Chapter 9 Utilization of Fish Waste for the Making of Fish Sauce

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	Increase the Value-Added Fish Production

1 Introduction

The agricultural production sector of developing countries has traditionally been given considerable attention by planners and policymakers, with relatively little focus on the development of agro-industry. Over the years, widespread failure of the survival of medium- to large-scale food-processing enterprises in developing countries has led to growing recognition of the need to foster the development of small-scale food industry. The role of the small-scale food processing as a subsector that can contribute significantly to the development of the rural economy is increasingly being realized (Dietz 1999).

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Biocatalyst Production Technology Division, Center for Bioindustrial Technology, Agency for the Assessment and Application of Technology, BPPT building 2, 15th floor, Jl. MH. Thamrin 8, Jakarta 10340, Indonesia e-mail: siswa59@yahoo.com Fishery is one of the agricultural sectors and can be regarded as a great and valuable source of proteins for both animal and human nutrition. Fish meal, fish sauce, surimi, and fish silage are traditional protein-based products. The FAO State of World Fisheries and Aquaculture report 2008 states that the aquaculture sector comprised about 3.06 million tons (or 56.0 %) of the world fishmeal production and 0.78 million tons (or 87.0 %) of the total fish oil production in 2006. Thus, the total amount of fishmeal and fish oil used in aquafeeds is estimated to have grown more than threefold between 1992 and 2006, from 0.96 million tons to 3.06 million tons and from 0.23 million tons to 0.78 million tons, respectively. Aquafeed manufacturers are increasing their use of fishmeal and fish oil at the expense of all other sectors (e.g., human consumption, industrial, and pharmaceutical).

Globally, the demand for and use of fishmeal has increased rapidly, especially in some of the emerging aquaculture countries in Asia. China is the single largest user of fishmeal. In 2004, it used 1.6 million tons, with 1.2 million tons being imported and the remainder coming from domestic production. Of this total amount, about 75 % was used for aquaculture production. The Asia-Pacific aquaculture sector uses about 2.4 million tons of fishmeal (equivalent to about 10.3 million tons of raw material) as a feed source. (GAFTA 2012). Fishmeal production in IFFO countries increased by 40 % during 2011 as catches in South America returned to normal levels. Northern European production fell as a higher percentage of catches was taken up by direct human consumption. Fresh fish prices in the local market have increased recently due to low supplies, particularly of fresh fish imported from Indonesia. The recent declining catches, escalating operating costs, and growing demand in the country of origin have reduced fish imports from neighboring Sumatra, Indonesia. Malaysia imported around 71 million tons of fresh fish last year, mainly from Thailand and Indonesia, a decline by almost 50 % compared with the amount imported in 2010 (IFFO 2011).

Over 6 million tons of fishmeal is produced worldwide each year from about 25–30 million tons of industrial fish. The demand is increasing with the growth in aquaculture and the price has been rising (Klickhardt 2006). Fish sauce is produced in a quantity of about 0.4 million tons each year (Dissaraphong et al. 2006). Fish silage is almost entirely used for feed. Norway is the major fish silage producer, producing about 0.14 million tons per year, mainly from aquaculture by-products (Rustad 2003).

There are promising opportunities in the upgrading of marine by-products and underutilized fish by using mild processing techniques to convert them into protein and peptide ingredients both to improve yield in traditional seafood and to be used as nutraceuticals or in functional foods.

2 Increase the Value-Added Fish Production

About 76 % of the global fish supply was used for human consumption in 2002 (FAO 2004). About 40 million tons were used for manufacturing products for direct human consumption. But up to 50–70 % of the fish may end up as co-products, as

the yield in filleting operations ranges from 30 to 50 % (Arason 2003; Kristinsson et al. 2006; Mackie 1982). About 6 million tons of trimmings and by-products from food fish processing are processed into fish meal (Fish meal Information Network 2007) and the rest is used in fish silage or as a fertilizer or discarded.

Great additional economic, nutritional, and environmental values can obtained by increasing the yield of raw material in fish filleting operations and in utilizing fish waste for producing by-product as fish sauce.

The meat, poultry, and fish industries in the USA and other parts of the world comprise up to 12 % of brine to modify both fresh retail and further processed products. This is done to improve quality, firmness, and juiciness, and to meet increasing consumers' demand for convenience food items while at the same time increasing yield. The brine is made up of water, salt, phosphates, and sometimes also other functional or flavor ingredients, like sodium lactate, polysaccharide gums, hydrolyzed whey, and soy proteins and modified starches (Xiong 2005).

Technological innovations in fish product development by utilizing fish waste need to be done in order to provide added value to small industries. A prospect which is very promising is the manufacture of fish sauce with high protein content, so that it can be used for the food industry.

3 Type of Fermented Fish

Fermented fish is a fish curing process done in traditional or modern fashions. In the traditional method, processing is done with salt and is left for some time, while in the modern approach, biopreservation, lactic acid bacteria is added to the fish in order for it to be fermented. This produces active antimicrobials such as lactic and acetic acid, hydrogen peroxide, and peptide bacteriocins. It can also produce the antimicrobial nisin, a particularly effective preservative.

In Indonesia, there are many different types of fermented fish, such as: (1) dried fish (jambal fermented fish, peda, ikan tungkai); (2) moist fish (bekasam, naniura, and picungan); (3) lumped pounded fish/shrimp (terasi); (4) semi liquid (kecap ikan, bekasang, and cincaluk).

3.1 Dried Fish

Peda is processed using mackerel (*Rastrelliger neglectus*) as the raw material. The basic method of peda processing is a salting process with two steps. The first salting step takes several weeks to develop its characteristic flavor and texture, and this is followed by a maturation phase. Salting is carried out using a 1:3 salt to fish ratio. Fish and salt are arranged in layers alternately in a salting tank. By the end of the process, the fish are soaked in saturated brine pickle, with coarse salt remaining at the bottom and the top covering the fish. This salting process normally takes 3 days. Maturation is performed by mixing the fish and salt thoroughly. The amount of salt

used is a one-third of the fish weight. During maturation, the fish are kept in bamboo baskets, with the process taking around 1-2 weeks (Rahayu 1992). Better products can be obtained by using eviscerated fish (Hanafiah 1987) and without salt addition in the second fermentation (Hanafiah 1987; Irianto 1990; Irianto and Brooks 1994). Microbiological studies on peda conducted by Hanafiah (1987) revealed that Grampositive cocci predominated and some were identified as lactic acid bacteria. The isolation of halotolerant bacteria from peda by Suwandi (1988) concluded that bacterial growth in peda was characterized as Gram-positive cocci, which were nonmotile, aerobes or facultative aerobes, catalase-positive, non-indole producers, and oxidase-negative. They can utilize citrate as the only carbon source, ferment glucose, and show proteolytic activity. Some of them are able to reduce nitrate. These bacteria can be classified as mesophiles and require a pH of 6–8. They show variations in salt tolerance and can be divided on the basis of salt tolerance as weak, moderate, and halotolerant bacteria. Idawati (1996) showed that lactic acid bacteria isolated from peda were homofermentative and heterofermentative Lactobacillus sp., Leuconostoc sp., and Streptococcus sp.

Jambal roti is processed from marine catfish (*Arius thalassinus*). In the processing, after being beheaded and eviscerated, the fish were washed and soaked in fresh water for 24 h and drained. The fish are arranged in a basin. Each layer of fish is sprinkled with salt and the bellies are also filled with salt. The total amount of salt used for salting is 30–35 % of the fish weight and the salting process takes around 24 h. After salting, the fish are freed from excessive salt and washed. Clean fish are soaked in fresh water for 15–20 min and subsequently split into a butterfly form. The split fish is sun dried for 3–5 days.

Ikan tukai is a traditional fermented fish product which is mostly processed from barracuda (*Sphyranea* sp.). The traditional processing method of ikan tukai is unique. Barracuda is washed and soaked in 20 % brine for around 2 h. After draining, the fish are dried for a day. Dried fish are then wrapped with taro leaves. The wrapped fish are kept underground for 2 days to allow fermentation and then sun dried until dry.

3.2 Moist Fish

Bekasam is a fermented fish product processed from freshwater fish by mixing with chilli and sugar. In bekasam processing, in spite of the salt addition, carbohydrate sources are also incorporated to stimulate the growth of lactic acid bacteria by decomposing into simpler compounds. Sources of carbohydrate used are cooked rice, roasted rice, and sticky rice.

A similar product is called naniura. In naniura processing, fish are firstly soaked in lemon juice or 25 % acetic acid solution for 3 h. After that, ground boiled rice is added and the fish are then packed and allowed to ferment. Another product similar to bekasam is picungan. This product is processed using marine fish. Picungan seeds are used as a carbohydrate source. The seeds are cut into small pieces before mixing with fish and salt.

3.3 Lumped Pounded Fish

Terasi is consumed in small quantities as a flavor. The product is not only for local consumption, but is also exported, mainly to the Netherlands and Suriname, in powder form. Terasi is usually made from planktonic shrimp "rebon" (*Atyas* sp. or *Mycis* sp.) (Budhyatni et al. 1982) or mixing waste shrimp shell and fish. Terasi can be processed in two ways: (1) with salt and (2) with mixing salt and ingredients.

In the processing of terasi with salt, firstly, rebon is washed, drained, and dried, until half dried. During drying, impurities such as small fish, mussel shells, and coral are removed. After that, semi dried rebon is sifted to separate sand and other undesirable materials. The rebon is then left overnight at ambient temperature and pounded the next day. During the first pounding, salt is added (around half of the total salt required during processing). The total amount of salt used in terasi processing is 2–5 % of the dry weight of rebon, which should be added as solution. Pounded rebon is sun dried and subsequently kept in a container at ambient temperature for 2–3 days. The stored rebon is then pounded for a second time, while the remaining salt is added. After that, the pounded rebon is sun dried and kept at ambient temperature for 2–3 days until soft. It is then ground by many passes through a meat grinder until it is fine. Fine rebon is formed in cubes or cylinders of 1 kg in weight and subsequently fermented for a week or more at ambient temperature (Yunizal 1998).

The other processing method of terasi with mixing salt and ingredients is similar to the processing method of terasi with salt. The difference is in the second pounding, where the salt solution is mixed with 2.5 % (w/w) coconut sugar and 2 % (w/w) tamarind prior to be added to fresh rebon. The additional ingredients accelerate the fermentation process (Yunizal 1998).

Microorganisms in terasi powder during ambient storage are those of *Lactobacillus* sp., *Staphylococcus* sp., *Bacillus* sp., and *Proteus* sp., but *Salmonella* sp., *Clostridium* sp., *Vibrio* sp., and *E. coli* are not found (Budhyatni et al. 1982).

3.4 Semi Liquid

Bekasang is a traditional product processed from the viscera of skipjack (*Katsuwonus pelamis*) and waste from cakalang fufu (smoked skipjack). The processing method of bekasang is as follows. Skipjack viscera obtained from processed cakalang fufu is washed and mixed with salt at a ratio of 2.5:1. The mixture of viscera and salt is kept for a week to allow fermentation. After fermentation ceases, the fermented viscera are boiled for 2 h and filtered using a gauze.

Cincaluk is the other traditional fermented fresh shrimp mixed with boiled rice and salt in a pan. For 1 kg of shrimp, the rice added is around 200–300 g, while the salt quantity is approximately 300 g. The pan is then sealed with the lid to avoid air entry and kept for 4 days until liquid release. After that, the mixture is put into bottles and eventually sealed tightly. Another method to process cincaluk is by mixing fresh shrimp with a ratio of 20:1:1 for tapioca flour, salt, and sugar. In its processing, shrimp are descaled and then washed. Tapioca flour is dissolved in water, gelatinized, and allowed to cool. Shrimp are then mixed thoroughly with salt, sugar, and gelatinized tapioca flour. The mixture is placed into washed bottles and sealed firmly and fermented at ambient temperature for 1-2 weeks.

Fish sauce is processed traditionally from oil sardine (*Sardinella lemuru*) by fermentation using high salt concentration.

4 Fish Sauce Processing

Although fermented fish sauce itself may not be directly used for a physiological functional food because of its high concentration of sodium chloride, the sauce may be useful as a source of biologically active substances, traditional food supplements in the diet, and are widely used around the world as condiments, as flavoring, material, and sometimes as a substitute for soy bean sauce (Watanabe et al. 2004). It was concluded that summer capelin may be successfully utilized for the production of fish sauce without added enzymes. Capelin harvested during the summer season was more suitable as a raw material than capelin harvested during the winter season, due to its higher proteolytic activity. Reduced salt content in processing may help increase the fermentation rate as well as improve nutritional properties by reducing the sodium content (Hjalmarsson et al. 2007). Many fermented fish products are prepared in different parts of the world and the method of processing depends upon various factors; availability of raw materials, consumers' preferences, and the climatic conditions of the region. In addition, fish sauce is a product that can be made cheaply from various fish raw materials, which are not normally used for food. Also, it is a clear brown liquid hydrolysate from salted fish (Amano 1962; Beddows 1985), and is a liquid seasoning commonly used in most parts of Southeast Asia. Fish sauce is called by different names, such as patis in the Philippines, shottsuru in Japan, budu in Malaysia, nam pla in Thailand, nước mắm in Vietnam, kecap ikan in Indonesia, yuilu in China, and ngapi in Burma, and it contains a mixture of amino acids and other protein degradation products. (Fukami et al. 2002).

Different countries have different recipes for making fish sauce. Nam pla (Thai fish sauce), which is the most dominant in the world market, is mainly produced from anchovies (*Stolephorus* sp.), mackerel (*Rastrelliger* sp.), and herring (*Clupea* sp.) (Wilaipan 1990).

4.1 Traditional Process

There are two major ingredients in fish sauce production, fish and salt. Traditionally, nam pla is produced by mixing one part salt with two or three parts fish and fermenting under static atmospheric conditions in an underground concrete tank at ambient temperature (30–40 °C) for up to 18 months. The process of salting the traditional fish processing will result in the loss of fish protein by 5 %, depending on the levels

of salt and the salting time. It is recommended that the added salt does not exceed 40 parts by weight of the fish. The supernatant from the fermentation tank is filtered and ripened under the sun for 2–4 weeks (Wilaipan 1990).

The other process can use marine and freshwater fish as a raw material for fish sauce production. Fish and sea salt are mixed at a weight ratio of approximately 2:1 for marine fish and 3:1 for freshwater fish. Traditionally, whole fish is dumped on a concrete floor, and the excess liquid is allowed to drain off. Fish are thoroughly mixed with salt. The mixture is then placed into a tank. Another layer of salt is spread on top. After 1 week, the fish may float and rise to the top of the fermentation tank. It is necessary to keep the fish immersed at all times, otherwise they will become rancid. While the salting progresses, hydrolysis of fish tissues by fish gut enzymes provides the necessary nutrients for bacterial fermentation to begin. The time needed for the full flavor of fish sauce to develop varies from 8 to 18 months at 30–35 °C (Owens and Mendoza 1985; Lopetcharat et al. 2001; Fukami et al. 2004). The finished product is a clear dark-brown color, with pH 5.5–5.8 and 20.5–27.4 % salt, and has a distinct aroma and flavor. The flavor and aroma determine its quality and these characteristics develop progressively as the fermentation process advances (Lopetcharat et al. 2001).

The ratio between salt and fish is very different, depending on the country, ranging from 1:6 to 1:2 (w/w). In Indonesia, the procedure of fish sauce processing is as follows. Whole fish were washed with 2 % saline solution and then chopped. The minced fish is inserted into a barrel and mixed with 5 % salt. Whole fish were washed with 2 % saline solution and then chopped up and put into the tank and mixed with 5 % salt. 60 % (v/v) of acetic acid was added to the mixture of salt and fish to pH 3–4. The addition of vinegar aims to reach medium acidic conditions (pH 3–4). Highly acidic conditions will increase the activity of the enzyme protease, which would break down proteins in the fish tissue.

The time required to make fish sauce depends on several factors, such as the type of acid. From various studies, the time required ranged from 4 to 10 weeks. The addition of protease enzymes from the outside can reduce the time required. According to Gilberg et al. (1984), the addition of the enzyme pepsin in the manufacture of fish sauce can reduce the processing time to get good quality fish sauce to just 3–6 h.

Microorganisms generally increase during the early fermentation stage and then decrease gradually as the fermentation time is extended (Saisithi et al. 1966; Ijong and Ohta 1995). Beddows et al. (1980) reported that budu (Malaysian fish sauce) produced in the presence of rifampicin does not have the unique aroma of budu. Microorganisms such as *Bacillus* and *Staphylococcus*, which were isolated from nam pla, kecap ikan or bakasang (Indonesian fish sauce), and patis (Philippine fish sauce), produced a significant amount of volatile acids (Saisithi et al. 1966; Ijong and Ohta 1995).

Fermentation takes place in tanks at temperatures slightly above room temperature for 1–5 days. After fermentation completed, filtering for solids separation was conducted. During the process of fermentation, competition among microorganisms occurs, with bacteria and microorganisms that survive participating in the process of fermentation. High amounts of salt causes a long fermentation time and the resulting soy products have a very salty taste. To overcome these problems various improvements have been made that is utilize proteolytic enzymes. The enzyme can work optimally with reducing concentration of salt and optimal fermentation conditions (Yunizal et al. 1982).

The liquid fish sauce can be added with sodium carbonate for a pH of 6–7 and be boiled and seasoned as needed. The product can also be add with a thickening agent such as carboxymethyl cellulose (CMC) before being packaged into bottles and becoming ready for the market.

4.2 Enzymatic Process

Making traditional fish sauce requires a relatively long time. Microorganisms producing protease enzyme adaptation takes a long time to be able to live in high salinity environments and other abnormal conditions.

During the fermentation process, the fish tissue undergoes hydrolysis by enzymes produced by microorganisms. The role of these enzymes is bond breaker polypeptides, to become simpler bonds. Microorganisms that develop during the fermentation of fish are not fully known. Nevertheless, estimated species of lactic acid bacteria such as *Laucosotic mesenterides*, *Pediococccus cerevisiae*, and *Lactobacillus plantarum* are growing. Some types of yeast are also thought to have evolved in the fermentation.

The addition of proteolytic enzymes before fermentation can shorten the fermentation time of fish sauce processing. In this case, it is no longer necessary to time the adaptation of microorganisms to produce enzymes that can hydrolyze proteins.

The high price of pure proteolytic enzymes becomes a constraint to produce fish sauce quickly, easily, and inexpensively. However, by utilizing the sap of papaya and pineapple extracts, the role of pure proteolytic enzymes can be replaced.

Papaya sap contained in the proteolytic enzyme is called papain. Papain has a high activity for hydrolyzing proteins. In the food industry, papain is used for retaining the freshness of beer, softening meat, and removing proteins in food, while in pineapples, especially young pineapples, bromelain is another proteolytic enzyme. Its ability to hydrolyze proteins is not dissimilar to papain.

However, fish sauce produced enzymatically has scent and color that are very different to the fish sauce made traditionally, although it is not very different in terms of nutritional content.

5 Quality of Fish Sauce

It is high in protein (as much as 10 %), and this protein is a complete one containing all the essential amino acids that the body requires for growth and regeneration. Top-quality fish sauce also contains a rich supply of B vitamins, particularly B 12,

Table 9.1 Nutrient contentbased on the quality of fishsauce (g/100 g)	Component	Quality I	Quality II
	Salt	28.4	28.4
	Protein	0.92	1.90
	Amino nitrogen	5.75	5.58
	Nitrogen organic	0.62	1.13

Table 9.2 Differences in the nutritional values of fish sauce on the different types of fish used

	Proximate, (%)					
Kind of fish used	Protein	Lipid	Carbohydrate	Water	Salt	
Petek (Leiognathus sp.)	5.57	0.84	0.46	88.07	5.04	
Kuniran (Upeneus sulphureus)	5.40	0.77	0.24	87.54	6.04	

pantothenic acid, riboflavin, and niacin. Other beneficial nutrients include calcium, phosphorous, iodine, and iron (Table 9.1).

According to Cole et al. (1985), good fish sauce is considered to have a protein content of 2.3 % and a pH below 6, but standard fish sauce in every country is different.

Whether fresh water or salt water, fish for making fish sauce are usually small ones that otherwise would have little value for consumption. Among the most common marine fish used are anchovies and a few related species of schooling fish, from 2 to 5 in. in length, found in plentiful supply in the rich gulf waters. Larger varieties of fish, such as mackerel and sardines, also make good fish sauce, but because they are relatively more expensive due to their value as a food fish, they are seldom used in the commercial production of fish sauce. For fish sauce to develop a pleasant, fragrant aroma and taste, the fish must be very fresh. The different types of fish used showed differences in nutritional value (Table 9.2).

The protein content of fish sauce from Petek (*Leiognathus* sp.) is higher than the protein content of fish sauce from kuniran (*Upeneus sulphureus*), which is caused by the different salt contents. Salt is known to inhibit the activity of enzymes in the process of breakthrough of the tissue into soluble protein. Some researchers reported that, the tissues of fish, especially the internal organs, contained a variety of enzymes. Some researchers reported that protease enzymes in the body tissues of fish are most active in acidic conditions (pH 3–4), while others are most active in base conditions (pH 7–8). Meanwhile, Cole et al. (1985) reported that endopeptidase enzymes and lipase are active at salt levels up to 15 %, and will decrease in activity if the salinity is increased. The results of the amino acid analysis of fish sauce protein shows there are 19 different amino acids, with the highest content being glutamic acid, amounting to 20.57 % of the total protein (Table 9.3). The high content of glutamic acid in the protein composition showed that fish sauce can use as a flavor enhancer.

During the process of making fish sauce, other flavor ingredients can be added. Thus, the created fish sauce can be adjusted to suit various tastes and it will increase the value of this product.

Table 9.3	Amino acids
compositio	on in fish sauce

No	Amino acids	% of total protein
1	Glycine	3.80
2	Alanine	1.10
3	Valine	6.24
4	Leucine	11.09
5	Isoleucine	5.33
6	Proline	9.78
7	Phenylalanine	5.51
8	Tyrosine	4.83
9	Tryptophan	0.60
10	Serine	4.58
11	Threonine	3.02
12	Methionine	2.75
13	Arginine	2.70
14	Histidine	6.42
15	Lysin	5.22
16	Aspartic acid	4.70
17	Glutamine	0.94
18	Glutamic acid	20.57
19	Asparaginic acid	0.55
-		

Isnawan et al. (2001)

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