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Hg, Zn and Cu levels in the muscle and liver of tiger sharks (*Galeocerdo cuvier*) from the coast of Ishigaki Island, Japan: Relationship between metal concentrations and body length

Tetsuya Endo ^{a,*}, Yohsuke Hisamichi ^a, Koichi Haraguchi ^b, Yoshihisa Kato ^c, Chiho Ohta ^d, Nobuyuki Koga ^d

^a Faculty of Pharmaceutical Sciences, Health Sciences University of Hokkaido, 1757 Ishikari-Tobetsu, Hokkaido 061-0293, Japan

^b Daiichi College of Pharmaceutical Sciences, 22-1 Tamagawa-Cho, Minami-Ku, Fukuoka 815-8511, Japan

^c Kagawa School of Pharmaceutical Sciences, Tokushima Bunri University, Sanuki, Kagawa 769-2193, Japan

^d Faculty of Nutritional Sciences, Nakamura Gakuen University, 5-7-1 Beppu, Johnan-Ku, Fukuoka 814-0198, Japan

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ABSTRACT

We analyzed Hg, Zn and Cu concentrations in the liver and muscle of tiger sharks (*Galeocerdo cuvier*) from the coast of Ishigaki Island, Japan. The Hg concentration in the muscle increased proportionally with body length in the tiger sharks, whereas that in the liver increased rapidly after maturity (defined by a length of over 2.7 m). Muscle Hg levels were higher than liver concentrations in immature sharks, with the inverse trend observed in mature sharks. Notably, the Zn and Cu concentrations in the liver tended to decrease with increasing body length. This rapid increase in hepatic Hg concentration concurrent with the onset of maturity in sharks may result from the continuous intake of Hg via food and the slower growth of mature sharks. The high concentrations of the essential metals Zn and Cu in immature sharks may be explained by the physiological demands related to rapid growth.

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1. Introduction

It is well-known that marine predators, particularly long-lived species such as tuna, swordfish, sharks and odontocetes (toothed whales, dolphins and porpoises), accumulate high levels of mercury (Hg) via the food web. These predatory fishes are important marine food resources, and the contamination levels of Hg in their edible parts (muscle), as well as the health risk involved in assimilating this potential toxin, are of great importance to consumers. The Hg concentrations in the liver are higher than muscle concentrations in tuna, swordfish (Storelli et al., 2005; Branco et al., 2007) and odontocetes (Endo et al., 2002, 2004, 2006). However, the available data on Hg levels in the liver and muscle of sharks are limited. The few studies completed thus far indicate a unique distribution pattern of Hg in those organs (Marcovecchi et al., 1991; Hornug et al., 1993; Branco et al., 2007). Positive correlations between Hg concentration in the muscle and body length (weight) have been reported for tuna and swordfish (Storelli and Marcotrigiano, 2001; Adams, 2004; Kojadinovic et al., 2006) and sharks (Walker, 1976; Lyle, 1984, 1986; Marcovecchi et al., 1991; Turoczy et al., 2000; Lacerda et al., 2000; Branco et al., 2004, 2007). Researchers hypothesize that the phenomenon is due to the

E-mail address: endotty@hoku-iryo-u.ac.jp (T. Endo).

increasing Hg burden in aging predators. Interestingly, the liver has historically escaped close scrutiny since it is not regarded as an edible organ. Therefore, only limited information is available regarding the relationship between Hg concentration in the liver and body length of this predator.

Cadmium (Cd) accumulation may be the highest in molluscs, particularly cephalopods, due not only to the inherent hierarchy of the marine food web but also to species-specific physiologic mechanisms (Honda, 1990; Das et al., 2003). Squid is a significant source of Cd for predatory fish and marine mammals (Honda, 1990; Holsbeek et al., 1998; Das et al., 2003). The concentrations of the essential metals zinc (Zn) and copper (Cu) in the liver and muscle of marine mammals and fish tend to decrease during the growth period (Honda et al., 1983; Storelli and Marcotrigiano, 2000; Zhang and Wang, 2005; Endo et al., 2007). Essential metal levels in the livers of marine mammals, as well as hepatic Hg and Cd concentrations, tend to increase after the growth period (Das et al., 2000; Endo et al., 2002, 2007). However, no studies focusing on fluctuating concentrations of these metals in the shark liver or the correlation of this change with body length have yet been reported.

Liver and muscle levels of toxic metals Hg and Cd, as well as essential metals Zn and Cu, vary in sharks not only with body length (age), but also with species, sex, feeding habits, habitat and season (Walker, 1976; Hornug et al., 1993; Lacerda et al.,

^{*} Corresponding author. Tel./fax: +81 133 23 3902.

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2000; Turoczy et al., 2000; McMeans et al., 2007). The complexity of the issue demands a large-scale survey with well-designated conditions to elucidate the body length-dependent changes in the concentrations of these metals in the liver and muscle of sharks. On August 31st and September 1st, 2007, a large-scale culling of sharks was undertaken off the coast of Ishigaki Island, Japan (see Fig. 1). We obtained liver and muscle samples from 42 tiger sharks (*Galeocerdo cuvier*), eight silvertip sharks (*Carcharhinus albimarginatus*), a bull shark (*Carcharodon leucas*) and a sandbar shark (*Carcharhinus plumbeus*), together with information on body size (length and weight) and gender for each sample.

Here, we report data gathered on Hg, Cd, Zn, Cu and iron (Fe) in the liver and muscle tissue obtained from tiger and silvertip sharks culled off the coast of Ishigaki Island, Japan. Our analysis elucidates the relationships between the metal concentrations in those tissues and body length; we also benefit from a comparison of data gathered on the bull and sandbar sharks in contrast to those for tiger and silvertip sharks from the same cull.

2. Materials and methods

2.1. Sampling

Liver and muscle samples from sharks culled off the coast of Ishigaki Island, Japan were collected on August 31st and September 1st, 2007. These samples from 52 sharks were stored at -20 °C until analysis.

2.2. Chemical analyses

Total Hg (Hg) in the shark samples was analyzed using a flameless atomic absorption spectrophotometer (Hiranuma Sangyo Co. Ltd., HG-1) after digestion with a mixture of HNO₃, HClO₄ and H₂SO₄ (Endo et al., 2002). Cd, Zn, Cu and Fe were analyzed using a Z-8100 Hitachi Polarized Zeeman flame atomic absorption spectrophotometer after digestion with HNO₃ and HClO₄ (Endo et al., 2002). As reported previously (Endo et al., 2002, 2004, 2007), DOLT-2 (National Research Council of Canada) and CRB 463 (BCR, European Commission) were used as analytical quality control samples for the determination of the metals. All metal concentrations are presented here on a wet weight basis, and expressed by mean \pm standard deviation (SD). Body size (total body length and weight) and gender of the sharks, as well as the analytical results for metals in the liver and muscle samples (mean of duplicate or triplicate analyses), are shown in Table 1.

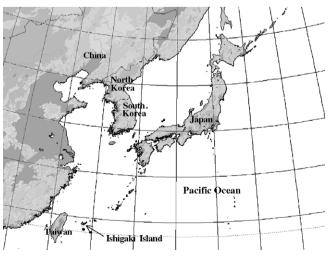


Fig. 1. Map of Japan.

2.3. Statistical analyses

The data were analyzed using a Student's *t*-test and Pearson's correlation coefficient test, using the Statcell program. The level of significance was set at p < 0.05.

3. Results and discussion

3.1. Body size and gender of analyzed shark species

Table 2 summarizes the body size and gender of the shark species included in this study and the analytical data for heavy metals. No particular sex-associated differences in body length, weight or metals profile in liver (n = 42) and muscle (n = 41) of the tiger sharks were observed. Among the 11 largest tiger sharks (above 2.29 m and 163 kg) shown in Table 1, however, nine were female and only two were male. The body size of females has been reported to be larger than that of males in five shark species caught in Australian waters (Lyle, 1984) and in four shark species caught off the coast of Brazil (De Pinho et al., 2002), probably due to faster growth in females. However, no detailed information on the growth of male and female tiger sharks is currently available. Nor were particular sex-associated differences observed in the silvertip sharks, although the number of male sharks in the sample population was limited (n = 2).

3.2. Correlation between total mercury concentration and body length

The highest concentrations of Hg in liver and muscle samples were found in the bull shark (28.1 and 3.65 μ g/wet g, respectively), which is a well-known predator (Table 2). The second highest concentration of Hg in liver samples summarized in Table 2 occurred in the sandbar shark (3.62 μ g/wet g, n = 1). The third highest Hg concentration occurred in the tiger shark liver samples: the average Hg concentration in the liver samples was 1.17 ± 3.14 µg/wet g (n = 42) with a wide range (0.11-20.1 µg/wet g), because the six largest tiger sharks (IG07-69, -33, -97, -30, -6 and -96) contained considerable levels of Hg (Tables 1 and 2). In contrast, the average Hg concentration in muscle samples from the tiger sharks was $0.78 \pm 0.29 \,\mu\text{g/wet g}$ (*n* = 41) with a relatively narrow range $(0.38-1.34 \mu g/wet g)$. Excluding the liver samples from the six largest tiger sharks, the average Hg level in the liver samples from this group $(0.33 \pm 0.19 \,\mu\text{g/wet g}, n = 36)$ was lower than that in muscle samples (0.70 \pm 0.22 μ g/wet g, *n* = 35). The average Hg level in liver samples from the silvertip sharks $(0.70 \pm 0.42 \,\mu\text{g/wet g})$ n = 8) was also lower than that in muscle samples $(1.80 \pm 0.45 \ \mu\text{g/wet g}, n = 8)$. In contrast, the Hg concentrations in liver samples from the bull shark and the sandbar shark (28.1 and $3.62 \mu g/wet g$) were higher than those in the corresponding muscle samples (3.65 and 1.66 μ g/wet g).

The relationships between Hg concentrations in liver and muscle samples and body length in the tiger sharks are shown in Fig. 2. As the body length of sexually mature tiger sharks may range from 2.3 to 2.9 m in males and from 2.5 to 3.3 m in females, and the average body length of a mature shark may be 3.3 m (Florida Museum Natural History, 2008), most of the tiger sharks analyzed in this study (1.19–3.20 m) appear to be immature animals.

The Hg concentration in the muscle samples from the tiger sharks increased proportionally with body length (p < 0.05). This phenomenon has been previously observed in the muscles of other shark species (Walker, 1976; Lyle, 1984, 1986; Marcovecchi et al., 1991; Lacerda et al., 2000; Turoczy et al., 2000; Branco et al., 2004, 2007). In contrast, the Hg concentration in the liver samples rapidly increased in sharks >2.7 m in body length. Extensive literature

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