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# Short Communication

# Fatty oil composition of *Osteobrama belangeri* (Val.) from Manipur

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The fatty oil content of *Osteobrama belangeri* and its chemical compositions were determined by the analysis of fatty acid methyl esters using gas chromatography (GC) and gas chromatography/mass spectrometry (GC-MS). A total of eighteen compounds were identified constituting about 96.1% and 96.7% of the crude head and body oils. Ten compounds were identified as major fatty acids in the oil and the main acids were oleic acid (38.3 and 35.1%), palmitic acid (27.3 and 23.9%), linoleic acid (12.6 and 21.8%) and octadecanoic acid (8.7 and 7.7%) besides two trace fatty acids.

Key words: Fatty oil composition, Osteobrama belangeri, oleic acid, palmitic acid.

## INTRODUCTION

Osteobrama belangeri (Valenciennes, 1844; family: Cyprinidae; Order Cypriniformes) locally known as Pengba, is a medium sized carp and one of the most preferred fish in the state. It is found in the rivers and lakes of Manipur, India. It is also found in Yunnan, China and Myanmar. It is moderate, highly venerable and categorically not evaluated by the Food and Agriculture Organization (FAO). It is threatened and endangered by extinction in the wild of the conservation assessment and management plan (CAMP) workshop conducted at the National Bureau Fish Genetic Research, Lucknow during 1997 (Molur and Walkar, 1998; Reddy, 2000; Suresh 2000; Menon, 2004). In the past, it formed a big fishery in Loktake Lake. However, this species has become rare due to the introduction of common carp; thus, efficient breeding technique for increasing its population was initiated in states. Among the other carps available in Manipur, O. belangeri almost doubled the price of any other carps consumed by the local people and thus has high commercial value. The literature survey on its fatty

oil analysis revealed that the fatty oil yield and its chemical composition were still not known. Therefore, there is this consideration to determine the fatty oil yield and chemical composition of its fatty oil, and identify the major chemical components.

# **MATERIALS AND METHODS**

The live fishes with weight of 23-27 g/fish were collected from the cultured pond of the institute, sacrificed and the fins were removed. The head of each fish was separated from body. The body (100 g) and the head (50 g) parts were finely grinded separately using home grinder.

#### Preparation of sample

The grinded head and body parts were separately taken in a beaker containing 500 ml water-methanol-chloroform mixture (1:1:2) and was stirred for one hour and repeated three times. Chloroform layer were separated, dried over anhydrous sodium sulphate and concentrates were removed. The head and body oils were found to be brownish and pale yellow in colour respectively with characteristic aroma. The percentage of the crude head and body oils were found to be 5.0% and 7.0% respectively based on the fresh weight basis, constituting about 12.0% of the oil.

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Table 1. Percentage composition of the fatty oil of Osteobrama belangeri.

| S/N | Fatty acid                   | Quantity (%) |          |
|-----|------------------------------|--------------|----------|
|     |                              | Head oil     | Body oil |
| 1   | Tetradecanoic acid           | 1.1          | 0.9      |
| 2   | Pentadecanoic acid           | t            | nd       |
| 3   | 9-hexadecanoic acid          | 2.0          | 1.1      |
| 4   | Hexadecanoic acid            | 27.3         | 23.9     |
| 5   | Heptadecanoic acid           | 0.8          | 0.2      |
| 6   | Linolenic acid               | 0.6          | 0.5      |
| 7   | Linoleic acid                | 12.6         | 21.8     |
| 8   | Oleic acid                   | 38.3         | 35.1     |
| 9   | Octadecanoic acid            | 8.7          | 7.7      |
| 10  | Tetracosanoic acid           | nd           | 2.6      |
| 11  | Archidonic acid              | 1.3          | 1.1      |
| 12  | 11-eicosenoic acid           | 1.1          | 1.0      |
| 13  | Eicosanoic acid              | 0.4          | 0.3      |
| 14  | docosahexaenoic              | 1.7          | nd       |
| 15  | 13-docosenoic acid           | nd           | 0.2      |
| 16  | Docosanoic acid              | 0.2          | 0.1      |
| 17  | Hexacosanoic acid            | nd           | 0.4      |
| 18  | Tetracosanoic acid           | t            | nd       |
|     | Percentage of oil identified | 96.1         | 96.9     |

GC/MS, gas chromatography-mass spectroscopy; nd, not detected; t, trace (lesser than 0.1%).

## Preparation of methyl esters of fatty oil

The fatty oil (1.0 ml) was dissolved in 20.0 ml of sulfuric acid-methanol mixture (1.0%) and heated for 2 h on a boiling water bath. The reaction mixture was then cooled and neutralized with saturated sodium carbonate till pH 7.0. Methyl esters were extracted with diethyl ether and ethereal layer of fatty acid methyl esters was dried using anhydrous sodium sulphate. Diethyl ether was removed on boiling water bath and the methylated mixture was cooled and stored at 4°C.

### Gas chromatography analysis

Gas chromatography (GC) was performed with a Clarus 500 Perking Elmer apparatus equipped with a flame ionization detector and a non-polar HP-1 (cross linked methyl silicone) with capillary column (30 m x 0.2 mm i.d., film thickness 0.33  $\mu m$ ). Chromatographic conditions were as follows: helium as carrier gas at a flow-rate of 1 ml/min; injection volume was 0.5  $\mu l$ ; injector temperature was 250°C respectively. The column temperature was held at 60°C for 5 min, and programmed at 3°C /min to 180°C; then 20°C /min to 280°C and was held for 20 min. The detector and injector temperatures were maintained at 250°C /min to 220°C.

#### Gas chromatography/mass spectrometry analysis

Analysis of the fatty acid methyl ester was performed on a gas chromatography/mass spectrometry (GC/MS) (varian Saturn, 2000) equipped with a Varian C.SVA-5MS capillary column (30 m x 0.25 mm i.d., film thickness 0.25  $\mu$ m). Chromatographic conditions were as follows: helium as carrier gas at a flow-rate of 1 ml/min (split mode); injection volume was 0.5  $\mu$ l; injector temperature was 250°C

respectively. The column temperature was held at 60°C for 5 min, and programmed at 3°C /min to 180°C and then 20°C /min to 300°C and held for 20 min with split mode injection (1:5). The column was coupled directly to the quadrupole mass spectrometer at EI mode at 70 eV with the mass range of 28-400 a.m.u range at 1 scan/s. Kovats retention indices were calculated using co-chromatographed standard hydrocarbons. The individual compounds (Table 1) were identified by mass spectra and by comparing their mass spectra with data available in the NIST 98 Library and literature (Adams, 2007).

## **RESULTS AND DISCUSSION**

Analysis of *O. belangeri* showed that it contained about 12.0% fatty oil, of which body oil was found to have about 7.0% and the head has about 5.0% crude fatty oils. The gas chromatography (GC) and gas chromatography/mass spectrometry (GC/MS) analysis of fatty acid methyl esters were carried out and the fatty acids identified were shown in Table 1, however ten compounds were found to be major in its oil (Figure 1).

A total of eighteen fatty acids, constituting about 96.1% and 96.7% of the crude head and body oils were identified and the main fatty acids were oleic acid (38.3 and 35.1%), palmitic acid (27.3 and 23.9%) and linoleic acid (12.6 and 21.8%) and octadecanoic acid (8.7 and 7.7%) besides two trace fatty acids.

Fourteen compounds constituting about 96.1% of the crude head oil were identified and the main fatty acids were oleic acid (38.3%), palmitic acid (27.3%) and linoleic

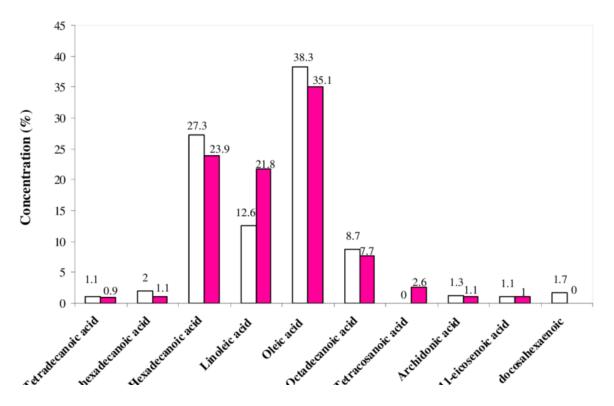


Figure 1. Major fatty acids of Osteobrama belangeri. A comparative study of the major fatty acids identified in head and body oils.

acid (12.6%), octadecanoic acid (8.7%) and 9hexadecanoic acid (2.0%) besides two trace fatty acids. Three minor fatty acids were found absent in the head oil: however they were identified and found to be present in body oil. Similarly, fifteen compounds constituting about 96.9% of the crude body oil were identified and the main aids were oleic acid (35.1%), palmitic acid (23.9%) and linoleic acid (21.8%), octadecanoic acid (7.7%), and tetracosanoic acid (2.6%). Three minor compounds namely tetracosanoic acid (2.6%), 13-docosenoic acid (0.2%) and hexacosanoic acid (0.2%) identified in body oil were found absent in the head oil. However, the results showed that the fatty acid composition of the head and body oils are similar but there is characteristic difference at the level of minor fatty acid content which showed that their presence or absence to their respective parts and thus, make the composition of the fatty oil noticeably different from one another. To our knowledge, this is the first report of the chemical composition of fatty oil of O. belangeri.

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