JOURNAL OF FOOD COMPOSITION AND ANALYSIS (2001) **14,** 453–460
doi:10.1006/jfca.2001.1005
Available online at http://www.idealibrary.com on IDEAL®

ORIGINAL ARTICLE

Total Mercury Content—Fish Weight Relationship in Swordfish (*Xiphias gladius*) Caught in the Southwest Atlantic Ocean

Eduardo Mendez*,[1], Horacio Giudice†, Alfredo Pereira†, Gustavo Inocente‡, and Dinorah Medina†

*Laboratorio de Electroquímica, Facultad de Ciencias, Universidad de la República, Iguá 4225 casi Mataojo, 11400 Montevideo, Uruguay, †Sección Control de Calidad, Departamento de Desarrollo Industrial, Instituto Nacional de Pesca, Ministerio de Ganadería, Agricultura y Pesca, Casilla de Correo 1612, Montevideo, Uruguay and ‡Sección Inspección Pesquera, Departamento de Desarrollo Industrial, Instituto Nacional de Pesca, Ministerio de Ganadería, Agricultura y Pesca, Casilla de Correo 1612, Montevideo, Uruguay

Received August 18, 2000, and in revised form March 7, 2001

The total mercury content in the edible part of swordfish (*Xiphias gladius*) was determined in 192 specimens ranging from 10 to 412 kg, in the period January 1997–December 1999. Total mercury values between 0.04 and 2.21 mg kg^{-1}, with a mean value of 0.62 $\pm$ 0.35 mg kg^{-1}, were obtained. Data were analyzed in two subsets. One of them, for specimens under 100 kg, yielded a mean value for total mercury content of 0.53 $\pm$ 0.02 mg kg^{-1}, and the other, for specimens above 100 kg, had a mean value of 0.94 $\pm$ 0.06 mg kg^{-1}. The existence of these subsets of data is related with the rate of mercury incorporation into swordfish flesh, in accordance with a potential model for total mercury bioaccumulation. Swordfish weighing below 100 kg can be considered safe for human consumption, according to international regulations.

Key Words: bioaccumulation; cold vapor atomic absorption spectrometry; mercury; swordfish (*Xiphias gladius*).

INTRODUCTION

The bioaccumulation of heavy metals in fish muscle is determined by different factors such as size, age, sex (Monteiro and Lopes, 1990), lipid content (Marcovecchio *et al*., 1988), and depth (Monteiro *et al*., 1996), making its assessment a difficult task in environmental research. However, the size of the fish can be taken as the main factor which determines the final levels of different metals, especially those which are accumulated during fish life, as the heavy metals. This fact is particularly relevant for fish which live in unpolluted areas, as the open oceans, because the metals accumulated in their flesh originate from the pollutant intake by small fishes which in turn are transferred through the food chain web.

Mercury can be considered a special case among the heavy metals, because its presence in marine water derives both from human activities and natural sources. In open oceans, the erosion of the sediments, where mercury is present as the sulfide salt, is the first step in the slow release of this metal in the water and atmosphere. Once

[1]To whom correspondence and reprint requests should be addressed: Instituto Nacional de Pesca, Casilla de Correo 1612, Montevideo, Uruguay. E-mail: emendez@fcien.edu.uy

0889–1575/01/050453 + 08 $35.00/0

dissolved in sea water, mercury is incorporated by small organisms by filtration, and transferred to higher trophic level fish through the food chain.

Swordfish (*Xiphias gladius*) is a carnivorous fish which inhabits Atlantic and Pacific oceans and large areas of the Mediterranean, Red and Black Seas (Love, 1980). The accumulation of mercury in this species has been widely recognized (Monteiro and Lopes, 1990; Vlieg *et al*., 1993). In addition, the presence of mercury in swordfish seems to be a fact independent of human pollution, since values in the range 0.45 and 0.9 $mg\,kg^{-1}$ were found in museum specimens caught between 1878 and 1909 (Miller *et al*., 1972), that is, before industrial activities began to pollute the ambient sea.

Since the Minamata accident in Japan in 1957, mercury concentration in fish has been thoroughly examined, and the incidence of mercury poison upon intake of contaminated fish is now very well established and known as the Minamata disease. In 1979, the FDA established an action level of 1.0 $mg\,kg^{-1}$ (wet basis) to regulate mercury content in commercial fish, and for those species which normally exceed this value, population is prevented from eating these fishes on a regular basis. Based on this, mercury is considered as a potential safety hazard for bonito, halibut, Spanish mackerel, king mackerel, marlin, shark, swordfish and bluefin tuna.

Swordfish fisheries have a relevant position among the tuna fisheries in Uruguay. Swordfish catch increased from 200 to 636 tons from 1992 to 1996, representing 67% of the tuna fisheries in that period. Most of the catch is exported, headed and gutted, to U.S.A. and Europe. In these countries, a tolerance limit for total (organic and inorganic) mercury in the edible flesh of 1.0 $mg\,kg^{-1}$ has been adopted. As this level is dependent on the fish weight, it is of interest to determine their relationship for quality control purposes.

In this work, the total mercury content in swordfish was determined for 3 years (January 1997–December 1999), and its relationship with fish weight was established.

MATERIALS AND METHODS

Samples

Swordfish was caught in the Southwest Atlantic Ocean from January 1997 to December 1999. Samples weighing 0.3–1 kg consisted of a transversal cut of *c*. 5 cm width at the cervical part of the body, and were sampled from commercial ships by the Veterinary Inspection staff of the National Fisheries Institute of Uruguay (Ministry of Livestock, Agriculture and Fishery, Montevideo, Uruguay). They were immediately sent to the laboratory, stored at $-30^{\circ}C$, and analyzed within the next three days. Prior to the analysis, samples were thawed, and a *c*. 200 g portion was homogenized in a Moulinex-type food processor.

Chemicals

All chemicals were of analytical grade and certified for low mercury content: H_2SO_4 (Fluka, 95–97%, Hg $<$ 0.005 ppm), HNO_3 (Fluka, 65%, Hg $<$ 0.005 ppm), $KMnO_4$ (Merck, Hg $<$ 0.05 ppm), NaCl (Fisher, total heavy metals as Pb $<$ 0.4 ppm), $NH_2OH{\cdot}HCl$ (J.T. Baker, Hg $<$ 0.005 ppm), and $SnCl_2.2H_2O$ (J.T. Baker, Hg $<$ 0.01 ppm). Tap water was distilled in a glass equipment and then purified to the nanopure level (resistivity $>$ 18 $M\Omega\,cm$) in a Barnsted equipment. Nanopure water was used throughout this work.

Instrumentation

A Perkin-Elmer 380 Atomic Absorption spectrophotometer was used, coupled with a Perkin-Elmer recirculating pump. A 15 cm length plastic cell, with 15 mm diameter quartz windows was attached to the burner head of the spectrophotometer. The cell was connected to the recirculating pump by Tygon tubing (Masterflex 6409-15), as well as the aerator tube attached to the BOD bottles. Mercury vapors were dried by passing through a tube containing Silica Gel Blue, placed between the exit of the vapors and the inlet of the cell. A mercury Electrodeless Discharge Lamp (EDL) was used at 40 mA, and was ignited with a Perkin-Elmer EDL Power Supply equipment set at 5 W. Every day, the tubing system and the cell were dried by recirculating air through a BOD bottle filled with Silica Gel.

Analytical Procedure

The technique of Hatch and Ott (1968) was used for sample digestion with slight modifications. One gram portion of the homogenized sample was accurately weighed in a 50 mL Teflon-lined screw-capped Pyrex tube, 5 mL of a 1:1 (v/v) mixture of concentrated H_2SO_4 and HNO_3 was added and the tubes were placed in a temperature-controlled water bath (Gallenkamp Model 20E) at 50°C for 2 h. The measurements were carried out by the Cold Vapor Atomic Absorption Spectrometric (CVAAS) method proposed by Uthe *et al.* (1970). The tubes were cooled under running water, 5 mL of 10% $KMnO_4$ aqueous solution was added, and the tubes contents were vortex mixed for 30 s. After allowing to stand for 30 min, 5 mL of 10% NaCl–10% $NH_2OH \cdot HCl$ aqueous solution was added and vortex mixed until disappearance of the pink color. The tubes contents were quantitatively transferred to a 300 mL BOD bottle with the aid of 100 mL of nanopure water. Alternatively, the wet digestion procedure was performed at room temperature for 24 h.

Mercury vapors were released from the solution by adding 5 mL of 10% $SnCl_2$ aqueous solution to the BOD bottle, and immediately capped with the aerator tube at an air flow rate of 1.5 L h^{-1}. The absorbance of the released vapors was monitored at 235.7 nm (slit width 0.7 nm) until a steady maximum was attained (usually in 2 min). The maximum absorbance was recorded, and the system was cleaned by bubbling in acidified permanganate aqueous solution. A complete cleaning of the whole system was performed periodically by recirculating through a BOD bottle with activated carbon specially designed for mercury adsorption.

A calibration curve in the range 0.01–2.0 ppm was designed every day from a 1.0 ppm working solution, prepared by appropriate dilution of a 1000 ppm Hg stock solution with 1 M H_2SO_4.

Glassware Cleaning

All glassware was rigorously cleaned before each analysis. Centrifuge tubes and BOD bottles were immersed in 6% NaOH in ethanol to eliminate any fat residue. When needed, ultrasound washing was done. Then, they were washed with tap water and immersed in a 10% HNO_3 aqueous solution for at least 24 h. After that, the glassware was thoroughly washed with tap water, distilled water and nanopure water. Clean glassware was stored wet and capped out of any dust, and was washed again with nanopure water immediately before use.

Data Analysis

Statistical relationship between total mercury content and fish weight was determined by least-squares regression analysis. Graphical deconvolution was performed using the Levemberg–Marquardt algorithm.

RESULTS

Analytical Procedure Performance

The digestion procedure was validated by carrying out the entire procedure of analysis with known additions of mercury (II) chloride. Three different levels were assayed in quintuplicate: 0.1, 0.5 and 1.0 ppm. The mean recovery, taking into account the range 0.1–1.0 ppm was $101 \pm 1\%$, as determined by the least-squares linear regression analysis procedure ($r = 0.9999$, $P < 0.001$).

The recoveries obtained with the digestion procedure used, indicated that no losses of mercury had occurred. This is one of the most important problems when carrying out mercury analysis, due to the high volatility of their compounds. The digestion procedure followed was essentially the one proposed originally by Uthe *et al.* (1970), but using capped tubes instead of Kjeldahl flasks. In addition, most of the procedure (weighing, digestion and reagent additions) was carried out in the same digestion tube, while vortex mixing favors the oxidation–reduction reactions involved. Alternatively, samples were digested at ambient temperature during 24 h, with the same results in the total mercury levels.

For the mercury levels usually found, a linear calibration curve was obtained ($P < 0.01$). The slope of the curve is directly related to the sensitivity of the method, and could be increased by increasing the EDL intensity. A working intensity of 40 mA provides a sensitivity in the order of 0.01 ppm which is sufficient for quality control purposes, with an increased lifetime of the lamp.

Normal Range for Total Mercury Content in Swordfish

A total of 192 specimens ranging from 10 to 412 kg were analyzed, yielding total mercury values between 0.04 and 2.21 $mg\,kg^{-1}$, with a mean value of 0.62 ± 0.35 $mg\,kg^{-1}$ ($P < 0.05$). From all the data analyzed, 14% were above the tolerance limit of 1.0 $mg\,kg^{-1}$, confirming that swordfish accumulates mercury in moderate to high levels, at least in the fish weight range usually commercialized.

The histograms of the values obtained were arranged in 0.20 $mg\,kg^{-1}$ intervals. The Gaussian fit to the histogram did not give an acceptable result. However, considering two simultaneous Gaussian fits, an excellent result was obtained (Fig. 1). These two normal distributions had mean values at 0.49 ± 0.02 and 1.04 ± 0.07 $mg\,kg^{-1}$ ($P < 0.05$).

Total Mercury Content in Relation to the Fish Weight

Data were adjusted to the potential model:

$$\text{Hg content (mg kg}^{-1}) = aW^b$$

where the Hg content is expressed on a wet basis, W is the weight of the headed and gutted fish in kg and a and b are constant parameters related to the fitted equation

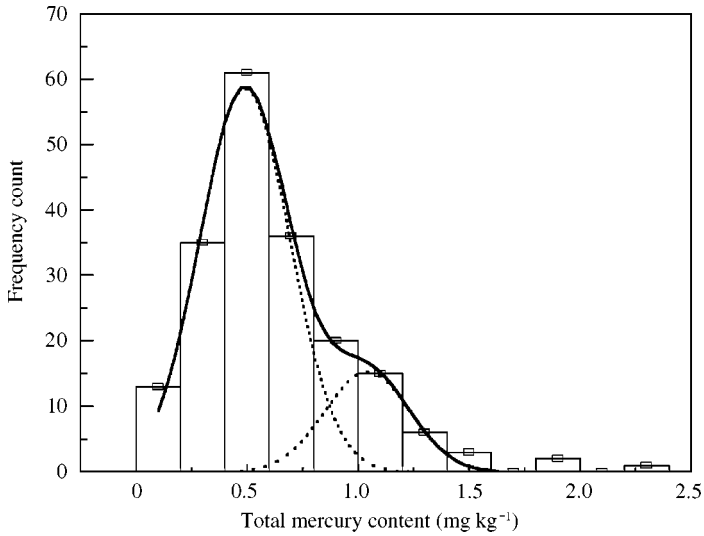

FIGURE 1. Histogram for total mercury content in swordfish (bars), arranged in 0.20 mg kg^{-1} intervals. Gaussian fit to the histogram (dot lines), and the sum of both gaussian curves (solid line).

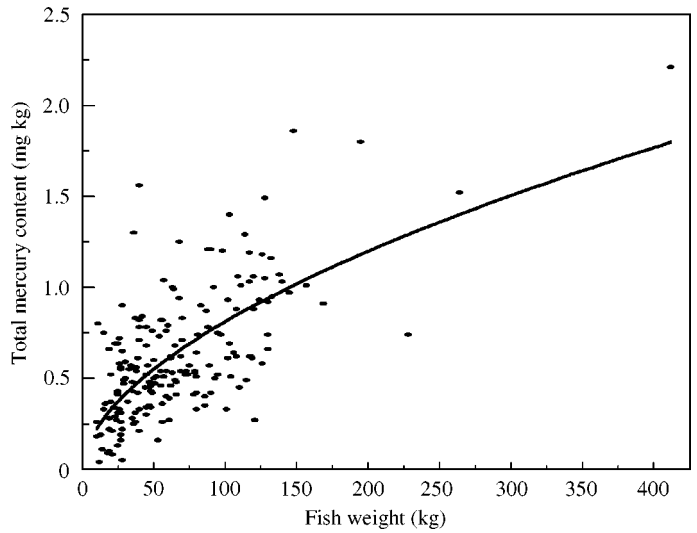

FIGURE 2. Plot of total mercury content (mg kg^{-1}) versus total fish weight ($n = 192$). The potential model is represented with the solid line.

(Fig. 2). The log–log transformation was fitted by the least-squares procedure, giving values for $a = 0.11 \pm 0.03$ and $b = 0.43 \pm 0.08$ ($P < 0.05$).

DISCUSSION

Swordfish, like other fish species, usually accumulates mercury as methyl mercury, while inorganic mercury usually represents a minor proportion of the total mercury

present in the tissues (Vlieg *et al.*, 1993). Methyl mercury does not occur naturally in water, and its presence in fish muscle is due to *in vivo* biochemical transformation or by ingestion of preformed methyl mercury along the food chain. Although methyl mercury toxicity is much higher than that of inorganic mercury, the total mercury content is usually considered for quality control purposes, provided that the digestion procedure is capable of releasing all the mercury from the biological matrix. In this sense, inorganic mercury (II) compounds can be used for calibration during the analytical procedure.

The normal values for total mercury content in swordfish obtained in this work indicate that moderate to high levels of mercury are present in specimens caught in the Southwest Atlantic Ocean, and that this level depends on the fish weight through a power relationship. This relationship was theoretically obtained by Norstrom *et al.* (1976) for methyl mercury bioaccumulation in fish muscle. According to this model, the coefficient b is directly related to the clearance order of the heavy metal, which measures the rate of mercury dilution or depletion by the species. The proposed model is based on the fact that the clearance capacity of mercury is independent of the metabolism and the temperature of the habitat, and that the influence of the fish weight on the clearance capacity is related to the fish growth, which determines the dilution of the mercury present in the muscle. In addition, the capacity of incorporation of mercury in fish through the diet is independent of the species (Norstrom *et al.*, 1976), but obviously will depend on the amount of dietary mercury. Thus, the actual content of mercury in swordfish depends on the mercury content of the diet and the size of the fish. Mercury being a natural contaminant, the presence of this metal in specimens caught in the pre-industrial period can be explained.

The derivative curve of the potential model represents the rate of mercury incorporation into the fish muscle (Fig. 3). Two different stages could be determined: (i) up to 50 kg, the rate of mercury incorporation decreased drastically from *c.* 0.011 to 0.006 mg kg^{-2} and, (ii) beyond 100 kg, the rate remains almost constant at

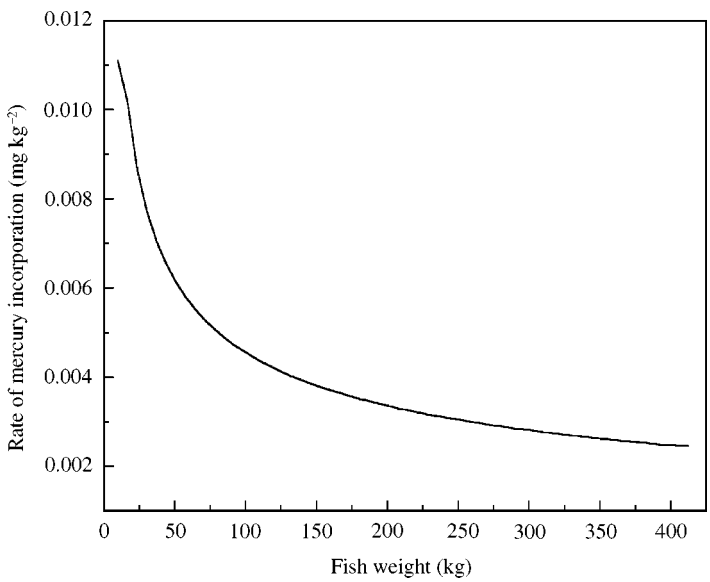

FIGURE 3. Rate of mercury incorporation, as a function of the total fish weight.

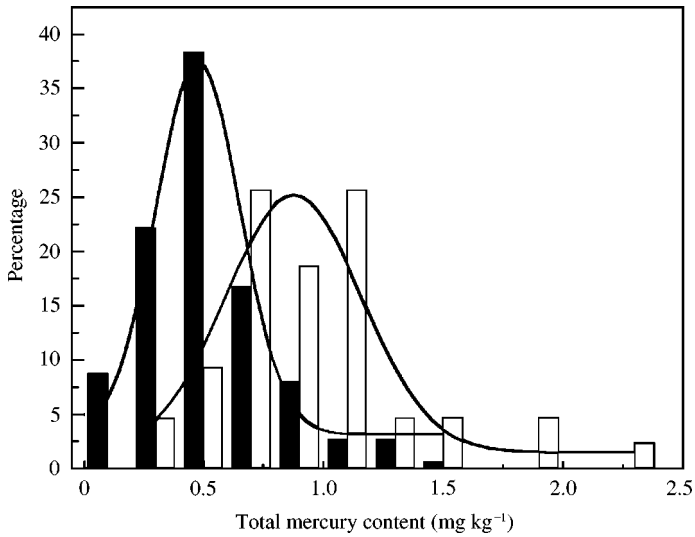

FIGURE 4. Histogram for total mercury content arranged in two subsets of data: for total fish weight (W) below (dark bars) and above (white bars) 100 kg.

c. 0.004 mg kg^{-2}. This difference in the mercury bioaccumulation in relation to the fish weight determines the two subsets in which the data are separated (Fig. 1). The first subset of data, with a mean mercury content of 0.49 mg kg^{-1}, corresponds to the mercury content for fish in a rapid growth, in such a way that the mercury ingested becomes diluted by the fast increase of the corporal mass. The second subset of data, with a mean mercury content of 1.04 mg kg^{-1}, corresponds to a slower increase in the fish body weight and, as a consequence, a lower dilution of the mercury content.

In order to confirm the above statements, the histograms corresponding to the mercury content for fish below and above 100 kg were obtained (Fig. 4). The mean values for the normal distributions obtained were 0.53 ± 0.02 mg kg^{-1} for $W <$ 100 kg and 0.94 ± 0.06 mg kg^{-1} for $W > 100$ kg ($P < 0.05$), which are in close agreement with the mean values obtained with the simultaneous fitting of two Gaussian distributions. These figures are in accordance with other already published studies (Monteiro and Lopes, 1990; Vlieg *et al.*, 1993).

Specimens usually commercialized in Uruguay are under 200 kg, with the highest frequency at around 50 kg. Mercury levels arranged according to the fish weight (Fig. 4) indicate that 6% of the specimens with $W < 100$ kg and 42% of the specimens weighing above 100 kg are beyond the tolerance limit of 1.0 mg kg^{-1}. Thus, swordfish weighing below 100 kg can be considered safe for human consumption, according to international regulations.

According to these results, during 1999, some samples from the same lot in which each specimen weighed under 100 kg were pooled and analyzed for mercury content, according to the sampling procedure suggested by the European Community. As can be seen in Table 1, all the lots analyzed yielded values under the tolerance limit of 1.0 mg kg^{-1}. Taking into account these results and guided by the institutional regulations, lots under the above-mentioned conditions (fish single weights under 100 kg) are now being pooled and analyzed as a single sample in our laboratory.

From the point of view of quality control, the construction of the bioaccumulation curve is recommended, as it would help in the assessment of some irregularities such

TABLE 1

Results for the analysis of different lots consisting of swordfish samples under 100 kg

Weight range (kg)	Number of samples in lot	Mercury content ($mg\,kg^{-1}$)
33–85	4	0.48
30–60	4	0.33
30–64	6	0.37
10–60	4	0.93
15–60	7	0.50
15–70	9	0.60
12–38	3	0.46
22–75	5	0.56
40–90	10	0.85
25–40	3	0.83

as contaminated fish stocks or fish lots. In our laboratory, the bioaccumulation curve for swordfish is being continuously updated, and similar curves are now being evaluated for other fishes which accumulate mercury, in particular, tuna species.

From a public health point of view, the consumption of fish with high mercury content does not represent a hazard, provided that it is not eaten on a regular basis. Public authorities should warn the population against the regular consumption of fish with high mercury levels.

ACKNOWLEDGEMENTS

The authors thank the Director of INAPE for giving the authorization to publish this work, and the Veterinary Inspection staff of INAPE for providing the samples. EM thanks the Universidad de la Republica for authorization to work at INAPE, and the PEDECIBA Programme.

REFERENCES

Hatch, W. R. and Ott, W. L. (1968). Determination of sub-microgram quantities of mercury by atomic absorption spectrophotometry. *Anal. Chem.* **40,** 2085–2087.

Love, R. M. (1980). *The Chemical Biology of Fishes.* Academic Press, London, 943 pp.

Marcovecchio, J. E., Moreno, V. J., and Pérez, A (1988). Total mercury levels in marine organisms of the Bahía Blanca estuarine trophic web. In *Metals in Coastal Environments of Latin America* (U. Seeliger, L.D. de Lacerda, and S. R. Patchineelam, Eds.), pp. 122–129. Springer–Verlag, Berlin.

Miller, E. E., Grant, P. M., Kishore, R., Steinkruger, F. J., Rowland, F. S., and Guinn, V. P. (1972). Mercury concentrations in museum specimens of tuna and swordfish. *Science* (New York) **175,** 1121–1122.

Monteiro, L. R. and Lopes, H. D. (1990). Mercury content of swordfish, *Xiphias gladius*, in relation to length, weight, age, and sex. *Mar. Pollut. Bull.* **21,** 293–296.

Monteiro, L. R., Costa, V., Furness, R. W., and Santos, R. S. (1996). Mercury concentrations in prey fish indicate enhanced bioaccumulation in mesopelagic environments. *Mar. Ecol. Prog. Ser.* **141,** 21–25.

Norstrom, R. J., McKinnon, A. E., and De Freitas, A. S. (1976). A bioenergetics-based model for pollutant accumulation by fish. Simulation of PCB and methylmercury residue levels in Ottawa river yellow perch (*Perca flavescens*). *J. Fish. Res. Board Can.* **33,** 248–267.

Uthe, J. F., Armstrong, F. A. J., and Stainton, M. P. (1970). Mercury determination in fish samples by wet digestion and flameless atomic absorption spectrophotometry. *J. Fish. Res. Board Can.* **27,** 805–811.

Vlieg, P., Murray, T., and Body, D. R. (1993). Nutritional data on six oceanic pelagic fish species from New Zealand waters. *J. Food. Comp. Anal.* **6,** 45–54.