

## Mercury Content in Sharks

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Prior to the occurrence of the Minamata disaster in Japan in which 46 people were killed and nearly 100 seriously injured by eating fish and shellfish contaminated by an alkylmercury compound (KURLAND et al. 1960) there had been few attempts to determine mercury levels in fish. However, RAEDER & SNEKVIK in 1941 reported levels in freshwater and marine fish from contaminated areas markedly higher than those found in uncontaminated waters. From 1964 to 1967, studies were conducted on fish in Sweden, and mercury concentrations of several ppm were reported, as was the observation that fish were found to contain mercury higher than that noted in waters from which they had been caught (VOSTEL 1972). Also, positive correlation between mercury concentrations in muscle tissue to total weight and age was confirmed (BACHE et al. 1971).

This paper reports the analytical results relating to total mercury concentrations in two species of sharks taken from Hawaiian marine waters.

### METHODS AND MATERIALS

Shark samples were obtained through the courtesy of the Hawaii State Department of Fish and Game in April, 1971, and were taken from the Pacific Ocean by long-line fishing within 20 mi of the coast of Hawaii. Twenty-six muscle and liver samples of the Tiger Shark, Galeocerdo cuvieri were analyzed from 13 individuals for total mercury content, as were 52 muscle and liver samples, representing 26 individuals of the shark, Carcharhinus limbatus.<sup>1</sup> All muscle samples (500-1500 g) were obtained from the median dorsal area, contained no skin or cartilage, and were maintained frozen until sample preparation for analysis by thawing, then homogenizing 250 g with a Waring blender. No water was added to the tissue. Liver samples were similarly prepared.

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<sup>1</sup>This species, included in a revision of the genus Carcharhinus by Garrick (1982), was previously thought to be restricted to the Atlantic Ocean, but is now recognized as being cosmopolitan in distribution.

Total mercury levels in both muscle and liver samples were analyzed by first using the digestion procedures of "wet ashing", followed by determination with cold vapor atomic absorption developed by HATCH & OTT (1968). Five grams of homogenate from each sample were digested in concentrated sulfuric-nitric acid (4 to 1) and further oxidized with concentrated nitric acid if charring occurred. Following digestion, excess oxidizing agents were reduced with a sodium chloride-hydroxylamine solution. Mercury ions ( $\text{Hg}^{++}$ ) were then reduced to elemental mercury ( $\text{Hg}^0$ ) with stannous sulfate solution, and the mercury vaporized in the flameless atomic absorption apparatus. Depending on mercury content, a 5 to 25 mL aliquot of the digest was placed in the sample reservoir of the "closed-loop" mercury analysis system together with 50 mL reducing solution plus sufficient distilled  $\text{H}_2\text{O}$  to bring the total volume to 100 mL. The system was then immediately closed, and the mixture stirred for 1 min after which airflow was initiated and maximum absorption at 2537Å noted. Standard operating conditions were employed, and mercury content calculated by comparison of sample responses to a standard curve derived from a series of  $\text{HgCl}_2$  standards. Subsamples were submitted for analysis to the WARF Laboratory, Madison, Wisconsin and to the Laboratory of Hygiene of the Wisconsin State Board of Health, Madison, Wisconsin for comparison purposes. The results showed good agreement with values obtained from our work to those obtained from the two independent laboratories. Maximum deviation was 0.05 ppm, which represented a difference of 6%.

Each shark was identified, measured and sexed (C. limbatus) when brought on board. Additionally, muscle and liver tissue samples were then secured, placed in polyethylene bags and placed in crushed ice.

## RESULTS AND DISCUSSION

Muscle and liver tissue samples were secured from each of 13 Tiger Sharks, Galeocerdo cuvieri and analyzed for mercury (expressed as ppm wet weight). A linear regression analysis was then conducted to determine if there was a correlation between mercury level and body length. As shown in Figure 1, a positive correlation was obtained in the regression of mercury concentration in both muscle and liver tissue to body length. Correlation coefficients for these relationships are shown in Table 1. A linear regres-

sion analysis was also conducted to determine if any correlation exists between mercury levels in muscle and liver tissue. Again, a positive correlation was observed (Table 2). Assuming that an increase in body length reflects a proportional increase in age, then it would appear that mercury is sequestered in slightly less amounts in liver vs muscle tissue in a gradual, additive manner. Other possible explanations exist such as alteration of food habits and environmental habitat shifts with increasing size. Unfortunately, we do not have the details of movement and food intake to speculate on the possible effects of food chain magnification.

As in the previous species, muscle and liver tissue samples were obtained from each of 26 sexed sharks (Carcharhinus limbatus) and analyzed for total mercury content. Again, a linear regression analysis was conducted (Figure 2), and a positive correlation found in respect to muscle and liver tissue Hg concentration to body length. Correlation coefficients of these relationships are shown in Table 3. A linear regression analysis was also conducted to determine if there was a correlation between mercury levels in muscle tissue with mercury levels in liver tissue (Table 4). Again, a positive correlation was noted. In addition, an attempt was made to determine if there was any statistically significant difference in the muscle tissue mercury levels between male and female sharks. A comparison of the average of the level of contamination (expressed as per inch/fish) in each of the individuals representing each sex was conducted. A one-factor analysis of variance was performed to determine significance of the difference between the two means. It was found that there was no significant difference in the level of total Hg between the sexes.

Finally, the same procedure was employed to determine if there was any significant difference in the level of mercury content between Galeocerdo cuvieri and C. limbatus (Table 5). From the high F-ratio it is apparent that the level of mercury contamination in C. limbatus was significantly higher than in G. cuvieri.

In both species of shark we observe a positive correlation between increase in body length and an increase in mercury concentration, as well as a similar parallel between mercury levels in both liver and muscle tissue. Again, we have little published information on the life history of C. limbatus which might shed some light on the higher Hg levels present as compared to the tiger shark. In this case, size is clearly not

Table 1. Correlation coefficients of relationships between total mercury in muscle and liver tissue and body length for Galeocerdo cuvieri.

No. of Data Pairs	Tissue	Correlation Coefficient	Slope	Y - Intercept
13	Muscle	0.846**	0.005	-0.136
13	Liver	0.714**	0.002	-0.039

\*\*p 0.01 Highly Significant

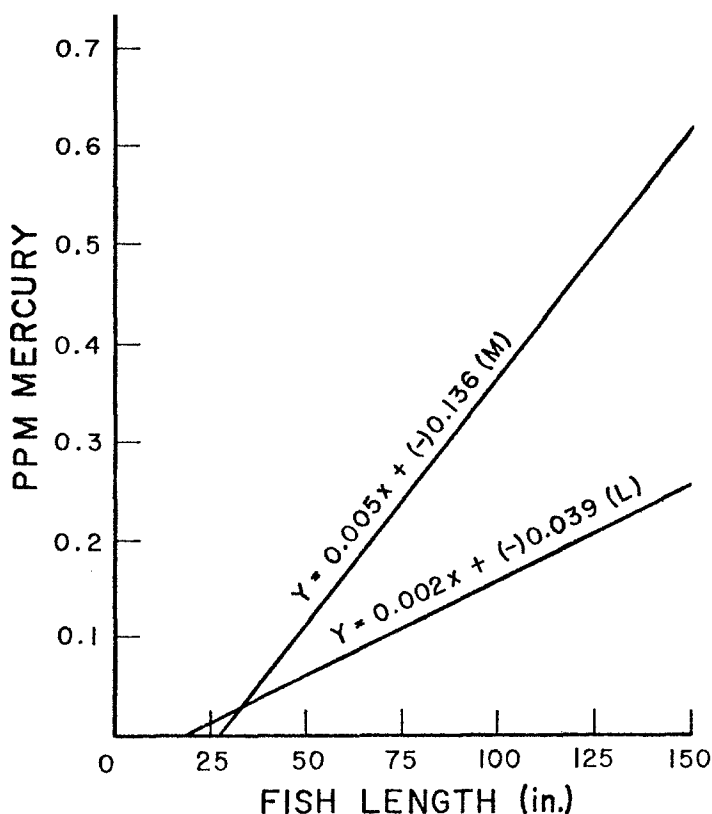


Figure 1. Regression curves describing the relationship of total mercury concentrations (ppm) in muscle (M) and liver (L) tissues of Galeocerdo cuvieri to body length.

Table 2. Data Summary: Regression of mercury concentration (ppm) in muscle tissue on mercury concentration (ppm) in liver tissue of Galeocerdo cuvieri.

No. of Data Pairs	Correlation Coefficient	Slope	Y - Intercept
16	0.872**	0.597	-0.033

\*\*p 0.01 Highly Significant

Table 3. Correlation coefficients of relationships between total mercury in muscle and liver tissue and body length for Carcharhinus limbatus.

No. of Data Pairs	Tissue	Correlation Coefficient	Slope	Y - Intercept
26	Muscle	0.762**	0.026	-0.658
26	Liver	0.642**	0.160	-6.993

\*\*p 0.01 Highly Significant

Table 4. Data Summary: Regression of mercury concentration (ppm) in muscle tissue on mercury concentration (ppm) in liver tissue for Carcharhinus limbatus.

No. of Data Pairs	Correlation Coefficient	Slope	Y - Intercept
26	0.753**	5.530	-2.466

\*\*p 0.01 Highly Significant

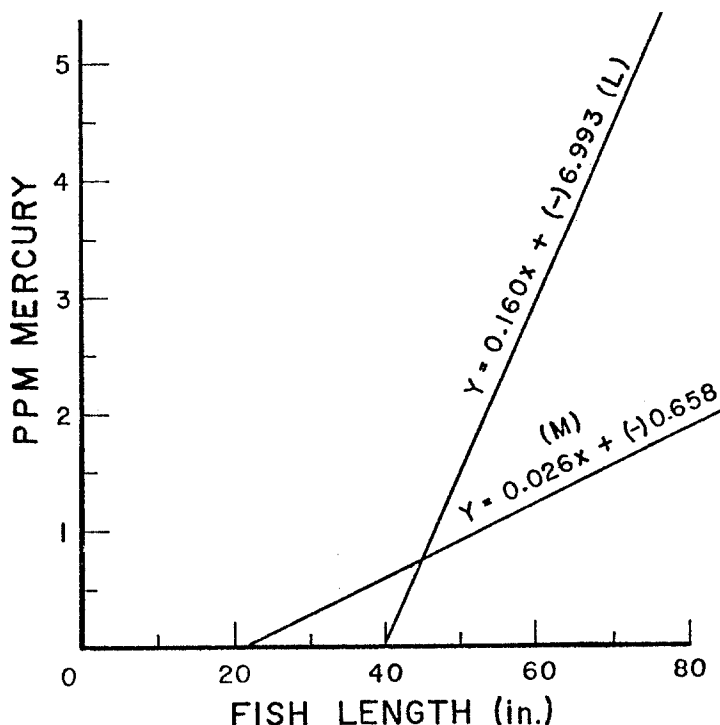


Figure 2. Regression curves describing the relationship of total mercury concentrations (ppm) in muscle (M) and liver (L) tissues of Carcharhinus limbatus to body length.

Table 5. Comparison of the level of total mercury contamination observed in the muscle tissues of Galeocerdo cuvieri and Carcharhinus limbatus. The mercury level in each fish (ppm) was divided by the fish length (in.); the mean of the quotients within each group was calculated.

Species	No. Samples Analyzed	Hg Conc. (ppm) Divided by Fish Length (in.) x 100			F-Ratio
		Range	Mean	Std. Dev.	
<u>G. cuvieri</u>	13	2.09-6.14	4.02	1.08	41.92**
<u>C. limbatus</u>	26	7.29-28.22	12.93	4.87	

\*\*p 0.01 Highly Significant

the issue. As the liver is very important in the metabolism and elimination of many toxicants, a higher concentration of mercury in the liver than in corresponding muscle tissue might suggest either recent exposure followed by concentration in the liver, or strong protein binding, perhaps in a similar manner as has been demonstrated with cadmium by LUCIS et al. (1970). From these data it would appear that in spite of considerable variability, the mercury levels in the livers of C. limbatus run slightly higher than corresponding muscle tissue whereas in G. cuvieri the opposite appears to be the case.

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