A process developed for preparation of partially hydrolysed and deodourised (PHD) fish flour, without solvent extraction is described. The PHD fish flour prepared from four varieties of trash fish, had creamy white colour and contained 87-92% protein and 7-8% available lysine. The products had shelf life of more than one year at room temperature, and caused no grittiness when incorporated in snacks and bakery products at 10% and 7.5% level respectively. The flour prepared from *Nemipterus* spp. had a PER 2.32 compared to 2.68 of raw fish meat and 2.5 of standard casein.

**INTRODUCTION**

Efforts are being made all over the world to develop edible fish flour or fish protein concentrate having good storage life and characteristics suitable for incorporation into domestic preparations (Moorjani and Lahiri, 1962; 1970; Hale, 1972; 1974). However, none of the methods developed so far could give a product which is satisfactory in all respects. By and large the major drawbacks of these products were the development of fishy odour during storage, grittiness and the carcinogenic nature of the residual solvent (Moorjani and Lahiri, 1962). Further, the product could not be mixed well with conventional foods. In this paper is presented a method for the preparation of partially hydrolysed and deodourised (PHD) fish flour which could be directly incorporated into snacks and other preparations without any objectionable features. Its characteristics, storage behaviour and suitability for incorporation into domestic snacks and bakery products have been discussed.

**MATERIALS AND METHODS**

PHD fish flour was prepared from the following individual species of fish viz., *lactarius* (*Lactarius lactarius*), croaker (*Otolithus* spp.), threadfin bream (*Nemipterus japonicus*) and ribbon fish (*Trichiurus lepturus*). These are low cost, non-fatty varieties of fishes available along Mangalore coast, with fat content ranging from 2-3%.
Dressed and washed fishes were fed to a deboning machine and the picked meat was placed in a thick muslin cloth fixed to a meat washing cradle and washed for 2-3 min. with a jet of cold water. The meat was then transferred to a steam jacketed kettle, boiled for 5-6 min. and transferred again to the washing cradle, where it was washed continuously with cold water till the wash water became clear; after which it was washed in hot water for 4-5 min. The rocking movement of the meat washing cradle kept the meat rolling to and fro while washing, thus facilitating a thorough washing of the meat. The meat was then pressed lightly in a hydraulic press to remove the free water and then minced in a power driven meat mincer. The minced meat was then partially hydrolysed overnight in a stainless steel container at room temperature with 1:1 (weight/volume) 1 N. hydrochloric acid. After hydrolysis the pH of the slurry was adjusted to 7.0 with 1 N. sodium hydroxide and the meat was washed alternatively with hot and cold water 4-5 times, till the wash water was clear. The washed meat was then pressed in a hydraulic press to remove as much water as possible and dried at 70°C in an electric drier to a moisture content of 5-6%. The dried material was then ground to 0.5 mm. particle size and treated with 1% acetic acid taken in requisite quantity of water. It was dried again to a moisture content of 5-6% and stored in airtight bottles or tin containers after cooling to room temperature.

Nitrogen was estimated by macro-kjeldahl method and multiplied by 6.25 for total protein. Fat was estimated by extracting the material with petroleum ether (60-80°C) in a soxhlet apparatus. Moisture was determined in an infrared moisture meter and the pH using an Elico pH meter. The volatile base nitrogen (VBN) was estimated by the Conway microdiffusion method as described by Beatty and Gibbons (1936). α-amino nitrogen was determined by the method of Pope and Steven (1939). Pepsin digestibility was estimated according to AOAC (1960) method. Available lysine was estimated by the Carpenter's (1960) method. Total plate count, E. coli, Coagulase positive Staphylococci and Salmonella were determined according to APHA methods (Anon, 1966).

Protein efficiency ratio (PER) of raw fish meat and PHD fish flour were determined according to the method of Chapman, et al., (1959) using male weaning albino rats (Vistar strain) weighing around 40 g. They were fed with iso-caloric and isonitrogenous diets at 15% protein level for four weeks. Since commercial casein obtained from the local dairy gave a very low value, (Chandrashekara, et al., 1962) the standard PER value of casein was taken for comparison.

RESULTS AND DISCUSSION

The proximate composition of fish flour prepared from four types of fishes is shown in Table I. Fish flour from lactarius and thread fin bream were comparatively better in colour and protein content than the other two. While the ash content was less than 1% in all samples, the fat content varied from 0.2 to 0.65% depending on the fat content of the fish. The protein content also varied from 87-93% for different types of fishes. The products when reconstituted in water and boiled, gave slight
TABLE I
PROXIMATE COMPOSITION OF PHD FISH FLOUR FROM DIFFERENT SPECIES OF FISH

<table>
<thead>
<tr>
<th>Fish flour from</th>
<th>lactarius</th>
<th>threadfin bream</th>
<th>croaker</th>
<th>ribbon fish</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protein %</td>
<td>91.87</td>
<td>92.85</td>
<td>87.48</td>
<td>87.24</td>
</tr>
<tr>
<td>Fat %</td>
<td>0.23</td>
<td>0.52</td>
<td>0.39</td>
<td>0.65</td>
</tr>
<tr>
<td>Ash %</td>
<td>0.89</td>
<td>0.52</td>
<td>0.89</td>
<td>0.23</td>
</tr>
<tr>
<td>Moisture %</td>
<td>5.98</td>
<td>5.23</td>
<td>5.98</td>
<td>8.50</td>
</tr>
<tr>
<td>Pepsin digestibility %</td>
<td>92.87</td>
<td>90.59</td>
<td>92.87</td>
<td>92.54</td>
</tr>
<tr>
<td>Available lysine (g./100g. protein)</td>
<td>0.02</td>
<td>7.98</td>
<td>7.08</td>
<td>7.63</td>
</tr>
</tbody>
</table>

TABLE II
NUTRITIONAL QUALITY OF PHD FISH FLOUR PREPARED FROM THREAD FIN BREAM

<table>
<thead>
<tr>
<th>Isocaloric diet containing</th>
<th>Protein content of the diet %</th>
<th>Initial body weight %</th>
<th>Gain in wt. at the end of four weeks g.</th>
<th>Total protein consumed during the period g.</th>
<th>PER</th>
</tr>
</thead>
<tbody>
<tr>
<td>Raw fish meat</td>
<td>15.63</td>
<td>37.75</td>
<td>115.50</td>
<td>43.06</td>
<td>2.68</td>
</tr>
<tr>
<td>PHD fish flour</td>
<td>15.51</td>
<td>41.00</td>
<td>83.24</td>
<td>35.93</td>
<td>2.32</td>
</tr>
</tbody>
</table>

smell of boiled fresh fish. The yield of fish flour from these fishes varied from 4-6%, based on the whole fish and 12-13% based on the picked meat. The pepsin digestibility was around 92% in all cases. The available lysine content was found to vary between 7 and 8%. The PHD fish flour had creamy white colour, had no grittiness when incorporated into products.

Storage behaviour of fish flours prepared from lactarius and ribbon fish were studied for one year. The parameters used were proximate composition, VBN and peroxide value. While there were no significant changes in the proximate composition, the VBN and peroxide values were negative throughout the period of storage.

Feeding experiments carried out with albino rats showed a protein efficiency ratio of 2.35 for fish flour prepared from
thread fin bream. The results of the experiment based on the values obtained with eight experimental animals are shown in Table II. The PER values are slightly low as compared to the PER of raw fish meat (2.68) but are quite comparable with that of standard casein (2.5).

Bacterial load determined at various stages of preparation and of the final product are shown in Table III. The data indicate that the very heavy load of bacteria observed in the raw picked meat was reduced gradually to nil at the stage of hydrolysis. The final product however showed a bacterial load of 400/g. of flour, which was due to subsequent contamination of the product during handling. However, the total count was much lower compared to the microbiological standards set for fish flours and fish protein concentrates (not exceeding 10,000/g.). Tests for the presence of E. coli and Coagulase positive Staphylococci and Salmonella were, however, negative in the final product.

A mild acid hydrolysis of fish meat was employed mainly for purposes of eliminating the coagulable and elastic properties of proteins. However, it also appears to have a favourable effect on the removal of fat apart from removing the residual bones responsible for causing grittiness in the final product. The extent of hydrolysis brought about was only marginal as judged by the α-amino nitrogen content of the meat before and after hydrolysis. In the case of thread fin bream the values were 7 mg./100 g. and 11.6 mg./100 g. of wet meat respectively and for croaker fish the corresponding values were 18.6 mg./100 g. and 23.3 mg./100 g.

Fish flour prepared from all the four varieties of fish were incorporated into snacks like spirals (chakkuli), sevu, diamond cuts, and bakery products like bread, biscuits and cakes, at 5-12% level. While 10% level of incorporation into snacks was found to be optimum and went without notice of fish flavour, it gave a slight fish flavour in bread, biscuit and cake. However, 7.5% level in bakery products was found to be quite suitable in all respects.

The method adopted does not aim at complete removal of fat from fish meat, since the approach for checking the flavour reversion (development of fishy odour) during storage was different from the earlier approaches. By repeated washing with hot and cold water, perhaps it is impossible to remove the bound lipids. It is also difficult to say, whether the partial hydrolysis adopted in the process can release the bound lipids to any great extent. However, the aim of the experiment was to remove fat as much as possible from the fish meat by washing with water and treat the final product with acetic acid to check the flavour reversion that occurs during storage. Observations of treated and untreated samples of fish flour, for over an year, showed that the untreated samples developed fishy odour in less than three months, while the treated samples kept well for more than an year without developing any fishy odour.

The process, apart from being simple is also efficient, since it gives an yield of 4-6% from whole fish and 12-13% based on picked meat. The maximum yield obtainable theoretically is 15-16% on picked meat, depending on the protein content.
of fish. The product has excellent keeping quality even when the fat content was 0.65%. However, it is desirable to reduce the fat content in the product to a level below 0.5% for long storage.

The washings of meat were collected and made use of for the preparation of "fish extract", similar to "meat extract", for use in microbiological media. The dressings of fish and the skin and bones obtained after picking the meat were made use of for making fish meal. The cost of fish flour prepared by this process works out to approximately Rs. 12.00 per kilogram, if fish is purchased at Rs. 500/- per tonne.

ACKNOWLEDGEMENT

The authors wish to record their sincere thanks to Dr. N. P. Patil, Director of Research, for his keen interest in this project and the encouragement given. We also thank Dr. N. L. Lahiry, for giving useful suggestions during the course of this study. The financial help given to this project by the Indian Council of Agricultural Research is gratefully acknowledged.

REFERENCES


Setty et al.: Partially hydrolysed and deodourised fish flour


National Oceanic and Atmospheric Administration, U. S. A.


