11	577	0.101	0.12	0.505	<0.001	0.046	0.02	0.08	
2.3	<0.05	0.002	0.165	0.002										
S1-C	*Hymenocephalus striatissimus* (2)	0.189	158	3.9	1.08	934	0.093	0.024	0.159	0.002	0.069	0.02	0.03	
4.0	<0.05	0.004	0.359	0.005										
	Macrurocyttidae													
7-A	*Zenion hololepis*	0.059	54.1	5.7	1.85	246	0.087	0.039	0.087	0.003	0.086	0.01	0.05	
1.1	<0.05	0.003	0.080	0.003										
	Myctophidae													
E3	*Ceratoscopelus warmingii* (1)	0.524	27.9	2.4	3.22	192	1.23	0.019	0.748	0.005	0.206	0.02	0.05	11
	<0.05	0.013	0.208	0.006										
E6	*Ceratoscopelus warmingii* (2)	0.579	46.0	2.2	3.25	305	0.262	0.031	0.992	0.008	0.184	0.02	0.05	13
	<0.05	0.010	0.188	0.009										
E8	*Ceratoscopelus warmingii* (3)	0.580	33.8	3.1	4.00	244	0.115	0.056	0.807	0.002	0.338	0.02	0.04	14
	<0.05	0.008	0.195	0.004										
2	*Diaphus problematicus*	0.072	25.1	2.5	2.09	150	0.090	0.043	0.785	0.001	0.182	0.01	0.04	
1.6	<0.05	0.002	0.091	0.002										
E8	*Diaphus regani*	0.250	15.9	1.9	2.44	261	0.139	0.019	0.761	0.001	0.114	0.01	0.04	
6.2	<0.05	0.006	0.099	0.002										
	Neoscopelidae													
14	*Solivomer arenidens*	0.149	170	5.2	2.50	318	0.096	0.056	0.481	0.001	0.112	0.02	0.04	
2.7	<0.05	0.003	0.218	0.002										
	Ophidiidae													
S1-C	*Glyptophidium japonicum*	0.109	68.1	2.4	2.20	690	0.065	0.021	0.197	0.004	0.080	0.02	0.07	
1.8	<0.05	0.007	0.365	0.006										
8-B	*Lamprogrammus niger*	0.146	125	2.0	2.66	241	0.122	0.027	0.518	0.003	0.159	0.02	0.06	
2.1	<0.05	0.007	0.245	0.004										
	Serrivomeridae													
3	*Stemonidium hypomelas*	0.052	153	2.7	2.41	192	0.067	0.022	1.08	0.001	0.181	0.01	0.03	
1.1	<0.05	0.001	0.231	0.001										
	Setarchidae													
3	*Ectreposebastes imus*	0.027	42.9	5.5	1.44	115	0.062	0.006	0.445	0.002	0.105	0.01	0.07	
0.47	<0.05	0.003	0.155	0.002										
S1-C	*Lioscorpius longiceps*	0.090	74.2	6.1	2.10	256	0.142	0.048	1.01	0.019	0.149	0.05	0.09	
1.4	<0.05	0.021	0.873	0.017										
	Sternoptychidae													
2	*Sternoptyx pseudodiaphana* (1)	0.056	59.4	3.3	1.71	171	0.188	0.060	2.89	0.001	0.128	0.01	0.02	
1.2	<0.05	0.002	0.155	0.002										
3	*Sternoptyx pseudodiaphana* (2)	0.061	63.7	3.6	1.87	190	0.183	0.067	1.67	0.001	0.192	0.03	0.02	
1.4	<0.05	0.003	0.101	0.002										
	Stomiidae													
2	*Chauliodus sloani* (1)	0.025	25.5	2.7	2.25	114	0.073	0.012	0.704	0.004	0.972	0.01	0.03	
0.45	<0.05	0.001	0.099	0.002										
3	*Chauliodus sloani* (2)	0.022	32.0	2.6	1.88	145	0.089	0.033	0.360	0.001	0.152	0.01	0.02	
0.49	<0.05	0.001	0.079	0.002										
4	*Chauliodus sloani* (3)	0.024	35.8	1.8	2.19	138	0.094	0.011	0.190	0.001	0.167	0.04	0.02	

0.46	<0.05	0.001	0.155	0.001									
E12	*Chauliodus sloani* (4)	0.222	11.0	1.6	2.28	169	0.098	0.016	0.635	0.001	0.135	0.01	0.03
5.2	<0.05	0.001	2.15	0.001									
3	*Idiacanthus fasciola*	0.032	28.6	3.2	2.22	129	0.154	0.030	0.828	<0.001	0.262	0.02	0.02
0.59	<0.05	0.001	0.702	0.002									
	Triglidae												
S1-B	*Pterygotrigla* spp.	0.165	17.2	9.3	1.76	770	0.098	0.087	4.29	0.001	0.211	0.01	0.05
3.3	<0.05	0.001	0.830	<0.001									
8-B	Unidentified	0.064	16.1	4.1	1.71	82.0	0.087	0.013	0.552	0.003	0.108	0.01	0.05
0.98	<0.05	0.003	0.603	0.002									

NA = No available information

Analytical quality was assessed using standard reference material DORM2 (Dogfish muscle; National Research Council, Canada). Recoveries of the elements ranged from 88% to 111% of the certified values. Concentrations of the trace elements were expressed on a dry weight basis (µg/g dw). In order to compare values with published data reported on a wet weight basis, values were converted to wet weight basis assuming moisture content of 75%.

Stable isotopes analysis

Sub samples from the homogenized samples were dried for 24 h at 60 °C and ground into powder with mortar and pestle. 1.0 mg powder samples were packed into 4 × 6 mm tin capsules for stable isotope measurements. Stable isotopes ratios of nitrogen and carbon were measured with a mass spectrometer (ANCAR-SL 20–20, Concern Ltd) coupled with an elemental analyzer. Isotopic ratios of carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) were expressed as the deviation from standards in parts per thousand (‰) according to the following conventional formular (Doi *et al*., 2005):

$$\delta^{13}C, \delta^{15}N = [(R_{sample} / R_{standard}) - 1] \times 1000 \ (‰),$$

where $R = {}^{13}C/{}^{12}C$ or ${}^{15}N/{}^{14}N^{o}$

Atmospheric nitrogen and belemnite, Pee Dee Belemnite (PDB) were used as the isotope standards for nitrogen and carbon, respectively. The analytical precision for the isotope analysis was ± 0.2‰ for both $d^{13}C$ and $d^{15}N$.

Statistical analysis

One-half of the value of the limit of detection of each element was substituted for those values below the limit of detection and applied in statistical analysis. Mann-Whitney *U*-test and Spearman's rank correlation coefficient were employed to measure the significance and correlation between trace element concentrations and stable isotope values. A *p*-value of less than 0.05 was considered to indicate statistical significance; all tests were two-tailed. Statistical analysis was performed using SPSS (version 12.0, SPSS Inc., Chicago, Il, USA) for Windows, 2003.

Results and discussion

Trace element accumulation

Concentrations of trace elements in the whole body of fish are shown in Table 1. Strontium was the most abundant element in the fish studied. The concentrations of Sr in this present study ranged from 24.5–934 µg/g dw (the highest value in *Hymenocephalus striatissimus*) (Table 1). This trend was also observed in other studies of whole body fish species in the Manila Bay of the Philippines (Prudente *et al*., 1997) and the East China Sea (Asante, 2005). The high Sr concentrations stem from the fact that the whole body of fish was homogenized and, chemically, strontium resembles calcium, which is known to accumulate in bones (Nielsen, 1986).

Neither Hg nor Pb was at levels likely to cause concern if they were to be consumed by human beings. Waterman (1987) suggested that the maximum level of Hg should not exceed 0.5 µg/g wet weight (ww) of fish. Mercury concentrations in all the 36 fish samples were below the detection limit of 0.05 µg/g dw (0.01 µg/g ww) (Table 1). Apart from the fact that the present specimens were smaller in size, mercury is generally known to accumulate with age. Mean Hg concentrations in commercial species of continental shelf are typically rather low (0.02–0.10 µg/g ww) (Brown & Balls, 1997), but there are marked exceptions, particularly among long-lived fish predators with concentrations of 1 µg/g ww or more (Topping & Graham, 1977). Lead concentrations in the species examined were predominantly low with about 90% of the specimens having levels less than 1 µg/g dw. The highest Pb concentration of 2.67 µg/g dw was found in *Bathygadus* spp.

Cadmium is toxic and, hence, elevated concentrations are a threat to marine biota. High Cd values encountered in some marine mammal species are diet related as a result of ingestion of cephalopods (Bustamante *et al*., 1998). Generally, concentrations of Cd were below 1 µg/g dw except in *Pterygotrigla* spp., *Sternoptyx pseudo-diaphana* and *Sternoptyx pseudodiaphana* which had 4.29, 2.89 and 1.67 µg/g dw, respectively. Chromium concentrations in the 36 samples ranged from 0.23–13 µg/g dw. The toxicity of Cr depends on the valency state of Cr in the compound (Merian, 1991; Mertz, 1987). Only 36% of the 36 fish samples had below 1 µg/g dw for Cr. Manganese concentrations were 261 µg/g dw (*Rouleina* spp., 112 µg/g dw (*Lamprogrammus niger*) and 99.3 µg/g dw (*Rouleina* spp.) while the rest ranged from 1.83–43.3 µg/g dw (Table 1). Selenium concentrations ranged from 1.6–9.3 µg/g dw. The highest concentration was found in *Pterygotrigla* spp..

The highest concentration of Zn detected was in *Hymenocephalus striatissimus* (143 µg/g dw). On the other hand, the highest Cu concentration was found in *Bathygadus* spp. (50.3 µg/g dw). Copper is essential for human health and its presence at these low levels could be considered desirable. Concentrations of Co ranged from 0.037–0.43 µg/g dw with *Caelorinchus kamoharai* (0.43 µg/g dw) and *Rouleina* spp. (0.42 µg/g dw) having the maximum levels. The maximum V concen-tration of 3.6 µg/g dw was found in *Cyclothone acclinidens*, followed closely by *Ceratoscopelus warmingii* (3.0 µg/g dw). Rubidium and Ba were accumulated by all the fish analysed, with concentrations ranging from 1.08–4.00 µg/g dw and 0.39–15 µg/g dw, respectively. The levels of Bi, Sb, In, Ag, Mo, Tl and Ga were low and not detected in some samples. It could be that these elements are low in the waters and or the sediments of the Sulu Sea.

Rouleina, the only species belonging to the family Alepocephalidae, recorded the highest concentrations of Mn and Ga, *Lioscorpius longiceps* belonging to the family Setarchidae, had the highest concentrations of Bi and In and *Bathygadus* spp., belonging to the family Macrouridae, showed the least concentration of Sr (Table 1). These variations among the species may be due to differences in metal assimilation and metabolic capacity, or may be indicative of species-specific accumulation.

Vertical distribution of trace elements

To understand the accumulation patterns of trace elements in relation to the habitation, the concentrations of trace elements were compared among habitation type of fish. The fish samples could be put into four groups on the basis of their habitation; mesopelagic, bathypelagic, shallow-demersal and deep-demersal. Higher concentrations of Sr, Zn, Cu, Se and Cd in fish appeared in deep-demersal species (Fig. 2), suggesting the pattern of vertical distribution of these trace elements in the water column in the Sulu Sea. The mean concentrations of Sr, V, Co, Cs and Mn from this study were comparable with those of the Manila Bay, the Philippines (Fig. 3).

While Zn and Rb concentrations were by a factor of half less than those of the Manila Bay, Pb and Cu concentrations were two times higher. However, it should be noted that background levels of trace elements in the marine environment can vary from region to region due to differences in local geology, and high contamination loads may, therefore, not be entirely due to increased loads of pollution.

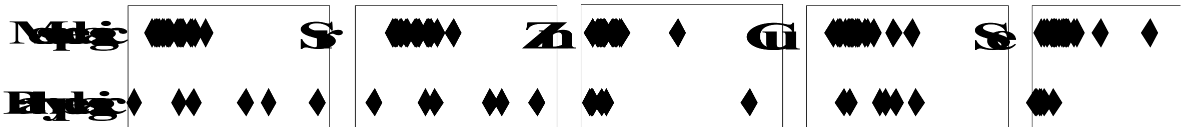

Fig. 2. Comparison of trace element concentrations in deep sea fish with their habitat from the Sulu Sea.

In general, concentrations of Zn and Cu from the Sulu Sea fish species were higher than fish from the North Lantau waters of Hong Kong (Fig. 3). Cadmium values from some fish samples from North Lantau waters of Hong Kong (< 0.9 – 23 µg/g dw) (Parsons, 1999a), were higher than those of the present specimens. However, there is an undeniably high degree of Cd pollution in Hong Kong (Parsons, 1999b). Comparing the results from this study to the dietary standards and guidelines for Hg, Pb, Cu and Zn in fish and shellfish of the Ministry of Agriculture, Fisheries and Food of the United Kingdom (MAFF, 2000), these levels were not high to warrant concern if they were to be consumed by humans (Fig. 3).

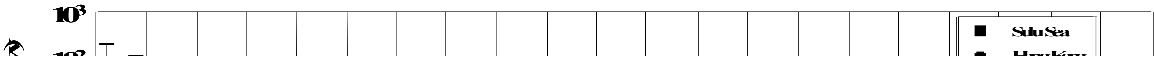

Fig. 3. Comparison of trace element concentrations (mean and range) in whole body of fish from Sulu Sea with studies in Hong Kong (Parsons, 1999a), Manila Bay (Prudente *et al.*, 1997) and MAFF Guideline values (MAFF, 2000). NA = Not available, ND = Not detected.

The general advisory limits for Zn and Cu in food are 50 and 20 µg/g ww, respectively (Kirk & Sawyer, 1991). When the zinc and copper concentra-tions in this study were converted into wet weight basis using the respective moisture contents, the levels in all the 36 samples were below these advisory limits.

In Hong Kong, the allowable limit of Pb in fish tissue is 24 µg/g dw (Parsons, 1999a) and considering this value, concentrations in all the 36 samples from the present study were low. Similarly, Cd concentrations were all below the 2 µg/g ww of safe Cd limit in seafood set by the Hong Kong Government (Parsons, 1998). However, 16.7% of the 36 fish samples had high Cr concentrations (with the highest value of 13 µg/g dw in *Malakichthys elegans*) when compared with Hong Kong's safe limit of 4 µg/g dw for Cr in sea food. This constitutes a health risk to humans as Cr is potentially toxic.

Stable isotopes ($\delta^{13}C$ and $\delta^{15}N$) analyses

A plot of δ^{15}N against δ^{13}C for the analyzed fish species from the Sulu Sea is depicted in Fig. 4. The demersal fish species showed a relatively higher trophic level than the pelagic ones. The comparatively lower δ^{13}C values of the pelagic species may suggest their coastal or pelagic source of food. Tables 2 and 3 show the calculated r and p values between trace element concentration and δ^{15}N and δ^{13}C values, respectively. Elements with significant p (< 0.05) and r values are shown in bold. Cadmium was not biomagnified as significant negative correlation ($p < 0.05$) was observed with δ^{15}N. This is in agreement with the fact that Cd is not biomagnified but diluted through trophic transfer. Copper, Mo, Ag and Sn also showed negative correlation with δ^{15}N though the correlations were not significant (Table 2). Manganese, Tl, Pb, Bi, In, Cs and As showed significant positive correlation ($p < 0.05$) with δ^{15}N (Table 2, Fig. 5), suggesting that these elements were biomagnified. To our knowledge, this is the first study reporting Tl biomagnification in fish. The biomagni-fication observed for As is contrary to the assertion that As does not biomagnify in the food chain (Maher & Butler, 1998; Eisler, 1994) but diluted by trophic transfer. This could be the first study reporting As biomagnification in fish, and further studies into this are required. Only Rb and Cs showed significant positive correlation with δ^{13}C (Table 3, Fig. 6), implying that Rb and Cs mainly originated from oceanic source since oceanic plankton has high δ^{13}C.

TABLE 2

Correlation and significant values of trace element concentrations against ^{15}N for all species from the Sulu Sea

Element	r *value*	p *value*
V	0.304	0.072
Cr	0.282	0.096
Mn	0.375	0.024
Co	0.218	0.201
Cu	– 0.135	0.434
Zn	0.032	0.855
Ga	0.107	0.534
As	0.562	< 0.001
Se	0.098	0.570
Rb	0.260	0.126
Sr	0.111	0.518
Mo	– 0.281	0.097
Ag	– 0.009	0.959
Cd	– 0.340	0.042
In	0.444	0.007
Sn	– 0.002	0.990
Sb	0.308	0.068
Cs	0.388	0.019
Ba	0.024	0.891

TI	0.358	0.032
Pb	0.335	0.046
Bi	0.3412	0.013

TABLE 3

Correlation and significant values of trace element concentrations against $\delta^{13}C$ for all species from the Sulu Sea

Element	r *value*	p *value*
V	– 0.057	0.740
Cr	0.230	0.178
Mn	0.193	0.259
Co	0.222	0.193
Cu	– 0.035	0.841
Zn	0.188	0.272
Ga	0.062	0.721
As	0.137	< 0.425
Se	0.293	0.083
Rb	0.342	0.041
Sr	0.116	0.502
Mo	– 0.152	0.377
Ag	– 0.183	0.285
Cd	– 0.028	0.870
In	0.225	0.188
Sn	– 0.088	0.611
Sb	0.153	0.374
Cs	0.359	0.032
Ba	0.155	0.367
TI	0.206	0.228
Pb	0.028	0.869
Bi	0.174	0.310

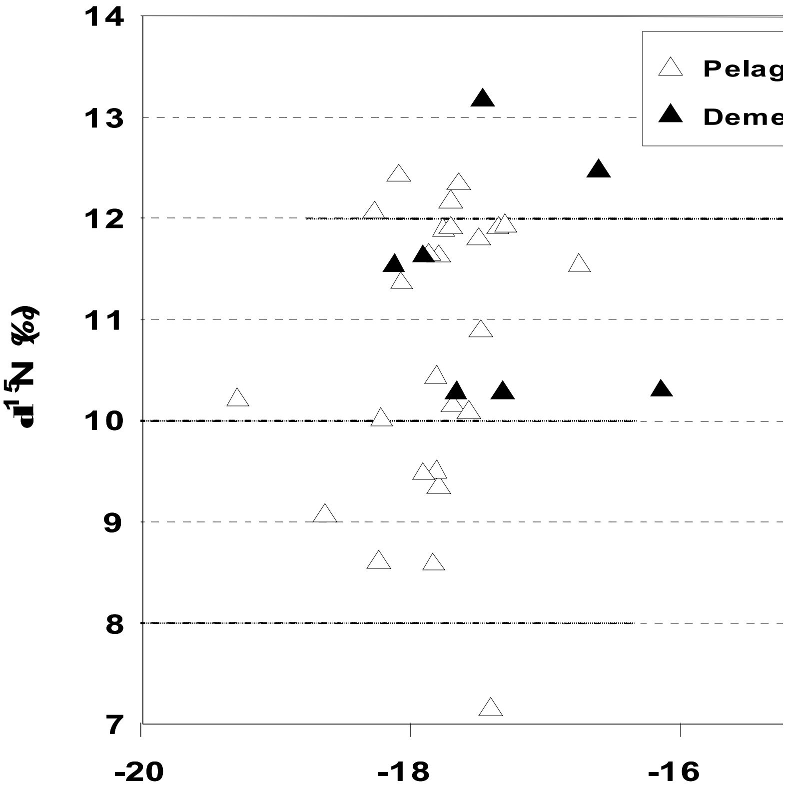

Fig. 4. δ15N and δ13C stable isotope plot of all analyzed species from the Sulu Sea.

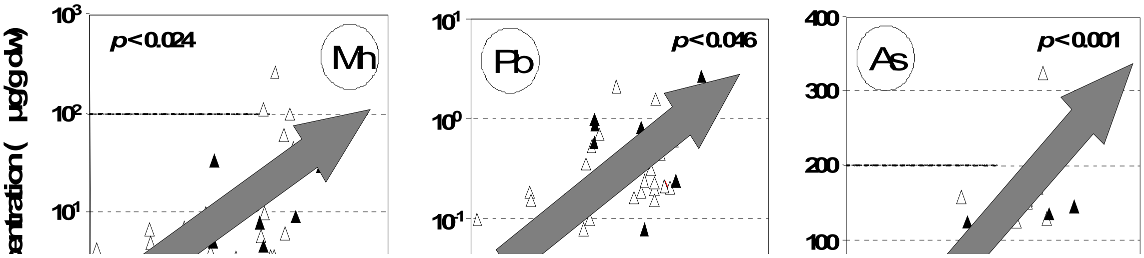

Fig. 5. Correlation between Mn, Pb and As concentrations and ä15N for all analyzed species from the Sulu Sea.

Conclusion

The study has provided information on the contamina-tion status of trace elements in deep sea fish from the Sulu Sea. The variations of trace elements among the species may be due to differences in metal assimilation and metabolic capacity or may be indicative of species-specific accumula-tion. Strontium was the most abundant element in the fish studied. The high Sr concentrations stem from the fact that the whole body of fish was homogenized and, chemically, strontium resembles calcium, which is known to accumulate in bones and hard tissues. Higher concentrations of Sr, Zn, Cu, Se and Cd in fish appeared in deep-demersal species, suggesting the pattern of vertical distribution of these trace elements in the water column in the Sulu Sea.

Manganese, Tl, Pb, Bi, In, Cs and As showed significant positive correlation ($p < 0.05$) with d^{15}N, suggesting that these elements were biomagnified. Rubidium and Cs showed significant positive correlation with d^{13}C, implying that Rb and Cs would originate from offshore waters as oceanic plankton has high d^{13}C.

Comparing results from this study to the dietary standards and guidelines for Hg, Pb, Cu and Zn in fish and shellfish for the Ministry of Agriculture, Fisheries and Food, UK, these levels were not high to warrant concern if they were to be consumed by humans. Although the sample sizes in this study were limited, the trace element concentrations found suggest that anthropogenic loading of toxic elements such as Hg, Cd and Pb to the Sulu Sea is relatively low. It may be inferred that fish from the Sulu Sea are not adversely affected by these toxicants and pose no risk for human health. However, 16.7% of the 36 fish samples had high Cr concentrations when compared with Hong Kong's safe limit of 4 μg/g dw for Cr in sea food. This constitutes a health risk to humans, as Cr is potentially toxic.

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