

## Comparison of Fatty Acid Composition of Some Tissues and Conversion Ratios of Stomach Containing Fatty Acids to Tissue Fatty Acids in *Barbus capito capito* Güldenstaed, 1773

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In this study, it was aimed to investigate the conversion ratios of stomach containing fatty acids to fatty acid profiles of muscle, gonad, gill, liver and adipose tissues and gastric contents in *Barbus capito capito* Güldenstaed, 1773. The significant differences were determined between gastric contents and adipose and gonad tissues in terms of total saturated fatty acids (SFA), similar saturated fatty acids values were found in the other tissues. The highest monounsaturated fatty acid (MUFA) amounts were found in adipose tissue. n-3 Polyunsaturated fatty acid (n-3 PUFA) and n-6 polyunsaturated fatty acid levels of muscle, gonad, gill and liver were higher than diet's n-3 and n-6 polyunsaturated fatty acids. However, n-3/n-6 polyunsaturated fatty acid ratio significantly increased in all tissues except adipose. Palmitic acid in total saturated fatty acids and oleic acid in total monounsaturated fatty acid were determined as the most abundant fatty acids. Total saturated fatty acids in the diet (26.95±1.68 %) augmented to 34.30 ± 2.57 % in gonad and decreased to 20.15±2.2 % in adipose tissue. Although monounsaturated fatty acid amounts in diet (35.82 ± 2.48 % decreased to 26.66 ± 2.48 %) in muscle, n-3 polyunsaturated fatty acid amount of diet (5.37 ± 2.04 %) increased to 24.2±1.89 % in muscle.

**Key Words:** *Barbus capito capito*, Conversion ratio, Fatty acids, Stomach content.

### INTRODUCTION

*Barbus capito capito* Güldenstaed, 1773, commonly called 'caner' in Turkey, occurs naturally in Caspian sea and Aral lake but particularly known as the fish of Aras basin. It is recommended to be the most proper species in terms of aquaculture due to its fast growth rate and white meat content<sup>1</sup>. Although there are some studies on meristic specialities of *B. capito capito*<sup>2,3</sup>, there is no previous report on qualitative characteristics of this species.

There has been augmented interest in the lipid and fatty acid (FA) composition of fish because of strong correlation between fish consumption and human health. It is known that fish lipids contain high levels of eicosapentaenoic acid (EPA),

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docosahexaenoic acid (DHA)<sup>4</sup> and there is inverse correlation of fish or long chain n-3 fatty acid (EPA and DHA) supplement consumption and coronary heart disease<sup>5</sup>.

The lipid content of fishes is highly variable between and within species, depending upon different biotic and abiotic factors<sup>6</sup>. Fatty acid composition of tissues gives valuable information on the dietary content of the fish<sup>7,8</sup>. The aim of present studies are (1) to investigate fatty acid composition of muscle, gonad, gill, adipose and liver and (2) to compare these values with the portion obtained from gastric content to determine the conversion ratios of dietary fatty acids to tissue's fatty acids in *B. capito capito*.

## EXPERIMENTAL

**Fish sampling:** Fish specimens were collected in Kale Creek, Aras basin, Erzurum, Turkey. Fishes were caught by means of cast nets. After capturing, fishes were transferred to the laboratory, tissue samples were excised and washed and stored at -20 °C until analyses.

**Analyses of fatty acids:** The preparation and analysis of fatty acid methyl esters (FAMES) from these fish were performed according to previous method<sup>9</sup>. A piece of muscle tissue obtained was added to 1 mL of 1.2 M NaOH in 50 % aqueous methanol with 5 glass beads (3 mm dia) in a screw cap tube and incubated in boiling water for 0.5 h. The saponified samples were cooled at room temperature for 25 min. They were acidified and methylated by adding 2 mL of 54 % 6 N HCl plus 46 % methanol and incubated at 80 °C for 10 min in a water bath. After rapid cooling, methylated fatty acids were extracted with 1.25 mL 50 % methyl-*tert*-butyl ether in hexane. Each sample was mixed for 10 min and the bottom phase was removed with a Pasteur pipette. The top phase was washed with 3 mL of 0.3 M NaOH. After mixing for 5 min, the top phase was removed for analysis. Following the base wash step, the FAMES is dried in anhydrous sodium sulfate and then transferred into a gas chromatography (GC) sample vial for analysis. Fatty acid methyl esters were separated by GC (HP6890, Hewlett Packard, Palo Alto, CA) with a fused-silica capillary column (25 m by 0.2 mm, pressure 9.00 psi, nominal initial flow 0.4 mL/min and average velocity 26 cm/s) with cross-linked 5 % phenylmethyl silicone (Agilent 19091B-102). The instrument conditions were as follows: carrier gas hydrogen, injector temperature 250 °C; flame ionization detector temperature 300 °C (the detector mode was constant make up flow, makeup gas type was nitrogen and makeup flow 30.0 mL/min); initial and final oven temperatures maintained at 170 and 300 °C, respectively, temperature increased at 5 °C min<sup>-1</sup> to 300 °C, which was held for 12 min and total run time was 38 min. The chromatograms with peak retention times and areas were produced on the recording integrator and were electronically transferred to the computer for analysis, storage and report generation. Peak naming was achieved through the use of a calibration standard FAME (Eucary Method 697110) containing nC9-nC30 fatty acids. Fatty acid methyl esters were identified on the basis of equivalent chain length data.

**Statistical analyses:** In the statistical analyses the data were subjected to ANOVA and the significant means were compared by Tukey's multiple range test<sup>10</sup> and presented as means  $\pm$  SEM. A p value less than 0.05 was accepted as statistically significant.

## RESULTS AND DISCUSSION

Fatty acid contents of the muscle, gonad, gills, adipose, liver and gastric content were presented in Table-1. Total saturated fatty acid, measured as  $26.95 \pm 1.68$  % in gastric content, was found to be increased in the gonads ( $34.30 \pm 2.57$  %) and decreased in the adipose ( $20.15 \pm 2.22$  %). In other words, total saturated fatty acid was increased by 26 % in the gonads and decreased by 23 % in the adipose. Nevertheless, the highest total monounsaturated fatty acid was detected in the adipose ( $53.56 \pm 3.29$  %) and the lowest in the muscle ( $26.66 \pm 2.48$  %), *i.e.* about twice (2.1) times of the muscle (Table-1).

As seen in the Table-1, the highest n-3 polyunsaturated fatty acid value was found in the muscle as  $24.20 \pm 1.89$  % and the lowest was seen in the adipose ( $4.87 \pm 2.50$  %), *i.e. ca.* 4.9 times of the adipose. Total n-6 polyunsaturated fatty acid was detected to be decreased in all tissues when compared to the dietary content. This indicates that the fish have converted n-6 polyunsaturated fatty acid to the other fatty acid types up to 40 % ratio.

n-3/n-6 Ratio, an important parameter in the comparison of fatty acid compositions of fish tissues, was found to be the highest in the muscle ( $1.14 \pm 0.09$ ) and the lowest was in the gastric content ( $0.21 \pm 0.1$ ). The most abundant fatty acid was palmitic acid (16:0) in the total saturated fatty acids and was measured as 64, 52, 53, 53, 68, 54 and 57 % in the adipose, gonad, liver, muscle, gill tissues and gastric content, respectively. Other important saturated fatty acids were lauric acid (12:0) and myristic acid (14:0). In the total monounsaturated fatty acids, oleic acid (18:1 n-9) was the most widespread fatty acid and adipose tissue had the maximum amount as 35.92 %.

An extensive amount of research has been conducted to determine the essential fatty acid requirement of fish and the relationship of diet with fish tissue. The reviews of the lipid requirements of fish have been provided by Castell<sup>11</sup>, Covey and Sargent<sup>12</sup> and Bell *et al.*<sup>13</sup>. In general, freshwater fish require dietary sources of polyunsaturated fatty acids of n-6 (linoleic acid, 18:2 n-6) and n-3 (linolenic acid, 18:3n-3) families for optimal growth<sup>14</sup>. This situation is different from marine fish. Because they are unable to elongate and desaturate fatty acids of the n-3 family in adequate quantity to satisfy minimum requirements. That is, EPA and DHA are known to play a vital role in marine fish<sup>12,15</sup>.

*B. capito capito* is an omnivorous freshwater fish species. According to Barus *et al.*<sup>16</sup>, stream barbell feeds mainly on bottom fauna. It also consume algae and higher water plants, while plant food predominates in more adult specimens (2-7 years of age). Lenhardt *et al.*<sup>17</sup> determined that main food in the diet of the stream barbell was *Chironomidae* although high quantity of *Simuliidae* and *Trichoptera*.

TABLE-1  
FATTY ACID PROFILES (% OF TOTAL FATTY ACIDS\*) OF SOME TISSUES OF *Barbus capito capito*

Fatty acid	n	Adipose	n	Gonad	n	Liver	n	Muscle	n	Gill	n	Stomach
12:0	4	0.71±0.29	3	1.91±0.34	-	-	-	-	-	-	2	0.82±0.41
14:0	4	4.02±0.58	3	3.56±0.67	7	2.08±0.44	-	-	-	-	6	2.28±0.67
16:0	4	12.91±1.93	3	18.03±2.23	7	14.61±1.46	6	17.29±1.57	7	17.27±1.46	7	15.72±1.46
18:0	4	1.52±1.01 <sup>c</sup>	3	12.21±1.16 <sup>a</sup>	7	5.61±0.76 <sup>b</sup>	6	10.32±0.82 <sup>a</sup>	7	13.52±0.76 <sup>a</sup>	7	5.06±0.76 <sup>bc</sup>
<b>ΣSFA</b>	<b>4</b>	<b>20.15±2.22<sup>c</sup></b>	<b>3</b>	<b>34.30±2.57<sup>a</sup></b>	<b>7</b>	<b>26.52±1.68<sup>abc</sup></b>	<b>7</b>	<b>25.04±1.68<sup>bc</sup></b>	<b>7</b>	<b>31.69±1.69<sup>ab</sup></b>	<b>7</b>	<b>26.95±1.68<sup>abc</sup></b>
16:1n7	4	13.81±1.15 <sup>a</sup>	3	8.55±1.33 <sup>b</sup>	7	7.71±0.87 <sup>b</sup>	3	6.69±1.33 <sup>b</sup>	7	5.09±0.87 <sup>b</sup>	7	5.51±0.87 <sup>b</sup>
16:1n9	3	0.81±0.04		-	3	0.67±0.04		-		-		-
18:1n9	4	35.92±1.54 <sup>a</sup>	3	20.07±2.36 <sup>bc</sup>	7	22.71±1.54 <sup>bc</sup>	6	18.33±1.66 <sup>c</sup>	7	15.17±1.54 <sup>c</sup>	7	26.18±1.54 <sup>b</sup>
18:1n9t	3	2.11±0.57 <sup>b</sup>	3	3.77±0.57 <sup>ab</sup>	7	4.55±0.37 <sup>a</sup>	2	5.29±0.7 <sup>a</sup>	7	5.18±0.37 <sup>a</sup>		-
20:1n9	3	1.64±0.38 <sup>c</sup>	3	3.42±0.38 <sup>ab</sup>	5	2.72±0.29 <sup>bc</sup>		-	4	4.25±0.33 <sup>a</sup>	6	2.03±0.26 <sup>bc</sup>
<b>ΣMUFA</b>	<b>4</b>	<b>53.56±3.29<sup>a</sup></b>	<b>3</b>	<b>36.89±32.81<sup>b</sup></b>	<b>7</b>	<b>37.61±2.48<sup>b</sup></b>	<b>7</b>	<b>26.66±2.48<sup>b</sup></b>	<b>7</b>	<b>27.88±2.48<sup>b</sup></b>	<b>7</b>	<b>35.82±2.48<sup>b</sup></b>
20:5n3	4	2.31±1.01 <sup>b</sup>	3	3.92±1.16 <sup>ab</sup>	7	2.99±0.76 <sup>ab</sup>	6	6.61±0.82 <sup>a</sup>	7	5.34±0.76 <sup>ab</sup>	7	2.74±0.76 <sup>b</sup>
22:5n3	3	0.62±0.77 <sup>b</sup>	2	2.32±0.95 <sup>ab</sup>	5	2.21±0.6 <sup>ab</sup>	3	5.32±0.77 <sup>a</sup>	5	3.99±0.6 <sup>ab</sup>	3	2.27±0.77 <sup>ab</sup>
22:6n3	4	1.15±1.23 <sup>c</sup>	3	6.31±1.42 <sup>cd</sup>	7	12.12±0.93 <sup>ab</sup>	6	16.44±1.01 <sup>a</sup>	7	10.46±0.93 <sup>bc</sup>	6	2.88±1.01 <sup>de</sup>
<b>Σn3</b>	<b>4</b>	<b>4.87±2.50<sup>c</sup></b>	<b>3</b>	<b>11.79±2.88<sup>bc</sup></b>	<b>7</b>	<b>18.67±1.89<sup>ab</sup></b>	<b>7</b>	<b>24.20±1.89<sup>a</sup></b>	<b>7</b>	<b>16.69±1.89<sup>ab</sup></b>	<b>6</b>	<b>5.37±2.04<sup>c</sup></b>
18:2n6	4	18.53±2.14 <sup>ab</sup>	3	11.52±2.47 <sup>bc</sup>	7	10.81±1.61 <sup>bc</sup>	6	11.68±1.74 <sup>bc</sup>	6	8.92±1.74 <sup>c</sup>	7	22.27±1.61 <sup>a</sup>
20:2n6	2	0.44±0.12		-	5	0.81±0.08		-		-		-
20:3n6	2	0.66±0.23		-	4	1.39±0.16		-		-	4	0.87±0.16
20:4n6	3	0.67±1.09 <sup>c</sup>	3	4.78±1.09 <sup>b</sup>	7	6.48±0.71 <sup>b</sup>	6	10.59±0.77 <sup>a</sup>	7	12.09±0.71 <sup>a</sup>	7	2.92±0.71 <sup>bc</sup>
<b>Σn6</b>	<b>4</b>	<b>20.45±2.18<sup>ab</sup></b>	<b>3</b>	<b>15.97±2.52<sup>b</sup></b>	<b>7</b>	<b>18.79±1.65<sup>ab</sup></b>	<b>7</b>	<b>22.35±1.65<sup>ab</sup></b>	<b>7</b>	<b>21.10±1.65<sup>ab</sup></b>	<b>7</b>	<b>25.66±1.65<sup>a</sup></b>
<b>n3/n6</b>	<b>4</b>	<b>0.24±0.16<sup>b</sup></b>	<b>3</b>	<b>0.73±0.14<sup>a</sup></b>	<b>6</b>	<b>1.01±0.1<sup>a</sup></b>	<b>7</b>	<b>1.14±0.09<sup>a</sup></b>	<b>7</b>	<b>0.88±0.09<sup>a</sup></b>	<b>6</b>	<b>0.21±0.1<sup>b</sup></b>

\*Values expressed as percentages of total fatty acids, -: Not detected, (a-b-c) means in a line with identical are not significantly different, values given as means ± SEM, (p < 0.05).

In the present study, dietary composition of *B. capito capito* is similar to the above data. In other words, 18:2 n-6 and 18:1 n-9 were found to be higher and 22:6 n-3 and 20:5 n-3 to be lower in diets than tissues. Nevertheless, 22:6 n-3 were 5.7 times higher and 20:5 n-3 2.8 times higher and n-3/n-6 ratio 4.7 times higher in the muscle tissue than diets. These findings may be attributed to high desaturase activity of freshwater fish<sup>4</sup>. Haliloglu *et al.*<sup>18</sup> found that different trout species fed with same diet at similar conditions. Furthermore, there are close relationships between the fish lipid composition and the diets of fish<sup>11,19</sup>. Present results also reveal a close relationship between adipose tissue with stomach content.

Generally, n-3/n-6 ratio changes between 1-4 in freshwater fish species and 5-14 in marine fish species<sup>20</sup>. The present results seem to be closer to those of freshwater fish species. In addition, the most important fatty acids were 16:0, stearic acid (18:0) and miristic acid (14:0) in the saturated fatty acids and 18:1 n-9 and 16:1 n-7 in the monounsaturated fatty acids for freshwater fish. Skuladottir *et al.*<sup>21</sup> and Aras *et al.*<sup>22</sup> reported similar results for total saturated fatty acids and that a large portion of muscle saturated fatty acids was palmitic acid in Atlantic salmon and *S. trutta macrostigma*.

*B. capito capito* had a higher desaturase activity than carnivore trout<sup>15,23</sup>. Therefore, the culture of omnivore fish gains a significant importance in terms of feeding of other fish species and nutrition of human beings and more detailed studies should be carried out to clarify these conditions.

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