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Biochemical quality and heavy metal content of fish meal and squid meal produced in Veraval, Gujarat

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ABSTRACT

The present study aimed to compare the biochemical quality including toxic heavy metal content of the fish meal and squid meal produced in commercial units in Veraval, Gujarat. Moisture and crude lipid contents were 10.9% and 4.1% for fish meal where as for squid meal the values were 9.2% and 3.1%, respectively. The crude protein content of both the meals was over 60%. Bulk density and water holding capacity was significantly higher for squid meal compared to fish meal. Significant differences ($p<0.05$) were observed for total volatile bases and lipid oxidation products such as peroxide value and free fatty acids, with higher values for squid meal compared to fish meal. Concentration of toxic heavy metals *viz*., cadmium (Cd), mercury (Hg), lead (Pb), copper (Cu) and nickel (Ni) were detected in both fish and squid meal where as mercury content was not observed in both the samples. Higher cadmium content was observed in squid meal (2.45 to 14.1 ppm) compared to fish meal (0.22 to 4.4 ppm) whereas lead content was higher for fish meal (4.6 to 18.2 ppm) compared to squid meal (0.46 to 1.77 ppm). Nickel content was in the range of 0.91 to 12.6 ppm in fish meal and 1.1 to 5.6 ppm in squid meal.

Keywords: Bulk density, Cadmium, Fish meal, Squid meal, Toxic metals

Introduction

Gujarat state has the longest coastal line of 1600 km with variety of aquatic fauna and ranks second in marine fish production in the country. Gujarat is also one of the leading seafood processing states, with a daily handling capacity of 14,877 t of seafood (Geethalaxmi *et al.,* 2011). Nearly 90% of fish caught is used for processing meant for export purpose, which generates high amount of finfish and cephalopod processing waste. From organised processing industry itself over 1.0 million tof waste is generated (Table 1) (Anon., 2005). The waste generated from fish processing industry poses a great threat to environment if proper measures are not taken for the disposal. Proper utilisation of fish processing waste in the form of fishmeal or squid meal will reduce environmental threat and at the same time can help aquaculture industry with alternative options for feed ingredients. Apart from the waste generated from fish processing industry, a large quantity of bycatch, which is normally called as *kutta* in Gujarat, consisting of juveniles of commercial fishes, small sized prawn, crab, small sized cephalopods and squilla are being utilised for the production of fish meal (Zynudheen *et al.,* 2004).

Table 1. Waste generated during fish and shellfish processing in India calculated from marine landings data (Anon., 2005)

Sources	Production (t)	Waste generated (t)
Fin fish	21,15,402	8,46,160 (40%)
Crustaceans	3,66,488	1,64,919 (45%)
Cephalopods	1,12,762	50,742 (45%)
Total	25,94,652	10,61,821

Fish meal is a ground solid product that is obtained by removing most of the water and some or all of the oil from fish or fish waste (Ruiter, 1995). The fish meal and oil industrieswere started in northern Europe and North America at the beginning of the 19th century. Fish meal is being used as a component of livestock feed for many years. It is popular because of its high nutritional value. It has high levels of essential amino acids such as lysine, which is often deficient in grain products that are the typical base for most animal feeds (Hall, 1992). It has high methionine and cysteine content and has high digestibility as well as biological value, also contains vitamins such as B_{12}, choline, niacin, pantothenic acid and riboflavin, and is a good source of calcium (Ca), copper (Cu), iron (Fe), phosphorous (P) and other trace minerals. Fish meal is also low in fibre and easy to produce (Keller 1990; Hall 1992).

Metal pollution in an area depends mainly on the geochemical composition of the coast as well as the anthropogenic activities in the sea. Information on bioaccumulation of heavy metals in marine organisms from open sea is not so common as is from coastal areas (Barber *et al.,*1972; Kureshy*et al.,* 1981; Prema *et al.,* 2006). Ichihashi *et al.* (2001) reported Cd, Pb, Ni and Cu in the liver of flying squid at levels of 106, 0.89, 1.5 and 920 ppm respectively. Bustamante *et al.* (2006) extensively studied the metal concentration in digestive glands of cuttlefish *Sepia officinalis* and Cd, Pb and Cu were 13.6, 2.2 and 362 ppm respectively. Lakshmanan (2003) observed trace metals in fish and fishery products and reported mercury values ranging between 0.02-4.2 ppm, cadmium between 0.32-18.6 ppm and lead between 0.27-19.9 ppm. Heavy metal concentrations are often determined by variables such as water contamination, mining activity and effluent treatment activities in the fishing region (Kulkarni, 2005). Many heavy metal ions like Hg^{2+}, $Pb2^{+}$, Cd^{2+}, Cu^{2+}, Ag^{+}, Au^{3+}, Pt^{4+}, and Ti^{+} are of concern because they are highly toxic and are present in a variety of waste streams that can potentially contaminate the environment. Therefore safe, efficient, and cost effective separation and prevention of these toxic metals are of great concern in modern day environmental scenario (De Rore *et al*., 1994). As the wastes generated from fish processing industry consists mainly of digestive parts, liver, kidney and skin of fish and cephalopods which are often contaminated with high levels of toxic heavy metals, the fish meal or squidmeal produced is likely to be contaminated with these heavy metals. Very little or no reported information is available regarding heavy metal contaminants of fish meal or squid meal. Hence the present study was undertaken with the objectives of studying heavy metal contamination as well as other biochemical and physical quality attributes of fish meal and squid meal.

Materials and methods

Fish meal and squid meal samples were collected from commercial establishments from Veraval, Gujarat during 2008 to 2010. Methodology used for the preparation of fish meal and squid meal is shown in Fig. 1. A total of 39 fish meal samples and 31 squid meal samples were analysed for trace metals and heavy metal content, and six samples were used for analysing biochemical and physical quality. The samples were collected in clean polythene bags and various trace metals like cadmium, nickel, lead, copper and mercury along with biochemical composition and chemical as well as physical quality parameters were analysed.

Processing waste
(Fish head, skin, gut, fins, gills, liver, kidney and scale for fish meal and skin, viscera, wings, cuttlebones, liver for squid meal) transported in HDPE boxes to meal processing unit

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Washing with potable water and mechanical chopping

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Cooking in stainless steel vessel at 150 ^{0}C for 3 h

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Pressing to separate press cake and press liquor

↓

Sun drying of press cake for 36 h

↓

Oven drying to adjust final moisture at 60 ^{0}C for 3 h

↓

Sorting

↓

Pulverising

↓

Packing in 50 kg polybags

Fig 1. Flow chart of the methodology for fish meal and squid meal preparation

Trace metal content analysis

Fish meal and squid meal samples were digested in teflon containers using a microwave digester (Ethos plus High Performance Microwave Labstation, Milestone, USA). Three grams of dried powdered sample was weighed in to 100 ml teflon vials and digested overnight with 7ml of pure nitric acid (AR grade, specific gravity: 1.38, Qualigens, India) and 3 ml of hydrogen peroxide. The microwave parameters were 700 W power for 1 h (40 min heating and 20 min ventilation). The digested contents were transferred to acid washed polypropylene bottles and made up to 25 ml with double distilled water and subjected to various metal analyses in Atomic Absorption Spectrophotometer (GBC 932AA, GBC Scientific Instruments, Australia) following the AOAC method (AOAC, 2000). Proximate composition of fish meal and squid meal were analysed as per AOAC (1990). Mercury (Hg) content was estimated using mercury analyser (MA5840, Electronic Corporation of India, Hyderabad) which works on the principle that mercury vapour (atoms) absorbs resonance radiation at 253.7 nm. Working conditions used for analysis of trace and heavy metals is

Table 2. Working condition for analysis by atomic absorption spectrophotometer

Heavy metals	Wave length (nm)	Slit width (nm)	Flame type (Support-Gas)	Sensitivity (μg g^{-1})
Cadmium	228.8	0.5	Air-Acetylene	0.009
Nickel	232.0	0.2	Air-Acetylene	0.04
Lead	217.0	1.0	Air-Acetylene	0.06
Copper	324.7	0.5	Air-Acetylene	0.025
Mercury	253.7	0.5	-	0.16

given in Table 2. Total volatile base nitrogen (TVBN) was determined by the Conway micro diffusion method (Conway, 1947). Peroxide value (PV) was determined iodometrically (AOAC, 1990). FFA was estimated as per AOCS (1990). Physical properties like water holding capacity (WHC) and bulk density were estimated as per the method of Arivawansa (2000).

Statistical analyses

Minimum of triplicate samples were used for each analysis. The results are expressed as mean ± standard error. One way ANOVA was carried out for trace metals in fish meal and squid meal samples and the significance was tested at 95% confidence level. All the statistical analyses were carried out using SPSS software (Version 10.00) (SPSS Inc., USA).

Results and discussion

Biochemical composition and biochemical quality

Biochemical composition and quality attributes of fish meal and squid meal are given in Table 3. Both squid meal and fish meal had good protein content of over 63%, indicating a very high quality protein supplement. However, the protein content of the fish meal and squid meal was observed to be less than the protein content of blue whiting, herring and capelin (Ariyawanza, 2000). The moisture content was around 10% in both squid meal and fish meal indicating the need for further increase in drying time in the production practices. The crude fat content was slightly higher (3.15 and 4.11% for squid meal and fish meal respectively) as the oil was not completely recovered in the process of fish meal or squid meal production. The ash content of both the meals were observed to be higher which could be due to contamination of meal with sand while drying as the fish meal and squid meal is dried under sun on mats placed on floor. The total volatile base nitrogen (TVB-N) content of squid meal was significantly higher ($p<0.05$) compared to fish meal which could be attributed to the inherent characteristics of squid. The fat oxidation products like peroxide value was observed higher for squid meal than fish meal. Hydrolytic rancidity as indicated by free fatty acid value was also higher for squid meal than fish meal.

Physical quality attributes of fish meal and squid meal

The physical quality parameters, particularly water holding capacity (WHC) and bulk density are very important attributes as they decide the floating ability of the pellets produced. Both fish meal and squid meal showed good WHC and bulk density with relatively higher values for squid meal (180.5% and 0.725 g ml^{-1}, respectively), compared to fish meal (140.24% and 0.6 g ml^{-1}, respectively) (Table 3). The WHC obtained for squid meal in the present study was higher compared to blue whiting, herring and capelin whereas that for fish meal it was slightly lower than the fish meal prepared from these fishes (Ariyawanza, 2000).

Table 3. Nutritional and biochemical composition of fish meal and squid meal (values are mean ± std. error, n=6)

Attributes	Fish meal	Squid meal
Moisture (%)	10.93 ± 0.89	9.21 ± 0.91
Crude protein (%)	63.88 ± 1.42	66.88 ± 2.13
Crude fat (%)	4.11 ± 0.28	3.15 ± 0.17
Total ash (%)	21.69 ± 1.19	20.96 ± 1.03
TVBN (mg N_2 100 g^{-1})	195.66 ± 2.17	414.86 ± 3.27
PV (meq kg^{-1})	11.31 ± 0.81	14.11 ± 0.71
FFA (% as oleic acid)	25.76 ± 0.92	38.08 ± 1.13
WHC (%)	140.24 ± 4.77	180.51 ± 2.98
Bulk density (g ml^{-1})	0.602 ± 0.04	0.725 ± 0.03

Trace metal content in fish meal and squid meal

Table 4 illustrates that the mean values of all the heavy metals in fish meal and squid meal are significantly different ($p<0.05$). The results indicated that the heavy metals, lead followed by copper and nickel predominated in fish meal whereas in squid meal, copper followed by cadmium formed the major share of heavy metals. Mercury content was absent in all the samples studied. The mean values for lead, nickel, copper and cadmium were 10.04, 5.73, 7.31 and 1.39 ppm for fish meal compared to 0.97, 2.58, 50.76 and 9.04 ppm in squid meal. One way ANOVA Indicated that in fish meal, lead and nickel were significantly higher ($p< 0.05$) compared to squid meal. Cadmium and copper were significantly higher ($p< 0.05$) in squid meal compared to fish meal samples. The principal pollutant transformation processes, microbial degradation or biotransformation may

Table 4. Trace and heavy metal content (ppm) of fish meal and squid meal (values are mean ± std. error, n=39 for fish meal and 31 for squid meal)

Heavy metals	Fish meal		Squid meal	
	Range	Mean values	Range	Mean values
Cadmium	0.22 - 4.40	1.39 ± 0.13*	2.54 - 14.11	9.04 ± 0.54*
Nickel	0.91 - 12.58	5.73 ± 0.54*	1.11 - 5.61	2.58 ± 0.21*
Lead	4.60 - 18.20	10.04 ± 0.43*	0.46 - 1.77	0.97 ± 0.006*
Copper	1.80 - 46.40	7.31 ± 1.30*	28.55 - 67.55	50.76 ± 2.01*
Mercury	ND	ND	ND	ND

*mean values are significant (p< 0.05) , ND = not detected

be the most efficient way for removal of chemical pollutants and their toxicity from the environment. Prasad *et al. (*2007) reported that heavy metals like Cd and Pb were 0.35-1.7 ppm, 2.25-5.28 ppm respectively in fish muscle of *Nemipterus japonicus* which was most commonly used for surimi production in India and wastes generated from this process are widely used for the production of fish meal. Heavy metals in squid and cuttle fish obtained from Mumbai coast and Gujarat coast from *FORV Sagar Sampada* had higher levels of Cd (ranged between 12.06 - 17.7 ppm) and other toxic metals (Lakshmanan, 2003). Murthy *et al.* (2008) reported that Cd was present in squid species in all the edible and non-edible parts and the values were above the maximum permissible limit of 1 ppm for export to the EU countries. The highest mean Cd concentration of 435.22 ppm was found in the liver, which ranged from 110.42 to 900.76 ppm. Hence, the utilisation of the pelagic cephalopod purpleback flying squid *Sthenoteuthis oualaniensis* landed along Gujarat coast for human consumption needs a cautious approach. High concentrations of heavy metals in the liver (also known as digestive gland, hepatopancreas or mid-gut gland) of ommastrephid squids have been reported earlier (Martin and Flegal, 1975; Smith *et al.,*1984). Murthy *et al.* (2009) also reported that higher Cd levels were encountered in commercial samples of cephalopods, which could be due to heavy industrial discharge along Gujarat coast and also current practice of soaking of cephalopods in chilled water; which could lead to elevation of cadmium levels due to rupture of internal organs. Irrespective of the species concerned, levels of Cu, Zn, Cd, Pb and Ni were more in liver followed by gills and muscle (Latha *et al.,* 2003). The higher levels of heavy metals observed in the present study could be attributed to the fact that the internal organs which forms the major heavy metal accumulating areas are also used in the preparation of fish/squid meal. The levels of cadmium content of squid meal was significantly ($p<0.05$) higher compared to fishmeal. The allowable limit of cadmium for aquatic feed is <3ppm as per EU norms. In fish meal, only 2 out of 39 samples were above the acceptable limits *i.e.*, >3 ppm, whereas in squid meal all the samples exceeded this limit. This might be because trace metals accumulate maximum in the organs such as liver/hepatopancreas, skin and gut particularly in cephalopods (Murthy *et al.,* 2008; 2009).

The results of the present study indicate that the fish meal and squid meal produced from Gujarat region has very good protein content of over 60% and less moisture content. Water holding capacity and bulk density of both fish meal and squid meal were observed to be higher which is beneficial for pellet production. The trace metals, particularly cadmium content were noticed higher for squid meal compared to fish meal. Care should be taken to reduce this trace metal contamination using quality raw material and by adopting Hazard Analysis Critical Control Point (HACCP) and Good Manufacturing Practices (GMP) in commercial production of fish and squid meal.

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