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Effects of season and location of catch on the fatty acid compositions of some Australian fish species

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Fatty acid compositions and lipid contents of five common table species of marine fish from temperate Australian waters were determined for samples taken during two seasons (four species) and three locations of markedly differing latitude (one species). Variabilities in total lipid extract and phospholipid fatty acid compositions were assessed between samples of each species, as well as between individuals of the same sample. These were then compared with data obtained for the other species analysed. The highest n3 polyunsaturated fatty acid (PUFA) content lipids were found in fish from colder waters (southern locations and spring season catches). The leaner species were found to have much more stable lipid characteristics with changes in the water temperature at sampling.

NOTATION

FA	Fatty acid
L	Lipid
M	Monounsaturated
n3	Omega-3
n6	Omega-6
P	Polyunsaturated
S	Saturated

INTRODUCTION

Numerous clinical and epidemiological studies over the past 40 years have correlated the long-term consumption of polyunsaturated fatty acids (PUFAs) of the n3 series with a reduced incidence of a range of common degenerative diseases, as well as an alleviation of some symptoms (Goodnight *et al.*, 1982; Kinsella, 1988; Harris, 1989).

Fish are the richest, widely available source of n3 PUFAs in the human diet. Their fatty acid compositions and contents, however, are known to vary significantly, even between individuals of the same species. A number of environmental factors, such as the nature and availability of food, and the season, location and year of catch, are believed to be important contributors to this variation (Gruger, 1967; Exler *et al.*, 1975). Despite the importance to humans of the nature of the fish lipids in their diet the majority of fish consumed today (87·7%, Hillcoat, 1990) are from catches of wild fish from the world's oceans, rather than from farmed fish fed on a controlled diet. Knowledge of the influence of sampling factors on lipid content and composition could enable a more informed selection of fish by consumers. However, despite the demonstrated importance of sampling factors in determining the nutritional quality of fish lipids, few authors have stated the season, year, water temperature or even precise location from which their samples were obtained.

Some authors (Reiser *et al.*, 1963; Ackman, 1980; Sinclair *et al.*, 1984; Evans *et al.*, 1986) have noted a correlation between low water temperatures and desirable lipid characteristics, such as high P/S and n3/n6 ratios. This is believed to be due primarily to variation in the dietary fatty acid profiles of the fish (Kayama *et al.*, 1963; Reiser *et al.*, 1963; Ackman, 1967; Gruger, 1967; O'Dea & Sinclair, 1982). It has also been demonstrated (Reiser *et al.*, 1963) that saturated fatty acids are not incorporated into fish tissue lipids at low water temperatures. Very little work has been carried out, however, to assess the extent of the variability in fish lipid content and fatty acid composition with changes in factors affecting water temperatures, such as the season and latitude of catch. This may be due to the fact that few species are common to wide ranges of latitude, as well as comparisons with existing data being made difficult due to the lack of reported sampling details.

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This study reports the fatty acid compositions and lipid contents of samples of five species of fish from temperate Australian waters. Four of these species were sampled during opposing seasons (spring and autumn), while one was sampled at three locations of differing latitude during the same season.

MATERIALS AND METHODS

Samples and their preparation

Each sample consisted of 10 fish of similar adult size taken from the same catch at a particular season and location, as detailed in Table 1a.

Fillets were removed and stored at −22°C prior to thawing and mincing of the white muscle only. Lipid was extracted as reported previously (Armstrong *et al.*, 1991), with calcium chloride (approximately 2 mg) added to the aqueous phase to ensure salting out of the more polar phospholipids into the organic (chloroform) phase. Care was taken throughout to avoid contact with plastics, so as to prevent contamination of gas chromatographic (GC) traces with plasticisers.

Isolation of the phospholipids was achieved using aminopropyl bonded phase cartridges (500 mg sorbent, Waters, MA) by the method of Kaluzny *et al.* (1985).

Fatty acid methyl esters (FAMEs) were formed from both the total lipid extracts and the isolated phospholipids (after neutralising the latter with methanolic sodium hydroxide) by reacting with methanolic sodium methoxide (0·5 M) at room temperature (Shehata *et al.*, 1970).

Gas chromatographic analysis and peak identification

Gas chromatographic analyses were carried out as reported previously (Armstrong *et al.*, 1991).

GC–MS was performed on an Hewlett-Packard gas chromatograph (HP 5890 Series II) with split injector (ratio 1 : 30) coupled to an Hewlett-Packard mass-selective detector (HP5971A) operating in the EI mode. The ion source was maintained at 190°C and data were collected at 0·9 scans/s. A DB-23 fused silica capillary column (30 m × 0·22 mm i.d. × 0·25 μm, J&W Scientific) was used. It was held at 175°C for 12 min and then heated at 5°C/min to 220°C, at which temperature it was held for a further 9 min. This temperature gradient was used for all GC runs performed. Detector and injector temperatures were set at 280 and 250°C, respectively.

Authentic mass spectra were obtained from FAMES purchased from the Sigma Chemical Co., and Alltech Assoc. Inc.

Data handling

Peak areas were corrected for relative theoretical response factors to FID (Bannon & Craske, 1988). FAMEs were then recorded as percentages of the total fatty acid content.

Mean fatty acid percentage compositions and coefficients of variability were calculated for individual fatty acids of each sample.

Comparisons between sets of fatty acid percentage area profiles were carried out using Principal Components Analysis (PCA) on standardised data matrices of all values exceeding 0·5–1·0% composition (depending on the number of data sets being compared and memory constraints). Two-dimensional scatterplots of the first two principal components were constructed from the results. The percentage of the total variances represented by these plots were significant and are stated in each case. The validity of clusters produced was tested by the construction of spider-web plots (not shown) to display all principal components whose eigenvalues exceeded 1·0—a criterion which was found consistently to require a larger number of components than other tests attempted (including the Scree test, Bartlett's significance test and selection of the number of components required to explain over 90% of the total variance). Scatterplots were also overlayed with data vectors representing the variables (fatty acids) to indicate correlation of each data point with particular fatty acids.

Table 1a. Fish sampling details

Species scientific name (common name)	Location of catch	Time of catch	Water temperature (°C)
Acanthopagrus australis (Black/Silver bream)	Sydney, NSW (33°53'S)	Spring 1989	16–20
		Autumn 1990	20–23
	Hervey Bay, Qld (24°40'S)	Spring 1990	24–25
	Lakes Entrance, Vic (37°50'S)	Spring 1990	12–14
Centroberyx affinis (Nannygai/Redfish)	Wollongong, NSW (34°25'S)	Spring 1989	16–18
		Autumn 1990	21–23
Zeus faber (John dory)	Sydney, NSW (33°53'S)	Spring 1989	16–20
		Autumn 1990	20–23
Zenopsis nebulosus (Mirror dory)	Ulladulla, NSW (35°21'S	Spring 1989	16–17
Genypterus blacodes (Ling)	Ulladulla, NSW (35°21'S)	Spring 1989	16–17
		Autumn 1990	20–23

Table 1b. Fatty acid methyl ester (FAME) peak identification

Peak order	Identity	Basis for identification	
		Mass spectrum	Retention time[a]
1	14:0	+	+
2	15:0i	+	–
3	15:0	+	+
4	16:0i	+	–
5	16:0	+	+
6	16:1n7	+	+
7	17:0i	+	–
8	17:1n?	+	–
9	17:0	+	+
10	17:1n8	+	–
11	U1 (18:0i?)[b]	–	–
12	18:0	+	+
13	18:1n9	+	+
14	18:1n7	+	–
15	U2 (18:1n5?)[b]	–	–
16	18:2n6 (LA)	+	+
17	U3 (18:2n3?)[b]	–	–
18	18:3n6	–	+
19	18:3n3 (LN)	+	+
20	U4[b]	–	–
21	18:4n3	–	+
22	22:0	–	+
23	20:1n9	+	+
24	20:1n7	+	–
25	20:1n5	+	–
26	20:2n9?[c]	–	–
27	20:2n6	+	–
28	20:3n6	–	+
29	20:4n6 (AA)	+	+
30	U5 (20:3n3?)[b]	–	–
31	20:4n3	+	–
32	20:5n3 (EPA)	+	+
33	22:1n11	+	+
34	22:1n9?[c]	–	–
35	22:2n6	–	+
36	22:2n3	–	+
37	22:3n3	–	+
38	22:4n6	+	+
39	21:5n3	+	–
40	22:5n3 (DPA)	+	–
41	22:6n3 (DHA)	+	+

[a] Compared with that of an authentic standard.
[b] U*x*, unknown peak *x* (U1–3 and U5 tentatively identified from relative positions compared with assignments made by other authors).
[c] Tentatively identified from peak position.

Two-tailed parametric (ANOVA) and non-parametric (Mann–Whitney) univariate tests were performed at the 95% confidence level (α = 0·05). Non-parametric comparisons of more than two data sets were made using Kruskal–Wallis single-factor analysis.

ANOVA and PCA calculations and plots were performed using a statistical software package (STATGRAPHICS, Statistical Graphics Corporation, Rockville, MD, USA).

RESULTS AND DISCUSSION

The lipid contents, fatty acid compositions and some nutritionally important values calculated from the latter are presented in Tables 2a and 2b for whole lipid extracts of all samples. Phospholipid fatty acid data are also given for Black bream and Redfish (Tables 3a and 3b). Means were calculated from the data obtained from the analysis of 10 individual fish from the one sample.

A total of 41 FAMEs were resolved and identified. These are listed in Table 1b, together with the method of identification used. Tentative identification of peaks for which no authentic standard was available was made by comparisons with the nature of surrounding peaks and the order of appearance established by other authors. The fatty acids determined were similar to those reported generally for fish. The nutritionally important n3/n6 and P/S ratios were also typical, while the low lipid contents meant that they were all classified as lean.

Lipid content and fatty acid composition

The data for total lipid extracts displayed large variabilities, even though the individual fish were of the same species, location and time of catch and were of similar adult size. This finding supports suggestions that individual fish should be analysed (Stansby, 1981; Brown *et al.*, 1989), rather than pooled samples. Sample sizes as large as is practicable should also be used, instead of the common practice of using only a few individual fish (Pearson, 1977; Pearson, 1978; Gibson, 1983; Gibson *et al.*, 1984; Fogerty *et al.*, 1986; Hearn *et al.*, 1987; Sinclair *et al.*, 1992).

The phospholipids of marine organisms usually have higher PUFA (Brockerhoff *et al.*, 1963) and lower monounsaturated fatty acid levels (Ackman, 1980) than the triglyceride fraction. PUFAs are preferentially incorporated and maintained in the phospholipids to ensure membrane permeability and fluidity even in low water temperatures.

The relative amounts of triglyceride and phospholipid in fish muscle depends on the tissue lipid content. Lean fish, such as those reported in this study, have their muscle lipids almost entirely comprised of phospholipids—storing the bulk of their triglyceride in the liver.

The extremely lean Ling, caught during spring, were found to have phospholipid fatty acid profiles very closely resembling those of their total lipids. This reflects the fact that their fatty acids were almost all bound in phospholipid molecules, thus allowing for little contribution to the total lipid profile by triglycerides.

The other species for which phospholipids were analysed contained more tissue lipid (and, therefore, more triglyceride) than the spring Ling sample. Accordingly, their phospholipid profiles revealed higher PUFA and lower saturated and monounsaturated fatty acid levels than the total bound fatty acid profiles.

Within samples displaying a fairly wide range of lipid contents, and thus triglyceride levels (namely, the spring samples of Black bream and Redfish), trends were found for increased saturates and monounsaturates and decreased PUFAs in fattier individuals. Again, these trends are consistent with greater trigly-

Table 2a. Fatty acid compositions of total lipid extracts (data given as mean (n = 10) percentage of total fatty acid mass (with % variabilities))

Peak elution order	Fatty acid identity	Black bream				Redfish		John dory		Ling		Mirror dory
		New South Wales		Victoria	Queensland	New South Wales		New South Wales		New South Wales		New South Wales
		Spring	Autumn	Spring	Spring	Spring	Autumn	Spring	Autumn	Spring	Autumn	Spring
1	14:0	1·56 (28·4)	4·71 (27·7)	3·54 (56·4)	1·64 (78·1)	2·29 (28·8)	4·57 (32·0)	1·50 (74·9)	2·00 (54·1)	1·78 (68·5)	1·93 (50·4)	2·94 (56·6)
2	15:0i	0·70 (24·5)	1·36 (33·8)	0·51 (48·4)	0·60 (31·5)	0·41 (10·5)	0·73 (44·2)	0·54 (37·7)	0·39 (59·2)	0·26 (125)	0·64 (53·1)	0·55 (48·6)
3	15:0	0·00 (—)	0·00 (—)	0·25 (126)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·02 (300)	0·00 (—)
4	16:0i	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)
5	16:0	17·4 (4·71)	28·9 (12·7)	22·3 (31·9)	19·7 (17·9)	17·8 (1·83)	22·0 (18·16)	24·2 (7·27)	27·8 (25·3)	23·0 (26·0)	24·8 (16·9)	25·3 (21·0)
7	17:0i	0·24 (78·6)	0·65 (63·8)	0·08 (300)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)
9	17:0	1·19 (21·1)	2·34 (44·4)	0·45 (300)	0·14 (174)	0·78 (12·2)	2·01 (80·3)	0·89 (23·3)	1·08 (25·0)	0·55 (182)	0·09 (203)	3·53 (204)
12	18:0	7·71 (13·9)	8·24 (12·6)	7·39 (24·6)	6·97 (10·8)	6·63 (9·3)	7·63 (13·9)	6·75 (8·78)	7·38 (24·2)	5·95 (12·8)	8·34 (16·3)	5·11 (15·0)
22	20:0	0·30 (25·3)	1·38 (110)	0·04 (201)	0·03 (300)	0·29 (16·0)	0·38 (57·5)	0·00 (—)	0·00 (—)	1·96 (130)	0·00 (—)	0·00 (—)
6	16:1n7	5·27 (18·2)	9·76 (19·9)	9·57 (28·9)	6·72 (28·6)	3·55 (24·2)	6·94 (17·7)	2·54 (45·9)	4·43 (33·5)	3·50 (37·6)	3·29 (61·4)	2·32 (46·3)
8	17:1n?	0·56 (44·5)	1·79 (81·2)	0·18 (26·1)	0·15 (214)	0·37 (17·5)	0·34 (95·9)	0·95 (8·74)	0·84 (26·9)	0·57 (72·9)	0·93 (174)	1·03 (38·7)
10	17:1n8	0·76 (73·2)	1·11 (69·8)	0·19 (128)	0·00 (—)	0·39 (13·8)	0·40 (66·6)	0·16 (200)	0·07 (300)	0·60 (107)	0·73 (196)	0·06 (300)
13	18:1n9	16·4 (28·5)	18·9 (14·3)	17·6 (27·0)	11·2 (28·1)	21·7 (22·9)	30·1 (8·80)	12·9 (9·68)	14·8 (20·2)	17·9 (12·2)	19·2 (14·5)	14·4 (26·9)
14	18:1n7	0·17 (15·8)	1·20 (51·0)	0·00 (—)	0·00 (—)	0·01 (300)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·42 (207)	0·00 (—)
23	20:1n9	0·44 (44·3)	0·12 (300)	0·05 (300)	0·05 (272)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)
24	20:1n7	0·61 (17·7)	0·39 (88·8)	0·29 (92·6)	0·06 (288)	2·24 (33·3)	4·51 (24·8)	0·46 (83·6)	0·80 (68·6)	1·01 (72·7)	0·98 (23·0)	5·21 (41·0)
25	20:1n5	0·54 (61·5)	0·37 (140)	0·17 (200)	0·16 (165)	0·11 (87·5)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)
33	20:1n11	0·63 (62·0)	1·08 (147)	0·89 (67·9)	1·09 (42·4)	1·64 (28·5)	2·04 (48·6)	0·04 (300)	1·14 (94·0)	0·35 (166)	0·45 (104)	1·48 (64·6)
34	22:1n9	0·89 (46·5)	0·63 (108)	1·39 (95·9)	0·10 (201)	0·00 (—)	0·02 (300)	0·00 (—)	0·00 (—)	0·40 (201)	0·00 (—)	0·27 (253)
19	18:3n3 LN	0·47 (61·3)	0·36 (159)	0·07 (203)	0·60 (165)	0·26 (64·0)	0·06 (200)	2·10 (40·0)	0·05 (300)	1·34 (123)	0·00 (—)	0·00 (—)
21	18:4n3	0·51 (46·4)	0·38 (107)	0·27 (138)	0·05 (201)	0·31 (59·6)	0·35 (61·5)	0·06 (300)	0·07 (300)	0·00 (—)	0·00 (—)	0·00 (—)
31	20:4n3	0·57 (38·1)	0·75 (91·4)	0·77 (128)	0·28 (214)	0·53 (27·7)	0·35 (54·0)	0·27 (84·1)	0·33 (54·2)	0·21 (111)	0·35 (74·7)	1·63 (29·1)
32	20:5n3 EPA	7·88 (24·6)	3·49 (28·2)	5·37 (32·3)	6·31 (20·0)	4·08 (13·9)	2·21 (16·5)	5·14 (13·7)	5·07 (27·6)	4·80 (28·1)	3·96 (39·1)	3·84 (13·7)
36	22:2n3	0·84 (36·7)	0·30 (75·4)	0·21 (130)	0·07 (167)	0·23 (52·2)	0·09 (133)	0·16 (135)	0·09 (13·1)	0·00 (—)	0·06 (300)	0·09 (300)
37	22:3n3	0·22 (61·7)	0·09 (300)	0·40 (207)	0·00 (—)	0·06 (173)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)
39	21:5n3	1·19 (33·6)	0·18 (106)	0·10 (59·6)	2·36 (40·9)	1·16 (37·2)	0·25 (84·1)	1·02 (13·0)	0·65 (49·6)	0·79 (74·3)	0·57 (80·0)	0·97 (75·0)
40	22:5n3 DPA	6·02 (17·4)	2·50 (29·2)	4·67 (47·1)	5·37 (24·7)	2·98 (10·4)	1·50 (35·3)	2·94 (15·4)	2·46 (28·0)	1·53 (45·5)	1·33 (31·4)	2·06 (29·2)
41	22:6n3 DHA	17·1 (30·4)	4·29 (40·2)	13·8 (44·4)	16·8 (35·9)	29·7 (24·6)	5·95 (31·7)	31·4 (17·2)	23·1 (26·7)	23·0 (41·4)	18·4 (38·4)	23·8 (33·8)
16	18:2n6 LA	1·78 (118)	0·00 (—)	0·84 (93·3)	1·66 (48·0)	1·05 (13·9)	1·39 (17·3)	1·44 (16·0)	0·84 (53·7)	1·29 (46·5)	1·25 (30·7)	1·46 (43·2)
18	18:3n6	0·18 (101)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·02 (300)	0·00 (—)	0·00 (—)
27	20:2n6	0·50 (20·7)	0·17 (130)	0·33 (72·0)	0·20 (67·8)	0·25 (11·7)	0·30 (86·5)	0·00 (—)	0·00 (—)	2·52 (265)	0·11 (12·8)	0·11 (170)
28	20:3n6	0·29 (59·4)	0·00 (—)	0·00 (—)	0·00 (—)	0·06 (127)	0·05 (200)	0·00 (—)	0·00 (—)	0·02 (300)	0·31 (266)	0·00 (—)
29	20:4n6 AA	4·50 (31·6)	1·87 (22·2)	4·61 (30·3)	12·2 (20·0)	1·91 (24·0)	1·02 (23·4)	3·87 (14·8)	3·17 (28·8)	3·78 (20·0)	4·65 (13·6)	2·35 (32·2)
35	22:2n6	0·00 (—)	0·00 (—)	0·04 (300)	0·26 (158)	0·05 (206)	0·07 (184)	0·00 (—)	0·00 (—)	0·00 (—)	0·05 (205)	0·00 (—)
38	22:4n6	1·70 (28·3)	0·80 (56·2)	0·98 (71·7)	2·40 (19·9)	0·50 (17·2)	0·09 (111)	0·42 (41·8)	0·22 (89·1)	0·69 (83·2)	0·40 (58·6)	0·40 (221)
26	20:2n9	0·58 (45·9)	0·33 (107)	0·42 (78·2)	0·08 (181)	0·25 (35·5)	0·02 (300)	0·00 (—)	0·00 (—)	0·00 (—)	0·04 (300)	0·11 (179)
11	U1 (18:0i?)	0·00 (—)	0·11 (201)	0·04 (300)	0·06 (300)	0·11 (83·0)	0·02 (300)	0·00 (—)	0·00 (—)	0·43 (154)	1·84 (61·8)	0·30 (300)
15	U2 (18:1n5?)	0·37 (42·6)	1·01 (45·4)	0·73 (105)	0·48 (120)	0·64 (53·5)	3·10 (42·3)	0·31 (109)	0·82 (65·3)	1·62 (51·8)	1·37 (57·1)	0·55 (145)
17	U3 (18:2n3?)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)	0·00 (—)
20	U4	0·00 (—)	0·00 (—)	0·03 (300)	0·05 (300)	0·31 (85·7)	1·47 (69·9)	0·00 (—)	1·85 (71·2)	0·00 (—)	3·58 (40·6)	0·11 (206)
30	U5 (20:3n3?)	0·00 (—)	0·43 (102)	0·35 (95·8)	1·87 (73·8)	0·07 (126)	0·00 (—)	0·00 (—)	0·12 (300)	0·09 (300)	0·00 (—)	0·03 (300)

Ux = Unknown peak x.

Table 2b. Important fatty acid and lipid composition factors for total lipid extracts (n = 10) (with % variabilities)

Factor	Black bream				Redfish		John dory		Ling		Mirror dory
	New South Wales		Victoria	Queensland	New South Wales		New South Wales		New South Wales		New South Wales
	Spring	Autumn	Spring	Spring	Spring	Autumn	Spring	Autumn	Spring	Autumn	Spring
%S	29·04 (5·68)	47·63 (12·0)	34·31 (31·8)	29·05 (17·4)	28·23 (2·62)	37·37 (17·5)	33·91 (8·08)	38·65 (23·3)	33·53 (25·3)	35·78 (15·2)	37·38 (18·2)
%M	26·21 (21·8)	35·31 (6·68)	30·57 (18·6)	19·54 (23·1)	30·0 (22·5)	44·36 (10·4)	17·00 (14·6)	22·11 (19·9)	24·34 (12·4)	26·01 (15·6)	24·79 (27·3)
%P	44·38 (14·7)	15·51 (28·4)	33·97 (31·2)	48·95 (16·7)	40·63 (18·5)	13·69 (19·9)	48·78 (4·92)	36·05 (23·0)	38·12 (26·1)	31·43 (27·0)	36·84 (25·9)
%n3	35·33 (21·1)	12·51 (33·6)	27·00 (33·0)	32·03 (24·0)	36·80 (18·9)	11·05 (21·8)	43·06 (11·6)	31·83 (23·0)	34·18 (32·4)	24·74 (35·1)	32·53 (27·7)
%n6	9·04 (25·3)	2·99 (20·2)	6·98 (27·1)	16·91 (14·5)	3·82 (15·5)	2·63 (16·2)	5·72 (12·1)	4·22 (26·8)	5·80 (25·0)	6·69 (23·0)	4·31 (29·8)
P/S	1·54 (18·0)	0·34 (43·2)	1·14 (46·9)	1·77 (31·5)	1·45 (20·5)	0·39 (36·5)	1·46 (16·7)	1·02 (42·2)	1·17 (41·7)	0·93 (38·7)	1·06 (45·6)
n3/n6	4·29 (37·7)	4·30 (31·1)	3·84 (19·4)	1·93 (26·3)	9·58 (6·24)	4·20 (15·0)	7·67 (19·5)	7·67 (13·4)	6·29 (47·7)	3·88 (37·0)	8·04 (29·8)
FA (mg/g)	10·10 (65·7)	12·30 (45·1)	4·54 (47·0)	2·00 (41·6)	6·44 (65·0)	4·99 (63·0)	1·28 (38·0)	2·26 (59·0)	3·12 (17·7)	1·34 (36·3)	5·44 (38·2)
%L	1·36 (38·1)	1·20 (49·8)	0·78 (31·5)	0·48 (18·0)	0·83 (47·2)	0·73 (44·7)	0·28 (17·0)	0·39 (22·9)	0·35* (16·2)	0·61 (56·6)	0·53** (10·5)

* n = 2 only.
** n = 3 only.

Table 3a. Fatty acid compositions of phospholipid extracts (data given as mean (n = 10) percentage of total fatty acid mass (with % variabilities))

Fatty acid	Black bream		Ling	
	Victoria	Queensland	New South Wales	
	Spring	Spring	Spring	Autumn
14:0	1·32 (31·2)	2·00 (28·4)	0·92 (96·5)	1·03 (27·0)
15:0i	0·27 (21·7)	0·28 (43·3)	0·60 (79·4)	0·20 (25·5)
16:0	16·9 (16·2)	12·7 (21·3)	23·3 (28·8)	15·2 (23·8)
17:0i	0·00 (—)	0·00 (—)	0·12 (155)	0·00 (—)
17:0	0·00 (—)	0·14 (156)	0·00 (—)	0·00 (—)
18:0	7·26 (19·9)	8·46 (19·0)	6·14 (9·18)	8·19 (16·4)
20:0	0·00 (—)	0·03 (153)	0·00 (—)	0·06 (75·5)
16:1n7	1·98 (29·7)	1·85 (30·7)	1·22 (55·3)	0·02 (201)
17:1n?	0·32 (29·6)	0·08 (267)	0·98 (35·0)	0·61 (36·7)
17:1n8	0·05 (300)	0·00 (—)	0·39 (84·3)	0·06 (206)
18:1n9	8·21 (16·6)	8·56 (9·51)	19·0 (13·2)	14·7 (13·5)
20:1n7	0·00 (—)	0·11 (126)	1·53 (30·0)	1·50 (25·0)
20:1n5	0·00 (—)	0·04 (300)	0·02 (300)	0·00 (—)
22:1n11	0·01 (300)	0·15 (162)	0·04 (300)	0·14 (90·5)
22:1n9	0·02 (300)	0·05 (201)	0·00 (—)	0·02 (200)
18:3n3 LN	0·00 (—)	0·07 (168)	0·00 (—)	0·00 (—)
18:4n3	0·00 (—)	0·01 (300)	0·00 (—)	0·14 (42·9)
20:4n3	0·31 (32·7)	0·36 (33·1)	0·16 (109)	0·19 (41·3)
20:5n3 EPA	8·80 (14·6)	6·32 (22·6)	5·29 (23·1)	5·46 (19·3)
22:2n3	0·28 (55·7)	0·12 (104)	0·02 (300)	0·05 (170)
22:3n3	0·34 (105)	0·64 (57·9)	0·24 (300)	0·11 (91·2)
21:5n3	2·13 (25·2)	2·81 (25·5)	1·37 (38·3)	1·48 (20·3)
22:5n3 DPA	7·07 (8·86)	8·07 (23·4)	1·98 (23·3)	2·04 (14·8)
22:6n3 DHA	31·1 (15·3)	28·0 (20·0)	30·3 (34·1)	41·0 (10·1)
18:2n6 LA	0·69 (18·3)	0·97 (21·3)	0·58 (37·2)	0·45 (37·3)
18:3n6	0·00 (—)	0·01 (300)	0·00 (—)	0·01 (71·5)
20:2n6	0·16 (43·4)	0·22 (54·5)	0·06 (205)	0·14 (61·0)
20:3n6	0·14 (43·1)	0·40 (28·0)	0·02 (300)	0·07 (67·8)
20:4n6 AA	9·83 (13·6)	14·9 (16·3)	5·02 (19·8)	5·76 (7·73)
22:4n6	2·19 (22·5)	3·53 (17·7)	0·61 (40·6)	0·71 (37·2)
20:2n9	0·13 (68·6)	0·03 (229)	0·00 (—)	0·02 (300)
U1 (18:0i?)	0·00 (—)	0·00 (—)	0·13 (125)	0·22 (54·3)
U2 (18:1n5?)	0·00 (—)	0·00 (—)	0·00 (—)	0·05 (300)
U3 (18:2n3?)	0·13 (223)	0·00 (—)	0·00 (—)	0·23 (58·1)
U5 (20:3n3?)	0·03 (207)	0·16 (57·6)	0·00 (—)	0·00 (—)

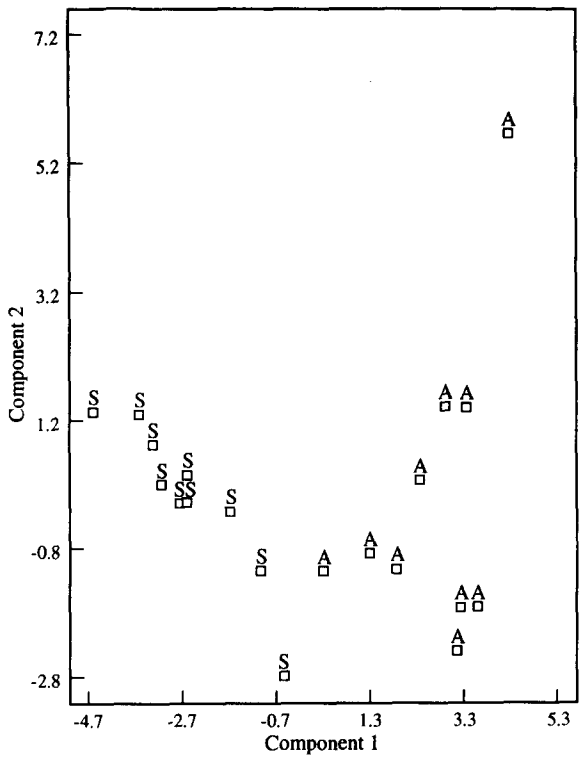
ceride contents in the fish with greater tissue lipid contents.

Variability between seasons

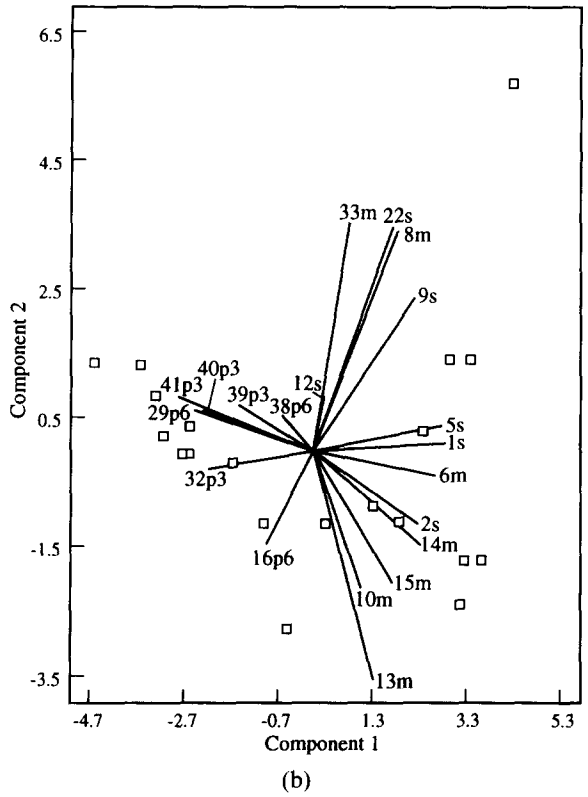
The fatty acid profiles of samples from the same species caught during different seasons were subjected to principal components analyses. Distinct seasonal clusters were obtained in the scatterplots of the first two principal components for the data of Black bream and Redfish (Figs 1(a) and 2(a)). When these scatterplots were overlayed with vectors representing the correlation of the variables with the components (forming the biplots shown in Figs 1(b) and 2(b)) it can be seen that both

Table 3b. Important fatty acid and lipid composition factors for phospholipid extracts (n = 10) (with % variabilities)

Factor	Black bream		Ling	
	Victoria	Queensland	New South Wales	
	Spring	Spring	Spring	Autumn
%S	25·76 (7·86)	22·50 (6·32)	31·09 (26·2)	24·67 (11·0)
%M	10·60 (18·5)	10·83 (12·3)	23·14 (13·7)	17·09 (10·3)
%P	63·17 (5·68)	66·49 (3·46)	45·65 (24·2)	57·73 (6·83)
%n3	50·19 (8·01)	46·62 (8·04)	39·41 (28·3)	50·61 (7·26)
%n6	12·98 (13·7)	19·86 (14·4)	6·23 (20·5)	7·12 (11·1)
P/S	2·48 (12·9)	2·97 (9·28)	1·64 (41·7)	2·38 (16·3)
n3/n6	3·95 (17·1)	2·41 (19·5)	6·66 (39·1)	7·17 (11·2)

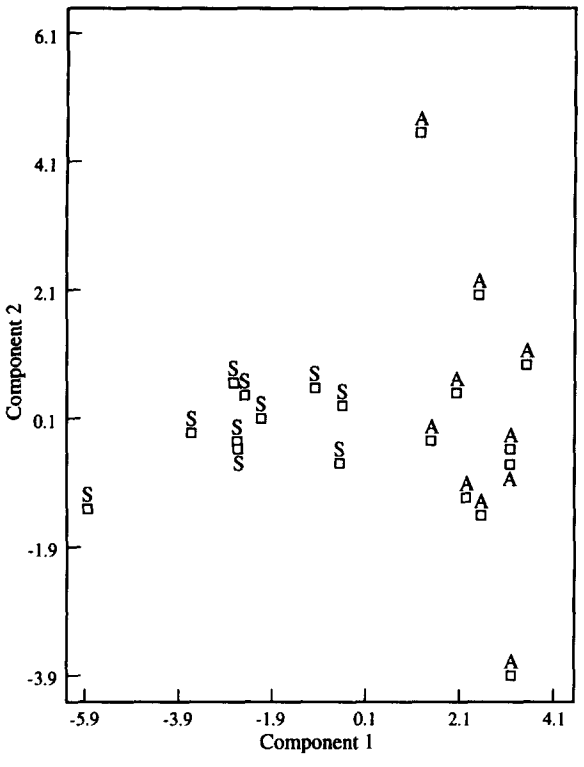


(a)

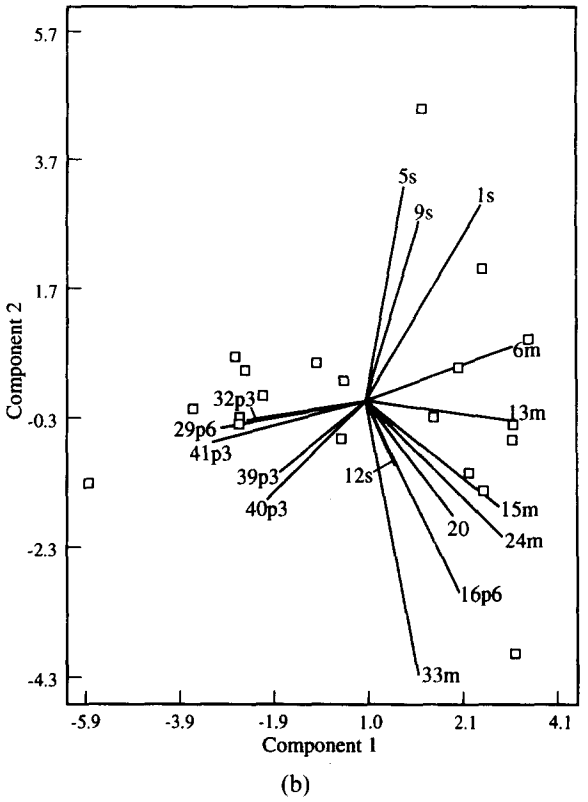


(b)

Fig. 1. PCA of total lipid extract fatty acids of Black bream sampled during autumn (a) and spring (S). Scatterplot (59·5% of total variance). (b) Biplot (numbers refer to elution order (Table 2): m = monounsaturated, p6 = n6 polyunsaturated, p3 = n3 polyunsaturated, s = saturated).



(a)



(b)

Fig. 2. PCA of total lipid extract fatty acids of Red fish sampled during autumn (A) and spring (S). (a) Scatterplot (64·6% of total variance). (b) Biplot (numbers refer to elution order Table 2): m = monounsaturated, p6 = n6 polyunsaturated, p3 = n3 polyunsaturated, s = saturated).

data sets show correlation of the spring fish with the major PUFAs, and the autumn samples with the monounsaturates and saturates.

The effect of varying the season of catch on factors believed to be of nutritional importance was investigated by univariate analysis (Table 4). The greatest number of inter-seasonal differences occurred in the Black bream and Redfish samples. Generally, the univariate analyses echoed the PCA conclusions that a shift toward greater degrees of saturation in the fatty acids had occurred during autumn.

The major factor affecting the fatty acid compositions of fish is their relative proportion in the diet (Yu & Sinnhuber, 1972; Stansby, 1981). This overrides fatty acid conversion within the fish. Although the species studied here were all carnivorous, the original source of their tissue fatty acids was marine algae and plankton. Seasonal variations are known to occur in the fatty acid composition of plankton (Lewis, 1962), which would cause a similar effect in the fish lying further along the food chain. The original cause of these changes is believed to be the variations in water temperature, with an increase in fatty acid saturation generally occurring during times of warmer water temperatures. The fish analysed in this study were sampled during spring (which follows winter, that has relatively cold water) and autumn (after exposure to warm summer waters). The observed shift towards a greater degree of saturation in autumn is consistent with the expected dietary changes.

PCA scatterplots did not form seasonal clusters for the fatty acid data of John dory or Ling (plots not shown), and fewer and less significant changes were determined by univariate analyses. This was likely to have been the result of their muscle lipids consisting almost entirely of phospholipid, which tends to maintain a relatively stable fatty acid composition even when the diet is altered.

In summary, the season of catch of fish for human consumption is really only a relevant factor when the species has some ability to store triglyceride within the muscle (as is indicated by tissue lipid contents of above approximately 0·7%).

Fattier individuals of such species will have lower P/S and n3/n6 ratios when caught after summer than their leaner counterparts.

Variability between locations

Samples of Black bream caught over a 13° latitude range (representing a 13°C range in water temperature) were analysed.

Principal components analysis performed on the total fatty acid profiles revealed some clustering between samples taken from Queensland and those from Victorian waters in the scatterplot of the first two principal components (Fig. 3). Both of these, however, overlapped with the Sydney sample, which was indicative of its intermediate latitude and milder water temperatures.

Table 4. Parametric and non-parametric statistical comparisons between spring (S, n = 10) and autumn (A, n = 10) samples

Quantity	Total fatty acids							
	Bream		Redfish		John dory		Ling	
	Mann–Whitney	ANOVA	Mann–Whitney	ANOVA	Mann-Whitney	ANOVA	Mann–Whitney	ANOVA
%S	S<A (p<0·001)	S<A (p=0·000)	S<A (p=0·002)	S<A (p=0·0006)	Accept (0·1<p<0·2)	Accept (p=0·148 8)	Accept (p>0·2)	Accept (p=0·518 2)
%M	S<A (0·002<p<0·005)	S<A (p=0·000 3)	S<A (p<0·001)	S<A (p=0·000 1)	S<A (0·005<p<0·01)	S<A (p=0·007 1)	Accept (p>0·02)	Accept (p=0·343 8)
%P	S>A (p<0·001)	S>A (p=0·000)	S>A (p<0·001)	S>A (p=0·000)	S>A (0·001<p<0·002)	S>A (p=0·000 9)	Accept (0·05<p<0·01)	Accept (p=0·078 7)
P/S	S>A (p<0·001)	S>A (p=0·000 0)	S>A (p<0·001)	S>A (p=0·000 0)	S>A (0·005<p<0·01)	S>A (p=0·017 2)	Accept (0·1<p<0·2)	Accept (p=0·127 0)
%n3	S>A (p<0·001)	S>A (p=0·000 0)	S>A (p<0·001)	S>A (p=0·000 0)	S>A (0·001<p<0·002)	S>A (p=0·001 3)	Accept (p=0·1)	Accept (p=0·059 0)
%n6	S>A (p<0·001)	S>A (p=0·000 0)	S>A (p<0·001)	S>A (p=0·000 1)	S>A (0·005<p<0·01)	S>A (p=0·003 2)	Accept (p>0·2)	Accept (p=0·223 0)
N3/n6	Accept (p>0·2)	Accept (p=0·983 0)	S>A (p<0·001)	S>A (p=0·000 0)	Accept (p>0·2)	Accept (p=0·998 1)	S>A (0·05<p<0·1)	S>A (p=0·032 7)
%AA	S>A (p<0·001)	S>A (p=0·000 0)	S>A (p<0·001)	S>A (p=0·000 1)	S>A (0·02<p<0·05)	Accept (p=0·067 7)	S<A (p=0·02)	S<A (p=0·016 9
%(DHA+EPA)	S>A (p<0·001)	S>A (p=0·000 0)	S>A (p<0·001)	S>A (p=0·000 0)	S>A (p=0·01)	S>A (p=0·006 9)	Accept (p>0·2)	Accept (p=0·219 5)
%L	Accept (p>0·2)	Accept (p=0·435 4)	Accept (p>0·2)	Accept (p=0·402 6)	S<A (p=0·02)	Accept (p=0·042 4)	—[a] —	— —

[a] Insufficient data available for Ling lipid comparisons.

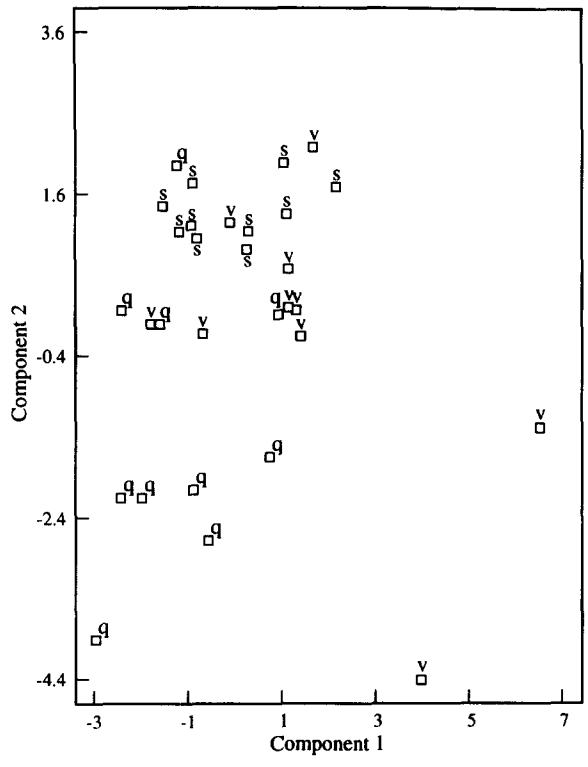


Fig. 3. PCA scatterplot of total lipid extract fatty acids of Black bream sampled from Queensland (q), Victoria (v) and Sydney (s) (44·3% of total variance).

Nutritional factors for the total lipid fatty acids extracted from these samples were compared (Table 5). As with the scatterplot, the greatest differences were found between the Victorian and Queensland samples. Lower n6 levels (mainly arachidonic acid (AA)) were found in the fish from colder (more southern) waters, as was also found by Naughton *et al.* (1983) and Sinclair *et al.* (1984). This resulted in greater n3/n6 ratios for samples from the two more southern locations. The Queensland fish were significantly higher in monounsaturates than the Victorian, while saturates were present at similar levels in all samples.

Table 5. Parametric and non-parametric statistical comparisons between Black bream sampled at Queensland (Q), Victoria (V) and Sydney (S) (n = 10 for each)

Quantity	Kruskal–Wallis	ANOVA
%S	Accept (p=0·613 2)	Accept (p=0·203 6)
%M	Q<V (p=0·001 9)	Q<V (p=0·000 6)
%P	Q>V (p=0·009 3)	Q>V (p=0·003 2)
P/S	Accept (p=0·061 3)	Q>V (p=0·027 7)
%n3	Accept (p=0·092 0)	Accept (p=0·105 5)
%n6	Q>S,V (p=0·000 0)	Q>S,V (p=0·000 0)
n3/n6	S<S,V (p=0·000 6)	Q<S,V (p=0·000 2)
%AA	Q>S,V (p=0·000 1)	Q>S,V (p=0·000 0)
%(DHA+EPA)	Accept (p=0·187 8)	Accept (p=0·178 5)

Principal components analyses of phospholipid fatty acid profiles failed to produce clusters for the sampling locations, indicating stable phospholipid compositions within this species over the 13° latitude of sampling.

These findings are contrary to those reported previously (Ackman, 1967; Sinclair *et al.* 1983), where increased PUFA contents and P/S ratios were noted for fish from colder waters. This is probably due to the fact that the water temperatures of the southernmost sampling (Victoria) were still mild (12–14°C). Reiser *et al.* (1963) subjected fish to water temperatures within a similar range to those encountered in this study (13–23°C) and concluded that there was no significant effect on the deposition or interconversion of PUFAs in the fish unless food deprivation was enforced. The higher PUFA contents and *P/S* ratios for the fish from Queensland waters are a result of the high n6, and in particular AA (2:4n6), contents compared with the samples from the two southern locations. This can be accounted for by considering that the food chain in this region is known to contain unusually high levels of AA (Johns *et al.*, 1979).

Based on the data presented in this report the consumption of Australian fish from more southern waters would be more highly recommended for human consumption (based on current nutritional knowledge).

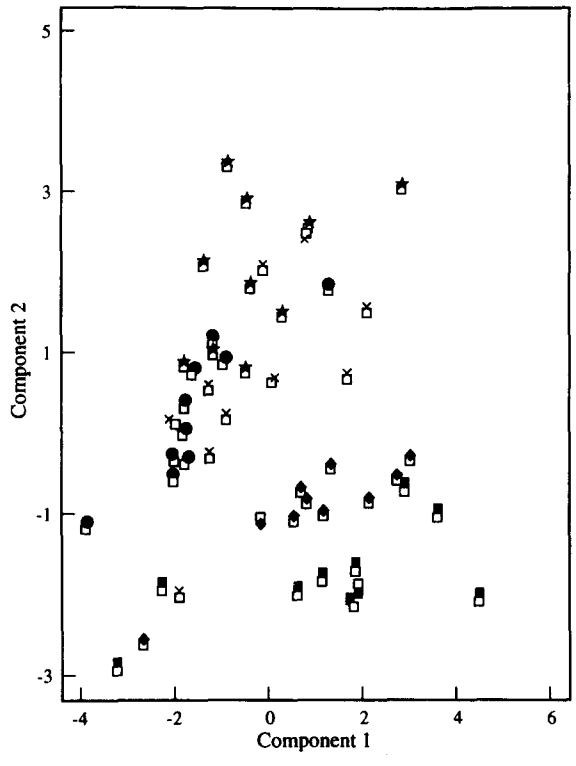


Fig. 4. PCA scatterplot of total lipid extract fatty acids of species sampled from Sydney during spring 1989 (43·9% of total variance). (★) Black bream, (♦) Redfish, (X) Ling, (●) John dory, (■) Mirror dory.

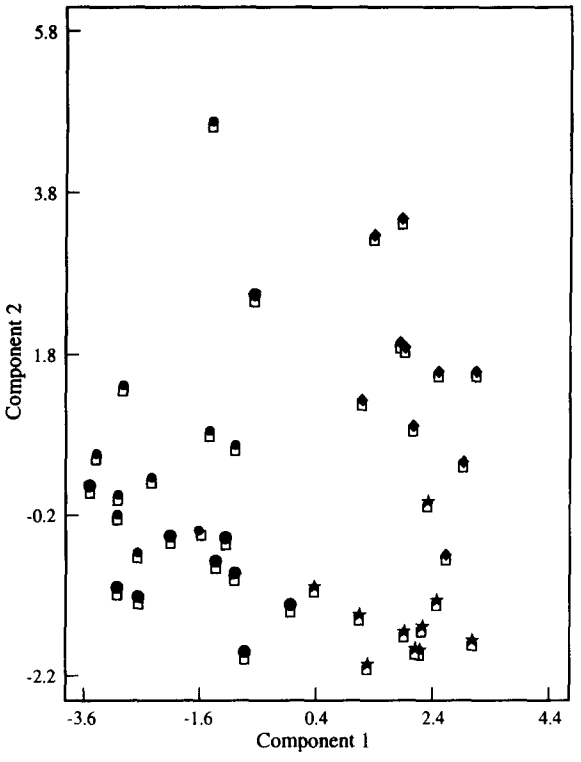


Fig. 5. PCA scatterplot of total lipid extract fatty acids of species samples from Sydney during autumn 1990) (53·8% of total variance). (★) Black bream, (♦) Redfish, (•) Ling, (●) John dory, (■).

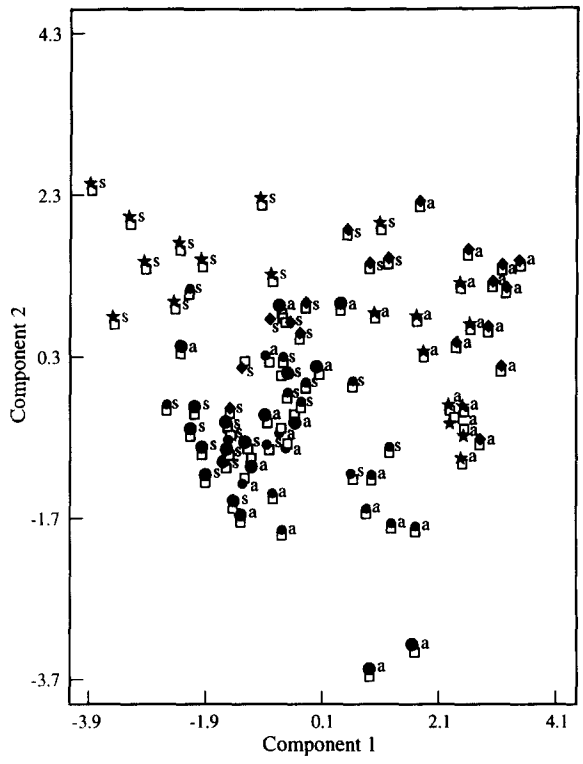


Fig. 6. PCA scatterplot of total lipid extract fatty acids of species samples during autumn (a) and spring (s) from Sydney (61·8% of total variance). (★) Black bream, (♦) Redfish, (•) Ling, (●) John dory, (■).

These fish had higher n3/n6 ratios than their more northern dwelling counterparts, and while *P/S* ratios were lower they were still generally acceptable.

Variability between species

The scatterplots yielded by PCA comparisons between fatty acid compositions of species sampled at similar locations during the same season are shown in Figs 4 and 5.

The spring fatty acid data (Fig. 4) produced much overlap, particularly between the extremely lean species (John dory and Ling). This indicates that the phospholipid compositions of the two species were similar, despite differences in dietary composition.

The autumn samples had more pronounced species-characteristic fatty acid profiles, as evidenced by the more distinct species clusters in the PCA scatterplot (Fig. 5). This sample was taken following summer, when food is generally more plentiful, which would enable a faster turnover in fatty acids bound in phospholipids, as well as the likelihood of greater triglyceride storage (as was evident by the greater lipid contents of the John dory and Ling during this season).

When species and seasonal fatty acid data were combined and analysed by PCA, pronounced differentiation between samples from the two seasons occurred (Fig. 6). For these data the season of catch, therefore, was more nutritionally relevant than the species, especially when selecting between moderately lean species.

CONCLUSIONS

Of the Australian fish sampled in this study those caught from more southern waters and during colder months had higher n3 PUFA contents in their lipids. They also had greater n3/n6 ratios, while maintaining similar *P/S* values, as compared with their more northern counterparts. Based on current belief that the nutritional benefits of fish consumption arise largely because of their high n3 PUFA contents it would appear that the colder water temperatures produced fatty acid compositions more favourable for human consumption.

When including lean fish (between approximately 0·7 and 2·0% lipid) in the diet, some consideration as to the species, location and season of catch may be justified. Certainly, when reporting fish lipid content and compositions, authors should state precise details of catch time and location, or prevailing water temperatures. Extremely lean species, however, have relatively constant lipid fatty acid compositions, and so may be consumed without regard to sampling details. It is for such extremely lean species, therefore, that fatty acid composition data can be most easily collected, and for which composition tables are most valid.

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