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Physicochemical Characteristics and Fatty Acid Profile of Smoked Skipjack Tuna (*Katsuwonus pelamis*) Using Coconut Fiber, Nutmeg Shell and Their Combination as Smoke Sources

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Abstract: : Study was carried out to assess the effect of different smoke sources (coconut fiber, combination of coconut fiber and nutmeg shell (1:1) and nutmeg shell on the physicochemical characteristics and fatty acid profile of smoked skipjack tuna (*Katsuwonus pelamis*). The lowest water content (56.26%), a_w value (0.96) and the highest protein content (38.95%) were recorded in the nutmeg shell-smoked skipjack fish. These results are different (P<0.05) from that obtained from smoking either using the coconut fiber only or the combination of coconut fiber and nutmeg shell. However the lowest fat content (2.13%) was recorded in the nutmeg shell-smoked skipjack fish and not different (P>0.05) from that obtained from other smoke sources. Although the ash content (2.30%) of the nutmeg shell-smoked skipjack fish is slightly higher (2.24%) than that obtained from the coconut fiber as smoke source and slightly lower (2.38%) than that of coconut fiber and nutmeg shell combination, these results are not different (P>0.05). The lowest SFA and MUFA contents were recorded in smoked skipjack fish of nutmeg shell smoking and different from (P<0.05) that of other smoking sources. However, the smoked skipjack fish of nutmeg shell smoking contained the highest PUFA content and different (P<0.05) from that obtained from the point of view of some physicochemical characteristics and fatty acid profile.

Keywords: Smoked skipjack tuna, Coconut fiber, Nutmeg shell, Physicochemical characteristics, Fatty acid profile.

Introduction

Fish smoking is a traditional fish preservation method which has been practiced for centuries and hot smoking with a temperature above 60°C is the traditional method commonly used in tropical countries. However, there is a wide variety of smoking methods in different countries as well as in regions within the same country. These different methods are due to different fish species and expected quality of end products^[1].

The presence of some phenols, formaldehyde and other compounds originated from the smoke penetrates into the fish flesh and acts as preservative to extend the shelf life of end products as well as to give a specific smoke flavour and taste^[2-4].

According to Kostrya and Pikielna^[5], a different smoke source produced also different complex smoke compounds that could consist of mixture of various volatile and non volatile compounds, such as phenol, syringol and guaiacol and its derivatives and affect the sensory traits, while Bower *et al.*^[6] and Gomez-Guillen *et al.*^[7] reported that the common smoking wood compounds were phenol which had an antioxidant function, organic acids, alcohol, carbonyl, hydrocarbon and nitrooxide during smoking process would stick onto fish surface and then penetrate into the fish flesh.

Skipjack tuna (*Katsuwonus pelamis*) locally known as "cakalang" is one of the most popular fish preserved by traditional smoking method in Bitung, North Sulawesi. The skipjack fish in this region are usually splitted in half in the form of butterfly and clipped on a bamboo frame. According to Hayward and Mosse^[8], in Ambon, bigger skipjack fish size was splitted, while smaller one was skewed on a bamboo frame before smoking over the burning wood on racks at a certain distance from smoke source. Yusnaini *et al.*^[9] noted that in North Maluku, *fufu* fish are smoked using kosambi (*Schleichera oleosa*) wood, but in North Sulawesi the same species are smoked using some different wood, including coconut (*Cocos nucifera*) shell or coconut fiber, while in Maluku asar fish are smoked using coconut fiber.

Isamu *et al.*^[10] reported that smoked skipjack tuna producers in Kendari city, Central Sulawesi, used coconut shell as smoke source and therefore similar physicochemical characteristics and organoleptic properties were found in the smoked skipjack samples obtained from those producers. While Sigurgisladottir *et al.*^[11-14,3,15] also reported that different smoke resources could affected the physicochemical and organoleptic characteristics of the end products. Abolagba and Melle^[14] reported that the use of semi-dry and dry rubber wood did not affect protein content of smoked Tilapia.

Indonesia is rich in natural woods and agricultural waste sources, such as teak (*Tectona grandis*) wood, coconut (*Cocos nucifera*) shell, coconut (*Cocos nucifera*) fiber, paddy (*Oryza sativa*) stem and corn (*Zea mays*) cobs, which are potential as smoke sources ^[16]. In North Sulawesi, agricultural waste, such as coconut shell, coconut fiber, nutmeg (*Myristica fragans*) shell and candlenut (*Aleuritus moluccana*) shell are abundantly available and these fuel sources are potential for smoke source in fish processing and preservation in this region.

However, there is only limited scientific information on the characteristics of smoked skipjack (*Katsuwonus pelamis*) fish produced in Bitung city, North Sulawesi, especially if smoked using coconut fiber, nutmeg shell or their combination. Therefore, this study was aimed to investigate the physicochemical characteristics and the fatty acid profile of smoked skipjack (*Katsuwonus pelamis*) prepared using different smoke sources.

Materials and Methods

Sample preparation.

Fresh skipjack (*Katsuwonus pelamis*) tuna and coconut fiber were purchased from local smoked fish processors, while nutmeg shells were bought from nutmeg farmers surrounding Bitung city, North Sulawesi. In this study, fresh fish of relatively same weight, approximately 2.5kg/fish after evisceration, were descaled, washed in running tap water, drained and then splitted in half (butterfly form) and skewed in bamboo frame before smoking. One hundred and fifty fish individuals were used for each smoking process on a smoking kiln (length of 6 m, width of 4 m and height from smoke source of 0.6 m), and the fish were smoked using coconut fiber, nutmeg shell and their combination (1:0; 1:1 and 0:1) as smoke sources for 180 minutes until the fish colour turned to silverish gold or yellowish gold. These smoked fish products were then analyzed to determine the physicochemical characteristics including water, protein, fat and ash content following methods described in AOAC ^[17], while the a_w value was measured using the method of Fuentes *et al.*, ^[18] and the fatty acid profile was determined using Gas Chromatography (GC 210A SHIMADZU) according to the method of AOAC ^[17].

Fatty acid profile analysis.

Samples for fatty acid profile analysis were prepared as follow: 10 g of sample were homogenized using 10 ml concentrated HCl then heated in a waterbath at 70°C until boiling about 30 minutes, cooled to room temperature before extracted with 25 ml diethyl eter and mixed using vortex before adding 25 ml petroleum benzene. After mixing with vortex the clear upper part of the solution was then put into 100 ml test tube and

evaporated in water bath at 60°C while flushing with nitrogen gas (N₂). Approximately 3 ml 0.5 N sodium methalonic was added and then continuously heated in a water bath at 60°C for approximately 10 minutes. After cooling up to room temperature a solution of 3 ml BF₃-CH₃OH 20% was added, reheated in a water bath at 60°C for 10 minutes and cooled, then *the methyl ester formed* was extracted using 1 ml heptane (vortex) and 2 ml concentrated NaCl was added. The upper part of the solution was taken and 1 micro liter of this solution was then injected in GC apparatus. The initial GC temperature set was 140°C and gradually increased 10° C/min. with the end column temperature was 260° C. The column used was RTX semipolar (length: 30 m) with Flame Ionization Detector (FID) and He as gas carrier.

Statistical analysis

Data obtained from three replications for proximate analysis in this study were analysed using One Way Analysis of Variance (ANOVA) and differences among means were determined by Least Significance Difference (LSD) using SPSS version 20 (Chicago, Illinois, USA), and 95% confidence level was used as indicator for statistical significancy. These data were presented as mean \pm standard deviation.

Results and Discussion

The physicochemical analysis results are presented in Table 1 and these show that the a_w values of smoked fish produced using different smoke sources are in the range of 0.96 to 0.99 with water content of 56.26% to 59.69%. Protein, fat and ash content of those samples are in the range of 36.06% to 38.95%, 2.13% to 2.39% and 2.24% to 2.38% respectively.

Table 1. The physicochemical of	characteristics (of smoked	fish produced	using	coconut	fiber,	nutmeg	shell
and their combination as smoke	e sources*.							

Smoked sources	a_{w}	Water content	Protein content	Fat	Ash
		(%)	(%)	content (%)	content (%)
Coconut fiber $(1:0)$	0.98±0.01 (b)	59.07±0.24 (b)	36.06±0.10 (a)	2.39±0.18 (a)	2.24±0.09 (a)
Nutmeg shell(0 : 1)	0.96±0.01 (a)	56.26±0.31 (a)	38.95±0.18 (c)	2.13±0.17 (a)	2.30±0.19 (a)
Coconut fiber and	0.99±0.00 (b)	59.69±0.30 (c)	36.78±0.33 (b)	2.34±0.09 (a)	2.38±0.07 (a)
nutmeg shell(1:1)					

*) Data in same column followed by same letter are not significant different (P>0.05)

The different a_w value and water content of smoked fish produced using different smoke sources are possible due to different smoke components and temperature obtained during the smoking process. Isamu *et al.*^[10] noted that different temperatures of the smoking process were caused by different cellulose and pentosan content of smoke sources used. Kostyra and Pikielna^[5] and Oduor-odote *et al.*^[15] also reported that different smoke sources used could affect the physicochemical and the sensory characteristics as well as the microbial quality of smoked fish products. Furthermore, Kumolu-Johson *et al.*^[19] stated that smoking time could affect the humidity in the kiln and an interaction of water vapour and fish flesh occured which reduced either the a_w value or the water content of the end products.

The a_w value of smoked sea bream (*Sparus aulata*) produced in Italia by smoking at $80 - 90^{\circ}$ C for 60 minutes was around 0.96^[20]. While Fuentes *et al.*^[18] reported that water content of the skipjack fish smoked using beech wood in Spain had a water content of 56.6% up to 66.2%, and according to Kardinal et al.^[21], water content of industrial specific skipjack smoked fish was less than 65%.

The highest protein is observed in the smoked fish processed using nutmeg shell (38.95%) and significantly different (P<0.01) from either samples smoked using coconut fiber (36.06%) or combination of coconut fiber and nutmeg shell (36.78%). Fuentes *et al.*^[18] reported that protein content of skipjack fish smoked using beech wood in Spain was in the range of 15.4% to 31.5%. While Isamu *et al.*^[10] observed that smoked skipjack fish traditionally produced in Kendari (Central Sulawesi) contained protein from 26.42% to 28.80% and Toisuta *et al.*^[22] also noted that protein content of smoked tuna was in the range of 16.75% to 24.08%. Huda *et al.*^[23] reported that smoked Indonesian catfish *Macrones nemurus* and *Cryptopterus micronema* contained 32.25% and 38.81% protein, respectively. However in this study protein content of the samples was found higher than that reported before, and it is possibly due to different fish species and freshness, nutrients available in its surrounding and smoking methods used.

Smoked fish produced using nutmeg shell as smoke source showed the lowest fat content (2.13%), but not significant different (P>0.05) from that of other samples. It is believed that nutmeg shell having a harder

texture than that of coconut fiber could give more heat and hence melted the fish fat, especially as the fish were splitted in half. Fuentes *et al.* ^[18] reported that in Spain, skipjack fish smoked using beech wood contained fat between 1.4% and 3.8%, while Huda *et al.*^[23] observed that fat content of Indonesian smoked catfish, *Macrones nemurus* and *Cryptopterus micronema*, was 32.06% and 8.02%, respectively. According to Bligh *et al.*^[24] and Gehring *et al.*^[25], the different fat content could be affected by some factors, such as fish species, age, smoking method, smoking time, nutrient, growth rate, migration as well as season.

The ash content determination of all smoked fish samples showed that there were no statistical differences (P>0.05) of skipjack fish smoked with the combination of coconut fiber and nutmeg shell as smoke source (2.38%), and nutmeg shell (2.30%) and coconut fiber only (2.24%). In recent study, Toisuta *et al.* ^[22] reported that ash content of smoked skipjack fish was in the range between 1.36% to 5.66%. Variations in ash content as reported by other workers are possibly due to differences between fish species and parts of fish used as samples ^[26]. According to Andrew ^[27], the ash content of smoked fish indicated that this product was a good source of minerals, such as calcium, calium, zinc, iron and magnesium.

The fatty acid profiles of samples smoked using coconut fiber, nutmeg shell and combination of coconut fiber and nutmeg shell are presented in Table 2. It was observed that those samples contained Saturated Fatty Acid (SFA) consisting of capric acid (C10:0), lauric acid (C12:0), myristic acid (C14:0), pentadecylic acid (C15:0), palmitic acid (C16:0), heptadecanoic acid (C17:0), stearic acid (C18:0), and arachidic acid (C20:0). While the Monounsaturated Fatty Acid (MUFA) consists of palmitoleic acid(C16:1), oleic acid (C18:1), ciseicosenoic acid (C20:1), erucic acid (C22:1) and nervonic acid (C24:1); and Polyunsaturated Acid (PUFA) consists of linoleic acid (C18:2n-6) gama linolenic acid (C18:3n-6), linolenic acid (C18:3n-3), arachidonic acid (C20:4n-6), eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). Toisuta *et al.*^[22] observed that by products (head, skin, intestine, liver and gonads) of skipjackfish (*Katsuwonus pelamis*) also contained Saturated Fatty Acids (SFA), capric acid (C10:0) ,lauric acid (C12:0) , myristic acid (C18:1n-9) and palmitoleic (C16:1), whereas Polyunsaturated Fatty Acids (PUFA) consists of oleic acid (C18:1n-9) and palmitoleic (C16:1), whereas Polyunsaturated Fatty Acids (PUFA) consists of SEPA (C20:5n-3), DHA (C22:6n-3), linoleic (C18:2n-6) and linolenic (C18:3n-3).

	Smoke sources				
Fatty acids	Coconut fiber	Coconut fiber and	Nutmeg shell		
	Cocollut Hoel	nutmeg shell (1 : 1).			
Capric acid (C10:0)	0.011 ± 0.002 b	0.007 ± 0.001 a	0.009 ± 0.001 a		
Lauric acid(C12:0)	0.062± 0.012 a	0.070± 0.016 a	$0.087 \pm 0.006 \text{ b}$		
Myristic acid (C14:0)	0.940± 0.048 a	1.116± 0.370 a	0.472 ± 0.074 a		
Pentadecylic acid (C15:0)	0.705± 0,361 a	0.551± 0.478 a	0.633 ± 0.023 a		
Palmitic acid (C16:0)	25.529± 0.408 b	23.824± 0.917 a	23.437± 0.604 a		
Heptacosanoic acid (C27:0)	1.729± 0.177 a	1.576± 0.236 a	1.521± 0.019 a		
Stearic acid (C18:0)	2.665± 0.422 a	3.583± 0.624 b	3.818± 0.083 b		
Arachidic acid (C20:0)	0.016 ± 0.004 a	0.018 ± 0.002 a	0.024 ± 0.002 a		
Total SFA	30.947 ± 0.980 a	30.015 ± 0.584 a	30.002± 0.780 a		
Palmitoleic acid (16:1n-7)	$3.680 \pm 0.761b$	3.353 ± 0.557 ab	2.276± 0.239 a		
Oleic acid (C18:1n-9)	10.545 ± 0.143 a	11.607 ± 1.866 a	12.169± 0.133 a		
Cis-eicosonoic acid (C20:1)	4.576 ± 0.113 b	4.760 ± 0.169 b	3.362 ± 0.038 a		
Erucic acid (C20:1)	28.204 ± 1.727 a	29.282 ± 1.976 a	27.166± 0.115 a		
Nervonic acid(C24:1)	0.366 ± 0.079 a	0.334 ± 0.049 a	0.437± 0.013 a		
Total MUFA	47.371± 1.237 ab	49.336 ± 1.945 b	45.562± 0.096 a		
Linoleic acid (C18:2n-6)	0.495± 0.101 a	0.457 ± 0.073 a	0.532± 0.015 a		
Linolenic acid(C18:3n-3)	0.913± 0.244 a	1.362 ± 0.217 b	1.296± 0.013 b		
Gama linolenic acid(C18:3n-6)	3.807± 0.751 a	3.959 ± 0.546 a	3.983± 0.090 a		
Arachidonic acid (C20:4)	0.400 ± 0.083 a	0.375 ± 0.057 a	0.446±0.004 a		
Eicosapentaenoic acid(EPA)	0.053 ± 0.012 a	0.694 ± 0.541 a	1.500± 0.214 a		
Docosahexsaenoic acid(DHA)	0.156 ± 0.034 a	0.063 ±0.098 a	0.200 ± 0.003 b		
Total PUFA	5.823 ± 0.721 a	7.017 ± 1.510 a	7.956±0.960 a		

Table 2. Fatty acid profile of smoked skipjack fish prepared using different smoke sources.

*) Data in same column followed by same letter are not significantly different (P>0.05)

The lowest SFA content (30.002%) was determined in the smoked skipjack fish samples prepared using nutmeg shell as smoke source and it is not significantly different (P>0.05) from those of either using coconut fiber combined with nutmeg shell (30. 015%) and coconut fiber only (30.947%). Ilow *et al.* ^[28] stated that SFA content of some smoked sea fish ranged between 24.2% and 28.0%. Hence, the SFA content of smoked skipjack fish samples in this study was observed higher than that reported by llow *et al.* ^[28].

While the highest MUFA content (49.336%) is observed in samples of smoked skipjack fish prepared using the coconut fiber and nutmeg shell combination as smoke source and it is significantly different (P<0.05) than those prepared using nutmeg shell (45.562%) but not different (P>0.05) from those smoked with coconut fiber only as smoke source (47.371%). The MUFA content of samples in this study is higher than that some sea fish species (26.0% - 39.8%) reported by llow *et al.*^[28].

Smoked skipjack fish prepared using nutmeg shell as smoke source showed the highest PUFA content (7.956%) and it is not significantly different (P>0.05) than those smoked using both the combination of coconut fiber and nutmeg shell (7.017%) and coconut fiber only (5.823%). These results are much lower than that reported by Ilow *et al.* ^[28] where PUFA content of some smoked fish species was in the range of 31.9% to 45.4%. According to Sinclair *et al.* ^[29] and Swastawati ^[30], differences in smoked fish flesh composition are due to different fish species and smoking methods. While Tenyang et al.^[31] noted that cooking or smoking could affect the catfish lipid through hydrolization or oxidation.

Conclusion

The present study showed that skipjack fish smoked using nutmeg shell contained lower water content, a_w value, fat content and SFA content, while protein and PUFA content were higher than that smoked with combination of coconut fiber and nutmeg shell (1 : 1) or coconut fiber only. Therefore, skipjack fish smoking with nutmeg shell gives the best smoked skipjack fish from the point of view of some physicochemical characteristics and fatty acid profile.

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