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# Fatty acid composition and cholesterol content of selected marine fish in Malaysian waters

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### Abstract

A consumer survey identified 10 species of most-preferred marine fish for daily consumption in Malaysia. Total lipids extracted from 10 species of the selected fish were analyzed for their total fat, fatty acids composition and cholesterol content. Most of the fish contained less than 6% lipid by weight and total cholesterol content was 37.1-49.1 mg/100g. The composition of fatty acids showed that total  $\omega$ -3 poly unsaturated fatty acids ( $\omega$ -3 PUFA; 29.7-48.4%) were the highest, followed by other PUFA (27.7-40.0),  $\omega$ -6 PUFA (11.0-20.0%), saturated fatty acid (3.63-11.4%), and finally, mono unsaturated fatty acid (MUFA; 1.37-9.12%). All samples showed a much higher content of  $\omega$ -3 PUFA when compared to standard menhaden oil. Most of the fish had a higher  $\omega$ -3/ $\omega$ -6 ratio (2.16-4.14) than the standard menhaden oil (2.03) except for Four Finger Threadfin (1.50), Indian Mackerel (1.67) and Striped Sea Catfish (1.78). The ratio of PUFA/saturated of the samples ranged from 5.49 to 25.2. © 2001 Elsevier Science Ltd. All rights reserved.

Keywords: Polyunsaturated fatty acid and cholesterol content; Marine fish; In Malaysia

# 1. Introduction

Polyunsaturated fatty acid (PUFA), especially the *n*-3 and n-6 PUFA, have been considered essential fatty acids and have been shown to have curative and preventive effects on cardiovascular diseases, neurodevelopment in infants, cancers and fat glycemic control (Conner, 1997; Kinsella, Lokesh, & Stone, 1990). Although it is generally recognized that PUFA composition may vary among species of fish, little attention has been paid to the PUFA composition of different species when selecting fish for diets. All fish are considered to be of similar nutritional value, and selection is chiefly based on availability, freshness, flavour and similar factors (Hearn, Sgoutas, Hearn, & Sgoutas, 1987). Results of clinical and epidemiological research suggest that eicosapentaenoic (EPA) and docosahexaenoic (DHA) acids, found only in fish and seafoods, have extremely beneficial properties for the prevention of human coronary artery disease (Leaf & Weber, 1988). Generally, the western diet is high in saturated fats and low in unsaturated. Fish components in daily

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meals can be considered as low. Layne et al. (1996) used a level of 3.5 mg/kg/day as the recommended level of PUFA intake. However, the US FDA stated that virgin and partially-hydrogenated menhaden oil is a Generally Recognised as Safe (GRAS) substance and it can be use as a PUFA supplement, for glycemic and Low Density Lipoprotein (LDL) cholesterol controls, provided that the daily intake of EPA and DHA does not exceed 3g/ day (FDA, 1997). To address the imbalance, some researchers have proposed an inclusion of a fish component in the diet (Lovegrove, Brooks, Murphy, Gould, & Williams, 1997), daily supplements of fish oil (Howe, 1996) or incorporation of stabilized fish oil in food products, such a margarine (Saldeen, Wallin, & Marklinder, 1998) and liquid foods (Kolanowski, Swiderski, & Berger, 1999). Therefore, when fish is suggested as a means of improving health, both fat content and the PUFA composition must be considered. Although it is generally recognized that PUFA composition may vary among species of fish, little attention has been paid to the composition of different species when selecting fish for diet (Hearn et al., 1987). Thus this study was carried out to determine fatty acid composition and cholesterol content of common marine fish.

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#### 2. Materials and methods

A short survey was conducted in an urban area to determine the types of fish most preferred by consumers (n=200) in their daily diet. Based on the survey, 10 species of marine fish were selected for analyses. The samples were obtained from local market and were procured at four different times to obtain four replicates. The fish were gutted, washed and filleted. The flesh were weighed (30g) and extraction of fat was performed by homogenising in 100 ml methanol extraction solvent (60% methanol:30% chloroform; Kinsella, Shimp, Mai, & Weihrauch, 1977). To determine the fatty acid composition, 0.2g of the extracted lipid was dissolved in 4 ml hexane and converted to methyl esters (FAME) by reaction with sodium methoxide (Timms, 1978). The resultant FAME on the top layer was pipetted out and dried to 0.2 ml with a stream of nitrogen, prior to gas chromatographic (GC) analysis. Menhaden oil (Sigma Co.) was used as standard PUFA and the identity of individual FAME was compared after conversion to equivalent chain length (Joseph & Seaborn, 1990). The menhaden oil was previously standardised with Sigma 189-19 standard PUFA mix.

Cholesterol was determined using a spectrophotometric method (Bohac, Rhee, Gross, & Ono, 1988). Routine GC analyses were performed on a Shimadzu GC-17A gas chromatograph equipped with FID and electronic pressure controller. The column used was SGE BPX-70 (60 m length×0.25 mm i.d.), split ratio 100:1. The analyses were performed isothermally at 200°C with helium, as carrier gas, flowing at 1.7 ml/min, injection port, 250°C and detector temperature, 260°C.

# 3. Results and discussion

The survey has identified 10 species of fish most commonly used in the everyday diet (Table 1). The Indian mackerel (local name: *kembung*) was the most popular fish (73% of respondents), due to its low cost and suitable size. The next group, with 40–50% response, comprised the Black Pomfret, Spanish Mackerel, Yellow Striped Scad, Silver Pomfret, Hardtail Scad and Whip-tail Stingray. Striped Sea Catfish, Fringescale Sardine and Four Finger Threadfin were probably eaten occasionally.

The fat and cholesterol contents of the fish are shown in Table 2. The species identified contain 1.46–5.77% fat. Based on fat content, most of the fish studied were lean fish, whereby the fat content was lower than 5% by weight (Bennion, 1980) except for Yellow Striped Scad which were medium-fat fish (5–10% fat by weight). The

Table 1 Types of fish preferred by consumers (n = 200)

Common name	Local name (species)	73.4 Respondent (%)	
Indian mackerel	Kembung (Rastrelliger kanagurta)		
Black pomfret	Bawal Hitam (Parastromateus niger)	49.7	
Spanish mackerel	Tenggiri papan (Scomberomorus commersonii)	47.5	
Yellow striped scad	Selar Kuning (Selarides leptolejus)	44.6	
Silver pomfret	Bawal Putih (Pampus argenteus)	40.7	
Hardtail pomfret	Cencaru (Magalapsis cordyla)	40.7	
Whiptail stingray	Pari (Gymnura spp)	40.1	
Striped sea catfish	Sembilang ( <i>Plotosus</i> spp)	16.9	
Fringescale sardine	Tamban (Clupea fimbriata)	13.6	
Fourfinger threadfin	Senangin (Eleutheronema tradactylum)	13.6	

Table 2
Total fat and cholesterol contents<sup>a</sup>

Species	Fat (g/100 g)	Cholesterol (mg/100 g) 41.1±0.38		
Spanish mackerel (Scomberomorus commersonii)	1.46±0.17			
Whiptail stingray (Gymnura spp)	$1.95 \pm 0.14$	$37.1 \pm 0.79$		
Fourfinger threadfin (Eleutheronema tradactylum)	$2.24 \pm 0.20$	45.9±1.74		
Striped sea catfish ( <i>Plotosus spp</i> )	$2.79 \pm 0.37$	$46.9 \pm 0.90$		
Black pomfret (Parastromateus niger)	$2.79 \pm 0.20$	$46.8 \pm 0.10$		
Silver pomfret (Pampus argenteus)	$2.91\pm0.11$	$40.3 \pm 0.50$		
Fringescale sardine (Clupea fimbriata)	$3.06 \pm 0.06$	$41.8 \pm 0.32$		
Hardtail scad (Magalapsis cordyla)	$3.08\pm0.11$	$46.6 \pm 0.86$		
Indian mackerel (Rastrelliger kanagurta)	$4.54 \pm 0.28$	$49.1 \pm 0.06$		
Yellow satriped scad (Selarides leptolejus)	5.77±0.52	47.3±0.31		

<sup>&</sup>lt;sup>a</sup> Numerical data from each column is expressed as average $\pm$ standard deviation (n = 4).

lean fish stores 50–80% of its fat in the form of triacylglycerol in liver and it is a source of fat-soluble vitamins, especially vitamin A and D (Jacquot, 1961). According to Feeley, Criner, and Watt (1972), these low-fat fish have a higher water content and, as a result, their flesh is whiter in colour. The fatty fish instead stores its fat in muscle tissue and the flesh is yellow, gray, pink or another colour (Gurr, 1992). Fat content

is influenced by species, season, geographical regions, age and maturity (Piggot & Tucker, 1990; Tsuchiya, 1961).

Table 2 also shows a trend whereby an increase in fat content is followed by increase in cholesterol levels, as exemplified by the values for Indian Mackerel and Yellow Striped Scad. However, statistically, the trend was not significantly correlated. The cholesterol content

Table 3
Fatty acid compositions of fish oils

Fatty acid	Species										
	Menhaden	Spanish mackerel	Sting ray	Four finger threadfin	Striped sea catfish	Black pomfret	Silver pomfret	Fringe scale sardine	Hardtail scad	Indian mackerel	Yellow striped scad
(a) Saturated fatt	y acid compo	sition (%)									
C14:0	5.97	1.16	0.78	2.42	2.48	2.07	1.63	1.52	1.35	1.04	1.82
C16:0	6.98	2.32	1.29	3.08	3.38	4.66	4.69	4.37	3.15	3.26	2.38
C17:0	0.35	0.58	0.26	0.44	0.90	0.52	0.61	0.76	0.45	0.98	0.53
C18:0	3.51	0.88	0.52	1.10	1.35	1.56	1.43	1.33	1.80	0.98	1.59
C20:0	2.94	0.29	0.26	0.22	0.68	0.52	0.20	0.76	0.45	0.81	0.26
C22:0	0.38	0.58	0.26	0.22	0.45	0.52	0.61	0.76	0.90	0.16	0.26
C23:0	0.42	0.29	_	0.22	0.45	0.52	_	0.38	0.90	0.16	0.26
C24:0	0.26	0.29	0.26	0.44	0.45	1.04	_	_	0.45	0.32	0.79
$\Sigma$ SATD.	20.81	6.39	3.63	8.14	10.14	11.39	9.17	9.88	9.45	7.71	7.89
(b) Monounsatura	nea jany acu 9.64	1.16	1.04	0.44	0.23	0.52	1.23	2.85	0.90	0.16	2.34
C16:1 C18:1	9.64 7.05	2.05	2.06	0.44	0.23	1.56	1.23	3.80	1.35	1.79	2.34 2.57
C20:1	2.34	0.58	0.26	0.22	0.23	1.04	0.20	0.95	0.90	0.98	0.26
C22:1	0.40	0.29	0.52	0.44	0.23	0.52	0.61	1.52	0.45	0.32	0.53
Σ MUFA	19.43	4.08	3.88	1.98	1.37	3.64	3.64	9.12	3.60	3.25	5.70
(c) ω-6 Polyunsat		acid (PUFA		ositions (%)							
C16:2ω6	0.53	_	0.26	_	0.45	_	0.20	_	_	0.16	_
C18:2ω6	10.6	9.65	12.4	18.0	13.2	9.84	12.0	8.74	7.66	16.5	9.08
C18:3ω6	1.79	0.58	1.55	0.66	0.68	1.04	0.82	1.32	0.90	1.95	0.79
C20:2ω6	0.16	_	0.26	0.22	0.68	0.26	-	_	1.35	0.16	0.52
C20:3ω6	0.36	0.29	0.26	0.22	0.45	0.78	0.20	0.19	0.45	0.33	0.26
C20:4ω6	0.47	0.29	0.26	0.22	0.68	0.52	0.60	0.19	0.45	0.33	0.26
C22:4ω6	0.11	_	0.52	_	_	0.26	_	0.19	-	0.16	0.53
C22:5ω6	0.21	0.29	2.58	0.44	1.80	0.78	0.20	0.38	0.45	0.49	0.53
Σ PUFA ω6	14.25	11.10	18.06	19.78	17.96	13.48	13.46	11.01	11.26	20.03	11.97
(d) ω-3 PUFA con	mpositions (9	26)									
C16:3ω3	9.06	4.68	5.67	8.13	7.43	4.15	7.14	4.16	3.60	6.19	4.23
C16:4ω3	0.10	0.58	_	0.44	0.23	1.56		0.19	1.35	0.16	0.26
C18:3ω3	0.68	0.58	0.52	0.44	0.68	4.52	0.61	1.32	0.90	2.44	0.79
C18:4ω3	1.36	0.29	0.26	0.22	0.45	0.52	0.61	0.19	0.45	0.98	0.26
C20:3ω3	0.93	3.22	7.22	3.52	4.05	1.56	0.61	1.89	2.50	2.12	2.38
C20:4ω3	0.35	0.29	0.26	0.22	0.23	0.52	0.61	0.19	_	0.16	0.26
C20:5ω3	8.54	5.85	5.15	5.93	6.76	5.18	0.82	4.35	5.76	4.72	3.97
C21:5ω3	0.72	0.29	_	_	0.23	_		0.38	_	0.16	_
C22:5ω3	0.92	3.80	2.32	0.88	1.58	3.11	2.46	0.89	2.70	1.95	1.85
C22:6ω3	6.20	23.3	17.5	9.89	10.4	9.36	18.9	17.3	28.6	14.5	27.3
$\Sigma$ PUFA $\omega$ 3	28.87	42.89	38.93	29.67	32.00	30.48	31.73	30.86	47.89	33.38	41.25
(e) Other types of	PIJFA com	nositions (%	á )								
C16:2	0.65	20.5	22.94	_	18.5	18.1	24.9	23.4	14.9	23.9	19.3
C16:3w4	10.6	_	_	28.57	-	0.52	_	0.38	-	0.16	-
C16:4w1	3.46	2.34	1.80	2.20	4.73	2.59	2.45	2.84	1.80	3.42	2.65
C18:2	1.18	8.77	9.54	8.79	10.4	9.33	8.98	6.10	9.71	7.82	10.1
C18:3w4	0.73	0.88	- -	0.22	0.45	- -	-	1.90	0.90	-	-
C18:4w1	-	0.38	0.26	0.22	-	_	_	0.38	0.45	_	0.26
		0.27	0.20	0.22				0.50	0.15		32.27

analysed ranged from 37.1 mg/100 g to 49.1 mg/100 g. These values were within the range found in Robert and Marcus (1990) findings (30–60 mg/100 g) in marine fish sample. Cholesterol content in fish is influenced by several factors, among them the PUFA content. According to Kinsella (1986), an increase in PUFA content will be followed by a decrease the cholesterol level.

Detailed fatty acid compositions are listed in Table 3; and total fatty acid contents are summarised in Table 4. Generally, the PUFA contents were much higher (56-92%) than the saturated fatty acids (3.63-11.4%) and MUFAs are lowest (1-10%). The trend is different when compared to an earlier study on fresh water fish, where the concentrations of MUFA were higher than the saturated and PUFA (Suriah, Teh, Osman, & Nik-Mat, 1995). Other researchers have also shown that freshwater fish have lower contents of PUFAs (Vlieg & Body, 1988). The difference can be attributed to the fact that freshwater fishes feed largely on vegetation and plant materials, whereas marine fish staple diets are mainly zooplanktons, rich in PUFAs. Menhaden oil, which is a marine fish oil, has been proposed by the USFDA (FDA, 1997), as a PUFA supplement. The concentrations of ω-3 PUFA (29.7%–48.4%) and other PUFA (27.7%–40.0%) in this study were found to be much greater than the standard menhaden oil (ω-3 PUFA, 28.9%; other PUFAs 16.6%00, whereas ω-6 PUFAs are 14.3%). The values for MUFA (1.37-9.12%) and saturated fatty acids (3.63–16.38%), in the fish analysed, were lower than menhaden oil (MUFA, 19.4%; saturated, 20.2%). This study has shown that

marine fish were richer in  $\omega$ -3 PUFAs (29.7–48.4%) than  $\omega$ -6 PUFAs (11.0–20.0%). Our earlier study (Suriah et al., 1995) and other investigators (O'Dea & Sinclair, 1982; Suzuki, Okazaki, Hayakawa, Wuda, & Tamaura, 1986), on freshwater and cultured fish, had shown pposite results, where the levels of  $\omega$ -3 PUFAs were lower than  $\omega$ -6 PUFAs. However, Wang, Miller, Perren, and Addis (1990) reported similar findings, in that marine fish were rich in  $\omega$ -3, especially DHA and EPA.

Fish oils have been proposed as antithrombotic dietary supplements. The rationale is that oils rich in C20:5ω-3 would provide substrates for the production of platelet anti-aggregatory factors, such as TXA<sub>3</sub> and PGI<sub>3</sub>, but concern has been expressed over the wisdom and efficacy of such proposals (Budowski, 1981; Goodnight, Harris, Connor, & Illingworth, 1982). While the 3 series prostanoids may be antithrombotic, there is no doubt that the 2 series prostaglandins, derived from C20:4ω-6, have a diverse and essential role to play in the metabolic processes of the body.

The contents of arachidonic acid (AA), EPA and DHA, of the fish analysed, ranged from 0.19–0.68%; 0.82–6.76% and 9.36–28.6%, respectively. Bowman and Rand (1980) reported that arachidonic acid (C20:4ω-6) is a precursor for prostaglandin and thromboxan which will influence the blood clot and its attachment to the endothelial tissue during wound healing. Apart from that, the acid also plays a role in growth. Fish that have higher contents of AA than menhaden oil (0.47%) were Striped Sea Catfish (0.68%), Silver Pomfret (0.60%)

Table 4
Total fatty acid contents (%) in fish oils

Species	Saturated	Mono- unsaturated	Poly- unsaturated ω-6	Poly- unsaturated ω-3	Other poly- unsaturated	Total poly- unsaturated
Black pomfret (Parastromateus niger)	11.4	3.64	13.5	30.5	30.6	74.5
Silver pomfret (Pampus argenteus)	9.17	3.27	13.5	31.7	36.3	81.5
Hardtail scad (Magalapsis cordyla)	9.45	3.60	11.7	48.4	27.7	86.9
Indian mackerel (Rastrelliger kanagurta)	7.71	3.25	20.0	33.4	35.3	56.8
Whiptail stingray (Gymnura spp)	3.63	3.88	18.0	38.9	34.5	91.5
Yellow striped scad (Selarides leptolejus)	16.1	11.4	12.0	41.3	32.3	85.5
Striped sea catfish (Plotosus spp)	10.1	1.37	18.0	32.0	34.0	84.0
Fourfinger threadfin (Eleutheronema tradactylum)	16.3	3.96	19.78	29.7	40.0	89.5
Fringescale sardine (Clupea fimbriata)	9.88	9.12	11.0	30.9	35.0	76.9
Spanish mackerel (Scomberomorus commersonii)	6.39	4.08	11.1	47.9	32.8	86.7
Menhaden oil	20.8	19.43	14.3	28.9	16.6	59.8

and Black Pomfret (0.52%). Based on an earlier study (Suriah et al., 1995), in cases, the contents of AA in marine fishes were lower than freshwater fishes.

The DHA contents of all the fish studied are higher than the amount found in menhaden oil (6.20%), and the highest level, found in Hardtail Scad (28.6%), was almost five times higher. The amounts of EPA in the species studied (0.82–6.76%) are lower than the menhaden oil (8.54%). Fish is a major component in daily Malaysian diet, and traditionally it is a major source of protein. It is estimated that fish contribute about 60–70% of protein intake (Malaysian Agricultural Directory & Index, 1993/1994). The annual per capita consumption of fish is estimated to be 39 kg, and is expected to increase to 45 kg in the year 2000 (Anon., 1995).

Piggott and Tucker (1990) suggested that the  $\omega$ -3: $\omega$ -6 ratio is a better index in comparing relative nutritional value of fish oils of different species. Table 5 shows that ratios of ω-3:ω-6 in oils extracted from the species studied. However, there is no recommended intake in terms of  $\omega$ -3:  $\omega$ -6 ratios but evidence in wild animals and estimated nutrient intake during human evolution suggest a diet ratio of 1:1 (Simopoulos, 1989). There was no correlation found between the ratio of  $\omega$ -6: $\omega$ -3 fatty acids and the polyunsaturated fatty acid. This is in line with what was reported by Iritani and Fujikawa (1982). This fact is further strengthened by the high ratio PUFA/Saturated (P/S ratio), which indicates a good supply of PUFA relative to saturated acids. The P/S ratios obtained from this study (5.30–25.2) were higher than that of the standard menhaden oil (2.87). The fish

Table 5 The polyunsaturated fatty acids/saturated (P/S) and  $\omega\text{-}3/\omega\text{-}6$  ratios of fatty acids from fish oils

Species	Ratio of P/S	Ratio of $\omega$ -3/ $\omega$ -6
Black pomfret	6.54	2.26
(Parastromateus niger)		
Silver pomfret	8.89	2.36
(Pampus argenteus)		
Hardtail scad	9.19	4.14
(Magalapsis cordyla)		
Indian mackerel	7.37	1.67
(Rastrelliger kanagurta)		
Whiptail stingray	25.2	2.16
(Gymnura spp)		
Yellow striped scad	5.30	3.45
(Selarides leptolejus)		
Striped sea catfish	8.28	1.78
(Plotosus spp)		
Fourfinger threadfin	5.49	1.50
(Eleutheronema tradactylum)		
Fringescale sardine	7.78	2.80
(Clupea fimbriata)		
Spanish mackerel	13.6	4.31
(Scomberomorus commersonii)		
Menhaden oil	2.87	2.03

identified in this study were found to be good sources of marine PUFA. Studies from Scandinavia, The Netherlands and Japan showed that people who eat fish about twice a week (240 g total weekly intake) have lower risks of heart attacks than people who rarely eat fish (Wardlaw, Insel, & Seigler, 1992). Thus, the suggestion to include 2–3 servings of fish per week (80–120 g), recommended for the Malaysian diet, would be sufficient to supply the required level of PUFAs , without the need for any supplementation.

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## References

Anon. (1995). 3rd National Agriculture Policy (1998–2010), Department of Fisheries, Ministry of Agriculture, Malaysia.

Bennion, M. (1997). Introductory Foods (7th ed.) MacMillan: New York, USA.

Bohac, C. E., Rhee, K. S., Cross, H. R., & Ono, K. (1988). Assessment of methodologies for calorimetric cholesterol assay. *Journal of Food Science*, 53 (6), 1642–1644.

Bowman, W. C., & Rand, M. J. (1980). *Textbook of Pharmacology* (2nd ed.). Oxford, UK: Blackwell Scientific Publication.

Budowski, P. (1981). Nutritional effects al n-3 polyunsaturated fatty acids. *Israel Journal of Medical Sciences*, 17, 223.

Conner, W. E. (1997). The beneficial effects of omega-3 fatty acids:cardiovascular disease and neurodevelopment. *Current Opinion in Lipidology*, 8, 1–3.

Feeley, R. M., Criner, D. E. C., & Watt, B. K. (1972). Cholesterol content of foods. *Journal of the American Dietetic Association*, 61, 134–148.

Food and Drug Administration. (1997). Federal register — rules and regulations. Food labeling: requirements for nutrient content claims, health claims, and statements of nutritional support for dietary supplements. 62 (108), 49859–49868.

Goodnight, S. H. Jr., Harris, W. S., Connor, W. E., & Illingworth, R. D. (1982). Polyunsaturated fatty acids, hyperlipidemia and thrombosis. *Artheriosclerosis*, 2, 87.

Gurr, M. I. (1992). Role of fats in food and nutrition (2nd ed.). London, UK: Elsevier Applied Science.

Hearn, T. L., Sgoutas, S. A., Hearn, J. A., & Sgoutas, D. S. (1987).Polyunsaturated fatty acids and fat in fish flesh for selecting species for health benefits. *Journal of Food Science*, 52 (5), 1209–1215.

Howe, P. R. C. (1996). Fish oil supplements and hypertension. *International Society for the Study of Fatty Acids and Lipids: ISSFAL Newsletter*, 34, 2–5.

Iritani, N., & Fujikawa, S. (1982). Competitive incorporation of dietary ω-3 and ω-6 polyunsaturated dietary acids into the tissue phospholipids in rats. *Journal of Nutritional Science and Vitaminology*, 28, 6214.

Jacquot, R. (1961). Organic constituents of fish and other aquatic animal foods. In G. Borgstrom, Fish as Food (Vol. 1, pp. 146–209). New York: Academic Press USA.

Joseph, J. D. & Seaborn, G. T. (1990). The analysis of marine fatty acids. In H. E. Stansby, Fish oil in Nutrition (pp. 40–72). New York: Van Nostrand Reinhold.

Kinsella, J. E. (1986). Food component with potential benefits: the n-3 polyunsaturated fatty acids of fish oils. *Food Technology*, 40(2), 89–97.

- Kinsella, J. E., Lokesh, B., & Stone, R. A. (1990). Dietary n-3 polyunsaturated fatty acids and amelioration of cardiovascular disease: possible mechanisms. *American Journal of Clinical Nutrition*, 52, 1–28.
- Kinsella, J. E., Shimp, J. L., Mai, J., & Weihrauch, J. (1977). Fatty acid content and composition of freshwater fin fish. *Journal of the American Oil Chemists Society*, 54, 424–429.
- Kolanowski, W., Swiderski, F., & Berger, S. (1999). Possibilities of fish oil application for food products enrichment with omega-3 PUFA. *International Journal of Food Science and Nutrition*, 50, 39–49.
- Layne, K. S., Goh, Y. K., & Jumpsen, J. A. (1996). Normal subjects consuming physiological levels of 18:3(n-3) and 20:5(n-3) from flaxseed or fish oils have characteristic differences in plasma lipid and lipoprotein fatty acid levels. *Journal of Nutrition*, 126, 2130–2140.
- Leaf, A., & Weber, P. C. (1988). Cardiovascular effects of n-3 fatty acids. New England Journal of Medicine, 318, 549–555.
- Lovegrove, J. A., Brooks, C. N., Murphy, M. C., Gould, B. J., & Williams, C. M. (1997). Use of manufactured foods enriched with fish oils as a means of increasing long-chain n-3 polyunsaturated fatty acid intake. *British Journal of Nutrition*, 67, 223–236.
- Malaysia Agricultural Directory & Index. (1993/1994). Major Agricultural Industries-Aquaculture (5th ed.). Pantai Maju Sdn. Bhd
- Mohsen, E. R. (1985). NIH Launching major research program on fish oils and health food. *Chemical News*, 6, 34–39.
- O'Dea, K., & Sinclair, A. J. (1982). Increased proportion of arachidonic acid in plasma lipids after 2 weeks on a diet of tropical seafoods. *American Journal of Clinical Nutrition*, 36, 868–872.
- Piggott, G. M., & Tucker, B. W. (1990). Effects of technology on nutrition. New York: Marcel Dekker.

- Robert, S. L., & Marcus, K. (1990). Omega-3 fatty acids in health and disease. New York: Marcel Dekker.
- Saldeen, T., Wallin, R., & Marklinder, I. (1998). Effects of a small dose of stable fish oil substituted for margarine in bread on plasma phospholipid fatty acids and serum triglycerides. *Nutr. Res.*, 18, 1483–1492.
- Simopoulos, A. P. (1989). Summary of NATO advanced research workshop on dietary w-3 and w-6 fatty acids: biological effects and nutritional essentiality. *Journal of Nutrition*, 199, 512–528.
- Suriah, A. R., Teh, S. H., Osman, H., & Nik-Mat, D. (1995). Fatty acid composition of some Malaysian freshwater fish. Food Chemistry, 54, 45–49.
- Suzuki, H., Okazaki, K., Hayakawa, S., Wuda, S., & Tamaura, S. (1986). Influence of commercial dietary fatty acids on PUFA of cultured freshwater fish and comparison with those of wild fish of the same species. *Journal of Agricultural and Food Chemitry*, 34, 58–60.
- Timms, R. E. (1978). Artefact peaks in the preparation and gas liquid chromatographic determination of methyl esters. *Australian Journal of Dairy Technology*, March 4–5.
- Tsuchiya, T. (1961). Biochemistry of fish oil. In G. Borgstrom, *Fish as Food* (pp. 1–58). New York, USA: Academic Press.
- Vlieg, P., & Body, D. B. (1988). Lipid contents and fatty acid composition of some New Zealand freshwater finfish and marine finfish, shellfish and roes. New Zealand Journal of Marine Freshwater Research, 22, 151.
- Wang, Y. J., Miller, L. A., Perren, M., & Addis, P. B. (1990). Omega-3 fatty acids in Lake Superior fish. *Journal of Food Science*, 55(1), 71–73.
- Wardlaw, G. M., Insel, P. M., & Seigler, M. F. (1992). Contemporary nutrition-issues and insights. St. Louis, USA: Mosby.