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Application of nutmeg shell pyrolysis-based liquid smoke for sea cucumber (*holothuria scabra*) processing

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Abstract : Traditional sea cucumber smoking generally uses hot smoke of wood fie in a smoke room. There have been no method and technological application to develop thesea cucumber smoking, such as the use of liquid smoke. The advantages of liquid smoke utilization in sea cucumber smoking are as follows: safer in wood usage as smoke source, environmental pollution can be reduced, and the product flavor can be controlled and consistent. This study was aimed at determining the optimum concentration and immersion time length in the smoke solution through experiment. The liquid smoke used was that of nutmeg shell pyolysis to yield the best flavor of smoked sea cucumber under immersion time of 30 min., 60 min., and 90 min., respectively. Results found that 30 minutes immersion in 5% liquid smoke solution yielded the lowest water content. The profile of sea cucumber fatty acid immersed for 30 min. in 5%, 10%, and 15% liquid smoke yielded palmitoleinic acid, palmitic acid, linoleiic acid, oleic acid, stearric acid, arachidic acid, and behenic acid.
Keywords : liquid smoke, sea cucumber, nutmeg shell, fatty acid profile.

Introduction

Sea cucumber is one of fisheries commodities potential to be utilized as food material, and even in dry form it has become one of the non-oil and gas export commodities.[1]The fisheries production potential in Indonesia is very high, but its utilization is still low so that the production development and fisheries product consumption opportunities are opened. Recently Indonesia has exported dry smoked product of sea cucumber to Hongkong, China, United States, Japan, and Singapore[2]

In developed countries, sea cucumbers are mostly used as high protein food material and in drug industries and other industries**.** North Sulawesi possesses sufficiently high potential of sea cucumber, but the quality is still low. It results from that their handling and processing are very traditional using simple equipment and have low hygienic condition.[3]

Sea cucumber is fisheries commodity known to quickly decompose. The quality declining process cannot be totally stopped but can be slowed down through processing and preserving techniques, one of which is smoking technique.[4]

Sea cucumber smoking in north Sulawesi Utara is generally conducted traditionally, the use of direct hot smoking method to preserve and to yield certain taste of the product.[5]Smoking sea cucumber and other food materials that is initially intended to prolong the product durability has developed to obtain certain appearance and taste of the food material. Several studies[67] have indicated that smoking is a preserving method that does not only increase the durability but also can give desired taste and color of the smoked product due to the presence of phenolic and carbonyl compounds.[8]The traditional smoked sea cucumber processors just apply the methods taught for generations and have not used better technology yet in order to increase the quality of the smoked sea cucumber products, such as the use of liquid smoked. In the traditional smoking, the smoked concentration, optimum smoking time, and smoking temperature are not consistent and difficult to control.[9]Besides, there is hazard risk potential to human health in relation with the presence of polycyclic hydrocarbon aromatic (PHA).[10] The PHA compounds can be formed through pyrolysis of wood. The most carcinogenic PHA is benzo(a)pirene.[11] The use of liquid smoke posseses several advantages: safe because it can reduce PHA, has antioxidant activity, and can inhibit bacterial growth. Smoking that can replace the direct smoking is liquid smoking method. Therefore, liquid smoking method needs to be applied.

The use of liquid smoke has more benefits than traditional smoking, since it is easily applied, and a faster process, gives typical characteristics of the end-product, such as aroma, color, flavor, and does not cause environmental pollution.[12]Besides, it gives even flavor, more efficient smoke material usage due to its repeatable utilization, can be used in various food materials, and carcinogenic compounds can be eliminated.Liquid smoke can be used in different ways, such as spray, immersion or directly mixed into the food.[13]

Several important factors need tobe considered in liquid smoke application through immersion method[14] are liquid smoke concentration, solution temperature, and immersion duration. The use of liquid smoke is one of the preserving methods to reduce problems in traditional smoking process. The liquid smoke is produced through smoke distillation in which gas-typed smoke is altered into a liquid form.[15]

Sea cucumbers possess good prospect and high economic value in either local or international markets due to its high nutritive content. Research findings indicate that sea cucumber contains 82% protein, 1.7% fat, 8.9% waterc, 8.6% ash, and 4.8% carbohydrate.[16]

Sea cucumbers are also potential to cure kidney disease, pneumonia, anemia, and prevent arteriosclerosis and body tissue aging. They have been utilized by coastal communities for long time as food material. This animal contains about 82% with complete amino acids, W-3-typed-unsaturated fat important for heart health, and relatively high eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), 25.69% and 3.69%, respectively. High EPA of the sea cucumber indicates an ability to quickly repair the tissue damage. Collagen content of the sea cucumber extract accelerates both internal and external wound healing after surgery, such as Caesarean section.[17]

Sea cucumbers have high nutritive content, but are easily damaged so that handling and processing are necessitated, one of which is through smoking method. Handling sea cucumber should be directed to food material demand in order to increase people's nutrition. Hence, preserving and processing are important ways to deal with rapidly decomposing fisheries products.[18]

Smoking is one of food processing methods lengthly known as one of the food product processing phases. The objective of smoking is not only to slow down destruction rate of the product, but to yield certain performance of the smoked product and typical flavor of the food material.[19] The traditional smoking recently used has weaknesses, such as uneven product quality, low durability, hazardous compound accumulation, environmental pollution, and fire risks. To deal with those weaknesses, for both smoked product quality and smoking process, the use of liquid smoke produced from nutmeg shell was done in sea cucumber smoking process.[20]Benefit of liquid smoke utilization more sensitive to aroma production, easily controlled aroma, applicable in various food materials, commercially acceptable for consumers, safer in licable in various methods, such asspray, immersion, or mixed directly in food.[21]

Materials and Method

Sample preparation of fresh sea cucumber (*Holothuria scabra)* and liquidsmoke. Twenty individuals of *H. scabra* were used. They were cleansed and removed the instestines, boiled for 10 minutes then cooled. They were immersed for 30 min., 60 min., and 90 min. in 5%, 10%, and 15% liquid smoke solution. The sea cucumber products were then dried in the oven at 80^{0}C. Physico-chemical analysis, $a_{w,}$employingmethod of Fuentes et al.[22], water content, protein, fat, ash content followed AOAC[23,] and fatty acid profile used Gas Chromatography (GC 210A SHIMADZU).

Sample Preparation

As much as 5g of sample was homogenized with 10 ml of concentrated HCL, put into 100-ml flask, and heated in the water bath at 70^0 C up to boiling for about 30 minutes. After cooled, it was extracted in 25 ml of Diethil Ether, then vortexed with 25 ml of Petroleum Benzene (40-60^0C). Vortex and separate the cear upper part into 100 ml flask, then evaporate in a waterbath at 60^0C through flowing nitrogen gas (N2). Moreover, about3 ml of 0.5 N methanolic sodium was added, closed, and heated in the Water Bath at 60^0C for about 10 min. After cooled, added with 3 ml of 20% BF3-CH30H solution, closed and heated in the waterbath at 60^0C for about 10 min., cooled, and extracted the formed methyl-esther with 1 ml of n-Heptan (Vortex), then added with 2 ml of saturated NaCl, yielding 2 layers. The upper layer was taken as much as 1µl and injected into GC in running condition at 140^0C programmed with increment of 10^oC/min. and the end column temperature of -260^0C,column length of 30m, column name of RTX-5-semipolar, He-carrying gas, and Flame Ionisasi Detector (FID).

Statistical Analysis

Data analysis applied parametric method of 0ne-Way ANOVA to determine the effect of 30 min., 60 min., and 90 min immersion in 5%, 10%, and 15% liquid smoke solution without drying in the oven on chemical composition of the sea cucumber. The analysis used version 20 SPSS (Chikago II, USA). The values were expressed in means±SD(Standard deviation), the significance was tested at $P<0.05$. This analysis used version 20-SPSS software (Chicago,IL, USA).

Results and Discussion

Mean values and standard deviation at the 30 min., 60 min., and 90 min. immersion in 5%, 10%, and 15% liquid smoke solution are presented in Table 1.

Table 1. Physicochemical characteristics of the smoked sea cucumber at different immersion duration and liquid smoke solution concentration.

Immersion duration	Liquid smoke concentration	Physico-chemical parameters				
		Aw	Water content (%)	Protein content (%)	Fat content (%)	Ash content (%)
30 min.	5 %	0.915	60.11±0.10	18.45±0.58	0.20±0.02	4.06±0.07
	10 %	0.922	76.07±1.01	16.18±0.01	0.17±0.01	5.60±0.13
	15 %	0.928	75.80±1.03	15.96±0.02	0.27±0.01	6.30±0.26
60 min.	5 %	0.933	78.91±0.33	15.21±0.02	0.38±0.02	4.27±0.19
	10 %	0.939	77.11±0.60	17.32±0.02	0.27±0.01	4.77±0.02
	15 %	0.941	77.16±0.89	14.67±0.01	0.26±0.01	4.58±0.03
90 min.	5 %	0.944	77.25±0.20	16.94±0.02	0.28±0.01	4.36±0.04
	10 %	0.946	76.23±0.31	9.46±0.02	0.16±0.02	10.31±0.20
	15 %	0.948	76.30±0.14	8.45±0.06	0.21±0.01	10.82±0.02

Chemical composition of the smoked sea cucumber using nutmeg-originated liquid smoke.

Water content. The lowest water content of the smoked sea cucumber using 30 min. immersion was found in 5% liquid smoke solution, 60.11%, andthe highest was recorded in 60 min. immersion 5% liquid smoke, 78.91%. The water content is relatively similar among immersion duration. Water content influences the texture of the end-product, solidly hard, dense, and tough.

The highest protein content was recorded at 30 min. immersion in 5% liquid smoke solution and the lowest at 90 min. immersion in 15% liquid smoke solution. The present study showedthat high protein content was dependent upon food, species, individuals within species, and between body parts.[24]Increased protein, fat, and ash content could result from decline in water content during the smoking process. Protein possesses negative correlation with water content meaning that each protein content increment is followed by declined watercntent. It could be said that 68% of protein content is determined by water content that is possibly due to denaturated protein.

The lowest fat content of the smoked sea cucumber was recorded at 90 min. immersion in 10% liquid smoke, 0.16% and the highest was found at 60 min. immersion in 5% liquid smoke solution, 0.38%. It could result from different immersion duration treatment.

The highest ash content was found at 90 min. immersion in 15% liquid smoke solution, 10.82% and the lowest at 30 min. immersion in 5% liquid smoke, 4.6%. Increased protein, fat and ash content could result from declined water content during smoking process. The higher the ash contained in a food material the higher mineral will be contained.[25]

Fatty acid profile

Gas Chromatography (GC) results of fatty acid profile analysis on *H. scabra* smoked through immersion in the nutmeg shell liquid smoke under different immersion duration and liquid smoke concentration are presented in Table 2.

Table 2. Fatty acid profile of smoked *H. scabra* using 5%, 10%, and 15% liquid smoke solution.

Immesion duration	Fatty acids	Liquid smoke solution		
		5%	10%	15%
30 min.	Palmitoleinic Acid	2.4	3.05	3.44
	Palmitic Acid	17.54	14.48	20.82
	Linoleic Acid	9.08	5.17	12.41
	Oleic Acid	43.66	61.51	47.04
	Stearic Acid	8.10	6.17	8.05
	Arachidic Acid	3.74	6.02	1.54
	Behenic Acid	3.56	6.46	1.84

Table 2 shows that *H. scabra* smoked using 30 min. immersion in 5%, 10%, and 15% liquid smoke contains Saturated Fatty Acid (SFA), the highest palmitic acid (C16:0), 20.82%, recorded in the sample immersed in 15% liquid smoke,stearic acid (C18:0), 8.10 %, in 5% liquid smoke, and arakhidic acid (C:20:0), 6.02%, in 10% liquid smoke.

Total Monounsaturated fatty acid (MUFA) of the smoked *H. scabra*,palmitoleic acid (C16:1) and oleic acid (C18) indicated that sample immersed in 10% liquid smoke solution contained the highest oleic acid, 61.51%.

Total Polyunsaturated fatty acid (PUFA) of smoked *H. scabra*,linoleic acid (C18:2n-6) and arakhidonic acid (C20:4n-6), showed that 30 min. immersion in 15% liquid smoke yielded the highest mean total PUFA in the form of linoleic acid, 12.41%.

The unsaturated fatty acid is more sensitive to heat with increased instability with its saturity level,[26] while mean fatty acid omega-3 content, particularly eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA),is 1.67% and 2.50%, respectively.

Conclusion

This study concluded that the smoked *H. scabra* smoked through 30 min. immersion in 5% liquid smoke had the lowest water content and the highest protein content among the immersion duration treatments. Fatty acid profile of *H. scabra* immersed for 30 minutesin5%, 10%, and 15% liquid smoke contained palmitoleinic acid, palmitic acid, oleic acid, stearic acid, arachidic acid, and behenic acid.

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