Research Article

Estimation of Fatty Acids, Amino Acids and Mineral Composition in *Osteobrama vigoressi* (Sykes,1839) from Nira River, Bhor.Maharashtra (India).

Nirbhay Sudhir Pimple*, Sanjay S. Kharat
Assistant Professor, Department of Zoology, Abasaheb Garware College, Karve road, Pune-4, India.
Head of Zoology Department, Principal, Modern College of Arts, commerce and Science, Ganeshkhind, Pune-1, India.
***Corresponding author's E-mail:** nirbhay.pimpale@yahoo.com

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ABSTRACT

Osteobrama vigorssi (Sykes, 1839) is an endemic species from Western Ghats of Maharashtra. The fish is been consumed in fresh as well as dry form by large population hence the study was conducted so as to determine Proximate, Amino acids, fatty acids and mineral composition from the muscles. Lipid content was 14.2% and moisture was 73%. Protein content was 57.9% Minerals included potassium (38%), phosphorous (23%), calcium (06%), sodium (06%), and magnesium (02 %); while zinc, iron, aluminum and copper were present in trace amounts. Eight essential amino acids with total amount of 1282.8µg/g were identified, and lysine formed the highest value, followed by leucine. The result obtained in the study shows that *Osteobrama vigorssi* is good source of proteins, minerals and essential amino acids and has high nutritional value.

Keywords: *Osteobrama vigoressi*, protein, carbohydrates, lipids, minerals, moisture, Essential amino acids, Atomic Absorption Spectrophotometer.

INTRODUCTION

Fresh water bodies throughout the globe are deteriorating in their physio-chemical status. This is due to the influx of sewage, industrial effluents, and agricultural discharge along with pharmaceutical waste which find their way towards these water bodies. These pollutants find their way in the aquatic fauna and ultimately in tissues of Fish. They interfere with the biochemical process of the organism.[1] "THE FISH AS FOOD FOR MANKIND" This need was felt only in the last few decades of 20th century [2, 3].Fish is known as a rich source of protein diet. Fish meat contains significantly low lipids and higher water than beef or chicken and is recommended over white or red meats.[4,5] There is increasingly demanded of fish for use as well as feed. However, information concerning the chemical composition of freshwater fishes in general is valuable to nutritionists concerned with readily available sources of low-fat, high-protein foods such as most freshwater fishes[6, 7].

Fish is rich in active protein which has a unique composition of amino acids, a high omega-3 polyunsaturated fatty acid content such as eicosapentaenoic acid (20:5 n-3, EPA) and docosahexaenoic (22:6 n-3, DHA), and fat-soluble vitamins as well as it represents a good source of micro- and macro-elements [8]; The shortage of α-linolenic acid (18:3 n-3, ALA) is responsible for neurological disorders and poor growth[9]. The presence of DHA and EPA have shown significant positive effect in preventing hypertension and cardiovascular diseases [10] and it has beneficial effects by improving defense mechanisms and also show anti-inflammatory activity of long-chain n-3 PUFAs.[11] Essential polyunsaturated fatty acids such as ALA, EPA and DHA are not synthesized in the human body and effectively synthesized only by aquatic organisms; therefore, humans can receive these essential fatty acids by marine and freshwater fishes[12, 13]. These fatty acids play an important role in the prevention and treatment of cardiovascular disease, autoimmune diseases, eye sight and the improvement of learning ability. Moreover, the measurement of some proximate profiles such as protein contents, lipids and moisture contents is often necessary to ensure that they meet the requirements of food regulations and commercial specifications[14]. Today the limited quantity of food calories is a great concern to many parts of the under developed world, but the quality, notably of proteins, is more crucial.

In recent years, the significance of polyunsaturated fatty acids especially the n-3 and n-6 fatty acids, has gained much attention because of their various biological activities in health and disease management. Recently, several investigations about meat quality and safety of fish from our fishponds were carried out[15, 16]. No such studies have been carried out on *Osteobrama vigorsii* (Sykes, 1839). Thus, it is important to assess the lipid content and fatty acid composition of Osteobramavigorsii *(sykes)* (Sykes, 1839). *Osteobrama vigorsii* (sykes,1839) is consumed by a large population and hence understanding the chemical composition and nutritional quality of this fish would help in formulation of balanced food products, it would provide us with the status of the nutritive value of the raw fish and its benefits to the consumer and would be of a great information for commercial fishery Unit. Detailed studies on physiology, genetics and general biology are therefore in a fish species very much relevant in order to put forward conservation protocols and to

propose newer and improved culture practices [17]. Hence, the study on the total lipid content and fatty acid composition was undertaken.

MATERIAL AND METHODS

Osteobrama vigorsii (Sykes, 1839) is an endemic species from Western Ghats of Maharashtra. 50 individuals of similar body weight and length were collected from Nira River located at Bhor (Maharashtra) at 18° 10' 0" N Latitude / 73° 51' 0" E longitude.

All fish specimens were washed, and placed in sterile universal bottles and kept at -20^{0}C .About 30 gms of fish muscle tissue was separated and used for the determination of different tests. Total protein content was estimated by Lowry's method [18], carbohydrate by Anthrone reagent method (Hedge & Hofreiter 1962) [19], lipid by Folch et al. Method [20]& Moisture and Ash by AOAC method[21]. The concentration of mineral elements was determined using Atomic Absorption Spectrophotometer (AAS) and calculated in ppm (µg/g dry weight). The muscle tissue was analyzed for its proximate composition.

Water Content (%)

Fish tissue has maximum amount of water. Fresh fish sample was sacrificed for the moisture estimation. Automatic moisture analyzer (IR 120, Denver, Moisture analyzer) was used for the estimation. Fish tissue was separated and 1 gm of the tissue was exposed to 100^0 C and then gradually the temperature was raised upto to 170°C until a stable was obtained. The moisture was estimated from the weight loss due to exposure to heating. The moisture was estimated in terms of percentage.

Formula:

$$\text{Water content (\%)} = \frac{\text{Initial weight} - \text{final weight}}{\text{Initial weight}} \times 1000$$

ASH

AOAC method [21] was used to determine the ash content from the fish sample under study. Fish tissues were cleaned washed and 5gms was taken in a previously ignited and weighed silica crucible. Then it was transferred to muffle furnace (Phoenix CEM Corporation, USA) the temperature was maintained at 600^0 C for 6 hours which resulted in the fish ash formation. The ash was allowed to cool and the weight difference was calculated.

Carbohydrate by Anthrone Reagent Method (HEDGE & HOFREITER 1962)

100 mg of fish tissue was hydrolyse by keeping in boiling water bath for three hours with 5ml of 2.5N Hydrochloric acid and then cool to room temperature. It was then neutralise with solid sodium carbonate until the effervescence ceases. The volume was graduated to 100ml and then centrifuged. 0.5 and 1ml aliquots were used for analysis. The standards were prepared by taking 0, 0.2, 0.4, 0.6, 0.8 and 1 ml of the working standard. The test tubes were graduated to 1ml by adding distilled water which was followed by adding 4ml anthrone reagent. The test tubes were heated in boiling water bath for eight minutes after cooling the reading was obtained for green to dark green colored absorption maximum at 630 nm.

Carbohydrate present in 100 mg of the sample= (mg of glucose/volume of test sample) X 100.

Total Lipid was Estimated by Folch Method

1gm of tissue was homogenized with chloroform/menthol (2/1) and the volume was made 20 times the volume of the tissue sample. (1 g in 20 ml of solvent mixture). The whole mixture was then agitated for 15-20min in an orbital saker at room temperature. The homogenate was filtered. The solvent was then washed with 0.2 volumes (4 ml for 20 ml) of water or better 0.9% NaCl solution. After vortexing some seconds, the mixture was centrifuged at low speed (2000 rpm) to separate the two phases. Remove the upper phase was removed by siphoning and kept it to analyze small organic polar molecules. The interface was rinsed one or two times with methanol/water (1/1) without mixing the whole preparation. After centrifugation and siphoning of the upper phase, the lower chloroform phase containing lipids was evaporated under vacuum in a rotary evaporator or under a nitrogen stream if the volume is under 2-3 ml.

Total Protein Estimation was carried out by standard Lowry's Method. 50 well plates were used to prepare the assay using Bovine Serum Albumin (BSA) as standard protein. The absorbance was measured at 595 nm.

RESULT AND DISCUSSION

Proteins, lipids Carbohydrates, moisture contents and Ash along were considered in evaluating the nutritional value of the species studied. *Osteobrama vigoressi* (Sykes, 1839)is consumed by large population in rural as well as urban region from the state of Maharashtra and Karnataka (India). The fish is consumed in fresh as well as dry form. The result obtained showed the nutritional elements of crude protein recording the highest values and lipid recording the lowest.

Table 1: Proximate composition (g/ 100g, dry weight) and caloric values of *Osteobrama vigoressi*. (Sykes, 1839).

Nutrients	*Osteobrama vigoressi*.
Ash	0.60
Carbohydrates	3.18
Lipids	10.78
Moisture	76.31
Protein	23.03

Values are expressed as ± Standard deviation

Table 2: The mineral constituents (ppm, g/g dry weight), and ratios (%) of *Osteobramavigoressi*.(Sykes, 1839).

Minerals	ppm, g/g dry weight and %	
Aluminum	13	>1
Calcium	2102	06
Copper	0.5	>1
Iron	22	>1
Magnesium	687	02
Phosphorous	6982	23
Potassium	11234	38
Sodium	2138	06
Zinc	58	>1
Total minerals	**21098.5**	

Table 3: Different groups of amino acids and total ammonia of *Osteobrama vigoressi.* (Sykes, 1839).The values are expressed as concentrations (µg/g dry weight) and ratios (%).

Amino acids	µg/g dry weight	%
Acidic amino acids	423	15.4
Amino acids with aromatic ring	243	10
Amino acids with OH- group	311	14.2
Amino acids with SH- group	123	03.3
Basic amino acids	544	16
Non-polar amino acids	987	32
Total amino acids	**2631**	
Total ammonia (µg/g)	36	

Table 4. Essential amino acids (EAA) profile of *Osteobrama vigoressi* (Sykes, 1839) values are expressed as (µg/g dry weight) in muscle tissues

Amino acids	µg/g dry weight
Leucine	235.1
Isoleucine	142.3
Valine	158.2
Threonine	147.1
Lysine	268.3
Histidine	098.4
Methionine	101.1
Phenylalanine	132.3
Total EAA	1282.8

This makes the fishes important living resources of dietary protein as other sea and freshwater fish [22, 23.] Protein estimation is an important tool in assessing various climatic and diseased conditions as it is highly useful in exploiting health status of animals [24]. High lipid fishes had less water and more protein than low-lipid fishes. This is in-line with the report of Steffens [25] that protein forms the largest quantity of dry matter in fish. The mineral constituents of *Osteobrama vigoressi* (Sykes, 1839) *were* studied. The result obtained showed considerable concentrations of potassium, phosphorus, sodium and calcium, which makes the fish as good source of minerals. The concentration of Potassium in the present study was high in comparison with other minerals. Few heavy metals analyzed were present, but within tolerable limits. They are in accordance with the study carried out by[1].The variation in different minerals recorded in. *Osteobrama vigoressi* (Sykes, 1839) could be due to the concentration of these minerals in the water body [26,] and the physiologically capacity of the fish to absorb and convert the essential nutrients from the diet or the water bodies where they live. This is supported by the findings of Ricardo et al. (2002), [27, 28 29.] The concentration of Zinc in the present study was 258 ppm, µg/g which is higher this may be due to zinc pollutants which enter the water body and find its way in fish tissue. Zinc is used in painting Ganesh Idol, which is later immersed in water after the festival. There by adding to high level of Zinc in water body. [30]The concentration of Zinc in the present study was 258 ppm, µg/g which is higher this may be due to zinc pollutants which enter the water body and find its way in fish tissue. Zinc is used in painting Ganesh Idol, which is later immersed in water after the festival. There by adding to high level of Zinc in water body. [31] Microelements are required for the biochemical process in trace amount, but they tend to become harmful when their concentrations in the tissues exceed the metabolic demands [32;] Microelements are required for the biochemical process in trace amount, but they tend to become harmful when their concentrations in the tissues exceed the metabolic demands [33.] Basic amino acids forms 17%,Acidic amino acids forms 15.6%,non-polar amino acids with aliphatic side chains constituted 15.1% ,amino acids with a sulphur group forms 04.3%, and amino acids with aromatic ring on the side chains forms 11.The present work would help to understand the composition of fish which is good sources of protein and minerals. It would be a very informative for commercial fishery science

CONCLUSION

The result obtained in the study shows that *Osteobrama vigoressi (*Sykes, 1839) is a good source of proteins, minerals and essential amino acids and has high nutritional value. Acidic amino acids forms 15.4%,non-polar amino acids with aliphatic side chains constituted 32% ,amino acids with a sulphur group forms 033%, Basic amino acids forms 16%, and amino acids with aromatic ring on the side chains forms 10%.The present work will help to understand the composition of fish which is good sources of protein and minerals. It would be a very informative for commercial fishery science.

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