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## FATTY ACID PATTERN DIFFERENCES AMONG INDIVIDUALS OF TWO ESTUARINE FISHES (*LEIOSTOMUS XANTHURUS* AND *MUGIL CEPHALUS*)

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**ABSTRACT** Ten individual fish of two estuarine species, spot (*Leiostomus xanthurus*) and striped mullet (*Mugil cephalus*), were analyzed for fatty acids. Fish of similar size were obtained from a single collection to minimize variability due to age, size, location and season. Analysis of variance (ANOVA) of each fatty acid provided statistically similar groups for each acid that existed among individual fish. Fatty acids in the striped mullet provided a greater number of statistically similar groups than those in spot, indicating greater variability among individual striped mullet, which probably reflected a greater diversity in the feeding regime for this species. ANOVA results within classes of fatty acids of both species indicated greater diversity in monounsaturated and polyunsaturated than saturated fatty acids. Eicosapentaenoic acid (EPA) showed more individual variability in both species than did docosahexaenoic acid (DHA). Dietary lipids and metabolic needs of the two species are distinct and may be the key factors in explaining individual differences observed in these two fish species.

### INTRODUCTION

Natural populations of fishes contain fatty acids and other nutritional components that are highly variable. Stansby (1981) has addressed some sources of variability in fatty acid composition of fish oils within a given species. Other studies have focused on individual species and variations with respect to age (Hayashi and Takagi, 1978), season (Ueda, 1976; Hayashi and Takagi, 1977, 1978; Gallagher *et al.*, 1989), size (Gallagher *et al.*, 1984) and geographical location (Addison *et al.*, 1973; Whyte and Boutillier, 1991), but have not addressed variations among individuals of these species. Assessing importance of these variables is dependent upon appraisal of individual variability, since inherent biochemical differences exist from fish to fish even when all other variables are minimized. It was essential that specimens be carefully selected of two species of coastal Gulf finfishes which differed little in size, development stage, weight or location of catch. This selection permitted specific examination of those fluctuations in individual fatty acids that may occur due only to individual differences. By using a non-random selection process for samples, results could not be used to characterize overall trends in the two species. However, it was felt that results would define some individual variations that are uniquely characteristic for these two fishes that would permit informative and useful comparisons to be made and that suggestions for these variability differences would be suitable.

Two species of coastal Gulf finfishes, spot and striped mullet, were chosen for assessing individual variability because they met several criteria. They represented fishes with different feeding regimes, they were collected easily

in large numbers from a given area in one catch, and they were numerous enough to permit selection of fish having little variation in size. Additionally, the biology of spot (Gunter, 1945; Dawson, 1958; Hodson *et al.*, 1981; Chestnut, 1983; Sheridan *et al.*, 1984) and striped mullet (Odom, 1966; Thompson, 1966) is well established, and both species are found abundantly in local coastal estuaries.

Spot is a dominant bottom fish and is considered to feed in schools over sand-mud bottoms on polychaetes, harpacticoid copepods, bivalves and possibly some detritus (Hildebrand and Schroeder, 1928; Darnell, 1958; Hodson *et al.*, 1981). Spot has a fairly small mouth and possesses gill rakers that permit retention of small food particles and prevent ingestion of relatively large food items such as fish, shrimp and crabs (Darnell, 1958; Hodson *et al.*, 1981; Chestnut, 1983; Sheridan *et al.*, 1984).

On the other hand, the striped mullet begins its life by eating planktonic plants and animals, but it changes its diet to include a broad range of detritus and plant material as it develops (Moore, 1974). It filter-feeds above organic muds containing microplant material and macroplant detritus (Odum, 1966, 1970), and it is generally considered to be a broad spectrum herbivore. Occasionally, however, carnivorous feeding has been observed in striped mullet (Bishop and Miglarese, 1978).

Fatty acids in marine dietary lipids, whether plant or animal, serve as an energy source for metabolism and provide polyunsaturated fatty acids (PUFA) essential for membrane structure and function. Lipids in muscle tissue of fish generally reflect those fatty acids obtained from the diet. Since these two species have a widely different natural diet, they provide an opportunity to examine individual variability within and between species.

## MATERIALS AND METHODS

### Collection

Striped mullet were collected on January 21, 1988 in the shallow estuary of Biloxi Bay, Mississippi. Spot were collected on April 13, 1988 at Ship Island, a barrier island 15 miles south of Gulfport, Mississippi, in the northeastern Gulf of Mexico. All fish were collected by gill net and maintained on ice until examined. Standard lengths were measured and weights recorded. Fish of approximately the same size were filleted and individual fish placed separately in polyethylene bags, flushed with N<sub>2</sub>, rapidly frozen and stored at -20°C. Average body weight of striped mullet was 230 g ( $\pm 12\%$  relative standard deviation: RSD) and average standard length was 221 mm ( $\pm 4.5\%$ RSD). Average body weight of spot was 147 g ( $\pm 5.2\%$ RSD), and average standard length was 174 mm ( $\pm 3.5\%$ RSD).

### Analytical Procedure

All solvents used in analysis were HPLC grade or analytical reagent grade. Standards were purchased from NuCheck Prep, Inc. (Elysian, MN). Fillets were homogenized using a Waring blender and 0.5 g aliquots weighed into screw-capped (Teflon-lined) centrifuge tubes (30 ml) and saponified at ambient temperature with ethanolic KOH under N<sub>2</sub> using a magnetic stirrer for one hour. Care was exercised in the volumes of saponifying mixtures used to keep the water level, derived from tissue, sufficiently high to prevent trans-esterification. Solvent ratios were those suggested by Nelson (1966). After dilution with distilled water, the neutral fraction was extracted with hexane. The remaining alkaline solution was acidified with 6N HCl, and free fatty acids were extracted with benzene. Benzene aliquots were combined and concentrated using a rotary evaporator. All evaporations were closely monitored to ensure that distillation temperatures did not exceed 25°C. Fatty acids were converted to methyl esters using 7% BF<sub>3</sub>-MeOH by the method of Metcalfe *et al.* (1966) modified to use ambient temperatures and a one-hour reaction period.

Identification of fatty acid methyl esters (FAME) was obtained by capillary gas chromatography (GC) using a Perkin-Elmer model Sigma 2000 gas chromatograph equipped with flame ionization detector and fitted with a 30 m x 0.25 mm i.d. fused silica capillary column coated with a 0.25  $\mu$ m film thickness of Dura Bond WAX (J & W Scientific) and operated with a split ratio of 100:1. The carrier gas, He, was maintained at 20 psi. Oven temperature was programmed at 90-250°C at a linear rate of 4°/minute. Data was processed using a Perkin-Elmer Sigma 10 data system with quantification of all compounds based on individual peak area response by GC compared to the

internal standard methyl tricosanoate. Quantitative data were corrected for differences in detector responses that were determined through analysis of authentic standards of each reported fatty acid. FAME were tentatively identified by comparison with retention times with those of authentic standards. Verification of identification on select samples was accomplished through gas chromatography mass spectrometry analysis conducted by National Marine Fisheries Service, Charleston Laboratory. Concentrations of individual isomers of PUFA were separately tabulated; separate isomers of monounsaturates (e.g. 18:1) were not reported.

### Sample Protection

Several precautions were taken to ensure that no degradation or other alteration of lipids occurred during extraction and saponification. All analytical steps were conducted at ambient temperatures, and samples were constantly flushed with N<sub>2</sub> to prevent oxidation. Further, as many steps as possible were conducted in a single extraction tube to reduce loss and degradation that occurs with sample transfer. All solvents were flushed with N<sub>2</sub> immediately before use to remove dissolved O<sub>2</sub> and to prevent oxidative degradation. Likewise, samples requiring storage were placed in sample bags which were flushed with N<sub>2</sub> before being frozen (-20°C). In addition, the antioxidant butylated hydroxytoluene (BHT) was added in a concentration of 0.005% (w/v) to extraction solvents to prevent oxidative degradation of unsaturated lipids.

### Data Analysis

One way analysis of variance (ANOVA) with *post facto* 95% confidence level range test (Statistical Graphics Corporation, 1988) was used to compare individual fatty acids as well as certain parameters derived from fatty acid data of individual fish. Similarity groups were established of individuals for each variable which were statistically indistinguishable ( $p < 0.05$ ). In addition, the number of groups was tallied as a further measure of individual variability.

## RESULTS

Figures 1 and 2 depict mean concentration of fatty acids in the samples of individual spot and striped mullet as well as mean % composition of total saturated, monounsaturated and PUFA. Figures 1 and 2 also include the standard deviations of the means of the ten individual fish and are shown by the dark bars in the graphs. Concentrations are shown in both wt% of the total fatty acids and

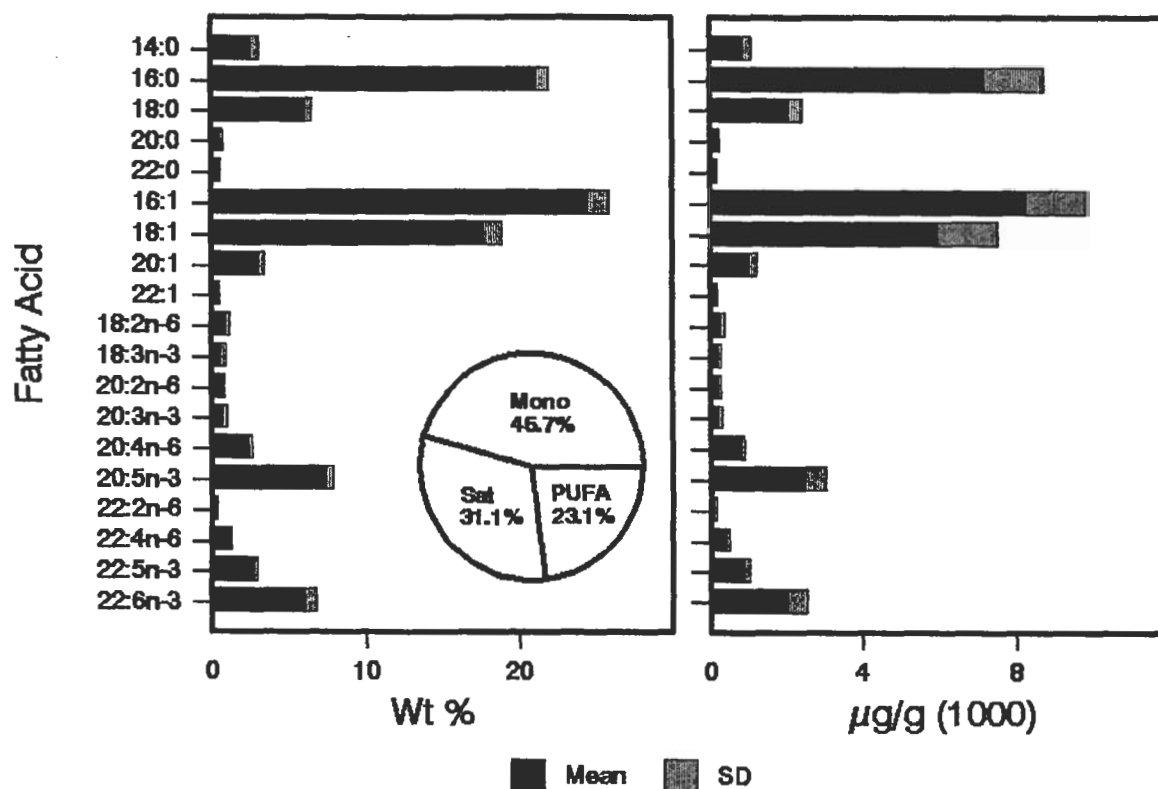


Figure 1. *Leiostomus xanthurus*. Distribution of fatty acids in spot. Empty bars to the left represent mean concentrations in wt % of total reported fatty acids of 10 individual fish. Bars to the right depict mean concentrations in µg/g (wet tissue). Gray bars are standard deviations computed on the mean of the 10 mean values for individual fish.

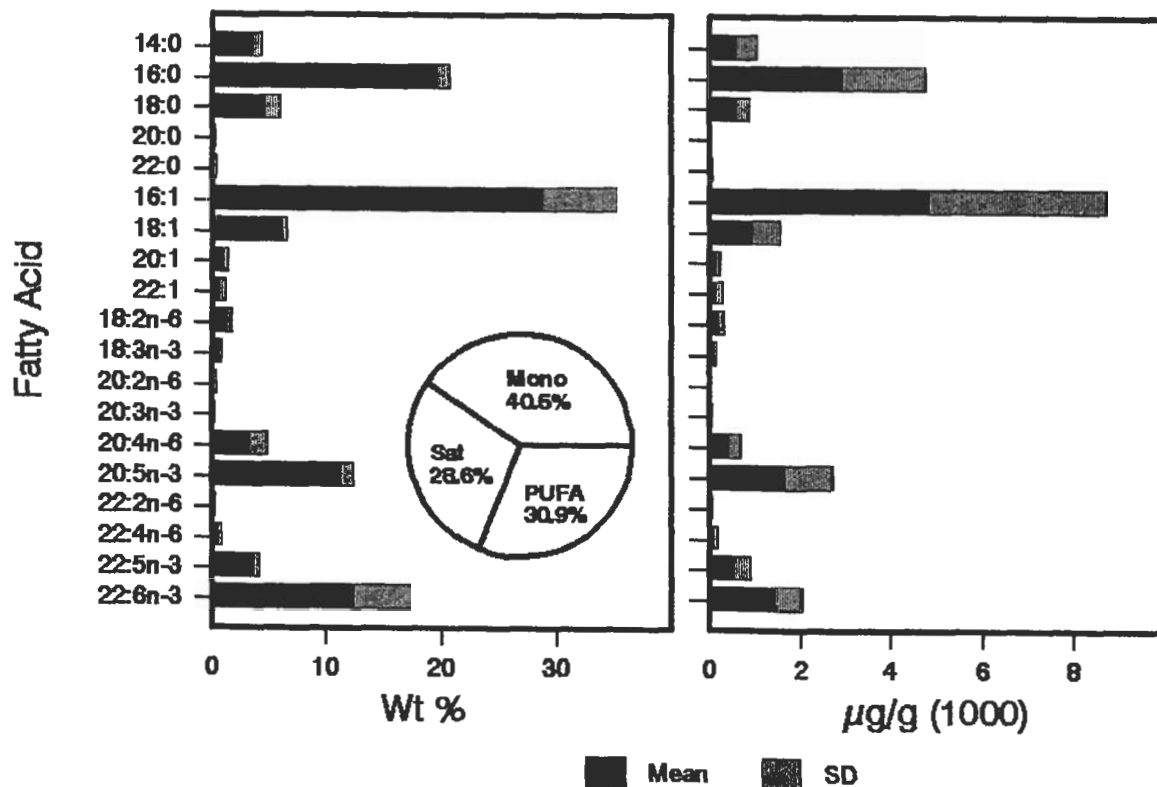


Figure 2. *Mugil cephalus*. Distribution of fatty acids in striped mullet. See caption for Figure 1.

in absolute concentrations of  $\mu\text{g/g}$  of wet tissue. Absolute concentrations are useful when assessing muscle tissue for nutritive value, particularly for omega-3 (n-3) content, since there is an increased interest in possible health benefits (Lands, 1986), while weight percent concentrations are useful in assessing biochemical significance of fatty acid distributions.

Tables 1 and 2 contain fatty acid data computed on a wt % basis for fatty acids in muscle tissue from spot and striped mullet, respectively. Also included are summations and ratios that are helpful in characterizing fatty acid profiles in finfish. Superscripts signify the statistically similar group(s) that each individual fish falls within for ANOVA treatment of each fatty acid or fatty acid parameter. At the end of each row is the number of groups produced by ANOVA examination of that fatty acid.

Individuals of both species varied in fat content. Striped mullet ranged from 1.82-6.38%, while spot ranged from 4.75-8.10%.

#### Fatty Acid Distribution in Spot and Striped Mullet

Fatty acid profiles (Figures 1 and 2) were similar from both species, particularly in content of saturated fatty acids. Hexadecanoic acid (16:0) was dominant, followed in decreasing order by octadecanoic acid (18:0) and tetradecanoic acid (14:0). The remaining saturated acids constituted less than one percent of the total fatty acids. The predominant monounsaturated acid in both fish was 16:1. Relative to 16:1, the contents of 18:1 and 20:1 acids were higher in spot than in striped mullet, whether expressed in wt% or  $\mu\text{g/g}$ . The two principal PUFA in both fishes were eicosapentaenoic acid (EPA, 20:5n-3) and docosahexaenoic acid (DHA, 22:6n-3). These n-3 PUFA constitute a higher percentage of the total fatty acids of striped mullet (23.7%) than the spot (13.5%), although in absolute concentration, these PUFA are enriched in spot (4,530  $\mu\text{g/g}$ ) relative to striped mullet (3,120  $\mu\text{g/g}$ ). A narrow range (2.25 to 2.86%) as well as low concentration of arachidonic acid (AA, 20:4n-6) was found in spot, whereas a wider range and higher concentrations (1.70-7.00%) were found in striped mullet.

#### Statistical Comparisons of Component Fatty Acids

Octadecanoic acid, 18:0, was the second most dominant saturated fatty acid in both spot and striped mullet. In spot, no significant difference in values was found among any of the individual fish (i.e. only one similarity group shown in Table 1). On the other hand, there were seven statistically similar groups for 18:0 in striped mullet (Table 2). Minor saturated components, 20:0 and 22:0 in striped mullet (22:0 in spot), showed no significant differ-

ences among any of the ten individual fish. Except for 22:1 in spot, each monounsaturate in both spot and striped mullet showed high diversity among individual fish (four to six similarity groups). Among the PUFA, there were more ANOVA similarity groups for EPA in both spot and striped mullet than for DHA, indicating a greater diversity of EPA than DHA in muscle tissue. In striped mullet, ANOVA treatment of arachidonic (20:4n-6), linolenic (18:3n-3) and octadecanoic acid (18:0) each produced seven similarity groups, the most diverse fatty acids in either fish.

#### Fatty Acid Classes

Figures 1 and 2 indicate that both fishes showed a prevalence of monounsaturates, with spot having 46% monounsaturated, 23% PUFA and 31% saturated, as compared to 40%, 31%, and 29%, respectively, for these fatty acid classes in striped mullet. The saturates for both fishes were less diverse than for either the monounsaturates or PUFA. Likewise, the average value of ANOVA similarity groups for individual saturated fatty acids was less than that found for either of the other fatty acid classes in both spot and striped mullet.

Fatty acids occurring in concentrations above 1% of total fatty acids showed a higher degree of individual variability than fatty acids occurring in less than 1% for both spot and striped mullet. The average number of similarity groups for all fatty acids whose concentrations are above 1% was 3.8 for spot and 5.0 for striped mullet, with 2.5 and 4.3 groups for fatty acids comprising less than 1%.

#### Fatty Acid Parameters

Total n-3/n-6 ratio showed little variation among individual spot with ANOVA, separating into only two statistically similar groups. Excluding individual spot No. 1, no distinction occurred among individuals (Table 1). Conversely, the separate sums of n-3 and n-6 fatty acids in spot were separated into five and three similarity groups, respectively. The n-3/n-6 ratio also varied less among individual striped mullet (four groups) than the separate total n-3 and total n-6 fatty acid parameters (five and seven groups, respectively). ANOVA treatment applied to total PUFA in both spot and striped mullet produced five similarity groups. In spot, the unsaturated/saturated and (EPA+DHA)/n-3 parameters produced two and four ANOVA groups, but in striped mullet, it was four and six groups. ANOVA treatment separated the calculated parameters of striped mullet into a larger number of groups than those of spot, demonstrating the higher degree of individual diversity for component fatty acids in the striped mullet.

TABLE 1

*Leiostomus xanthurus*. Fatty acids in spot. Entries are means for three replicate analyses of homogenized muscle tissue from each of 10 individual fish. Values in parentheses are % relative standard deviations. Entries in rows sharing the same superscript letter are not statistically different (p<0.05) and are referred to as similarity groups; group numbers refer to numbers of similarity groups computed for each fatty acid.

	No. 1	No. 2	No. 3	No. 4	No. 5	No. 6	No. 7	No. 8	No. 9	No. 10	Groups
	Concentration in wt %										
14:0	1.84 (2.48) <sup>a</sup>	2.21 (2.19) <sup>b</sup>	2.82 (1.29) <sup>ad</sup>	2.24 (1.89) <sup>b</sup>	2.78 (1.79) <sup>od</sup>	3.00 (2.81) <sup>d</sup>	3.56 (4.63) <sup>a</sup>	2.70 (3.69) <sup>o</sup>	2.81 (0.527) <sup>ad</sup>	2.16 (3.83) <sup>b</sup>	5
18:0	21.4 (0.754) <sup>ab,o</sup>	22.2 (3.53) <sup>o</sup>	20.5 (0.868) <sup>ab</sup>	21.3 (1.39) <sup>ba</sup>	21.5 (1.36) <sup>bo</sup>	20.0 (2.47) <sup>a</sup>	20.3 (2.69) <sup>a</sup>	20.6 (2.94) <sup>ab</sup>	21.4 (1.71) <sup>ab,o</sup>	22.6 (1.60) <sup>o</sup>	3
18:0	6.60 (1.78)	6.31(5.30)	6.07 (9.75)	6.56 (6.62)	5.53 (1.28)	6.76(8.74)	5.61 (12.3)	6.53 (5.78)	6.04 (6.06)	5.88 (1.02)	1
20:0	0.745 (2.34) <sup>ab,o</sup>	0.453 (41.3) <sup>ab</sup>	0.963 (30.2) <sup>o</sup>	0.812 (0.515) <sup>bo</sup>	0.640 (16.0) <sup>ab,o</sup>	0.836 (30.4) <sup>ab,o</sup>	0.476 (44.4) <sup>ab</sup>	0.578 (31.6) <sup>ab,o</sup>	0.325 (2.32) <sup>a</sup>	0.307 (10.1) <sup>a</sup>	3
22:0	0.503 (24.3)	0.553 (4.28)	0.607 (10.3)	0.629 (5.08)	0.520 (14.9)	0.607 (10.7)	0.606 (12.3)	0.395 (18.1)	0.455 (40.2)	0.514 (4.91)	1
16:1	24.6 (1.99) <sup>bo,d</sup>	24.8 (1.733) <sup>bo,d</sup>	25.7 (0.349) <sup>de,f</sup>	21.8 (0.799) <sup>a</sup>	23.7 (3.42) <sup>bo</sup>	23.5 (1.66) <sup>b</sup>	26.0 (1.48) <sup>el</sup>	26.8 (3.37) <sup>f</sup>	25.0 (1.41) <sup>o,de</sup>	23.3 (1.27) <sup>ab</sup>	6
18:1	18.0 (1.34) <sup>de</sup>	19.3 (0.331) <sup>a</sup>	16.6 (1.41) <sup>ab</sup>	17.0 (6.07) <sup>ab,od</sup>	17.4 (2.09) <sup>bo,d</sup>	17.0 (3.26) <sup>ab,od</sup>	16.9 (1.32) <sup>ab,o</sup>	16.5 (1.01) <sup>a</sup>	17.7 (1.38) <sup>o,d</sup>	20.3 (0.719) <sup>f</sup>	6
20:1	3.44 (2.10) <sup>od</sup>	2.90 (10.1) <sup>ab,o</sup>	3.31 (1.38) <sup>bo,d</sup>	3.78 (3.04) <sup>d</sup>	2.98 (9.40) <sup>ab,o</sup>	3.38 (9.13) <sup>od</sup>	3.12 (9.65) <sup>ab,o</sup>	2.76 (10.7) <sup>ab</sup>	2.60 (1.86) <sup>a</sup>	2.49 (3.09) <sup>a</sup>	4
22:1	0.516 (28.9)	0.405 (28.9)	0.606 (6.63)	0.517 (28.3)	0.535 (12.4)	0.509 (31.5)	0.484 (13.1)	0.368 (10.6)	0.458 (28.1)	0.582 (10.4)	1
18:2n-6	1.40 (3.82) <sup>b</sup>	0.730 (29.8) <sup>a</sup>	1.18 (21.2) <sup>ab</sup>	1.22 (8.29) <sup>ab</sup>	0.889 (14.2) <sup>ab</sup>	0.933 (26.5) <sup>ab</sup>	0.764 (32.7) <sup>a</sup>	1.03 (24.0) <sup>ab</sup>	0.712 (8.08) <sup>a</sup>	0.837 (6.35) <sup>a</sup>	2
18:3n-3	0.845 (2.51) <sup>ab,o</sup>	0.449 (52.1) <sup>ab</sup>	0.765 (7.25) <sup>ab,o</sup>	1.32 (1.98) <sup>o</sup>	0.505 (31.2) <sup>ab</sup>	0.633 (34.8) <sup>ab</sup>	0.406 (59.8) <sup>a</sup>	1.10 (51.8) <sup>bo</sup>	0.300 (0.900) <sup>a</sup>	0.420 (15.1) <sup>a</sup>	3
20:2n-6	0.877 (5.24) <sup>o</sup>	0.685 (20.5) <sup>ab,o</sup>	0.872 (4.55) <sup>bo</sup>	0.963 (1.20) <sup>o</sup>	0.646 (16.7) <sup>ab,o</sup>	0.787 (24.9) <sup>bo</sup>	0.623 (33.7) <sup>ab,o</sup>	0.641 (35.1) <sup>ab,o</sup>	0.384 (3.58) <sup>a</sup>	0.479 (19.3) <sup>ab</sup>	3
20:3n-3	1.07 (7.27) <sup>de</sup>	0.332 (13.7) <sup>ab</sup>	1.01 (5.13) <sup>o,de</sup>	1.18 (3.32) <sup>a</sup>	0.731 (16.3) <sup>bo,d</sup>	0.859 (26.6) <sup>o,de</sup>	0.674 (38.0) <sup>bo,d</sup>	0.619 (34.6) <sup>ab,o</sup>	0.356 (7.47) <sup>ab</sup>	0.206 (18.2) <sup>a</sup>	5
20:4n-6	2.50 (2.34) <sup>bo,d</sup>	2.28 (3.95) <sup>ab</sup>	2.27 (2.04) <sup>ab</sup>	2.43 (2.97) <sup>bo,d</sup>	2.36 (3.60) <sup>bo</sup>	2.64 (1.94) <sup>od</sup>	2.25 (4.59) <sup>ab</sup>	2.86 (9.95) <sup>d</sup>	2.47 (3.42) <sup>bo,d</sup>	2.01 (7.45) <sup>a</sup>	4
20:5n-3 (EPA)	7.36 (1.44) <sup>ab</sup>	6.70 (2.10) <sup>a</sup>	6.96 (3.00) <sup>ab</sup>	8.01 (3.02) <sup>d</sup>	7.45 (2.23) <sup>bo,d</sup>	8.06 (5.02) <sup>od</sup>	8.22 (4.55) <sup>d</sup>	6.88 (3.57) <sup>ab</sup>	7.41 (3.09) <sup>bo</sup>	7.21 (1.41) <sup>ab</sup>	4
22:2n-6	0.343 (24.3) <sup>ab</sup>	0.298 (7.41) <sup>b</sup>	0.232 (32.9) <sup>a</sup>	0.312 (19.2) <sup>a</sup>	0.247 (26.2) <sup>a</sup>	0.193 (41.4) <sup>a</sup>	0.249 (50.6) <sup>a</sup>	0.164(24.7) <sup>a</sup>	0.216 (44.1) <sup>a</sup>	0.558 (4.62) <sup>a</sup>	2
22:4n-6	1.04 (15.0) <sup>a</sup>	1.17 (2.41) <sup>ab</sup>	1.27 (4.28) <sup>ab</sup>	1.25 (6.95) <sup>ab</sup>	1.28 (7.84) <sup>ab</sup>	1.09 (26.6) <sup>ab</sup>	1.30 (7.98) <sup>ab</sup>	1.07 (6.66) <sup>ab</sup>	1.41 (4.55) <sup>b</sup>	1.19 (3.47) <sup>ab</sup>	2
22:5n-3	2.25 (4.39) <sup>a</sup>	2.57 (1.80) <sup>ab</sup>	2.68 (2.92) <sup>bo</sup>	2.98 (5.80) <sup>od</sup>	3.07 (2.70) <sup>d</sup>	2.89 (6.97) <sup>bo,d</sup>	2.73 (5.11) <sup>bo,d</sup>	2.55 (4.86) <sup>ab</sup>	3.06 (6.74) <sup>od</sup>	2.24 (3.55) <sup>a</sup>	4
22:6n-3 (DHA)	4.60 (0.637) <sup>a</sup>	5.80 (2.09) <sup>b</sup>	5.59 (2.82) <sup>o</sup>	5.78 (3.80) <sup>b</sup>	7.16 (0.729) <sup>o</sup>	6.58 (6.64) <sup>o</sup>	5.81 (5.54) <sup>b</sup>	5.96 (4.65) <sup>b</sup>	6.87 (2.58) <sup>o</sup>	6.67 (1.26) <sup>o</sup>	3
ΣPUFA	22.3 (1.39) <sup>ab</sup>	21.0 (2.30) <sup>a</sup>	22.8 (1.36) <sup>bo</sup>	25.5 (3.03) <sup>a</sup>	24.3 (3.21) <sup>o,de</sup>	24.7 (1.51) <sup>de</sup>	23.0 (2.80) <sup>bo</sup>	22.9 (4.12) <sup>bo</sup>	23.2 (2.27) <sup>bo,d</sup>	21.8 (1.22) <sup>ab</sup>	5
Σsaturates	31.1 (1.41) <sup>ab</sup>	31.7 (0.967) <sup>ab</sup>	31.0 (1.38) <sup>ab</sup>	31.5 (0.629) <sup>b</sup>	31.0 (0.704) <sup>ab</sup>	31.0 (1.99) <sup>ab</sup>	30.6 (1.98) <sup>a</sup>	30.8 (0.627) <sup>a</sup>	31.0 (1.99) <sup>ab</sup>	31.5 (1.02) <sup>ab</sup>	2
Σmonounsaturates	46.6 (0.912) <sup>d</sup>	47.4 (0.200) <sup>d</sup>	46.2 (0.161) <sup>od</sup>	43.1 (1.38) <sup>a</sup>	44.4 (1.05) <sup>bo</sup>	44.4 (0.880) <sup>ab</sup>	46.5 (0.397) <sup>d</sup>	46.4 (1.09) <sup>d</sup>	45.8 (0.235) <sup>od</sup>	46.7 (0.514) <sup>d</sup>	4
Σunsat/Σsat	2.21 (1.04) <sup>ab</sup>	2.16 (1.58) <sup>a</sup>	2.23 (2.08) <sup>ab</sup>	2.17 (0.405) <sup>ab</sup>	2.23 (1.02) <sup>ab</sup>	2.23 (1.53) <sup>ab</sup>	2.28 (2.99) <sup>b</sup>	2.25 (1.22) <sup>ab</sup>	2.22 (2.71) <sup>ab</sup>	2.17 (1.14) <sup>ab</sup>	2
Σn-6	5.13 (3.02) <sup>o</sup>	4.00 (11.0) <sup>ab</sup>	4.55 (4.94) <sup>ab,o</sup>	4.92 (2.03) <sup>bo</sup>	4.14 (7.19) <sup>ab,o</sup>	4.56 (11.2) <sup>ab,o</sup>	3.88 (12.9) <sup>a</sup>	4.69 (15.1) <sup>ab,o</sup>	3.78 (0.614) <sup>a</sup>	3.88 (7.24) <sup>a</sup>	3
Σn-3	17.2 (2.48) <sup>a</sup>	17.0 (1.05) <sup>a</sup>	18.3 (2.23) <sup>ab,o</sup>	20.5 (3.30) <sup>a</sup>	20.2 (2.55) <sup>de</sup>	20.1 (3.51) <sup>de</sup>	19.1 (2.73) <sup>bo,d</sup>	18.2 (2.01) <sup>ab,o</sup>	19.4 (2.62) <sup>o,de</sup>	17.9 (0.433) <sup>ab</sup>	5
Σn-3/Σn-6	3.35 (5.33) <sup>a</sup>	4.31 (10.2) <sup>ab</sup>	4.03 (3.97) <sup>ab</sup>	4.18 (1.51) <sup>ab</sup>	4.89 (5.28) <sup>b</sup>	4.49 (15.5) <sup>ab</sup>	5.01 (13.6) <sup>b</sup>	3.96 (15.6) <sup>ab</sup>	5.13 (2.44) <sup>b</sup>	4.64 (7.49) <sup>b</sup>	2
(EPA+DHA)/n-3,%	41.4 (2.97) <sup>ab</sup>	47.4 (0.344) <sup>od</sup>	43.0 (0.283) <sup>b</sup>	40.0 (0.376) <sup>a</sup>	47.2 (1.18) <sup>od</sup>	45.8 (1.61) <sup>o</sup>	42.1 (1.98) <sup>b</sup>	48.5 (0.883) <sup>d</sup>	48.1 (0.990) <sup>d</sup>	48.4 (1.05) <sup>d</sup>	4

TABLE 2

*Mugil cephalus*. Fatty acids of striped mullet. Entries are means for three replicate analyses of homogenized muscle tissue from each of 10 individual fish. Values in parentheses are % relative standard deviations. Entries in rows sharing the same superscript letter are not statistically different ( $p < 0.05$ ) and are referred to as similarity groups; group numbers refer to numbers of similarity groups computed for each fatty acid.

	No. 1	No. 2	No. 3	No. 4	No. 5	No. 6	No. 7	No. 8	No. 9	No. 10	Groups
	Concentration in wt %										
14:0	2.52 (5.54) <sup>a</sup>	4.14 (2.20) <sup>a,d</sup>	4.43 (1.78) <sup>a</sup>	3.99 (1.85) <sup>a,d</sup>	2.85 (8.12) <sup>ab</sup>	4.30 (1.93) <sup>a,d</sup>	4.39 (5.02) <sup>a</sup>	2.89 (4.83) <sup>ab</sup>	3.01 (0.930) <sup>b</sup>	3.91 (2.77) <sup>b</sup>	5
16:0	22.0 (5.10) <sup>a</sup>	18.8 (1.68) <sup>a</sup>	19.0 (1.99) <sup>a</sup>	20.0 (1.68) <sup>ab</sup>	18.4 (5.21) <sup>a</sup>	19.6 (0.467) <sup>a</sup>	20.0 (0.910) <sup>ab</sup>	19.1 (3.72) <sup>a</sup>	19.1 (1.92) <sup>a</sup>	21.4 (1.28) <sup>ab</sup>	3
18:0	7.19 (2.22) <sup>a</sup>	2.99 (5.07) <sup>a</sup>	3.49 (3.81) <sup>ab</sup>	4.40 (3.61) <sup>a,d</sup>	6.19 (1.75) <sup>f</sup>	3.66 (2.36) <sup>b</sup>	3.94 (7.14) <sup>b,c</sup>	5.55 (3.50) <sup>a</sup>	5.71 (0.323) <sup>a,f</sup>	4.46 (6.25) <sup>d</sup>	7
20:0		0.255 (3.65)	0.251 (3.60)	0.231 (2.69)	0.354 (3.35)	0.219 (1.10)	0.341 (45.6)	0.345 (3.72)	0.237 (2.15)	0.215 (8.01)	1
22:0		0.585 (3.07)	0.479 (26.7)		0.644 (3.62)					0.427 (22.0)	1
16:1	18.2 (9.48) <sup>a</sup>	39.1 (0.957) <sup>f</sup>	35.3 (1.12) <sup>a</sup>	27.6 (1.22) <sup>bd</sup>	21.1 (2.94) <sup>a</sup>	34.8 (0.275) <sup>b</sup>	30.5 (0.702) <sup>d</sup>	26.1 (6.77) <sup>bc</sup>	25.8 (7.04) <sup>b</sup>	28.9 (2.61) <sup>a,d</sup>	6
18:1	5.67 (1.06) <sup>ab</sup>	5.87 (7.28) <sup>b</sup>	6.66 (4.65) <sup>bd</sup>	5.68 (1.43) <sup>ab</sup>	6.17 (1.99) <sup>bd</sup>	6.57 (1.38) <sup>cd</sup>	6.69 (3.98) <sup>cd</sup>	5.17 (3.32) <sup>a</sup>	6.08 (3.09) <sup>ba</sup>	6.73 (3.05) <sup>d</sup>	5
20:1	0.991 (0.469) <sup>o</sup>	1.20 (10.9) <sup>d</sup>	0.945 (2.36) <sup>ba</sup>	1.07 (3.99) <sup>cd</sup>	2.10 (2.09) <sup>f</sup>	1.04 (0.378) <sup>bd</sup>	0.773 (10.8) <sup>b</sup>	0.441 (1.88) <sup>a</sup>	1.09 (11.1) <sup>cd</sup>	0.775 (4.15) <sup>b</sup>	4
22:1		1.30 (4.84) <sup>a</sup>	0.874 (28.2) <sup>d</sup>	0.393 (8.45) <sup>ab</sup>	0.732 (0.968) <sup>cd</sup>	1.83 (4.53) <sup>f</sup>	0.631 (20.7) <sup>bd</sup>	0.297 (5.48) <sup>a</sup>	0.423 (31.8) <sup>ab</sup>	0.504 (8.65) <sup>ab</sup>	6
18:2n-6	0.959 (7.71) <sup>ab</sup>	1.30 (10.3) <sup>a</sup>	1.23 (6.62) <sup>ba</sup>	1.71 (5.08) <sup>d</sup>	0.860 (10.0) <sup>a</sup>	0.865 (3.75) <sup>a</sup>	2.27 (9.14) <sup>a</sup>	1.68 (12.3) <sup>d</sup>	1.47 (1.89) <sup>cd</sup>	1.69 (1.90) <sup>d</sup>	5
18:3n-3	0.606 (5.27) <sup>ba</sup>	0.452 (3.81) <sup>ab</sup>	0.518 (14.6) <sup>ab</sup>	1.36 (0.863) <sup>a</sup>	0.553 (0.377) <sup>ab</sup>	0.373 (0.660) <sup>ba</sup>	0.783 (14.9) <sup>a,f</sup>	0.678 (1.92) <sup>d</sup>	0.879 (12.0) <sup>f</sup>	0.722 (10.9) <sup>da,f</sup>	7
20:2n-6	0.407 (6.37) <sup>a</sup>	0.147 (3.97) <sup>ab</sup>	0.228 (1.70) <sup>ab</sup>	0.288 (29.8) <sup>ba</sup>	0.571 (15.9) <sup>f</sup>	0.108 (13.0) <sup>a</sup>	0.390 (18.9) <sup>da</sup>	0.316 (11.6) <sup>a,da</sup>	0.251 (13.8) <sup>ab</sup>	0.147 (27.0) <sup>ab</sup>	6
20:3n-3	0.362 (4.60) <sup>ab</sup>	0.423 (15.1) <sup>ba</sup>	0.385 (2.15) <sup>ba</sup>	0.317 (5.48) <sup>ab</sup>	0.435 (4.05) <sup>e</sup>	0.318 (2.54) <sup>ab</sup>	0.433 (27.2) <sup>o</sup>	0.292 (11.7) <sup>ab</sup>	0.278 (1.15) <sup>a</sup>	0.300 (1.44) <sup>ab</sup>	3
20:4n-6	4.90 (1.81) <sup>f</sup>	3.15 (0.809) <sup>d</sup>	3.53 (2.34) <sup>a</sup>	3.09 (2.84) <sup>d</sup>	7.00 (1.07) <sup>g</sup>	1.70 (0.672) <sup>a</sup>	1.84 (4.29) <sup>a</sup>	3.68 (3.46) <sup>e</sup>	2.72 (4.20) <sup>o</sup>	2.15 (1.56) <sup>b</sup>	7
20:5n-3 (EPA)	13.4 (1.30) <sup>a</sup>	9.85 (2.10) <sup>a</sup>	9.85 (1.46) <sup>a</sup>	11.4 (0.354) <sup>ad</sup>	11.7 (2.19) <sup>d</sup>	12.8 (0.374) <sup>a</sup>	10.9 (1.69) <sup>ba</sup>	10.8 (3.12) <sup>ba</sup>	11.8 (3.34) <sup>d</sup>	10.7 (1.93) <sup>b</sup>	5
22:2n-6	0.157 (1.45) <sup>a</sup>		0.205 (23.6) <sup>a</sup>	0.105 (19.8) <sup>a</sup>	0.145 (20.7) <sup>a</sup>	0.613 (26.4) <sup>b</sup>	0.170 (43.5) <sup>a</sup>	0.0883 (12.5) <sup>a</sup>	0.0496 (14.1) <sup>a</sup>		2
22:4n-6	0.689 (2.79) <sup>cd</sup>	0.942 (5.25) <sup>a,f</sup>	0.887 (22.1) <sup>da</sup>	0.477 (8.10) <sup>ab</sup>	1.13 (6.49) <sup>f</sup>	0.623 (1.78) <sup>bc</sup>	0.453 (16.4) <sup>ab</sup>	0.474 (17.0) <sup>ab</sup>	0.325 (12.9) <sup>a</sup>	0.267 (0.619) <sup>a</sup>	6
22:5n-3	3.94 (10.9) <sup>o</sup>	3.53 (3.52) <sup>ab</sup>	3.80 (0.877) <sup>ba</sup>	3.69 (3.05) <sup>ba</sup>	4.96 (6.42) <sup>d</sup>	3.89 (0.423) <sup>o</sup>	3.28 (1.79) <sup>ab</sup>	2.99 (3.52) <sup>a</sup>	3.41 (5.63) <sup>ab</sup>	3.66 (2.45) <sup>ba</sup>	4
22:6n-3 (DHA)	18.0 (11.4) <sup>o</sup>	6.00 (4.51) <sup>a</sup>	7.24 (1.01) <sup>a</sup>	13.7 (3.35) <sup>b</sup>	13.6 (9.46) <sup>b</sup>	5.50 (1.39) <sup>a</sup>	11.6 (1.13) <sup>b</sup>	18.6 (5.37) <sup>o</sup>	16.9 (4.03) <sup>o</sup>	12.5 (2.32) <sup>b</sup>	3
ΣPUFA	43.4 (5.53) <sup>a</sup>	25.8 (1.13) <sup>a</sup>	27.9 (0.249) <sup>a</sup>	36.2 (1.14) <sup>o</sup>	41.0 (4.55) <sup>da</sup>	26.8 (0.554) <sup>a</sup>	32.1 (0.0809) <sup>b</sup>	39.7 (3.89) <sup>cd</sup>	38.1 (3.60) <sup>cd</sup>	32.1 (1.74) <sup>b</sup>	5
Σsaturates	31.7 (4.41) <sup>o</sup>	26.7 (1.24) <sup>a</sup>	27.6 (1.55) <sup>a</sup>	28.6 (1.44) <sup>ab</sup>	28.5 (4.37) <sup>ab</sup>	27.7 (0.236) <sup>a</sup>	28.6 (0.605) <sup>ab</sup>	27.9 (2.39) <sup>a</sup>	28.1 (1.37) <sup>a</sup>	30.4 (0.836) <sup>ba</sup>	3
Σmonounsaturates	24.9 (7.00) <sup>a</sup>	47.5 (0.573) <sup>o</sup>	43.8 (0.549) <sup>f</sup>	34.7 (0.782) <sup>cd</sup>	30.0 (2.18) <sup>b</sup>	44.3 (0.147) <sup>f</sup>	38.6 (0.274) <sup>a</sup>	32.0 (5.97) <sup>bc</sup>	33.4 (5.28) <sup>o</sup>	37.0 (2.18) <sup>da</sup>	7
Σunsat/Σsat	2.16 (6.58) <sup>a</sup>	2.74 (1.69) <sup>d</sup>	2.53 (1.18) <sup>cd</sup>	2.44 (1.98) <sup>ba</sup>	2.46 (6.04) <sup>o</sup>	2.46 (0.958) <sup>o</sup>	2.42 (0.623) <sup>ba</sup>	2.54 (3.33) <sup>cd</sup>	2.51 (1.91) <sup>o</sup>	2.23 (1.16) <sup>ab</sup>	4
Σn-6	6.42 (2.22) <sup>f</sup>	4.61 (2.63) <sup>o</sup>	5.19 (2.22) <sup>d</sup>	5.19 (2.21) <sup>d</sup>	8.58 (1.10) <sup>g</sup>	3.29 (4.90) <sup>a</sup>	4.68 (2.96) <sup>o</sup>	5.77 (3.48) <sup>o</sup>	4.50 (4.23) <sup>o</sup>	3.99 (0.127) <sup>b</sup>	7
Σn-3	36.9 (6.58) <sup>o</sup>	21.2 (1.87) <sup>a</sup>	22.7 (0.444) <sup>a</sup>	31.0 (1.69) <sup>ad</sup>	32.4 (5.86) <sup>d</sup>	23.6 (0.334) <sup>a</sup>	27.5 (0.425) <sup>b</sup>	33.9 (4.07) <sup>da</sup>	33.6 (3.51) <sup>da</sup>	28.1 (1.96) <sup>ba</sup>	5
Σn-3/Σn-6	5.76 (7.56) <sup>o</sup>	4.61 (4.33) <sup>b</sup>	4.37 (4.42) <sup>ab</sup>	5.97 (3.94) <sup>o</sup>	3.78 (6.42) <sup>a</sup>	7.18 (4.96) <sup>d</sup>	5.88 (3.41) <sup>o</sup>	5.88 (2.45) <sup>o</sup>	7.48 (0.733) <sup>d</sup>	7.05 (1.90) <sup>d</sup>	4
(EPA+DHA)/n-3, %	84.9 (0.460) <sup>a</sup>	74.8 (0.447) <sup>a</sup>	75.3 (0.844) <sup>a</sup>	81.1 (0.604) <sup>o</sup>	78.1 (0.276) <sup>b</sup>	77.9 (0.186) <sup>b</sup>	82.1 (0.186) <sup>cd</sup>	86.9 (0.139) <sup>f</sup>	85.4 (0.441) <sup>a</sup>	82.4 (0.348) <sup>d</sup>	6

## DISCUSSION

The polyunsaturated fatty acids in all fish lipids (both n-3 and n-6) are derived solely from the diet, but ultimately are of plant origin. In general, plants synthesize all of their fatty acids, and phytoplankton is the basic food in the aquatic field. Those species that feed directly on plant material (phytoplankton and algae) reflect those plant fatty acids, while higher order carnivores accumulate n-3 and n-6 PUFA contained in their prey which have progressed through the food chain from the original plant source (Sargent, 1976; Sargent and Whittle, 1981). Red and brown macroalgae found in both the northern and southern hemispheres are rich in arachidonic acid and EPA (Jamieson and Reid, 1972). Dunstan *et al.* (1988) reported high concentrations of both EPA and arachidonic acid in finfishes who feed on macroalgae in temperate Australian waters which is consistent with findings of Evans *et al.* (1986); high levels of both fatty acids were also observed with the striped mullet in this study. Gibson *et al.* (1984) reported fatty acids in 24 Australian finfishes, of which only the members of the mullet family (Mugilidae), whiting, turbot and leatherjacket had higher EPA concentrations than DHA. A diet containing macroalgae may help explain the elevated levels of both arachidonic acid and EPA in the striped mullet.

The pronounced variability in the fatty acid levels in individual striped mullet is most likely due to inclusion of detrital material in the diet, rather than the macroalgae. Organic detritus in estuarine waters and sediments is composed primarily of small amorphous aggregates which may originate from several sources, including benthic microalgae, phytoplankton, microbes and aggregates of dissolved organic carbon excreted or leached from plants and animals as well as salt marsh plants (Boesch and Turner, 1984). Organic carbon in estuarine sediments is extremely variable (Lytle and Lytle, 1985) and would account for the more highly variable diet of striped mullet which is derived in large measure from sedimentary organic matter.

Spot feed almost exclusively on invertebrates, primarily marine polychaetes and small bivalves. Because of their selective feeding habits, their diet is more consistent than the diet of striped mullet, particularly those feeding in the same areas. Marine polychaete worms, a dietary item of spot but not mullet, contain high concentrations of n-3 PUFA with EPA (20:5n-3) concentrations much higher than DHA (22:6n-3) (Lytle and Lytle, 1990a). Similarly, EPA concentrations were higher than DHA concentrations in the individual spot. Over 90% of 40 species of Gulf finfishes analyzed in our laboratory (Lytle and Lytle, 1990b) contained higher concentrations of DHA than EPA. Spot was one of the exceptions.

Saturated fatty acids, both individually and as a class, are conservative, i.e. are relatively constant and in this case demonstrate little fluctuation in level and distribution among individuals of either spot or striped mullet. On the other hand, the monounsaturates, both individually and as a class, exhibited a wider variation among the individual fish for both species. Individual striped mullet showed a considerable range in 16:1 concentrations; again, this could be a result of the broad spectrum of plant and detrital material in the diet. The narrower range of concentrations of 16:1 among individual spot may reflect the consistent invertebrate diet.

Arachidonic acid, the major n-6 PUFA found in both spot and mullet, was one of the most variable constituents in mullet, producing seven statistically similar groups with four groups in spot. That variation provides strong evidence that this n-6 PUFA is a non-conservative component in both species. High proportions as well as high variability of arachidonic acid are characteristic of tropical Australian marine fish and shellfish (Gibson, 1983; Sinclair, 1983). However, significant levels have been reported in some northern hemisphere fish (Kinsella *et al.*, 1977; Gunstone *et al.*, 1978; Gibson *et al.*, 1984; Gooch *et al.*, 1987).

In summary, the results of this study, based upon a small but selective group of fish, indicate that each constituent fatty acid as well as fatty acid class varies in individuals within a species of marine fish, even when all environmental and physiological effects are minimized. The extent of individual fish variation differs between the two species that were studied, with striped mullet showing much greater variability in fatty acid composition and lipid content than did individual spot. Diet is most likely the primary cause of variations in individual fish, and a more diverse diet probably accounts for the accentuation in individual variability in striped mullet. It is possible that samples collected from other locales or during another season would have shown entirely different trends. This can only be established from more definitive investigations on the composition of fish diets under a variety of fish collection conditions.

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