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SOME NUTRITIONALLY IMPORTANT FATTY ACIDS IN SEVEN VARIETIES OF FISH EATEN IN SRI LANKA

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SUMMARY. The component acids of lipids extracted from the edible portions of seven marine species *Sardinella longiceps, Rastrelliger kanagurta, Thunnus albacares, Scomberomorus commersoni, Caranx stellatus, Carcharhinus* sp. and *Raja mamillidens* and one fresh-water species *Tilapia mossambica* have been determined by gas-liquid chromatography. Palmitic (C 16:0), palmitoleic (C 16:1), oleic (C 18: 1), eicosapentaenoic EPA (C 20: 5 n—3) and docsahexaenoic, DHA (C 22: 6 n—3) were the most abundant fatty acids. The small pelagics have a higher content of EPA and DHA and a higher P/S ratio than the larger and more expensive pelagic species.

INTRODUCTION

Fish oils and their constituent omega - 3 polyunsaturated fatty acids, eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) have been shown to

1. decrease plasma triglyceride, very low density lipoproteins (VLDL) and low density lipoprotein (LDL) levels
2. inhibit atheromatous changes in arteries and
3. to exhibit a host of "membrane effects" many of which are beneficial (1,2).

Although the fatty acid content of fish consumed in temperate regions is known (3,4), no such analysis has been carried out on local fish. This is a report of an analysis of the fatty acid composition of eight varieties of fish commonly consumed in Sri Lanka.

MATERIALS AND METHODS

The eight varieties studied and the months of the catch are indicated in Table 1.

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TABLE 1. Total lipid content of edible portions of fish in g per 100 g fillet (mean and range)

Species	Total lipid in g/100 g fillet Sample 1	Sample 2
1. *Sardinella longiceps* Sardine	6.3 5.9—6.5 (July 1988)	4.9 4.8—5.0 (January 1989)
2. *Rastrelliger kanagurta* Indian mackerel	3.8 3.6—4.0 (August 1988)	3.4 3.3—3.5 (December 1988)
3. *Thunnus albacares* Yellow fin tuna	2.4 2.3—2.6 (August 1988)	2.5 2.3—2.7 (November 1988)
4. *Scomberomorus commersoni* Spanish mackerel	2.4 2.2—2.5 (September 1988)	2.3 2.2—2.5 (January 1989)
5. *Caranx stellatus* Trevally	1.3 1.2—1.5 (September 1988)	1.3 1.2—1.5 (January 1989)
6. *Carcharhinus sp.* Grey dog shark	0.8 0.7—0.9 (September 1988)	0.9 0.6—1.0 (January 1989)
7. *Raja mamillidens* Skate	0.9 0.8—1.0 (January 1989)	0.9 0.9—1.0 (February 1989)
8. *Tilapia mossambica* Tilapia	1.9 1.7—2.0 (December 1988)	1.8 1.6—2.0 (January 1989)

The date of catch is given within brackets.

The fish were collected from the fish landing sites, packed in ice and brought to the laboratory withing 2 or 3 hours of landing. The number of fish taken for analysis varied with the size of the species. Within each species, fish of approximately the same size and age were selected. The head, tail, fins, viscera, eggs and skin (except in the Indian mackerel and the sardine) were removed. Fish fillets minus bone were cut into small pieces and mixed before taking aliquots for analysis.

Extraction and purification of fish oil

Aliquots of fish fillet were minced for 2 min, and the total lipid content determined by the method of Bligh and Dyer (5). The lipid solution was placed in a rotary evaporator and the $CHCl_3$ evaporated to obtain the lipid material, which was stored at —18°C in n-hexane flushed with nitrogen gas to prevent oxidation.

Packed-column gas-liquid chromatography

The n-hexane was evaporated and the dried lipid sample used to prepare methyl esters of the fatty acids according to the method of Christie (6). The content and composition of the methyl esters were analysed using a Shimadzu Series G C-7 AG chromatograph equipped with a flame ionization detectorand using a glass column (4mm i.d. x 2.6.m) packed with SP 2340, and Shimadzu -R- 112 chart recorder at a chart speed of 0.5 mm/min. The operating conditions were: column temperature 190°C, injection temperature 250°C, detector temperature 250°C, carrier gas nitrogen 40ml/min.

Capillary column gas-liquid chromatography

The fatty acid methyl esters (FAME) were prepared from each lipid sample by treating with boron-trifluoride in methanol and the completion of conversion to the esters checked by thin-layer chromatography using the method of Christie (6).

The FAME of each lipid sample was then analysed for content and composition of the different fatty acids, using a Shimadzu series GS—9AG gas chromatograph equipped with a flame ionization detector and a Perkin-Elmer R-100 chart recorder, at a chart speed of 10 mm/min. A SUPELCOWAX 70 fused silica capillary column (30 mm x 0.32 i.d.) was used: column temperature 200° C, injection temperature 250°C, detector temperature 250°, carrier gas helium 21.5 ml/min. hydrogen 30 ml/min., air 240 ml/min.

GLC identification.

The saturated fatty acids were identified by comparison with retention times of mixtures of methyl esters C 14 : 0, C 16 : 0, C 18: 0, C 20: 0 and 22: 0 reference standards.

The methyl esters of the unsaturated acids were identified by direct comparison of the retention times of methyl esters, using cod-liver oil as a reference standard, according to the method of Ackmann (7) and Holman (8). Equivalent chain-length values were used as a basis for confirming the identity of the acids, according to the procedure recommended by Christie (9). From a plot of the logarithms of the retention times against the number of carbon atoms, nearly linear relations were observed for homologous series.

To obtain quantitative data the area and concentrations of the peaks on the chromatograms were integrated electronically by the Systems Instrument, model 5000 E integrator

RESULTS

The total lipid content of each variety analysed is given in Table 1. Three samples of each species were analysed. Only one extraction of each sample was necessary. A second extraction with $CHCl_3$ indicated the presence of negligible amounts of fatty acids in the residue of the first extraction. Table 1 shows that sardines caught during seasons 6 months apart, from the same fishing grounds, have different total lipid contents, raising the possibility of there being variations with the seasons. Table 2 gives the fatty acid composition of the fish oils as determined by packed-column gas chromatography. There is a wide variation in the fatty acid composition of the fish oils. The major components in all species are plamitic (C 16 : 0), palmitoleic (16 C : 1), stearic (C 18 : 0), oleic (C 18 : 1), archidonic (C 20 : 4), eicosapentaenoic (C 22 : 5) and decosahexaenoic (C 22 : 6).

TABLE 2. Fatty acid composition of lipids of fish analysed by the packed column GLC from sample 1 (g fatty acid per 100 g total fatty acids).

Species	c14:0	c15:0	c16:0	c16:1	c18:0	c18:1	c18:2	c18:3	c20:0	c20:3	c20:4	c20:5	c22:4	c22:5	c22:6	others
Sardinella longiceps Sardine	14.98	1.01	29.75	15.15	6.06	9.84	tr	tr	tr	—	2.71	5.68	tr	tr	14.14	0.63
Rastrelliger kanagurta Indian mackerel	2.14	1.01	22.91	10.34	9.06	8.51	tr	tr	tr	tr	2.55	10.11	—	5.04	22.92	5.41
Thunnus albacares Yellow fin tuna	4.17	1.01	20.48	10.82	6.48	7.81	0.56	0.62	—	0.1	5.81	9.81	0.89	6.41	21.94	3.09
Scomberomorus commersoni Spanish mackerel	3.00	—	24.53	10.81	10.21	10.92	1.50	0.28	—	—	4.61	8.42	1.70	3.86	18.92	1.24
Caranx stellatus Trevally	3.51	1.01	23.81	6.08	10.41	8.40	1.10	0.31	tr	tr	2.40	4.19	1.98	6.83	27.05	2.92
Carcharhinus sp. Grey dog shark	1.91	0.92	20.41	4.82	15.21	11.82	0.24	0.22	1.10	—	10.41	4.07	1.06	5.86	20.70	1.93

TABLE 3. Fatty acid composition[a] of 8 species of fish analysed by the capillary column GLC from sample 2 (g fatty acid per 100 g total fatty acid)

Species	Saturated Fatty Acids						Monounsaturated Fatty Acids										
	c14:0	c15:0	c16:0	c18:0	c20:0	c22:0	c16:1 n-7	c16:1 n-5	c18:1 n-9	c18:1 n-7	c18:1 n-5	c20:1 n-11	c20:1 n-9	c20:1 n-7	c22:1 n-11	c22:1 n-9	c24:1 n-9
Sardinella longiceps Sardine	3.8	0.4	30.7	8.7	tr	tr	5.9	tr	3.9	2.2	1.9	tr	tr	tr	—	—	tr
Rastrelliger kanagurta Indian mackerel	1.1	0.3	21.8	8.0	1.5	0.4	8.6	0.5	4.4	2.6	tr	0.4	0.2	0.2	0.1	tr	0.6
Thunnus albacares Yellow fin tuna	2.5	0.5	21.2	10.5	0.5	0.2	3.7	0.3	10.0	3.3	0.1	0.2	0.4	0.2	0.1	0.1	0.5
Scomberomorus commersoni Spanish mackerel	1.2	0.2	33.1	10.6	tr	2.0	10.8	tr	10.8	3.8	tr	tr	0.3	tr	0.1	tr	0.5
Caranx stellatus Trevally	3.5	0.8	27.6	9.1	0.1	0.1	5.5	0.1	15.9	2.5	tr	0.1	0.5	0.2	0.1	0.1	0.3
Carcharhinus sp. Grey dog shark	1.7	0.2	17.9	12.9	tr	0.1	2.1	0.1	11.7	3.7	—	0.2	0.5	0.1	—	0.1	0.5
Raja mamillidens Skate	0.9	tr	14.9	14.8	tr	tr	3.9	0.9	12.4	8.7	tr	tr	1.5	tr	tr	tr	0.3
Tilapia mossambica Tilapia	2.0	0.4	20.6	6.1	tr	—	4.0	0.2	15.0	3.7	0.2	0.5	1.0	0.1	tr	tr	0.4

(Continued)

TABLE 3. Fatty acid composition[a] of 8 species of fish analysed by the capillary column GLC from sample 2 (g fatty acid per 100 g total fatty acid)

Species	Polyunsaturated Fatty Acids														others[b]
	c18:2 n-6	c18:3 n-3	c18:4 n-3	c20:2 n-6	c20:3 n-6	c20:3 n-3	c20:4 n-6	c20:4 n-3	c20:5 n-3	c21:5 n-3	c22:4 n-6	c22:5 n-6	c22:5 n-3	c22:6 n-3	
Sardinella longiceps Sardine	tr	1.1	tr	tr	tr	—	2.6	tr	7.3	tr	1.1	tr	1.9	27.8	0.7
Rastrelliger kanagurta Indian mackerel	1.8	1.7	1.8	0.3	0.2	tr	2.1	0.6	8.7	0.7	tr	1.3	2.1	19.3	8.7
Thunnus albacares Yellow fin tuna	1.3	0.7	2.6	0.2	0.1	0.2	3.3	0.5	6.7	0.2	0.3	1.5	1.5	20.5	6.1
Scomberomorus commersoni Spanish mackerel	1.5	0.7	0.8	tr	—	—	2.3	—	4.5	tr	tr	0.9	0.9	13.5	1.5
Caranx stellatus Trevally	0.1	0.4	0.6	0.2	0.2	0.1	2.2	0.3	3.9	0.3	0.3	1.0	2.0	13.7	8.1
Carcharhinus sp. Grey dog shark	1.3	0.3	0.4	0.6	0.5	0.5	6.8	0.5	2.3	—	5.5	3.7	2.0	19.5	4.3
Raja mamillidens Skate	0.7	0.3	tr	—	0.3	—	5.6	—	2.1	—	5.2	3.8	3.5	20.0	0.2
Tilapia mossambica Tilapia	5.5	4.6	0.6	0.6	1.5	1.8	2.5	1.5	1.2	0.3	1.8	0.3	3.9	11.5	8.2

n denotes position of first double bond from methyl end of the fatty acid.
a minor fatty acids (less than 0.1 %) not included
b minor fatty acids and unidentified acids.

The concentration of polyunsaturated acids (PUFA) of the omega 3 (or n-3) and omega 6 (or n-6) series vary with the species (Table 3). DHA (C 22:6, n-3) is present in the highest concentration and EPA (C 20: 5, n-3) in significant amounts in all species tested.

Table 4 indicates the concentration of total saturated and poly-unsaturated acids (PUFA) in the oils and polyunsaturated to saturated or the P/S ratio. The Spanish mackerel (S. thora) and the trevally (S. paraw), the most sought after fish among Sri Lankans, have the lowest P/S ratios.

Table 5 gives the concentration of the five nutritionally important polyunsaturated fatty acids, expressed both as mg per 100g fillet and 100 g as purchased. In calculating the amount of each acid in the fish as purchased in the open market, it is assumed that only a portion of the fish purchased consists of fillet : namely, in whole sardine, Indian mackerel and tilapia, 40%, in the mid-cut of the yellow fin tuna, spanish mackerel and the gray dog shark, 85%, in trevally, mid-cut 75% and 50% in the skate. All the species have relatively low contents of linoleic and alpha-linolenic acids. The rich sources of EPA and DHA are the sardine (S. salaya), the Indian mackerel (S. hurulla), the Spanish mackerel (seer) and the yellow fin tuna (S. kelawella).

TABLE 4. Content of total saturated acids, unsaturated acids and polyunsaturated acids, acids with 4,5 and 6 double bonds and total acids in the n-3 and n-6 series, in fish lipids (as % of the total acids) from sample 2 and the ratio, polyunsaturated to saturated acids

Species	Total n-3	Total n-6	Total 4-ene	Total 5-ene	Total 6-ene	Total saturated fatty acids	Total unsaturated fatty acids	Total PUFA	P/S ratio
Sardinella longiceps Sardine	38.1	3.7	3.7	9.2	27.8	43.6	55.7	41.8	0.96
Rastrelliger kanagurta Indian mackerel	34.9	5.7	4.5	12.8	19.3	33.1	58.2	40.6	1.23
Thunnus albacares Yellow fin tuna	32.9	6.7	6.7	9.9	20.5	35.4	58.5	39.6	1.12
Scomberomorus commersoni Spanish mackerel	20.4	4.7	3.1	6.3	13.5	47.1	51.4	25.1	0.53
Caranx stellatus Trevally	21.3	4.0	3.4	7.2	13.7	41.2	50.7	25.3	0.61
Carcharhinus sp. Grey dog shark	25.5	18.4	13.2	8.0	19.5	32.8	62.9	43.9	1.34
Raja mamillidens Skate	25.9	15.6	10.8	9.4	20.0	30.6	69.2	41.5	1.36
Tilapia mossambica Tilapia	25.4	12.2	6.4	5.7	11.5	29.1	62.7	37.5	1.29

TABLE 5. Content of some nutritionally important polyunsaturated fatty acids, as mg per 100 g of fish fillet and of the fish as purchased (AP)—sample 2

Species	C 18:2, n-6 (linoleic)		C 18:3,n-3 (alpha linolenic)		C 20:4, n-6 (arachidonic)		C 20:5, n-3 (eicosa pentaenoic)		C 22:6, n-3 (docosa hexaenoic)	
	Fillet	A P	Fillet	A P	Fillet	A P	Fillet	A P	Fillet	A P
Sardine	tr	tr	54	22	127	51	358	143	1363	545
Indian mackerel	61	25	58	2	71	29	296	118	656	263
Yellow fin tuna	33	28	18	15	83	70	168	142	513	436
Spanish mackerel	35	29	16	14	53	45	104	88	311	264
Trevally	1	1	5	4	29	22	51	38	178	134
Grey dog shark	12	10	3	2	61	52	21	18	176	132
Skate	6	3	3	1	50	25	19	10	180	90
Tilapia	99	40	83	33	45	18	22	9	207	83

DISCUSSION

Total lipids

The concentration of lipid in the fillets of fish is small, varying from 0.8% in gray dog shark to 6.3% in sardine (Table 2), values that differ markedly from those reported by Gopakumar and Nair (10) in Cochin, India, namely 18% for seer, 3% for mullet, 0.8% for tuna and 2.2% for caranx. The fat content of fish would depend on their feeding habits and other environmental factors. The sardine has almost double the amount of lipid found in other species.

Fatty acid composition.

The packed column gas chromatography gives quantitative information on satuarted and unsaturated acids grouped according to chain-length. Capillary gas chromatography provides more accurate identification and quantification of the different fatty acids.

The results of both methods of analyses show that the most abundant saturated acids are palmitic (C 16: 0) and stearic (C 18: 0). The Spanish mackerel and sardine have the highest concentration of palmitic, while stearic is found in high concentration in the skate, shark, Spanish mackerel and tuna. These two acids are abundant in most dietary fats.

As in other naturally occurring fats, oleic acid (C 18: 1, n–9) is the most abundant monounsaturated acid in fish oils, but its concentration is less than in vegetable oils, the highest being in trevally (15.9%) and the lowest in the sardine (3.9%) and Indian mackerel (4.4%).

The distribution of polyunsaturated fatty acids (PUFA) in fish oils is different from that in vegetable oils. Whereas the most abundant PUFA in oils from soya, sesame, corn and sunflower are linoleic (C 18:2 n-6) and to a lesser extent alpha-linolenic (C 18:3 n-3), these two acids are found in relatively low concentration in fish oils. The amount of linoleic (5.5%) and alpha linolenic (4.6%) in tilapia are low when compared with values ranging from 40 to 45% of linoleic in the vegetable oils mentioned.

In contrast to vegetable oils, fish oils are rich in other PUFA which give rise to prostaglandins and their derivatives and play an important role in the coagulation of blood. EPA (C 20:5, n-3) is found in most fish oils in higher concentration than arachidonic, AA (C 18:4, n-6), an acid abundant in animal fats. The highest concentration of EPA is in the small pelagics (sardine and Indian mackerel). A PUFA found in high concentration in all the oils studied is DHA (C 22: 6, n-3). The sardine has the highest concentration (27 8%), the other oils having 11-20%.

When the concentration of these acids in fish as purchased is considered (Table 5) the fish that provide high amounts o" EPA plus DHA, per 100 g purchased, are the sardine (688 mg EPA), the yellow fin tuna (578 mg), the Indian mackerel (380mg) and the Spansih mackerel.

The British National Formulary (No. 19, March 1989) recommends a daily dose of 1.64 g EPA plus 1.09 g DHA in concentrated fish oils, for reducing plasma triglycerides in patients with severe hypertriglyceridaemia judged to be at special risk of ischaemic heart disease. These amounts of EPA and DHA could be obtained from 1.15 kg. sardine, 1.39 kg. Indian mackerel, 1.15kg yellow fin tuna and 1.86 kg of Spanish mackerel. Pressure cooking of small fish such as the sardine and Indian mackerel also permits the consumption of the entire fish, including bones which will add a considerable amount of calcium to rice and vegetable diets that are deficient in this mineral.

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