

FPC'S QUALITY VIRTUALLY THE SAME AS ITS RAW MATERIAL'S QUALITY

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Several years ago, the National Marine Fisheries Service (then the Bureau of Commercial Fisheries) began a program designed to produce a satisfactory protein concentrate from whole fish. The purpose was to help alleviate the protein malnutrition that affects much of the world's population and to provide an economic stimulus to the U. S. fishing industry. A high-quality protein is needed to supplement the vegetable proteins, which are the world's principal source of this important nutrient.

The use of fish for this purpose is ideal because the seas abound in unutilized species. The concept of FPC is not new. Knobl (1967) reviewed descriptions of several processing methods. Chemical methods have used isopropyl alcohol as a solvent (Guttman and Vandenheuvel, 1957; Dambergs, 1959; Power, 1964; and National Marine Fisheries Service, 1966).

Isopropyl alcohol has been shown highly efficient in removing lipid and water from raw material. A product containing high-quality protein can be obtained by isopropyl alcohol extraction of various species of fish.

Thus far, few investigations have compared the chemical composition and nutritive quality of the raw material with that of the FPC processed from it. So we have not known whether the solvent extraction and later processing produces any significant change in quality from that of raw material.

This report presents results of a comparison of FPC's chemical composition and nutritive quality with the same properties of the raw fish used as starting material.

The FPC data were obtained in early stages of National Marine Fisheries Service

program, when a batch cross-current method of extraction was used. Although the present method is a straightforward stage-wise countercurrent method, we have found no difference between the two methods of extraction with respect to chemical composition and nutritive value. The principal difference is in the efficiency and economy of solvent usage. For this report, the FPC data are representative of FPCs prepared by isopropyl alcohol extraction of red hake regardless of extraction method.

WHAT WE FOUND

The chemical and nutritive properties of freeze-dried whole fish and FPC (fish protein concentrate) made from red hake (*Urophycis chuss*) were studied. Samples of whole fish were collected during July, October, and November 1964, and January 1965. The products were analyzed for proximate composition, amino acid concentration, and protein efficiency ratio (PER).

The samples of freeze-dried whole fish showed only slight changes in chemical composition, notably in lipid content, which ranged from 9.8% to 14.2% (dry weight). The PERs ranged from equal to casein to PERs significantly better than casein (July, October, and January).

The FPC samples prepared from same catches of fish showed only slight differences in concentration of crude protein; this ranged from 86.5% to 89.9%. The PERs of these samples also ranged from equal to casein (October and November) to PERs significantly better than casein (July and January).

With the exception of the October period, FPC's nutritive quality did not differ significantly from raw material's.

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EXPERIMENTAL PROCEDURES

Raw Material

The whole red hake was chosen as raw material for this study because it is an underutilized species and readily available to this laboratory. The fish were caught near Block Island, off Rhode Island. The hake were taken directly from net as soon as they arrived on deck and placed immediately on ice. About 100 to 200 pounds of hake from each catch were brought to the laboratory, where they were stored in a freezer at -40 C. The fish were used in a series of experiments within 3 weeks.

Product Preparation

A. Freeze-Dried Whole Fish

Because the nutritive value of raw fish is difficult to evaluate in feeding trials, we chose arbitrarily to freeze-dry the fish. This technique would provide a product suitable for testing--yet one likely to be as similar to raw material as possible in chemical and nutritive characteristics.

Twenty-pound samples of the frozen whole raw hake were placed into liquid nitrogen and ground, first through a Rietz^{1/} extractor, and then through a Rietz disintegrator in a stream of liquid nitrogen. The ground particles, collected in liquid nitrogen, were loaded into freeze dryer in an excess of liquid nitrogen; then they were freeze-dried at pressure of 300 to 500 μ for 24 hours. During this period, the temperature of the platen was kept at 40 C. When drying was completed, the vacuum was broken with nitrogen. Then the samples were stored under nitrogen at -20 C.

B. Fish Protein Concentrate

Hake used for FPC production were from same catch as those used for freeze drying. Each production run consisted of 20 pounds of whole hake ground through a .25-inch end plate.

The solvent used for each test was fresh 91% (v/v) isopropyl alcohol. It was mixed with fish in a ratio of solvent to fish of 2 to 1. The slurry was stirred briefly and then transferred to extraction unit. After 30 minutes, the mixture was pumped to a basket centrifuge for separation of solids. The second

extraction was made by running hot solvent distillate through the solids in centrifuge basket while centrifugation continued. This extraction was continued for 30 minutes at distillate temperature of 78 C. Centrifugation continued for another 15 minutes. The third extraction consisted of a continuous extraction of solids with isopropyl alcohol. (Described by Brown & Miller, 1969.) The liquid was removed continuously from extractor and evaporated, and the condensate pumped back to the extractor. After final separation of solids, the extracted material was desolventized under vacuum at 40 C. for 16 hours. The dried product was ground through a Wiley mill equipped with a screen having 0.5-mm openings.

Product Analysis

A. Chemical Analysis

Moisture, ash, and crude protein were determined by procedures of Association of Official Agricultural Chemists (1965). Lipids were determined by method of Smith, Ambrose, and Knobl (1964); amino acids by method of Moore, Spackman, and Stein (1964); and available lysine by method of Carpenter (1960).

B. Nutritive Evaluation

Samples of freeze-dried fish were fed in amount desired (ad libitum) to male albino rats (Charles River CD strain), randomly allotted to groups of 8 rats each. The samples were added to a nutritionally adequate basal diet at 10% level of crude protein (Campbell, 1960). The gain in weight and amount of food consumed were recorded each week for 4 weeks; the PER was then calculated.

RESULTS

Freeze-Dried Whole Fish

A. Chemical Analysis

Table 1 shows the proximate composition of freeze-dried whole fish at each sampling period. The freeze-dried raw material from each catch varied in its concentration of lipid and, to a slight extent, in its concentration of crude protein. In general, the concentration of lipid was lowest in October sample--9.8%. In contrast, samples of July, November, and January catches were 14 to 15%. Thus, the

^{1/}Trade names are used merely to simplify descriptions; no endorsement of the products is implied.

Table 1. Proximate composition of freeze-dried whole red hake (*Urophycis chuss*) caught during 1964-65

Date of catch	Crude protein	Lipid	Ash	Moisture
	-----Wt %-----			
<u>1964</u> : July	74.5	13.1	12.5	3.01
October	76.7	9.7	13.1	2.74
November	75.0	14.2	12.7	2.88
<u>1965</u> : January	75.5	13.7	11.4	3.36

Note: The crude protein, lipid, and ash are reported on the basis of dry weight.

concentrations of crude protein and ash in October fish were slightly higher than these concentrations in the other samples.

Table 2 shows amino acid composition and available lysine concentration of freeze-dried samples. A comparison of all amino acids recovered, as percent of total crude protein, revealed that 83% (October) to 92% (January) of protein can be accounted for. Undoubtedly, other components of the nitrogen--such as various amines, ammonia, urea, creatine, taurine, and anserine--could make up this difference because there is a concentration factor from raw wet fish to freeze-dried product.

Thompson and Farragut (1965) reported an observation with whole alewives (*Alosa pseudoharengus*). They postulated that considerable metabolic energy is used when spawning commences, and that excretory processes of fish do not keep pace. So metabolic nitrogen products build up in the body.

In our study, although physiological condition of fish was unknown, the fish caught in October exhibited a slightly higher concentration of total nitrogen (crude protein) and lower concentration of amino acids than did other samples. The concentrations of amino acids did not reveal any marked changes during sampling periods; rather, they reflected changes related to an increase in total recovery as percent of the protein. The values obtained for available lysine fluctuated from period to period and, apparently, did not reflect any trend.

B. Nutritive Evaluation

Table 3 gives the mean total weight gain, food consumed, and protein efficiency ratio values from feeding trials of freeze-dried ground hake. The differences within each catch of fish were not significant, but differences between catches were highly significant ($P < .01$). The freeze-dried fish used in diets prepared from November sampling resulted in lower PERs than did other samples. To check accuracy of these data, we did another experiment (Nov. 1964B). The results confirmed previous test: protein quality was comparable to casein. The PERs obtained with diets made of freeze-dried fish from July, October, and January were better than those obtained with casein.

FISH PROTEIN CONCENTRATE

A. Chemical Analysis

Table 4 shows proximate composition of FPCs. The crude protein ranged between 85.5 and 88.9%. Very little difference was found in protein concentrations of FPCs produced from fish in July, October, and November--but concentration increased in FPC made from January fish.

Values for total residual lipid in FPCs show that removal of lipids by solvent-extraction procedure was not affected by the fish's physiological state. However, the composition of the residual lipids, which was not determined, may reflect differences.

Table 2. Amino acid analysis and available lysine of the standard reference samples: freeze-dried whole red hake (Urophycis chuss)

Amino acid	Data for red hake caught on:			
	July 1964	October 1964	November 1964	January 1965
	-----% of protein-----			
Available lysine	6.84	6.99	6.58	7.35
Total lysine	7.32	7.36	7.35	7.63
Histidine	1.91	1.78	1.91	1.92
Arginine	5.85	5.77	5.89	5.96
Aspartic acid	8.74	8.50	9.07	9.50
Threonine	3.82	3.68	3.98	4.07
Serine	3.86	3.75	4.00	4.08
Glutamic acid	12.67	12.62	13.64	14.05
Proline	4.52	4.09	4.27	4.57
Glycine	7.02	7.13	7.48	7.70
Alanine	6.08	5.67	6.11	6.50
Valine	4.61	4.31	4.59	4.88
Methionine	2.70	2.65	2.91	3.02
Isoleucine	3.86	3.71	4.00	4.21
Leucine	6.42	6.17	6.76	7.07
Tyrosine	2.76	2.62	2.83	3.01
Phenylalanine	3.57	3.31	3.70	3.82
Total	85.71	83.13	88.49	91.99

Table 3. Mean weight gain, food consumed, protein efficiency ratio (PER), and adjusted PER of animals fed freeze-dried ground whole red hake caught during 1964-65

Date of catch	Group	Tests	Rats/test	Weight gained		Food consumed		Protein efficiency ratio ^{1/}	
				Mean	Standard error	Mean	Standard error	Mean	Standard error
1964: July		6	8	159.7	2.5	377	2.0	3.39	0.04
October		6	8	157.8	2.1	411	2.4	3.58	0.04
November	A	2	8	131.8	2.0	419	3.1	2.90	0.03
	B	2	8	145.4	2.9	378	3.7	3.08	0.04
1965: January		1	8	158.6	3.2	390	5.7	3.46	0.05

^{1/} The protein efficiency ratios were adjusted to a protein efficiency ratio of casein equal to 3.00.

Table 4. Proximate composition of FPC produced from whole fish caught during 1964-65

Date of catch	Crude protein	Lipid	Ash
1964: July	86.2	0.4	14.2
October	85.5	0.1	14.7
November	86.1	0.3	14.6
1965: January	88.9	0.2	12.9

Note 1: The data are on the basis of dry weight.

Note 2: Crude protein is calculated on the basis of N x 6.25.

The concentration of ash remained relatively constant (14%) for July, October, and November samples of FPC, but concentration in January sample decreased, which reflected the increase in protein.

To compare proximate composition data of FPC with those of freeze-dried whole fish, we had to place them on a moisture-free and lipid-free basis. When this was done, the values of crude protein in freeze-dried fish showed higher concentration in January than in other months. The same was true of ash concentration.

Table 5 lists results of amino acid analyses and determinations of available lysine concentrations in FPCs produced from raw fish caught during sampling periods. These data

also show differences in percentage recovery, ranging from 96.0 to 102.0% of the protein. Most concentrations of amino acids either increased or decreased according to the recovery. Lysine, histidine, arginine, and proline, however, remained relatively constant. The concentration of available lysine fluctuated from period to period with no apparent trend.

In general, the major difference between amino acid concentration of FPCs, compared with that of freeze-dried samples, is the greater recovery, as percent of protein, from FPC samples.

B. Nutritive Evaluation

Table 6 shows results of feeding FPC diets to laboratory animals.

Table 5. Amino acid analysis and available lysine of FPC's produced from fish caught in different months of the year

Amino acid	Data for FPC produced from fish caught on:			
	July 1964	October 1964	November 1964	January 1965
-----% of protein-----				
Available lysine	8.09	7.87	8.19	7.70
Lysine	8.28	8.23	8.31	8.31
Histidine	2.01	1.90	1.95	1.93
Arginine	6.70	6.78	6.69	6.77
Aspartic acid	9.75	9.67	10.61	10.49
Threonine	4.28	4.21	4.58	4.56
Serine	4.32	4.23	4.69	4.65
Glutamic acid	14.38	14.39	15.83	15.62
Proline	5.10	5.13	5.13	5.18
Glycine	7.52	7.62	8.46	8.72
Alanine	6.49	6.26	6.93	7.56
Valine	5.13	4.74	5.33	5.34
Methionine	3.11	3.22	3.44	3.43
Isoleucine	4.28	4.23	4.68	4.63
Leucine	7.16	7.16	7.88	7.85
Tyrosine	3.10	3.25	3.41	3.24
Phenylalanine	3.86	3.98	4.32	4.21
Total	95.47	95.00	102.24	102.35

Table 6. Mean weight gain, food consumed, protein efficiency ratio (PER), and adjusted PER of animals fed diets of FPC produced from raw fish caught in various periods of the years 1964-65

Date of catch	Group	Tests	Rats/test	Weight gained		Food consumed		Protein efficiency ratio ^{1/}	
				Mean	Standard error	Mean	Standard error	Mean	Standard error
-----g-----									
1964: July		6	8	151.8	2.2	365	3.9	3.34	0.04
October		6	8	139.0	2.0	412	2.9	3.09	0.04
November	A	2	8	130.3	4.0	418	9.2	2.85	0.07
	B	2	8	141.5	3.8	363	6.4	3.12	0.05
1965: January		1	8	154.0	8.6	363	12.0	3.62	0.12

^{1/} The protein efficiency ratios were adjusted to a protein efficiency ratio of casein equal to 3.00.

According to PER values obtained from feeding trials, the nutritive values of protein in FPCs processed from same catch of fish did not differ significantly. But a highly significant difference ($P < .01$) occurred in FPCs made from different catches. The FPCs processed from July and January fish were better than those prepared from October and November fish. Furthermore, the two latter groups were comparable only with casein, whereas the former two FPCs were better than casein. Compared with freeze-dried whole fish, the differences in PER from catch to catch were similar, with exception of October fish. In this instance, the freeze-dried sample had a significantly higher PER than FPC. At this time, we are unable to explain these results.

Chemical analysis revealed little information that could be related to nutritional data.

CONCLUSIONS

The results of tests over 6 months indicated certain aspects of FPC's nutritional value are affected by the raw material. Both freeze-dried samples of fish and FPCs prepared from same fish were found to have PERs comparable with casein, whereas PERs of other samples were superior to casein. Chemical analyses of the samples failed to yield any clues as to cause of this difference.

A longer study would have been useful in providing more detailed information on seasonal changes. However, this study has shown that, with one exception, the quality of FPC is no different from quality of raw material (or, at least, of freeze-dried raw material). Furthermore, it has shown that processing with isopropyl alcohol had no significant effect on nutritive value of raw fish.

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