



Utilization of *Gambusia (Affinis affinis)* For Fish Sauce Production

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Abstract

Fish sauce made from *Gambusia (Affinis affinis)* is evaluated in this study. Whole *Gambusia* samples were carefully washed, well mixed with 25% (w/w) salt and incubated at room temperature for five months (from January up to the end of May, 2006). Salted fish was manually drained using cheese cloth to separate supernatant liquor to keep out salted fish. The fish sauce obtained was composed of 65.97% moisture, 12.37% crude protein, 1.56% lipid, 19.33% ash and 9.08% sodium chloride content. In addition, the pH value was 6.08 and the nonessential amino acids (3,864 mg/100 ml) were higher than the essential amino acids (AAs) ones (2,172 mg/100 ml). Based on the biochemical criteria, nutritional value as well as microbial aspects indicated that *Gambusia* fish could be successfully utilized for fish sauce production as value added fish product.

Keywords: *Gambusia (Affinis affinis)*, fish sauce, biochemical criteria, nutritional value, microbial aspects.

Sivrisinek Balığı (*Affinis affinis*) Balık Sosu Üretimi İçin Kullanımı

Özet

Çalışmanın amacı, sivrisinek balığından (*Affinis affinis*) balık sosunun üretilmesi ve bunun besinsel değerlendirmesinin belirlenmesidir. Sivrisinek balığına ait örnekler dikkatlice yıkanmış, %25 (w/w) tuz ile iyice karıştırılmış ve beş ay boyunca (2006 yılı Ocak ayından Mayıs ayının sonuna kadar) oda sıcaklığında tutulmuştur. Tuzlu balığın bulunduğu kapta oluşan suyu uzaklaştırmak için tülbent kullanılmış ve elle süzme yapılmıştır. Elde edilen balık sosu; %65,97 nem, %12,37 ham protein, %1,56 lipid, %19,33 kül ve %9,08 sodyum klorür içermektedir. pH değeri 6,08 ve esansiyel olmayan amino asitler (3.864 mg/100 ml) esansiyel amino asitlerden (AA) (2.172 mg/100 ml) daha yüksek bulunmuştur. Biyokimyasal kriterlere göre besin değeri ve mikrobiyal durumu göz önüne alındığında, sivrisinek balığının katma değerli balık ürünü olarak balık sosu üretimi için başarılı bir şekilde kullanılabilir.

Anahtar Kelimeler: Sivrisinek balığı (*Affinis affinis*), balık sosu, biyokimyasal kriterler, besin değeri, mikrobiyal etki.

Introduction

Many fermented fish products are prepared in different parts of the world and the method of processing depends upon various factors, viz., availability of raw materials, consumers preference and the climatic conditions of the region (Al-Jedah *et al.*, 2000). 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 brown, liquid seasoning commonly used in most parts of Southeast Asia; it is called by different names in different countries and it contains a mixture of amino acids and other protein degradation products (Ijong

and Ohta, 1995; Ruiz-Capills *et al.*, 2000; Gildberg, 2001; Sanceda *et al.*, 2001; Aquerreta *et al.*, 2001; Fukami *et al.*, 2002; Ichimura *et al.*, 2003). 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, are widely used in 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 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). The *Gambusia* fish is a small freshwater species and it is not normally used for human consumption. Therefore, the main objectives of this work were to utilize *Gambusia (Affinis affinis)* as by-catch for sauce production, also to set a standard for proximal analysis and nutritional value for fish sauce produced as value added fish product for Egyptian consumers.

Materials and Methods

Fresh raw *Gambusia Affinis affinis* (5.28±1.75 cm length and 1.63±0.88 g weight) were obtained from El-Qanatir EL-Khairia Fish Research Station, National Institute of Oceanography and Fisheries (NIOF) during harvesting season of 2005. The samples were well mixed with 25% (w/w) finally refined sodium chloride (Bono salt, Saltines Co.) by gloved hands. They were packed in polyethylene bags, tightly sealed and put into screw caps-hard plastic containers. After that, all containers were incubated at room temperature for 150 days (from January to the end of May, 2006). After that, salted fish was manually drained using cheese cloth to separate fish sauce to keep out salted fish.

Analytical Methods

The moisture, crude protein (TN×6.25), total lipid and ash content were analyzed using standard methods (AOAC, 1995). pH value (pocket-sized pH meter), total volatile base nitrogen (TVB-N) content and Thiobarbituric acid (TBA) number were determined (Pearson, 1976). Trimethylamine nitrogen (TMA-N) was measured (AOAC, 1995). Individual amino acids were determined using reverse phase HPLC; hydrolysis, derivatization and analysis were performed according to the Pico-Tag method (Millipore Co-operative, 1987). Tryptophan was not determined. Values of amino acids were expressed as (mg/100 ml). For microbiological analysis, 10 g of sample were transferred aseptically to 90 ml of sterile 0.1% (w/v) peptone water. Serial decimal dilutions in 0.1% peptone water were prepared and triplicate 1ml of appropriate dilutions was poured on selective agar plates. Plate count agar (PCA, Oxoid) for total bacterial count (mesophilic bacteria), PCA (Oxoid) contained 10% salt for halophilic bacteria count and Potato-Dextrose Agar (PDA, Oxoid) for yeasts and molds count were incubated at 37°C for 48 hrs (FAO, 1979).

Microbiological results were converted into log₁₀ CFU/ml.

Statistical Analysis

Then results (triplicate determinations) of chemical composition, biochemical criteria and microbial aspects were expressed as means ± SE (using SPSS 10).

Results

Proximal Analysis

Table 1 shows the compositional parameters of raw *Gambusia*. Proximal analysis of whole *Gambusia* samples (on wet weight basis) had the content of 73.42% moisture, 14.59% crude protein, 6.56% lipid and 4.81% ash.

Fish Sauce

Proximal analysis of fish sauce was (on wet wt. basis) composed of 65.97% moisture, 12.37% crude protein, 1.56% lipid, 19.33% ash and 9.08% sodium chloride content as shown in Table 2.

Biochemical Criteria

The values of pH, TVB, TMA and TBA of *Gambusia* sauce are shown in table (3). The pH value of fish sauce was 6.08. On the other hand, TVB-N and TMA-N contents were 107.80 and 0.75 mg /100 ml, respectively. Concerning TBA, value was 1.55 mg Malonaldehyde/1000 ml sauce Table 3.

Table 1. Proximal analysis (Mean± SE) of whole *Gambusia* fish

Constituent	%
Moisture	73.42±0.05
Crude protein	14.59±0.62
Lipid	6.56±0.79
Ash	4.81±0.13

Table 2. Proximal analysis (Mean± SE) of fish sauce

Constituent	%
Moisture	65.97±0.09
Crude protein	12.37±0.77
Lipid	1.56±0.31
Ash	19.33±0.08
Sod. chloride	9.08±1.03

Table 3. Biochemical criteria (Mean± SE) of fish sauce

Criterion	Value
pH	6.08±0.02
TVB-N (mg /100 ml)	107.80±3.95
TMA-N(mg /100 ml)	0.75±0.40
TBA (mg MA/1000 ml)	1.55±0.16

Amino Acids (AAs) Composition

Amino acids (AAs) composition of Gambusia sauce is shown in Table 4. Total AAS was 6,036 mg/100 ml; 2,172 mg/100 g as essential AAS and 3,864 mg/100 ml nonessential AAS.

Microbiological Aspects

On the other hand, microbiological count of TBC, HBC and yeasts and molds were 2.0, 2.30 and 2.10 log₁₀ cfu/ g sauce, respectively (Table 5).

Discussion

The compositional parameters of raw Gambusia (Table 1) are quite similar of male capelin-by products except ash content (2.3%) used for fish sauce production (Gildberg, 2001). A great variety of raw materials can be used for sauce production provided that the content of proteolysis enzymes is sufficient to give efficient solubilization and protein hydrolysis. Most frequently fish sauce is made from small pelagic species like anchovies and sardines (Amano, 1962; Lee, 1990; Saisithi, 1994).

On the other hand, the results of chemical composition of fish sauce (Table 2) are in agreement with those reported by Cho *et al.* (2000); Park *et al.* (2001) who reported that the range of chemical composition of fish sauce was 61.40-79.20%

moisture, 0.9-13.70% crude protein and 18.20-25.80% ash content.

The value (6.08) of pH in this study (Table 3) was slightly increased compared to those (4.66-5.91) reported by Cho *et al.* (2000) and Aquerreta *et al.* (2001) and it is in agreement with those reported by Ijong and Ohta (1995), Cho *et al.* (1999), and Park *et al.* (2001). The high pH value of sauce may reflect bacterial activity during fermentation and probably as a consequence of the accumulation of basic compounds (Aquerreta *et al.*, 2001). Concerning TVB, content was laid in range 14.1-338.6 mg TVB-N /100 ml which were reported by Aquerreta *et al.* (2001), Ruiz-Capillas *et al.* (2000). The content of TVB-N might be attributable to the rate of hydrolysis of fish muscles by fish enzymes and microbial activity during fermentation under the lower salt concentration (Ijong and Ohta, 1995). Whereas the content of TMA (Table 3) was lower than (2.63–60.5 mg/100 ml) found by Oh, (1999) and Ruiz-Capillas *et al.* (2000). In addition, TBA figures in this work are lower than those (13.8-66.5 MA/kg) found by Chang *et al.* (1994) and Aquerreta *et al.* (2001).

AAs are major contributors to the palatability of fish sauce and which derived as result breakdown of fish proteins as affected by enzymes action. In general, AAs composition in this work depends mainly on the amount of salt added and fish source used for sauce production. The nonessential amino acids 3,864 mg/100 (nonessentials) are higher than 2,172 mg/100g (essentials). The results obtained are in disagreement with those reported by Ijong and Ohta (1995) and Shih *et al.* (2003) who reported that the essential amino acids were higher than the nonessential AAs.

Furthermore, the results of microbial load in Table 5 were confirmed with Dissaraphong *et al.* (2006) who reported that microbial load of fish sauce decreased with increase fermentation period, possibly caused by high concentration of salt. They added to halophilic bacteria counts decreased gradually during fermentation and were lower than 1 log CFU/ml after 4 and 5 months, respectively compared with the count of halophilic bacteria in Table 5. Those micro-organisms might contribute to the hydrolysis of proteins as well the flavor development of the fish sauce obtained.

In conclusion, fish sauce contains an important content of protein and essential amino acids and it is safe for human consumption as well. Therefore, fish gambusia can be utilized as raw material for fish sauce production.

Table 4. Amino acids composition (mg/100 ml) of fish sauce

Amino acids	mg /100ml	Amino acids	mg /100ml
Essential:		Nonessential	
lysine	63	Aspartic	1248
Therionine	129	Glutamic	965
Valine	142	Cystine	441
Methionine	556	Tryptophan	*n.d.
Arginine	211	Serine	679
Isolucine	29	Praline	179
Leucine	241	Glycine	260
Phenylalanine	214	Alanine	92
Histidine	111		
Tyrosine	476		
Total (a)	2172	Total (b)	3864
Grand (a + b)	6036		

*n.d.: not determined

Table 5. Microbiological aspects (log₁₀ cfu/ml) of fish sauce

Microbiological aspects	log ₁₀ CFU /ml
Plat count agar (PCA)	2.0±0.06
Halophilic bacteria (HB)	2.30±0.11
Yeasts and molds	2.10±0.02

References

- A.O.A.C. 1995. Official Methods of Analysis, 16th ed. Association of Official Analytical Chemists International, Arlington, Virginia, USA.
- AL-Jedah, J.H., Ali, M.Z. and Robinson, P.K. 2000. The inhibitory action of spices against pathogens that

- might be capable of growth in a fish sauce (mehiawah) from the Middle East. *International J. Food Microbiology*, 57(1/2): 129-133.
- Amano, K. 1962. The influence of fermentation on the nutritive value of fish with special reference to fermented fish products of South-East Asia. In: E. Heen and R. Kreuzer (Eds.), *Fish Nutrition*, Fishing News Books, London, 180 pp.
- Aquerreta, Y., Astiasaran, I. and Bello, J. 2001. Use of exogenous enzymes to elaborate the Roman fish sauce garum. *J. of the Science of Food and Agriculture*, 82(1): 107-112.
- Chang, C.M., Ohsima, T. and Koizumi, C. 1994. Changes in the composition of free amino acids, organic acids, and ripening of "hatahata-zushi", a fermented fish product of sandfish (*Arctoscopus japonicus*) and boiled rice. *Journal of the Science of Food and Agriculture*, 66: 75-82.
- Cho, Y.J., Im, Y.S., Lee, K.W., Kim, G.B. and Choi, Y.J. 1999. Changes of components in salt-fermented northern sand lance, *Ammodytes personatus* sauce during fermentation. *J. of the Korean fisheries Society*, 32(6): 693-698.
- Cho, Y.J., Im, Y.S., Park, H.Y. and Choi, Y.J. 2000. Quality characteristics of Southeast Asian salt-fermented fish sauces. *J. of the Korean Fisheries Society*, 33(2): 98-102.
- Dissaraphong, S., Benjakul, S., Visessanguan, W. and Kishimura, H. 2006. The influence of storage conditions of tuna viscera before fermentation on the chemical, physical and microbiological changes in fish sauce during fermentation. *Bioresource Technology*, 97: 2032-2040.
- FAO 1979. *Manuals of food quality control, 4-Rev.1-Microbiological Analysis*, Food and Agriculture Organization of the United Nations, Rome, 14 pp.
- Fukami, K., Ishiyama, S., Yaguramaki, H., Masuzaw, T., Nabeta, Y., Endo, K. and Shimoda, M. 2002. Identification of distinctive volatile compounds in fish sauce. *J. Agric. Food Chem.*, (50): 5412-5416.
- Gildberg, A. 2001. Utilisation of male Arctic Capelin and Atlantic cod intestines for fish sauce production-evaluation of fermentation conditions. *Bioresource Technol.*, 76: 119-123.
- Hjalmarsson, G.H., Park, J.W. and Kristbergsson, K. 2007. Seasonal effects on the physicochemical characteristics of fish sauce made from capelin (*Mallotus villosus*). *Food Chemistry*, 103: 495-504.
- Ichimura, T., Hu, J., Quaaaita, D. and Mayymas, S. 2003. Angiotensin.1-Converting enzyme inhibitory activity and insulin secretion stimulative activity of fermented sauce. *J. Bioscience and Bioengineering*, 96(5): 496-499.
- Ijong, F.G. and Ohta, Y. 1995. Amino acid composition of bakasang, a traditional fermented fish sauce from Indonesia. *Lebensm. Wiss. U.-Technol.*, 28: 236-237.
- Lee, C.H. 1990. Fish fermentation technology- a review. In: P.J.A. Reilly, R.W.H. Parry, L.E. Barile, (Eds.) *post harvest technology, preservation and quality of fish in South-East Asia*, Echanis Press, Manila: 1-13.
- Millipore Co-Operative 1987. *Liquid Chromatographic Analysis of Amino Acid in Foods Using a Modification of the PICO*.
- Oh, K.S. 1999. Quality characteristics of salt-fermented and sandlance sauce. Anchovy sauce. *J. Korean Fish. Soc.*, 32(3): 252-255.
- Park, J.N., Fukumoto, Y., Fujita, E., Tanaka, T., Washio, T., Otsuka, S., Shimizu, T., Watanabe, K. and Abe, H. 2001. Chemical Composition of Fish Sauces Produced in Southeast and East Asian Countries. *Journal of food composition and analysis*, 14: 113-125.
- Pearson, D. 1976. *The Chemical Analysis of Food*. Churchill, New York, London: 374-410.
- Ruiz-Capillas, C., Gillyon, C.M. and Horner, W.F.A. 2000. Determination of volatile basic nitrogen and trimethylamine nitrogen in fish sauce by flow injection analysis. *Eur. Food Res. Technol.*, 210: 434-436.
- Saisthi, P. 1994. Traditional fermented fish: fish sauce production. In: A.M. Martin, (Ed.), *Fisheries Processing: Biotechnological Application*, Chapman and Hall, London: 111-131.
- Sanceda, N.G., Suzuki, E. and Kuratal, T. 2001. Development of normal and branched chain volatile fatty acids during the fermentation process in the manufacture of fish sauce. *Journal of the Science of Food and Agriculture*, 81: 1013-1018.
- Shih, I.L., Chenb, L.G., Yu T.S., Chang, W.T. and Wang, S.L. 2003. Microbial reclamation of fish processing wastes for the production of fish sauce. *Enzyme and Microbial Technology*, 33: 154-162.
- Watanabe, F., Michihata, T., Takenaka, S., Kittaka-Katsura, H., Enomoto, T., Miyamoto, E. and Adachi, S. 2004. Purification and characterization of corrinoid compounds from Japanese fish sauce. *J. Liquid chromatography& Related Technologies*, 27(13): 2113-2119.