

Nutritional Evaluation of Fish Solubles

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This paper reports the results of a preliminary study on the biochemical composition and nutritional characteristics of fish solubles from oil sardines (*Sardinella longiceps*) and white tailed pink perch (*Nemipterus japonicus*). The nutritional quality of sardine solubles has been evaluated by feeding trials using albino rats also. The studies have shown that compared to a control group of rats whose diet had casein as the sole source of protein, a group of rats in whose diet dried sardine solubles replaced half of the casein, had a noticeably higher growth rate. This higher growth rate was not prominent in the early stages of growth (4-7 weeks). But, during later stages (7-10 weeks), solubles incorporated diet supported a distinctly higher growth rate. This effect was more pronounced in female rats (17% over the control group) compared to the male rats (4% over the control group). Fish solubles are found to be poor sources of essential amino acids. Thus, the observed increase in growth rate is higher than the rate expected from the amino acid make up. This probably supports the view expressed by workers elsewhere that fish solubles contain some unidentified growth factors.

Fish meal industry in India, is still in its infancy. Higher quality fish meal, made under hygienic condition in sophisticated fish meal plants is a valuable protein supplement in poultry and pig rations. Development of fish meal industry is very essential for ensuring full utilisations of our increasing marine landings. In countries like Norway the fish meal industry handles roughly two thirds of the total fish landed (Sobstad, 1977). No authentic figures for India are available in this respect. Kamasastri & Rao (1962), Madhavan (1969) and Devadasan & Venkataraman (1979) have made some attempts to study the status of fish meal industry in the country. Recent statistics (MPEDA 1983) shows a downward trend in the meagre export of fish meal from India.

The wet reduction process is the usual method adopted for fish meal manufacture in modern plants. In this process the fish is first cooked to facilitate separation of the lipids from the protein matter. The cooked fish is then pressed in special presses. The press liquor contains roughly 20-25% of the proteins, which may under some conditions go as high as 45%. In addition to

protein, the press liquor carry fat and many other valuable nutrients also, especially the B vitamins. But often due to the expenses involved in concentrating the press liquor (after oil separation), this "stick water" is wasted and only the press cake is further dried to make fish meal. This is an avoidable wastage of valuable nutrients. The "fish solubles" in this stick water can be concentrated and added back to the press cake before final drying to produce the so called 'whole meal'. The high nutritive value of the 'solubles' justify the extra expenditure involved in concentration of the stick water. But systematic studies bringing out this high nutritional quality of fish solubles are essential to convince the producer and the consumer.

Oil sardine and white tailed pink perch (*Kilimeen*) are two potential sources for our fish meal industry along the south-west coast of India. Oil sardine forms the main raw material for the few modern fish meal plants in this region. These plants recover the oil from the stick water but discard the stick water without recovering the solubles.

Fish solubles have received a lot of attention in recent years. Joseph Soares *et al.* (1972) have reviewed the work in this field exhaustively. Some unidentified growth factors have also been reported in fish solubles (Mason *et al.* 1961, Mason, 1969) which has created increased interest in the study of their nutritional characteristics. But no systematic study has been reported on the nutritional value of solubles from Indian fishes. This paper reports results of a preliminary investigation on the nutritional quality of the solubles from oil sardine and white tailed pink perch.

Materials and Methods

Solubles from oil sardines and kilimeen were prepared as follows:

Whole fish procured fresh from the local landing centres were washed well, and cooked in steam at 1.05 kg/cm² pressure for 10 min. The stick water oil phase along with some suspended materials was separated from the meal cake in a basket centrifuge. The stick water phase was allowed to stand overnight in a cold room to facilitate separation and removal of the oil phase. The stick water, after separation of the oil, was concentrated initially using super heated steam. After the initial concentration, the "condensed solubles" were dried and powdered by heating under vacuum. The press cake was separately dried and mixed well with the dried solubles in the ratio 3:1 to make the "whole meal." In the case of kilimeen, dressed meat was used for making the meal.

Moisture, protein and fat were determined by standard methods (AOAC, 1975). Ashing was done in a muffle furnace at 550°C and contents of sodium, potassium and calcium in the samples were determined by flame photometry using a Systronics model flame photometer (Type 121-1). Phosphorous was determined by the method of Fiske & Subba Row (1925). Other minerals were estimated by atomic absorption spectrophotometry using a Varian techtron atomic absorption spectrophotometer (model No. 1100). Amino acid analysis was done in a Technicon NC 2p amino acid analyser.

Four weeks old albino rats were used for the feeding trials for nutritional evaluation.

Solubles from oil sardine only were used for feeding trials. The control group of rats were fed on a casein based diet with casein as the sole source of protein. The test group of rats were given a diet in which half the casein was replaced by solubles from sardines. Diets for both groups were identical in all other respects (composition of diets given in Table 3). The final casein based diet had a protein content of 12.5% on dry weight basis. The test diet had a protein content of 12.1% on dry weight basis. The effect of each type of diet on male and female rats was separately studied. The rats were put on the respective diet, in metabolism cages for 8 days for adaptation. Food and water were given *ad libitum*. After the adaptation period, weight gain was noted every 5th day. Nitrogen retention and utilization were studied during two 10 days periods at two stages of growth namely, 36-45 days old stage and 61-70 days old stage by quantitative collection of faeces and urine and by noting the quantity of feed given and residual feed left. The protein consumed, absorbed and utilized were calculated from the data on nitrogen contents of feed, residual feed, faeces and urine. Urine was collected in bottles containing about 5 ml of dilute sulphuric acid to minimise loss of volatile basic nitrogen. The residual feed, faeces and urine collected over the two 10 day periods were separately analysed for nitrogen content by the microkjeldahl procedure.

Results and Discussion

The dry matter contents of stick water from kilimeen and oil sardine were 9.6% and 7%, respectively. Protein contents of the two samples of stick water were 7.2% and 5.5% respectively. In commercial practice the stick water after concentration to a reasonably low water content is added to the press cake before final drying to prepare whole meal. In this study, however we tried to dry the stick water into a dry product under vacuum before mixing with press cake. Addition of solubles to press cake meal brings down the fat level and increases the protein content of the press cake meal. Sardine and kilimeen solubles had 72% and 70% protein respectively with a moisture content of 6% and 2.05%.

Table 1 gives the amino acid composition of the solubles from sardine and kilimeen and also those of the dried press cake and whole meal from sardine. Solubles from both fishes are rich in glycine, alanine and glutamic acid but are generally poor in most of the essential amino acids except lysine. Arginine content is also reasonably high. Kilimeen solubles were found to be poor in cystine, methionine, valine and tyrosine. Most of the taurine in sardine appears in the solubles fraction. According to Monson (1969) taurine can be one of the unidentified growth factors in fish solubles. Sardine solubles are found to be rich in glycine which is an important amino acid in poultry rations.

Table 2. *Content of inorganic elements in fish solubles and fish meal samples*

	Sardine solubles	Sardine meal	Sardine whole meal	Kilimeen (dressed) solubles
Values expressed as g/100 g dry samples				
Sodium	5.06	0.91	2.12	2.73
Potassium	5.29	1.42	2.50	5.90
Calcium	2.19	5.94	4.50	2.70
Phosphorus (Inorganic)	1.10	2.30	2.02	1.32

Table 1. *Amino acid composition of the fish solubles and fish meal samples % amino acid in the acid hydrolysate*

	Fish solubles from oil sardines	Press cake meal from oil sardines	Whole meal (Sardines)	Solubles from dressed Kilimeen
Taurine	2.35	Trace	0.60	—
Aspartic acid	5.30	6.60	6.30	5.58
Threonine	1.90	2.70	2.60	1.87
Serine	5.70	5.60	5.12	3.78
Glutamic acid	12.12	12.53	12.37	15.78
Proline	4.73	4.10	4.27	4.54
Glycine	19.10	10.94	13.00	16.15
Alanine	13.44	10.14	10.90	24.65
Cystine	2.90	4.60	4.20	0.91
Valine	2.10	5.06	4.32	0.80
Methionine	2.19	2.30	2.13	0.25
Isoleucine/leucine	7.30	6.90	7.01	7.60
Tyrosine	1.25	3.30	2.90	0.39
Phenyl alanine	2.17	3.10	2.80	1.58
Histidine	3.02	3.76	3.72	2.12
Lysine	8.00	12.75	11.60	10.30
Arginine	5.92	5.60	5.37	4.70

(Values are average of four batches analysed)

The ash content of the dried solubles ranged from 16–17%. Sardine solubles is found to be a good source of all the minerals (Table 2). It has a copper content of 1.57 mgm%. According to Mason *et al.* (1961) the necessary inorganic constituents for promoting growth include copper and molybdenum.

Values expressed as mg per 100 g dry samples

Magnesium	99.00	220.00	188.00	—
Iron	29.00	88.00	72.00	5.70
Zinc	3.46	17.71	15.06	—
Copper	1.57	1.40	1.44	—
Manganese	0.248	1.70	1.32	—

Table 3. *Composition of the diets given*

	Control %	Test %
Casein	15.0	7.5
Sardine solubles	—	7.5
Starch	40.0	40.0
Sucrose	32.0	32.0
Ground nut oil	5.0	5.0
Salt mixture		
(Hebbel <i>et al.</i> 1937)	4.0	4.0
dl Methionine	0.2	0.2
Vitamin mixture		
(Chapman <i>et al.</i> 1959)	1.0	1.0
Cellulose	2.8	2.8

Solubles are said to be very rich in B vitamins. But in the present study we could not take up the estimation of the vitamins. Table 4 gives the protein intake and weight gain and also the calculated protein efficiency ratio value (PER) of the two diets for male and female rats separately at different periods of growth. Incorporation of fish solubles increased the intake of feed. During

4-7 weeks of age (1-3 weeks in the feeding data) however, fish soluble incorporated diet did not show any increase in PER, compared to the control casein diet. But during subsequent days (7-10 weeks of age i.e. 4-6 weeks in the feeding data) soluble incorporated diet was found to give higher weight gain per g of protein consumed. This trend was seen in males as well as in females (17% increase in females and 4% in males compared to the control diet). Yanase & Arai (1967) also have reported similar observations in their studies using female rats fed on a diet containing solubles from jack mackerel, saury, Alaska pollack etc. as one of the protein sources. They found the growth rate increased by 7.4% over the group on casein based control diet when Alaska pollack solubles replaced part of the casein in diet as protein source. They attributed this to the unidentified growth factor present in the fish solubles. These authors had earlier found that male rats do not respond favourably when casein was replaced isonitrogenously by fish solubles, fish meal etc. However in their studies using female rats they also found a difference in the rats' response to incorporation of fish solubles in the diet at two different stages of growth.

Isonitrogenous replacement of casein by sardine solubles was found to result in rats losing weight. So this experiment had to be abandoned. Hence fish solubles cannot be recommended as the sole source of proteins in the diet on account of its poor content of essential amino acids. When half the casein in the control diet is replaced by

sardine solubles there is a noticeable increase in PER especially towards the later stages of growth. The effect was a little more pronounced in female rats compared to male rats. During the entire period of the experiment the growth rate was improved by 17% for female rats and 4% for male rats by the test diet over the casein diet. Sobstad has reported the study of Laksesswala (Sobstad 1977) that for getting optimum growth, a mixture of 20% solubles with 80% press cake meal is ideal. Bjorn (1973) suggested a mixture of 30% solubles with 70% meal as the ideal combination. In our studies, instead of press cake meal, casein at 50% (w/w) level was mixed with solubles to get good growth.

Table 5 gives the figures for digestibility and nitrogen utilisation for the diet in male and female rats at two different stages of growth.

Apparent digestibility of the proteins of the control casein diet decreased from 90% in the initial stages of growth to 87% in later stages in females and from 89% to 85% in males. In both cases, incorporation of solubles decreased the protein digestibility. But with increasing age, digestibility of the proteins of solubles incorporated diet showed gradual improvement in males as well as females. Newport's (1976) studies using pigs have also shown that replacement of milk protein with fish protein reduces the apparent digestibility. But he has not reported any effect of age on this digestibility values in pigs. Huber & Slade (1967) and Raven

Table 4. Comparison of the protein efficiency ratios of the two diets

Sex and average weight of rats	Diet	Weight gain weeks			Protein intake weeks			PER weeks		
		1-3	4-6	1-6	1-3	4-6	1-6	1-3	4-6	1-6
Female 44 g	Casein	42.8	14.3	57.1	18.4	14.8	33.2	2.33	0.97	1.72 (100)
"	Casein + sardine solubles	64.2	24.0	88.3	26.7	17.3	44.0	2.40	1.39	2.01 (117)
Male 58 g	Casein	59.8	19.3	79.1	24.0	17.7	41.7	2.49	1.09	1.89 (100)
"	Casein + sardine solubles	66.8	35.5	102.3	29.2	22.8	52.0	2.30	1.56	1.97 (104)

Table 5. *Comparison of the digestibility and nitrogen utilization of the two diets*

Sex and average weight of rat	Diet	Apparent digestibility of protein		Protein utilised as % of intake		Protein utilised as % of absorbed	
		Weeks		Weeks		Weeks	
		1-3	4-6	1-3	4-6	1-3	4-6
Female							
44 g	Casein	90	87	83	80	92.8	92.12
"	Casein + sardine solubles	82	84	73	78	88.9	96.40
Male							
58 g	Casein	88	85	82	78	92.43	92.00
"	Casein + sardine solubles	83	87	70	81	84.20	92.90

(1972), however, using calves, found a low digestibility of fish protein at 1-3 weeks of age which improved with age. The results of the present study show that digestive enzyme in growing rats are not able to digest fish solubles protein efficiently. However, after the initial growth period, digestive enzymes appear to become more powerful as is seen from the improved digestibility of fish solubles protein at later stages of growth.

Utilisation of the protein (as percentage of intake) of the control casein diet decreases in male as well as female rats when they grow older. Tagle & Donoso (1965) also found the apparent NPU of casein to be reduced from 49 to 34 in 41 days and 133 days old rats. Replacement of half the casein in diet with fish solubles initially reduced the utilization in growing rats. However with increasing age the utilization of proteins of solubles incorporated diet shows gradual improvement in males as well as females as can be seen from Table 5. A similar trend is seen in the figures for protein utilized as percentage of the absorbed protein also. These figures suggest that the amino acid make up of the solubles incorporated diet does not meet the full requirements in rats in initial stages of growth. However after this initial stage, the amino acid requirements appear to undergo a change as is evident from the improved utilization of the proteins of solubles incorporated diet at this stage. At this age, the requirements are met better by the solubles incorporated diet in males as well as females. Probably at this age the amino acids of fish solubles effectively

supplement those of casein or the requirement for the various essential amino acids may not be that much critical as in the young ones. Reports in the literature regarding the requirement of amino acids with respect to age are conflicting Forbes & Yohe (1955), Hartsook & Mitchell (1956) Henry & Kon (1957).

The study thus shows that sardine solubles are poor in essential amino acids but are rich in amino acids like glycine, alanine, glutamic acid etc. In growing male as well as female rats, utilization of the protein is reduced when solubles partially replace casein in diet. However with increasing age, utilization improved in the case of solubles incorporated diets, probably due to changed amino acid requirements during later periods of growth. The data collected show that female rats utilize the proteins of fish solubles better than the males. However age seems to be the main factor causing variations in the utilization of the protein. The growth rate of rats fed on solubles incorporated diet is much higher than the rate expected from such a protein, deficient in essential amino acids. This probably supports the suggestion that fish solubles contain unidentified growth factors.

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