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Methyl Mercury and Omega-3 Fatty Acids in Fish: Health Risk vs Benefits of Consumption

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ABSTRACT

The consumption of fish containing omega-3 fatty acids can result in several protective health effects including a reduced risk of cardiovascular disease, stroke, and diabetes, *etc*. These protective effects may be challenged by the presence of mercury (Hg) in the muscle tissue of fish. Mercury can increase the risk of cardiovascular disease and impede neurological development. Fish represent the main source of exposure to Hg for the general population, and large predatory fish such as swordfish, yellowfin tuna and black marlin (Indo-Pacific Black marlin) have the highest levels of Hg contamination. With provisional tolerable weekly intake (PTWI) of 1.6 µg/kg set by regulatory agencies for Methyl Mercury (MeHg) an adequate balance of risk and benefits through fish consumption is currently a nutritional/environmental health key issue. As a result, to choose the most suitable species of fish by considering the levels of omega-3 and MeHg, the consumption frequency, and the meal size are essential aspects to balance benefits and risk. However, the levels of contaminants were only determined in one species of freshwater fish and four species of marine fish, which was a limiting factor to establish recommendations concerning human consumption of fish.

1. Introduction

Sri Lanka is a small tropical Island in the Indian Ocean off the southern tip of an India and having an Exclusive Economic Zone (EEZ) area of 517,000 km^2. Capture fisheries produced 293,170 t and total fisheries production including aquaculture was 452,890 t in 2015 [1]. Fish can be finfish, shellfish (mollusks and crustaceans), or any other form of marine or freshwater animal life that can be used for human or domestic animal consumption [2, 3]. It is a competitive alternative source for other meat products. As an important nutrient, protein plays a pivotal role in human life of man and nation. Hence, fish are known as a rich source of nutritional quality; relatively low content of saturated fat and cholesterol, high in long-chain Poly-Unsaturated Fatty Acids (PUFAs), high biological value protein, vitamins (especially very important source of vitamin D) and minerals such as iodine, calcium, phosphorus, sodium, potassium, and magnesium [2, 3, 4]. The flesh of fresh fish is the most common source of high protein food and it plays an important role in human nutrition in Sri Lanka.

Omega-3 (n-3) Fatty Acids (FAs) are the most important essential PUFAs and help to maintain the proper and balanced human health. The main n-3 FAs are α-linolenic acid (ALA, C18:3 ω3), eicosapentaenoic acid (EPA, 20:5 ω3) and docosahexaenoic acid (DHA, 22:6 ω3). EPA & DHA are considered as main marine-derived FAs while ALA is considered as derived from plant oils, poultry and eggs derived FAs [5]. A small amount of EPA and DHA can be synthesized in the body from ALA; however, most are acquired directly from dietary sources. As evidence, decreased blood concentrations levels of omega-3 FAs have been associated with several neuropsychiatric conditions, including Alzheimer disease, schizophrenia, and

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depression. Sahar and Robert [4] reported that studied individuals who eat fish once a week or more, had a 60% lower risk of developing Alzheimer disease than those who consumed fish less frequently. Dietary intake of fish and omega-3 FAs has been associated with a lower risk of Alzheimer's disease and stroke. Several studies have been founded to find out an inverse association between fish and n-3 FAs consumption particularly with respect to their apparent reduction in the risk of coronary heart disease [6] and higher intake of DHA has been associated with decreased systolic and diastolic blood pressure [4]. Various organizations in worldwide such as World Health Organization (WHO), American Heart Association (AHA), European Food Safety Authority (EFSA), etc. have made several recommendations for dietary intake of omega-3 FAs [7]. The recommended amount of omega-3 FAs (EPA+DHA) for healthy adults is 0.3-0.5 g/day while that value is approximately 1 g/day for at high risk of developing Coronary Heart Disease (CHD) [5].

One of the potential barriers of frequent consumption of certain fish species is the possible toxicity risk to human health caused by contaminants [2]. Recently, the perception of fish as healthy food has been challenged by less favorable information regarding safety risks, more specifically the potential adverse health impacts of chemical contaminations in wild fish. Fish are the major source of human exposure to contaminants such as methylmercury (MeHg), polychlorinated biphenyls (PCBs), dioxins, organochlorine pesticides, and other environmental contaminants [8]. Bouzan *et al.* [9] and Harvard Center for Risk Analysis (HCRA) published the results of various studies remarking that although some fish contain MeHg, which may harm the developing fetus, fish consumption among the general population should be encouraged because of the nutritional benefits.

Mercury (Hg) is a highly reactive heavy metal with no known physiological activity in living beings. Exposure to toxic levels of Hg results in neurologic and renal damage, but the consequences of long-term exposure to low levels of Hg are poorly understood [10]. Mercury is rarely found as a free element in nature, but its elemental form is emitted from coal-burning electric power plants and used in chlorine production, dental amalgams, thermometers, and batteries. After release into the air, it cycles from rain into streams, lakes, and oceans where it is converted by microorganisms into organic MeHg. Smaller amounts of inorganic Hg that are naturally available in the environment may also be converted to MeHg by these microorganisms. When these microorganisms are ingested, Hg bioaccumulates in the food chain from smaller creatures to larger predators, with tissue concentrations depending on the level of local contamination and the size, lifespan, and predatory nature of each creature [11]. Thus, total Hg (T-Hg) levels tend to be higher in large, long-lived predators (e.g., swordfish, yellowfin tuna) and lowest in short-lived species (e.g., Tilapia) [12, 13]. Within the above works, authors studied the T-Hg level in fish tissues and its relationship with the size of fish. On the other hand, risk association of Hg, specially MeHg, reference dose defined as a Tolerable Daily Intake (TDI) or Tolerable Weekly Intake (TWI) assigned by the several organizations such as WHO, EFSA, and Food & Drug Administrative of United Status (US-FDA) [14].

To understand the possible health consequences of alternative risk management action, it is necessary to quantify potential health benefits and risk associated with fish consumption patterns. During past few years, a number of investigations have been conducted to quantitatively compare the risks of exposure to chemical pollutants in fish associated with health benefits of essential and omega-3 fatty acid consumption [5,15], but no research conducted in Sri Lanka to understand the country scenario. The objective of this paper to evaluates the levels of benefits of essential FAs and MeHg levels in selected fish species.

2. Material and Methods

A total of 529 fish were collected from commercial landing site in Sri Lanka during the period of 2009-2014 and covered the all wet and dry seasons (for freshwater fish) and throughout the country (for marine fish) belonging to five fish species; yellowfin tuna (*Thunnus albacares*, n=140), swordfish (*Xiphias gladius*, n=176), Tilapia (*Oreochromis mossambicus*, n=145), skipjack tuna (*Katsuwonus pelamis*, n=44), black marlin (*Istiompax indica*, n=24). Length and weight of fish were measured. Then approximately 250 g of the edible portion of the dorsal area was obtained from large fish (yellowfin tuna, skipjack tuna, black marlin, and swordfish) and whole fish of Tilapia were packed in separate polyethylene bags. Samples were transported in a cooler to the analytical chemistry laboratory/NARA and stored at -20 °C in the deep freezer until further analysis.

2.1 Analysis of mercury:

All T-Hg values obtained here from the author's previous studies and detail of the analytical method were explained on those manuscripts [13, 16]. The MeHg levels were extrapolated from the T-Hg values. For the exposure assessment of Hg, MeHg was considered for calculation and evaluation. Some

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studies reported that T-Hg in fish was entirely in the form of MeHg [17]. However, throughout this study, we assumed that MeHg level to be 90% of T-Hg [14].

2.2 Analysis of fatty acids:

The muscle samples of fish were analyzed for their lipid contents and fatty acid profiles as composite samples. All the samples were analyzed as duplicates. Total lipids in muscle tissue samples were extracted using the method described by Bligh and Dyer [18]. According to the lipid content of each composite sample, the Bligh and Dyer extract was used for the Fatty Acid Methyl Ester (FAME) generation. Capillary Gas Chromatograph (GC) (GC-2014 Shimadzu, Kyoto, Japan) was used to determine the fatty acid profiles.

The gas chromatograph was equipped with a Fused Silica DB-Wax capillary column (105 m) from Restek PA and Flame Ionization Detector (FID). Helium was used as a carrier gas at 14 psi. The initial temperature of the column was set at 160 °C and finally increased to 240 °C at the rate of 3 °C min^{-1}. The detector temperature was set at 270 °C, while the temperature at the injection port was maintained at 240 °C. The qualitative analysis was done with the base of the retention time of Qualmix Fish-S FAME mix (Larodan, Sweden) while the quantitative analysis was performed with Heptadecanoic acid as an internal standard. Methyl linolenate, methyl eicosapentaenoate, methyl docosatetraenoate, and methyl docosahexaenoate were taken into account for calculating to the omega-3 fatty acids.

2.3 Quality control:

Each sample was analyzed in duplicates. The blanks and quality control samples were analyzed in the same manner as samples. All chemicals and standards were used in high purity grade (AR) and purchased from Sigma Aldrich. To quantify the T-Hg by AAS, a calibration curve was constructed with 5 standards whereas fatty acid methyl esters (FAME) analysis by GC consisted with an Internal Standard (IS) of heptadecanoic acid ($C_{17}H_{34}O_2$). The accuracy of analytical procedure was analyzed using several certified quality control materials from Food Analysis Performance Assessment Scheme (FAPAS, UK), and the laboratory participated into two proficiency testing programs of the FAPAS for T-Hg within the studied period. The Z values of the proficiency testing programs were within a satisfactory range (PT-07115-2009, Z =0.1, and PT-07215-2014, Z = 0.0).

The recovery results of quality control materials proved the suitability of the method for Hg, total fat, and FAs analyses. The limit of quantification of Hg was 0.07 mg/kg while the fatty acid percentage when <0.15%, were reported it Not Detectable (ND). The results of quality control materials are listed in table 1 (A & B).

Table 1: List of certified reference materials used in the present study and the results obtained (1-A: Hg and fat 1-B: fatty acids).

1-A

QC-Material and type	Matrix	Analyte & Unit	Certified value	Obtained value ±SD
T-0774-QC	Canned fish muscle	Hg, µg/kg	19.9	20.01±0.70
T-07192-QC	Canned crab meat	Hg, µg/kg	95.68	94.47±6.10
T-0188-QC	Canned meat meal	Fat, %	7.96	7.91±0.22

1-B

Analyte	Assigned Value (%)	Range (%)	Obtained Value (%)	Recovery (%)
Alpha Linolenic Acid, C18:3 n-3	2.16	1.82-2.51	2.33	107.87
Eicosapentaenoic Acid, C20:5 n-3	7.41	6.67-8.15	6.71	90.55
Docosahexaenoic Acid, C22:6 n-3	7.81	7.03-8.59	7.06	90.40
Linoleic Acid, C18:2 n-6	6.57	5.92-7.23	5.95	90.56
Eicosadienoic Acid, C20:2 n-6	0.52	0.31-0.73	0.41	78.85
Arachidonic Acid, C20:4 n-6	0.54	0.32-0.75	0.48	88.89
Oleic Acid, C18:1 n-9 cis	19.3	17.37-21.23	18.05	93.52
Erucid Acid, C22:1 n-9	0.65	0.39-0.91	0.64	98.46
Nervonic Acid, C24:1 n-9 cis	0.52	0.31-0.73	0.42	80.77

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2.4 Risk benefits analysis:

The risk from the consumption of fish was calculated based on equation given by Ouédraogo and Amyot [19], based on Provisional Tolerable Weekly Intake (PTWI) and endpoint used for fish contaminants with cumulative effects (equation 1).

$$A = W\ X\ \frac{I}{C} \dots\dots\dots\dots\dots\dots\dots\dots\dots (1)$$

Where, A = amount (g) of a fish that can be safely eaten weekly basis (PTWI), W = average body weight of Sri Lankan adults (average for male and female, here 58 kg), I = tolerable weekly intake of MeHg; 1.6 µg/kg body weight, [20] and C= MeHg concentration in fish (µg/g w.w). Addition to that, in this study, we estimated health risk vs benefits of consuming commonly use five fish species which was characterizing a hazard quotient (HQ) [21]. Based on the previous literature, fish with HQ<1 means that health benefits from fish consumption while HQ>1 means the risk [21].

$$HQ = \frac{R.EFA\ X\ C.MeHg}{C.EFA\ X\ RfD.MeHg\ X\ AW} \dots\dots (2)$$

Where, R.EFA= recommended a daily dose of essential FAs from the diet, 500 µg/day [21], C.MeHg= MeHg content of fish muscle, C.EFA= EPA+DHA content of muscle tissue of fish (mg/g, ww), RfD.MeHg = reference dose for MeHg; (0.1 µg/kg/day) and AW= average weight of an adult individual, set to 58 kg [22].

3. Results and Discussion

The mean T-Hg concentration of studied fish species, derived value of MeHg concentration and omega-3 FAs amount are given in table 2. The highest average MeHg concentration was recorded from the swordfish (0.81 mg/kg) that was the approximately seven-fold higher than the lowest average recorded species, skipjack tuna. The swordfish, marlin, and yellowfin tuna are known to feed on pelagic fish and invertebrates, particularly squid thus inhabit in a high level of the food chain [23]. A high concentration of Hg accumulation was found in higher trophic level fish compared to low trophic level species like skipjack tuna and Tilapia [14]. Comparatively low levels of MeHg observed in skipjack tuna as it is comparatively a fast-growing, short-lived fish in the medium levels of marine food chains [16]).

According to the results in table 2, marine fish species have a high content of essential FAs, (methyl Linolenate, methyl eicosapentaenoate, methyl docosatetraenoate, and methyl docosahexaenoate) which were significantly higher than the freshwater species (Tilapia). Hence, the studied species can be indicated as a valuable food source for human nutrition. The omega-3 content of these species are given in table 3 and ranged from 0.0201 to 0.400 g/per serving (85 g or 3 oz). The omega-3 content of the fish varied with the water temperature where they grow and their food sources *etc*. However, general species-specific pattern of fatty acid composition for fish species can be identified [15].

Table 2. Mean and standard deviation (SD) of T-Hg (mg/kg), methyl Hg (mg/kg) and omega-3 FAs (g/kg) concentration of dorsal muscle of studied fish species (values given in wet weight basis)

Fish Species	T-Hg±SD (mg/kg)*	MeHg (mg/kg)	omega-3 FA ±SD, g/kg
Yellowfin tuna	0.30±0.18	0.27	4.70±0.25
Indo-Pacific black marlin	0.49±0.37	0.44	2.70±0.31
Swordfish	0.90±0.52	0.81	3.45±0.27
Skipjack tuna	0.13±0.06	0.12	3.38±0.34
Tilapia	0.26±0.23	0.23	2.37±0.41

*references: [12, 13,16]

Table 3. Omega-3 content of the seafood species used for mercury analysis is given in grams of EPA and DHA per 85 g (3 oz) serving of seafood.

Fish Species	Omega-3 content (g/85 g)
Yellowfin tuna	0.400
Indo-Pacific black marlin	0.230
Swordfish	0.293
Skipjack tuna	0.287
Tilapia	0.201

The risk-benefit analysis of fish can be assessed by comparing MeHg content in fish consumed and the number of meals required to fulfill the health benefits of the omega-3 FAs monthly basis. This concept will help the consumers to determine which species of fish should be favorably for consumed (table 3). The omega-3 requirement was calculated based on 2 scenarios; (i) person without CHD, the omega-3 requirement consider as a 0.3 mg/day (ii) person with CHD, the omega-3 requirement consider as 1 mg/day. If the omega-3 serving/month is lower than the allowable servings/month based on MeHg content, those species are preferable for human consumption. As an example, a healthy person with no signs of CHD need to consume yellowfin tuna at a rate of 10 servings per month to fulfill the omega-3 requirement, however, based on the MeHg concentration, it should not have exceeded the 7 servings per month. Considering all studied fish species, only skipjack tuna can be consumed for the group of no risk at CHD, without exceeding mean MeHg levels (Table 4).

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Table 4. Servings rate per month of fish species that should be consumed by individuals both with and without CHD to obtain maximum health benefits, PTWI amount and safe serving rate considering the MeHg and HQ value.

Fish Species	Omega-3, no CHD (servings/month)	Omega-3, CHD (servings/month)	A (PTWI), g/week	Measured MeHg limit (servings/month)	HQ
Yellowfin tuna	10	32	299	**7**	6
Indo-Pacific black marlin	17	56	183	**4**	16
Swordfish	13	44	100	**2**	23
Skipjack tuna	13	45	691	16	4
Tilapia	19	64	345	**8**	10

*in here, the bold number appears in the "Measured MeHg limit" columns; the MeHg limit would be exceeded before obtaining the recommended amount of omega-3s, serving size; 198 g (7 oz), the bodyweight of the person; 58 kg

Omega-3 FAs essentially should be in the diets and it was explained by several researchers [24]. Fish provide considerable amounts of EPA+DHA necessary for a healthy life but can be contaminated with MeHg and other organic and inorganic pollutants. Therefore, obtaining omega-3 FAs from other fish species placed in low levels in the food chain and other sources like single cell oil also can play a major role. By contrast, this will lead to sustainable fisheries through well-managed harvesting strategies. In this decade, numbers of artificially omega-3 enriched foods such as eggs, chicken, some naturally omega 3 rich foods such as flaxseed, flaxseed oil, soybean oil, and walnuts, and omega-3 enriched infant formulas such as powder milk and cereals reach to market as heart-healthy products [15].

In the present study, HQ represents the risk-benefit ratio for fish consumption due to MeHg and omega-3 intake respectively. Considering the HQ value, all studied species were associated with risk and generally appeared the not appealing for human health due to a significant amount of omega-3 and high amount of MeHg. As in a general way, here we also considered health benefit/risk assessment mainly based on the MeHg concentrations in raw products, even though most products are consumed after cooking. Some researchers highlighted that MeHg bioavailability depends on the selenium (Se) level and it may help to inhibit the absorption of MeHg [25]. However, different metal accumulated in different levels into different species and body parts [21], and it is necessary to provide a complete picture regarding risk information to people allowing them to select and consume a diversity of fish for a healthy life.

4. Conclusion:

Although some researchers highlighted and questioned the benefits of the fish consuming both high levels of Hg and omega 3 fatty acids, there is no doubt about the beneficial effects, especially to the reduction of CHD. There are some fish species such as skipjack tuna in low T-Hg (<0.15 mg/kg), and comparatively high omega-3 fatty acids while some fish species like swordfish are high in T-Hg (>0.5 mg/kg), are especially not rich in omega 3. Accordingly, based on this viewpoint that avoiding the consumption of fish higher in T-Hg will negotiation nutritional intake of omega-3 fatty acids from fish reflects an incomplete understanding of its composition. Considering the value of MeHg and HQ, studied fish species are associated with the risk of health hazard in Sri Lankan consumers. Therefore, adults should consider the serving amount and frequency per month especially when they consume apex predators such as swordfish, Indo-Pacific black marlin, and yellowfin tuna. Meantime, it is necessary to conduct the comprehensive nutritional and toxicological study, risk analysis, and risk communication program based on this finding.

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References

1. NARA. Sri Lanka fisheries yearbook, available online www.nara.ac.lk. Colombo, Sri Lanka: NARA. 2016.
2. Jose LD, Ana B, Roser MC & Juan ML. Benefits and risk of fish consumption, II RIBEPEIX, a computer program to optimize the balance between the intake of omega-3 fatty acid and chemical contaminants. Toxicology, 2007, 230, 227-233.
3. Mohamed ABS, Brahim M, M'hamed E. & Abu TAA. Seasonal variation of fatty acids in muscles of Tunisian swordfish (*Xiphias gladius*, L.1758). Journal de la Societe Chimique de Tunisie, 2010, 12, 129-137.
4. Sahar, SAMS & Robert AG. Levels of omega 3 fatty acids in Australian seafood. Asia Pacific Journal of Clinical Nutrition. 2008. 17, 385-390.
5. Smith KL & Guentzel JL. Mercury concentrations and omega-3 fatty acids in fish and shrimp: Preferential consumption for maximum health benefits. Marine Pollution Bulletin.2010, 60, 1615-1618.
6. Alan HS. Public health guidance on cardiovascular benefits and risks related to fish consumption. Environmental Health, 2007, 6, 1-4.
7. Kris-Etherton PM, Grieger JA & Etherton TD. Dietary reference intakes for DHA and EPA. Prostaglandins, leukotrienes and essential fatty acids, 2009, 81, 99-104.
8. Turunen AW, Männistö S, Kiviranta H, Marniemi J, Jula A, Tiittanen P, Suominen-Taipale L, Vartiainen T & Verkasalo PK. Dioxins, polychlorinated biphenyls, methyl mercury and omega-3 polyunsaturated fatty acids as biomarkers of fish consumption. European Journal of Clinical Nutrition, 2010. 64, 313.
9. Bouzan C, Cohen JT, Connor W, Kris-Etherton P, Gray G, König A, Lawrence R,Savitz D & Teutsch S. A quantitative analysis of fish consumption and stroke risk. American Journal of Preventive Medicine. 2005. 29, 347-352.
10. Eliseo G, Inmaculada SG, Pieter VV, Peter B, Antti A, Jorge GA, Jeremy DK, Rudolph AR, Jose M & Frans JK. Mercury, fish oil and the risk of myocardial infarction. New England Journal of Medicine. 2002. 347, 1747-1754
11. Dariush, M. Fish, mercury, selenium and cardiovascular risk: current evidence and unanswered questions. International Journal of Environmental Research and Public Health, 2009, 6, 1894-1916.
12. Jinadasa BKKK, Ahmad SBN, Edirisinghe EMRKB. & Wickramasinghe I. Mercury content in yellowfin tuna (*Thunnus albacares*) and swordfish (*Xiphias gladius*) and estimation of mercury intake. Journal of Food Security, 2014, 23-26.
13. Jinadasa BKKK. & Edirisinghe EMRKB. Assessment of heavy metals (Cadmium, Lead and Total Mercury) in Tilapia *sp*. in Sri Lanka. Food Additives and Contaminants: Part B, 2014, 90-94.
14. Strandberg U, Palviainen M, Eronen A, Piirainen S, Laurén A, Akkanen J & Kankaala P. Spatial variability of mercury and polyunsaturated fatty acids in the European perch (*Perca fluviatilis*) – Implications for risk-benefit analyses of fish consumption. Environmental Pollution.2016. 219, 305-314.
15. Mahaffey KR, Sunderland EM, Chan HM, Choi A L, Grandjean P, Mariën K, Oken E, Sakamoto M, Schoeny R, Weihe P, Yan C-H & Yasutake A. Balancing the benefits of n-3 polyunsaturated fatty acids and the risks of methylmercury exposure from fish consumption. Nutrition Reviews, 2011, 69, 493-508.
16. Mahaliyana AS., Jinadasa BKKK, Liyanage NPP, Jayasinghe GDTM & Jayamanne SC. Nutritional composition of skipjack tuna (*Katsuwonus pelamis*) caught from the oceanic waters around Sri Lanka. American Journal of Food and Nutrition, 2015, 3, 106-111
17. Sevillano-Morales JS, Cejudo-Gómez M, Ramírez-Ojeda AM, Cámara Martos F & Moreno-Rojas R. Risk profile of methylmercury in seafood. Current Opinion in Food Science, 2015, 6, 53-60.
18. Bligh EG & Dyer WJ. A rapid method of total lipid extraction and purification. Canadian Journal of Biochemistry and Physiololgy, 1959, 37, 911-917.
19. Ouédraogo O & Amyot M. Mercury, arsenic and selenium concentrations in water and fish from sub-Saharan semi-arid freshwater reservoirs (Burkina Faso). Science of the Total Environment, 2013, 444, 243-254.

20. JECFA. Summary of conclusion of the 72 of the joint FAO/WHO expert committee on food additives. Rome, Italy: FAO/WHO. 2010.

21. Gladyshev MI, Sushchik NN, Anishchenko OV, Makhutova ON, Kalachova GS & Gribovskaya IV. Benefit-risk ratio of food fish intake as the source of essential fatty acids vs. heavy metals: A case study of Siberian grayling from the Yenisei River. Food Chemistry, 2009, 115, 545-550.

22. MHNIM.Non communicable diseases risk factor survey Sri Lanka 2015. Ministry of health nutrition and indigenous medicine, Sri Lanka. 2015.

23. Gobert S, Pasqualini V, Dijoux J, Lejeune P, Durieux EDH & Marengo M. Trace element concentrations in the apex predator swordfish (*Xiphias gladius*) from a Mediterranean fishery and risk assessment for consumers. Marine Pollution Bulletin, 2017, 120, 364-369.

24. Corte CD, Iasevoli S, Strologo AD, Sanseviero M & Nobili V. Chapter two - Omega-3 fatty acids and fatty liver disease in children. In: Toldrá, F. (ed.) Advances in Food and Nutrition Research. Academic Press. 2018.

25. Afonso C, Costa S, Cardoso C, Coelho I, Castanheira I, Lourenço H, Gonçalves S, Oliveira R, Carvalho ML, Martins MF, Bandarra, NM & Nunes ML. Bioaccessibility in risk-benefit analysis of raw and cooked seabream consumption. Journal of Food Composition and Analysis, 2016, 68, 118-127.