

Mercury and Selenium Levels in Striped Dolphins Caught off the Pacific Coast of Japan

Kazuomi ITANO, Shin'ichiro KAWAI, Nobuyuki MIYAZAKI,*
Ryo TATSUKAWA** and Toraya FUJIYAMA***

Osaka City Institute of Public Health and Environmental Sciences,
8-34, Toho-cho, Tennoji-ku, Osaka 543, Japan

*Department of Zoology, National Science Museum,
3-23-1, Hyakunin-cho, Shinjuku-ku, Tokyo 160, Japan

**Department of Environment Conservation, Ehime University,
3-5-7, Tarumi, Matsuyama 790, Japan

***Department of Marine Sciences, University of the Ryukyus,
Nishihara-cho, Okinawa 903-01, Japan

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We examined the distribution of mercury and selenium in fifteen tissues of striped dolphins (*Stenella coeruleoalba*). The total mercury level in the mature dolphins showed differences among the tissues and was highest in the liver. The total mercury concentration in most tissues increased with age, and reached a constant level at 20 to 25 years of age. The total mercury level in the tissues was not appreciably different among mature males, pregnant females, lactating females and resting females. In the muscle of mature individuals, the total mercury level of striped dolphins collected in 1977 and 1980 was appreciably higher than that of those collected 1978 and 1979. Methylmercury showed less variation in concentration among the tissues. The ratio of methylmercury to total mercury in muscle decreased with growth after about 10 years of age when the increase of methylmercury stopped. Selenium levels in the dolphins increased with age as total mercury levels did. High correlation coefficients were found between the total mercury and selenium levels in spleen, muscle, pancreas and liver. The concentrations of total mercury in the various tissues of immature dolphins were much lower than those of mature ones.

Seals and dolphins as higher-order carnivores have been reported to concentrate much mercury from fish and shellfish, and to be able to tolerate it with no apparent symptoms of poisoning.^{1~3)} It has also been reported that the total mercury concentration in the livers of marine mammals is significantly correlated with that of selenium.^{2~4)} A similar correlation was observed in the livers of seals from fresh and brackish water.⁵⁾ Koeman *et al.*⁶⁾ suggested that selenium in marine mammals might have a protective effect against the toxic action of mercury compounds.

Seals from industrial areas, or from areas adjacent to these areas, have been reported to contain higher levels of mercury than those from other areas.⁷⁾ Thus, marine mammals appear to be a useful indicator of the level of

mercury accumulated in the environment. However, little has been reported about the mercury contents of tissues in marine mammals off the coasts of Japan, except for a report by Arima and Nagakura⁸⁾ on the muscle of toothed cetaceans. In this paper, the levels of total mercury and methylmercury are reported in various tissues and at different growth stages of striped dolphins, *Stenella coeruleoalba* (Meyen, 1833), collected off the Pacific coast of Japan. The relationship between mercury and selenium concentrations was also examined.

MATERIALS AND METHODS

Fifty-five striped dolphins were caught by a driving method at Kawana on Izu Peninsula in October 1977 and

at Taiji on Kii Peninsula in December 1978, 1979 and 1980. The samples taken from different tissues were stored at -20°C and homogenized before analysis. Females having ovaries with a corpus luteum or albicans and males having testes of 15.5 g or more were classified as mature animals.⁹⁾ The age was estimated from the number of growth layers in the dentine or cementum of the teeth.¹⁰⁾

Total mercury¹¹⁾: Ten to one hundred milligrams of samples were weighed in a quartz glass boat and covered with a sodium oxide-calcium oxide mixture and activated alumina as the agent for ashing. Total mercury was analyzed using a semi-automated flameless atomic absorption spectrophotometer. The recovery of total mercury was more than 95%. **Methylmercury¹²⁾:** Two to five grams of samples were homogenized with 15 ml of 30% sodium chloride solution and mixed with an equal volume of 6 N hydrochloric acid. Methylmercury chloride was extracted with benzene, transferred to a cysteine solution and re-extracted with benzene. Methylmercury in the benzene extract was analyzed by a gas chromatograph equipped with an electron capture detector. All the values measured were corrected for the average recovery of 70% in this procedure. **Selenium¹¹⁾:** Two to five grams of samples were digested in a concentrated nitric-perchloric acid mixture. Selenium was complexed with 2,3-diaminonaphthalene after interfering elements were masked with disodium ethylenediaminetetraacetate. The selenium complex was extracted with cyclohexane and determined fluorometrically. The recovery of selenium was approximately 90%. The concentrations of total mercury, methylmercury and selenium were expressed as $\mu\text{g/g}$ fresh weight.

RESULTS

Mean levels of total mercury, methylmercury and selenium in various tissues

Table I shows the concentrations of total mercury, methylmercury and selenium in various tissues of mature dolphins (more than 8.5 years of age). The liver showed the highest total mercury level (maximum: $475 \mu\text{g/g}$). The variability of total mercury concentration was larger than that of methylmercury. The concentration of methylmercury was slightly higher in the liver (maximum: $10.6 \mu\text{g/g}$) than in the other tissues, but the methylated fraction of mercury present in liver was the lowest (maximum: 5.6%). The concentrations of selenium were also the highest in liver (maximum: $127 \mu\text{g/g}$), followed by spleen, kidney, lung, pancreas, cerebrum, second stomach, muscle, heart, first stomach, large intestine, diaphragm, blubber and cerebellum, in that order. The order of selenium levels was almost the same as that of total mercury levels.

The concentrations of total mercury, methylmercury and selenium in eight different tissues of immature dolphins (aged between 1.6 to 2.2 years) are shown in Table II. The

TABLE I. TOTAL MERCURY, METHYLMERCURY AND SELENIUM LEVELS IN TISSUES OF MATURE STRIPED DOLPHINS COLLECTED FROM 1977 TO 1980

Tissue	$\mu\text{g/g}$ (Fresh tissue, mean \pm S.D.)			
	N	T-Hg	M-Hg	Se
Blubber	18	3.5 ± 1.4	0.34 ± 0.19 (9.7)	1.4 ± 0.8
Muscle	26	15.2 ± 8.0	5.3 ± 0.8 (35)	2.8 ± 2.2
Heart	15	6.1 ± 1.6	3.7 ± 1.2 (61)	2.7 ± 1.2
Pancreas	14	15.2 ± 10.8	1.5 ± 0.5 (9.9)	4.3 ± 3.1
Spleen	14	22.6 ± 16.1	2.3 ± 0.5 (10)	5.8 ± 2.7
Kidney	14	14.7 ± 6.6	3.2 ± 0.8 (22)	5.6 ± 2.2
Liver	15	205 ± 102	7.0 ± 1.8 (3.4)	48.6 ± 28.7
First stomach	15	7.2 ± 2.5	2.2 ± 0.5 (31)	2.2 ± 0.8
Second stomach	15	8.2 ± 4.2	2.0 ± 0.7 (24)	3.5 ± 1.3
Large intestine	15	6.1 ± 2.0	1.7 ± 0.7 (29)	2.0 ± 0.6
Lung	15	17.1 ± 14.0	1.3 ± 1.3 (7.6)	5.1 ± 2.5
Diaphragm	15	10.1 ± 4.5	4.6 ± 2.1 (46)	1.8 ± 1.0
Cerebrum	7	9.7 ± 6.2	1.1 ± 0.3 (11)	3.7 ± 2.4
Cerebellum	7	2.3 ± 1.2	0.81 ± 0.15 (35)	0.85 ± 0.38
Blood	9	1.2 ± 0.2		0.87 ± 0.19

N, number of samples analyzed; T-Hg, total mercury; M-Hg, methylmercury; Se, selenium. In parentheses, the percentages of methylmercury to total mercury are shown.

TABLE II. TOTAL MERCURY, METHYLMERCURY AND SELENIUM LEVELS IN TISSUES OF IMMATURE STRIPED DOLPHINS COLLECTED IN 1977 AND 1978

Tissue	$\mu\text{g/g}$ (Fresh tissue, mean \pm S.D.)					
	N	T-Hg	N	M-Hg	N	Se
Blubber	6	0.12 ± 0.18	4	0.022 ± 0.015 (18)	6	2.22 ± 0.72
Muscle	6	1.27 ± 0.22	4	1.22 ± 0.24 (96)	6	0.88 ± 0.25
Heart	4	1.30 ± 0.49	3	1.08 ± 0.66 (83)	4	3.08 ± 0.84
Pancreas	5	0.47 ± 0.09	4	0.38 ± 0.08 (81)	5	1.44 ± 0.34
Spleen	5	0.77 ± 0.28	4	0.56 ± 0.14 (73)	5	3.34 ± 0.82
Kidney	5	3.38 ± 1.30	4	1.22 ± 0.69 (36)	5	5.82 ± 0.94
Liver	6	5.81 ± 2.76	4	1.65 ± 1.01 (28)	6	1.97 ± 1.03
First stomach	5	0.95 ± 0.22	4	0.80 ± 0.10 (84)	5	1.62 ± 0.72

See the footnote to Table I.

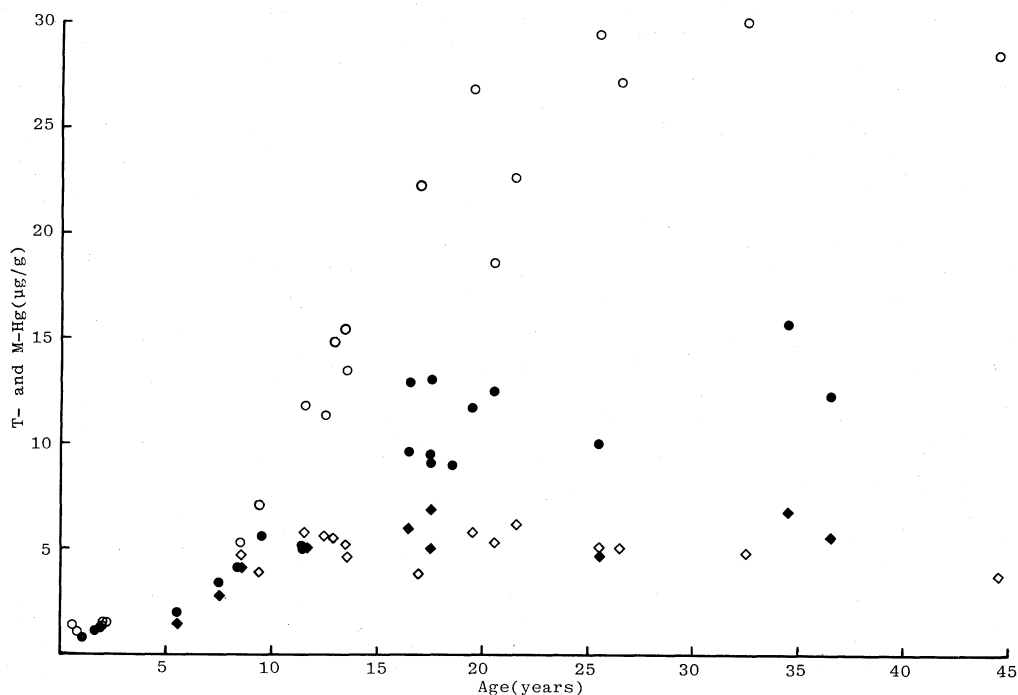


FIG. 1. Changes of Mercury Levels in Muscle of Striped Dolphins with Age.

○, total mercury (1977 and 1980 groups); ●, total mercury (1978 and 1979 groups); ◇, methylmercury (1977 and 1980 groups); ◆, methylmercury (1978 and 1979 groups).

total mercury level was slightly higher in liver (maximum: $9.35 \mu\text{g/g}$) than in the other tissues. The ratios of methylmercury to total mercury in blubber, liver and kidney were lower than those of the other tissues. The selenium level was slightly higher in kidney (maximum: $6.90 \mu\text{g/g}$) compared with those in other tissues.

Changes in mercury and selenium levels with age

Total mercury levels in the muscle and liver increased with age (Figs. 1 and 2). The concentration in muscle leveled off above 20 years of age. The muscle concentration of total mercury showed a difference between the two groups of samples over 10 years of age. The constant level of total mercury in the muscle of

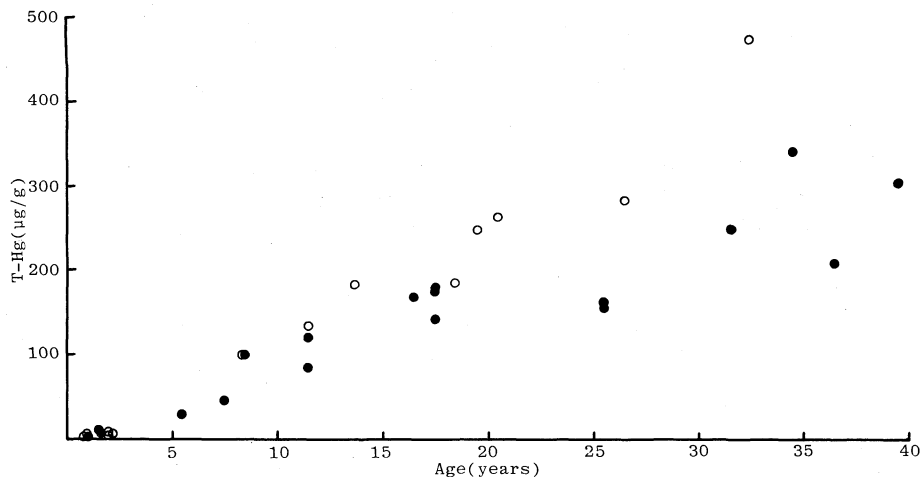


FIG. 2. Changes of Total Mercury Levels in Liver of Striped Dolphins with Age.

○, 1977 group; ●, 1978 and 1979 groups.

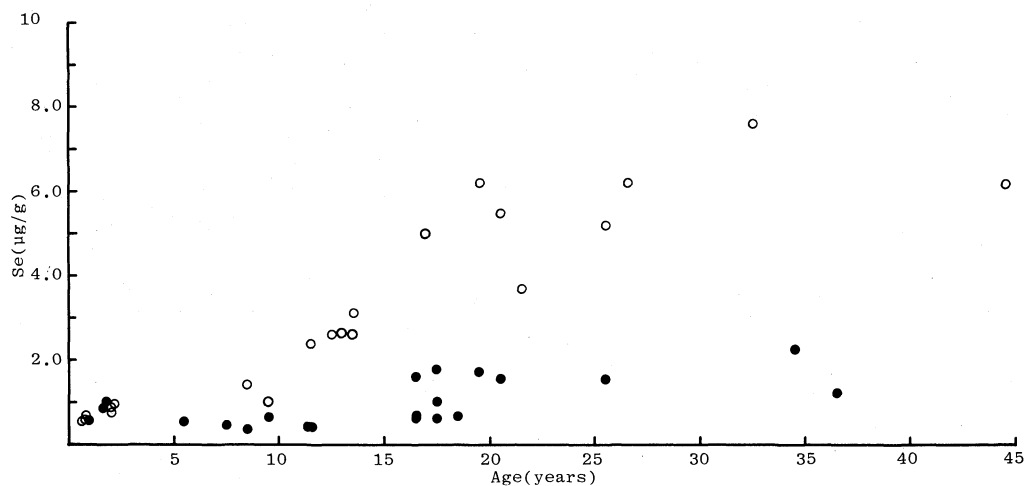


FIG. 3. Changes of Selenium Levels in Muscle of Striped Dolphins with Age.

○, 1977 and 1980 groups; ●, 1978 and 1979 groups.

the 1977 and 1980 samples was about twice as high as that of the 1978 and 1979 ones. However, in other tissues, the levels of total mercury were not appreciably different between the two groups. As shown in Fig. 1, the methylmercury concentration reached a constant level (about $6 \mu\text{g/g}$) in muscle over 10 years of age, and did not show any significant differences among the samples collected in different years. No appreciable differences in the total mercury level were found among mature males, pregnant females, lactating

females and resting females.

The changes of selenium levels in the muscle and liver with age are presented in Figs. 3 and 4. In muscle, an increase of the selenium concentration with age could be observed more clearly in the 1977 and 1980 samples than in the 1978 and 1979 ones. The selenium level in the liver of the 1977 samples was also slightly higher than that of the 1978 and 1979 samples. The level in the blubber of the 1977 samples remained constant from the calf stage, while the level in the blubber of the 1978

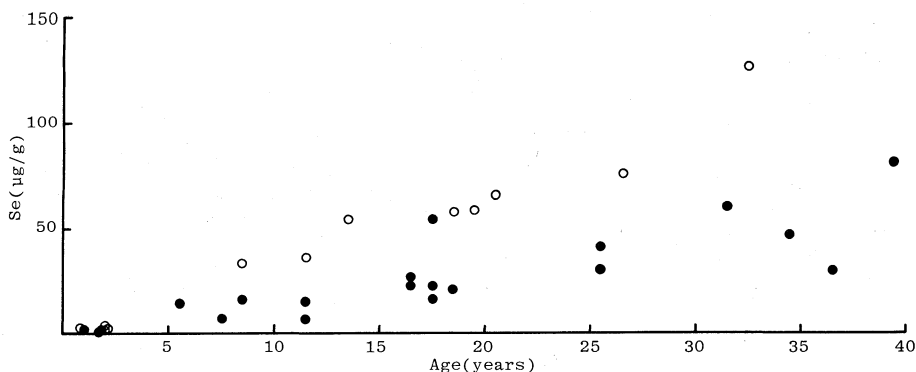


FIG. 4. Changes of Selenium Levels in Liver of Striped Dolphins with Age.

○, 1977 group; ●, 1978 and 1979 groups.

TABLE III. RELATIONSHIPS BETWEEN TOTAL MERCURY AND SELENIUM LEVELS IN TISSUES OF MATURE STRIPED DOLPHINS

Tissue	N	r Value	Equation	p Value
Blubber	22	0.14	Not significant	<0.6
Muscle	31	0.94	$Y = 3.51X + 5.17$	<0.001
Heart	15	0.74	$Y = 1.14X + 2.95$	<0.01
Pancreas	15	0.89	$Y = 4.04X + 1.17$	<0.001
Spleen	16	0.99	$Y = 5.00X + 6.12$	<0.001
Kidney	20	0.39	Not significant	<0.1
Liver	20	0.81	$Y = 2.66X + 87.67$	<0.001
First stomach	15	0.52	$Y = 1.65X + 3.62$	<0.05
Second stomach	15	0.29	Not significant	<0.3
Large intestine	15	0.75	$Y = 2.51X + 1.07$	<0.01
Lung	15	0.63	$Y = 3.56X - 1.85$	<0.02
Diaphragm	15	0.61	$Y = 2.80X + 5.14$	<0.02
Cerebrum	7	0.77	$Y = 2.03X + 1.67$	<0.05
Cerebellum	7	0.82	$Y = 2.66X + 0.07$	<0.02

X and Y show selenium and total mercury levels, respectively. p value, the mathematical probability estimated from Student's *t* test.

samples slowly decreased with age (data not shown). In other tissues, no significant difference was found between the two samples. However, the selenium concentration in the blubber of the 1978 samples was higher in pregnant females than in lactating females and mature males (data not shown).

Correlation between mercury and selenium

Correlations between total mercury and selenium contents in various tissues of mature dolphins are shown in Table III. Correlation coefficients higher than 0.7 were observed in eight tissues. The regression equations of total

mercury on selenium are also presented in Table III. The relationships between total mercury and selenium in muscle and liver are shown graphically in Figs. 5 and 6. The atomic weight ratio of mercury to selenium is 2.54. Therefore, the slope values of the equations for spleen, pancreas and muscle were above 1.0 on a molar basis.

DISCUSSION

Kasuya¹³⁾ and Miyazaki *et al.*¹⁴⁾ conducted detailed biological studies on striped dolphins distributed off the Pacific coast of Japan. It is

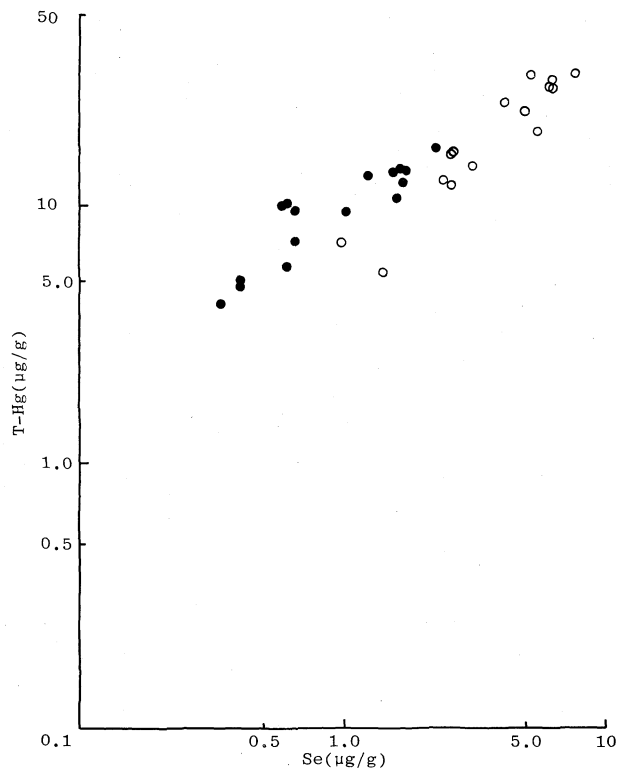


FIG. 5. Relationship between Total Mercury and Selenium in Muscle of Mature Striped Dolphins. ○, 1977 and 1980 groups; ●, 1978 and 1979 groups.

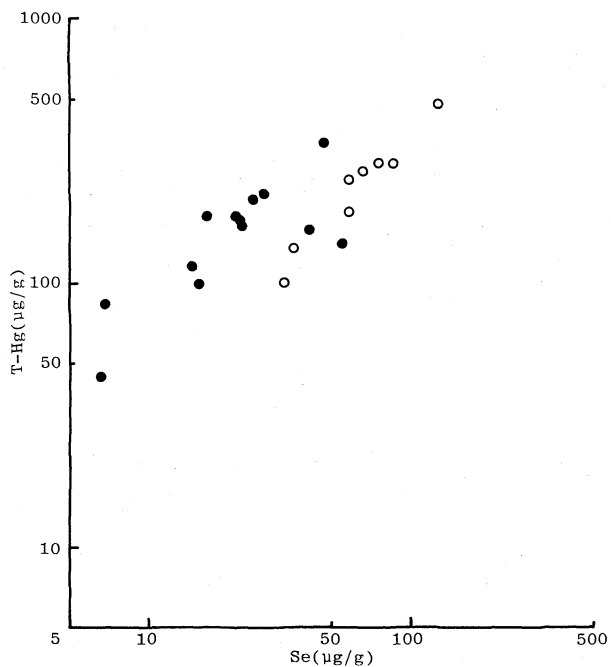


FIG. 6. Relationship between Total Mercury and Selenium in Liver of Mature Striped Dolphins. ○, 1977 group; ●, 1978 group.

one of the toothed cetaceans whose age determination has been well studied.

The total mercury concentrations in muscle, liver and blubber of adult gray and common seals off eastern Canada^{15,16)} were 1~2, 10~50 and 0.1~0.2 $\mu\text{g/g}$ wet weight, respectively. In an adult gray seal from an industrial area,⁷⁾ the concentrations in muscle and blubber were 5.9 and 1.4 $\mu\text{g/g}$, respectively. These values were lower than those of striped dolphins. Fur and nails of gray seals contain higher levels of mercury than muscle does.¹⁵⁾ These tissues do not exist in dolphins. The fur and nails of seals might contribute to excretion of mercury as hair does in humans. However, livers and brains of adult common and harbor seals from mercury-polluted areas contained 100 to 800 and 5 to 30 $\mu\text{g/g}$ of mercury, respectively.^{2,7)} These values were comparable with those of striped dolphins (209~475 and 1.5~19.8 $\mu\text{g/g}$). The concentration of total mercury in muscle, liver, kidney and cerebrum of harbor porpoises from Canada³⁾ were somewhat lower than those of striped dolphins. Differences in total mercury concentration between harbor porpoises and striped dolphins appear to arise from differences in specimen age, length of life and food level of mercury.

Miyazaki *et al.*¹⁴⁾ suggested that the striped dolphin usually migrates along the Pacific coast of the Kii Peninsula to Hokkaido following the movement of the Kuroshio current. In the striped dolphin, the total mercury level in muscle was found to be appreciably different between the 1977 and 1980 samples, and the 1978 and 1979 samples. The mercury levels in muscle of 1978 and 1979 samples were also slightly higher than those of the 1974 samples reported by Arima and Nagakura.⁸⁾ As the 1974 samples were collected at Kawana, as were the 1977 samples, mercury levels in the muscle appear to be independent of the location where the specimens were collected. This implies that the mercury content in food affects the mercury level accumulated in the muscle of striped dolphin. However, the appreciable difference of mercury levels in the

muscle among the groups collected in different years remains unexplained.

The muscle of marine fishes usually contains most of the mercury as methylmercury and a constant level of selenium exceeding the content of mercury.¹¹⁾ About 80% of the total mercury in the muscle of seals is present in the methylated form.^{6,15)} For the muscle of striped dolphins below 10 years of age, 70 to 100% of the total mercury was found in the methylated form. However, the content of methylmercury in the muscle of striped dolphins was saturated above 10 years of age and the ratio of methylmercury to total mercury decreased with age (minimum: 20%). A similar tendency has been observed in the ordinary muscle of the blue marlin¹⁷⁾ and the muscle of short-finned pilot whales.¹⁸⁾ The low ratios of methylmercury to total mercury in the liver, kidney and cerebrum of toothed cetaceans^{3,18)} and harbour seals¹⁾ are comparable with the values in striped dolphins, but the ratio in liver is slightly higher in some seals¹⁹⁾ than in striped dolphins. Since most of the methylmercury introduced into the stomach of mice was absorbed in the gastrointestinal tract, though only a small percentage of inorganic mercury is absorbed,²⁰⁾ almost all of the mercury taken up in the striped dolphin is methylmercury. The methylmercury taken up is transferred to the tissues *via* the blood.⁷⁾ The above information suggests that the striped dolphin may have a demethylation mechanism. It has been observed that in harp seals dosed orally with methylmercury chloride demethylation takes place in the liver and kidney.²¹⁾

The correlation coefficients between total mercury and selenium in muscle and spleen of striped dolphin were above 0.9. In other marine mammals, such as seals and other species of dolphins, the only tissues showing values higher than 0.9 were the liver and brain where mercury mostly exists in the inorganic form.^{2,4,5)} The regression slopes of total mercury on selenium in liver and muscle were comparable with the results of Koeman *et al.*²⁾ and Arima and Nagakura.⁸⁾

The critical organ for methylmercury is the

brain. Total mercury concentrations in the cerebrum of older striped dolphins and the brain of adult common and gray seals¹⁾ were above 10 $\mu\text{g/g}$. Berglund and Berlin²²⁾ and Suzuki²³⁾ showed that the methylmercury concentration in the brain associated with toxic manifestations in some terrestrial mammals was around 10 $\mu\text{g/g}$. It was noted that the critical level of methylmercury in the brain increases with the coexisting selenium.²⁴⁾ Jernelöv *et al.*²⁵⁾ and Ohi *et al.*²⁶⁾ reported that large portions of the mercury in brain of minks and rats fed methylmercury and selenium were found as methylmercury. Berlin²⁷⁾ revealed that elemental mercury could be transmitted to the brain where it was oxidized to mercuric mercury. Kosta *et al.*²⁸⁾ reported that with mercury levels of up to 13 ppm in the brain, workers exposed to mercury vapor did not exhibit obvious clinical signs of intoxication, and that the mercury contents in brain were closely correlated with the selenium contents. Almost all mercury in the blood of striped dolphins exists as the methylated form.²⁹⁾ It is as yet unknown whether or not the brain of marine mammals has a demethylation process. However, it appears that the relatively low ratio of methylmercury to total mercury and relatively high level of selenium we observed in the cerebrum of striped dolphins represent protection mechanisms against mercury poisoning.

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