

Nutritional and Techno-Functional Properties of Fish Protein Powder (FPP) from Underutilized Small Fish (*Barbus paludinosus*) Species

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To cite this article:

Bezuayehu Gutema Asefa. Nutritional and Techno-Functional Properties of Fish Protein Powder (FPP) from Underutilized Small Fish (*Barbus paludinosus*) Species. *Journal of Food and Nutrition Sciences*. Vol. 9, No. 3, 2021, pp. 84-88. doi: 10.11648/j.jfns.20210903.13

Received: April 21, 2021; **Accepted:** June 7, 2021; **Published:** June 15, 2021

Abstract: Around a billion people in the world have inadequate protein intake, leading to reduced growth and development. In growing interest to find alternative protein sources, several studies have been carried out on new protein ingredients, including fish protein powder (FPP), single cell protein, and leaf proteins. FPP, extracted from either underutilized species or byproducts of the fish industry, is one of such alternatives and have been studied for a couple of decades. It contains a concentrated protein intended for human consumption. In view of assessing the suitability for further processing and product development, profiling the nutritional and techno-functional properties of FPP produced from different fish species is pertinent. This study evaluated the proximate composition, some mineral content and techno-functional properties of FPP extracted from small barbus (*Barbus paludinosus*) species. A mince of gutted fish was used to extract the protein powder using isopropyl alcohol. The average moisture, protein, ash and fat content were found 3.07%, 72.38%, 24.37%, and 1.9%, respectively. The mineral analysis of the FPP has showed high concentration of Phosphorous, Calcium, Potassium and Sodium. Techno functional attributes of the FPP were also comparable with FPP products from other species. Generally, protein powders from underutilized fish (*Barbus paludinosus*) could be used to formulate nutritionally improved products with satisfactory techno functional properties.

Keywords: Fish Protein Powder, *Barbus paludinosus*, Nutrition, Proximate Analysis, Techno Functional Properties

1. Introduction

Globally, one of the major challenges in nutrition until today is protein-energy malnutrition (PEM): i.e., deficiency of energy derived from proteinaceous foods [1]. Around a billion people in the world get insufficient protein from their diet, leading to reduced growth and development. Several factors, including low nutrient density of food products and lack of diversity in the staple diet have vital role in downgrading the nutritional quality of foods in developing countries [2]. In growing interest to alleviate PEM, searching for alternative protein sources have been underway since several decades. Several new protein ingredients, including fish protein powder (FPP), single cell protein, and leaf proteins have been used as a source of protein energy in a nutritious diet [1].

Composition, functional properties and applicability of different protein sources have been studied by scientists in a response to the growing demand for food proteins [3]. FPP, extracted from either underutilized species or byproducts of the fish industry, is one of such alternatives and have been studied for a couple of decades. FPP is a dried and shelf stable product, intended for human consumption. It contains highly concentrated protein than the original fish flesh, and it is an excellent source of highly digestible amino acids [4, 5]. FPP has been used for numerous applications in food industries as a functional and nutritional property enhancer. For instance, FPP have been used to enhance functional properties by serving as a binder, dispersing agent and emulsifier in a food industry [6, 7]. Besides, fish protein powder has shown importance to improve protein quality of diet specifically for pre-school children and infants under the

age of five [8, 9].

In view of the importance of exploiting the wasted and underutilized fishery resources for the production of FPP, several researchers evaluated the nutritional and techno functional characteristics of FPP extracted from different fish species. The aim of the current study was to characterize the biochemical composition and techno functional attributes of FPP prepared using underutilized fish (*Barbus paludinosus*) species, obtained from lake Ziway, Ethiopia.

2. Materials and Methods

2.1. Fish Protein Powder (FPP)

Small barbus (*Barbus Paludinosus*) species were harvested from Lake Ziway using a monofilament gillnet. A total of 72 fish with average weight and an average length of 10.83 ± 6.19 g and 10.00 ± 1.93 cm, respectively were used for the experiment. The fish were gutted, washed thoroughly, and minced before the extraction process.

Extraction of FPP was performed following a procedure described in Sikorski and Naczek, 1981 [10]. The minced fish was transferred into a 250 ml Erlenmeyer flask placed in a water bath which has been set at 25°C. The first stage of extraction was done using isopropanol alcohol for 50 minutes at the mentioned temperature. For the second stage of extraction using isopropanol alcohol, the temperature has been raised to 75°C and maintained for additional 90 minutes. In the last stage of extraction, azeotropic isopropanol alcohol has replaced the isopropanol and the extraction was completed in 70 minutes at 75°C. In between the three extraction processes, the mixture has been centrifuged to separate the supernatant. The final supernatant fraction was dried and milled into a fine powder before packaging in a polyethylene bag.

2.2. Evaluation of Nutritional Value

Proximate composition and functional properties were analyzed in triplicate while the mineral content was analyzed in duplicate. Results were reported in average \pm standard deviation.

2.2.1. Determination of Protein Content

Crude protein was determined by the Kjeldahl method

$$\text{Weight of fat} = (\text{weight of container} + \text{extracted fat}) - (\text{weight of container})$$

$$\text{Fat content (\%)} = \left(\frac{\text{Mass of fat extracted (g)}}{\text{Weight of original sample (g)}} \right) \times 100$$

2.2.4. Determination of Ash Content

Clean crucibles were placed under a muffle furnace at 550°C for one hour. Crucibles were moved from the furnace to desiccators and cooled to room temperature before weighing. Five grams of the sample was added to dried silica crucibles. Placed in a muffle furnace and the temperature was held at 550°C overnight. The crucibles were taken to desiccators and cooled to room temperature. The cooled crucible containing the samples were then weighed

according to AOAC, 2005 [11]. A gram of sample, 12 ml of concentrated H_2SO_4 , and 1g of catalyst was added to the digestion flask. It was digested at 400°C for 2^{1/2} hours. After digestion was completed, the content in the flask was diluted by 50 ml of water and neutralized using 40% of 40ml NaOH. Then, Kjeldahl distillation unit was used to distill off the ammonium. The distilled ammonium was trapped using a 2% boric acid mixed with indicator solution containing methyl blue and methyl red. Lastly, the ammonium borate complex was titrated using standardized 0.1 N HCL. The amount of consumed HCL was recorded and total crude protein was calculated as total nitrogen.

$$\%N = \frac{(\text{ml titrated} - \text{blank}) \times N \times \text{Eq. wt of Nitrogen}}{1000 \times Wt} \times 100$$

$$\% \text{ Crude Protein} = \%N \times 6.25$$

Where, Wt = weight of the sample; N= concentration of sulfuric acid

2.2.2. Determination of Moisture Content

The moisture content of the sample was analyzed following AOAC (2000) [12]. Firstly, all the drying dishes were heated at 105°C for 1 hour in a drying oven. Then the dishes were taken to a desiccator to cool. The weight of the drying dish (W_1) and five grams of samples (W_2), were recorded. The drying procedure was carried out for 3 hours at 105°C in an oven. Then, the samples were taken out and kept in a desiccator at room temperature. The cooled samples were weighed and put back into the drying oven for 1 hour. The procedure repeated until constant weight was achieved (W_3).

$$\text{Moisture (\%)} = \left(\frac{W_2 - W_3}{W_2 - W_1} \right) \times 100$$

2.2.3. Determination of Fat Content

Total lipid was extracted by a diethyl ether solvent system [13]. About 5 g of sample was used to extract the fat. The weighted sample was added to a 250 ml round bottom flask with 50 ml diethyl ether, and extraction took place for about 4 hours at 110°C in the Soxhlet extractor. Then, removal of the organic solvent was done at 40-60°C under reduced pressure using a rotary evaporator. Finally, the weight of fat was calculated using the following equations.

immediately [12].

$$\text{Ash (\%)} = \left(\frac{\text{Weight of ash}}{\text{Weight of sample}} \right) \times 100$$

2.3. Determination of Functional Properties and Quality Criteria

2.3.1. Water / Oil Absorption Capacity

Determination of water/oil absorption was carried out using a procedure described by Bencini 1986 [14]. A one gram of FPP sample was weighed into clean conical

graduated tube. The powder was mixed thoroughly for 30 seconds by adding 10 ml distilled water. The mixture was held for 30 minutes at room temperature and centrifuged for 30 minutes at 5000 rpm. After completing the centrifugation, the volume of the free water (supernatant) or oil was read directly from the graduated centrifuge tube. Conversion of the absorbed water to weights in grams was done by multiplying using the density of oil i.e., 0.894 g/ml, and the density of water i.e., 1 g/ml. The oil/water absorption capacities were then expressed in grams of oil/water absorbed per gram of flour sample [14].

$$\text{Absorbed water} = \text{total water} - \text{free water}$$

2.3.2. Bulk Density

The bulk density of FPP samples were determined using a procedure of Onkuwa 2005 [15]. Ten grams of sample was weighed in a 10 ml measuring cylinder. Then a constant volume was achieved by repeatedly tapping the cylinder at the bottom. Finally, the packed volume was registered and calculation of the bulk density was done by using the following formulae.

$$\text{Bulk density (g/ml)} = \frac{\text{weight of sample (g)}}{\text{volume of sample (ml)}}$$

3. Results and Discussion

Table 1. Proximate composition, functional properties of FPP.

Parameters	FPP
Moisture content (%)	3.07 ± 0.06
Crude ash (%)	24.37 ± 0.14
Crude protein (%)	72.38 ± 0.85
Crude Lipid (%)	1.90 ± 0.14
Total phosphorous - TP (%)	4.34 ± 0.22
Calcium - Ca (mg/kg)	31,066.7 ± 242.30
Sodium - Na (mg/kg)	578.3 ± 26.85
Potassium - K (mg/kg)	1445.8 ± 29.40
Bulk density (gm/ml)	0.54 ± 0.11
Water holding capacity (%)	334 ± 1.60
Oil holding capacity (ml/gm)	1.43 ± 0.11

3.1. Proximate Composition

From the data presented in table 1, the moisture, crude ash, crude protein, and crude lipid were 3.07 ± 0.06, 24.37 ± 0.14, 72.38 ± 0.85, and 1.90 ± 0.14 in FPP, respectively. Similar moisture values were obtained in various studies [16-18]. The obtained moisture value is generally accepted based on FAO's standard, which declares < 10% as an acceptable level for FPP [16]. Similarly, comparable results in crude protein and crude lipid were found from FPP extracted from catfish (*Pangasius hypophthalmus*) [19]. According to Sathivel *et al.*, (2004) [20], a protein content ranging from 63 to 81.4% was found from different parts of herring. The crude ash content in our finding was 24.37 ± 0.14%, which is remarkably higher than previous reports [16, 17, 19]. This could result because, in all above-mentioned studies, flesh edible parts of the fish were used to extract the FPP, while whole gutted fish was used in our study, which could consequently raise the

total ash content due to the presence of the fish frame and head as part of the extract. From the proximate analysis, it can be concluded that FPP is a highly proteinaceous ingredient with low fat and moisture content.

3.2. Mineral Content

The mineral content of FPP can be used to further elaborate on the nutritional importance in a food. As shown in Table 1, 43.46 ± 0.22, 31.07 ± 2.42, 0.58 ± 0.26, and 1.44 ± 0.29 g/kg of P, Ca, Na, and K were obtained respectively. All the recorded mineral contents broadly vary compared to a previous finding by Goes *et al.*, (2015) [21], who found 5.13 ± 0.66, 7.09 ± 0.92, 6.43 ± 0.11, and 1.39 ± 0.06 g/kg respectively for P, Ca, Na, and K. The observed difference compared to the finding by Goes and the team could be due to the variation in species and part of the muscle tissue used for the analysis. In their research, the FPC was prepared from muscle tissue of the fish, while the whole fish was used in the current study. It was also stated that the mineral in FPP may vary depending on the raw material used for the extraction [7, 22].

3.3. Functional Properties

3.3.1. Bulk Density

The obtained bulk density of FPP was 0.54 (g/ml). Higher bulk density ranging from 1.34 to 4.72 g/ml were reported in other studies [18, 23, 24]. The observed variation in the bulk density of FPP could be due to the difference in processing method used to prepare the protein powder. All the FPP used in the previous findings were dried using either freeze dried or spray dried, while in the current study, oven drying method were applied. The variation in the bulk density could therefore be due to the different driers used in the studies. The bulk density of powder is known to be influenced by several factors such as design of dryer and dryer configuration [25]. A high bulk density is important and desirable as it can significantly reduce costs of packaging and transportation [25].

3.3.2. Water Holding Capacity

The water holding capacity found in the current study is 334% which is comparable with other researchers' findings [18, 23, 24], in which the WHC fall in the range between 253.15% to 359.21%. In food product development, the capacity of proteins in preventing water expel from the three-dimensional structure i.e., water holding capacity, is crucial for texture development [26]. Protein ingredients having both high and low WHCs may have their own consequences. The former may dehydrate other ingredients in a food system, while the later could be unstable in humid storage [3].

3.3.3. Oil Holding Capacity (OHC)

The OHC of the protein powder was 1.43 ml/g. This finding was similar to the finding by Lone *et al.*, 2015 [27], who obtained the same result from protein concentrate extracted from Traut fish protein isolate. In other study, OHC of 2.43 ml/g was reported from freshly minced meat protein concentrate [28]. Since OHC is mainly depend on the

available non-polar amino acids in the protein structure and side chains, the observed low OHC could be related with the amount of non-polar amino acids in the protein structure of the FPC [27]. OHC of proteins are known to influence the organoleptic properties typically mouthfeel and flavor. During a development of food products such as sausage, cakes, batters and mayonnaise, high OHC is vital [29].

4. Conclusion

Proximate composition, some mineral content and techno-functional attributes of FPP produced from underutilized fish (*Barbus paludinosus*) species were studied. The FPP was rich with high quality protein, minerals and showed good techno functional properties. Broadly high concentration of Phosphorous, Calcium, Potassium and Sodium were found in this study compared to previous findings which could be due to inclusion of whole fish except the viscera during FPP extraction. Protein powders from underutilized fish (*Barbus paludinosus*) could be used for formulation of nutritionally improved products with acceptable techno functional properties. Future research on micronutrient composition, shelf-life evaluation and economics aspect of the protein powder would be relevant. Furthermore, comprehensive research is necessary to assess the potential of other underutilized fish species in making quality fish protein powder.

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