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Submission date: 10-Sep-2019 10:10AM (UTC+0700)

Submission ID: 1169981242

File name: Jurnal_Internasional_tahun_2018.pdf (220.2K)

Word count: 3453

Character count: 18731

Contamination Concentration in Smoked Golden Sandfish (*Holothuria Scabra*) using Nutmeg Shell Coconut Shell Smoking Materials

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Abstract : Sandfish are one of the marine export commodities that should immediately be developed its processing method. It is important due their high economic value in international markets, and one of those is golden sandfish *Holothuria scabra*.

Sandfish smoking is generally conducted in traditional method using hot smoked from burning wood in a smoking room. The process usually used any kind of wood as smoke material depending upon the wood availability. Smoking can be defined as a penetrating process of volatile compounds into the sandfish from burning wood in order to gain products of specific taste and aroma, long shelflife from their anti-bacterial content and to inhibit the enzymatic activity.

Smoking is a processing or preservation method using treatment combination of drying and administration of natural chemicals from combustion of natural fire wood.

Sandfish smoking from burning wood for preservation originates from human civilization, in which it could yield products of desired taste and aroma¹.

Several compound contents, such as phenol, formaldehyde, and other compounds, coming from penetrated smoke into the meat and functioning as preservatives are believed to yield good taste and typical aroma and can give longer shelflife²³. This study was aimed at knowing the microbiological and chemical contamination content in the smoked sandfish, *H. scabra*, using smoking materials of nutmeg shell and coconut shell.

Smoking in general still uses smoking materials of coir, coconut shell and several wood types. There is also dry nutmeg shell, wastes of nutmeg fruit processing. This material can be used as smoking material to yield specific smoked sandfish product. Results showed that the sandfish smoked with nutmeg shell contained 25.14% water, 60.67% protein, 2.02% fat, 16.38 % ash, while those smoked with coconut shell contained 24.44% water, 64.97% protein, 2.04% fat, and 16.81% ash. The sandfish smoked with nutmeg shell held 0.033 mg/kg mercury, 1.79 mg/L Cd and 3.50 mg/L Pb, while those smoked with coconut shell held 0.042 mg/kg mercury, 0.84 mg/L Cd, and 7.71 mg/L Pb. The ALT analysis found 9.09×10^1 colonies/g in the sandfish smoked with nutmeg shell and 1.36×10^2 colonies/g in those smoked with coconut shell. *Escherichia coli* was found <3 APM/g in the sandfish smoked with both smoking materials.

Keywords : nutmeg shell, coconut shell, smoking, golden sandfish, microbiological pollution, chemical pollution.

Netty Salindeho / International Journal of ChemTech Research, 2018, 11(07): 308-313.

DOI= <http://dx.doi.org/10.20902/IJCTR.2018.110736>

Introduction

Sandfish are one of the fisheries product commodities potential to use as food material, and even dry sandfish have become one of the non-oil and gas commodities. The potential of fisheries production in Indonesia is very high, but low utilization, so that the opportunities for fisheries product and consumption development are widely opened. Indonesia has exported sandfish in dry smoked form to Hongkong, China, United States, Japan, and Singapore⁴.

In developed countries, sandfish are largely used as high protein-food material and in drug industries as well. In North Sulawesi, despite sufficiently high potency, the quality is still low. Handling and processing is very traditional using traditional equipment, and low sanitation and hygiene, so that the end products are not enough to meet good standard requirements⁵. Sandfish are an easily rotten fisheries commodity. Quality decline process cannot be totally terminated, and processing and preservation techniques can only slow down the process, one of which is smoking⁶.

Sandfish processing in North Sulawesi is generally done traditionally using direct hot smoking method in order to preserve and to give some taste. According to⁷, sandfish and other food materials smoking that is initially intended to prolong the product shelflife has been developed to obtain certain appearance and smoke taste of the food materials. Some studies have shown that smoking of various food products is preservation method that not only can increase durability but also yield desired taste and color of the smoke products due to presence of phenolic and carbonyl compounds. Phenol has anti-oxidant activity inhibiting oxidative rancidity. All compounds contained in the smoke contribute to determination of smoked meat flavor characteristics. During smoking, the smoke is absorbed by the product surface and water in the smoked meat product. Aldehyde, ketone, phenol, and organic acids of the smoke have bacteriostatic power or bacteriacide in the smoked meat. Beside combination of heat and smoke, surface dehydration, protein coagulation, and resin deposition of formaldehyde and phenol condensation are effective chemical and physical inhibitors against microorganism growth and penetration into the smoked meat⁸.

The quality decline process cannot be entirely terminated, but slowed down through processing and preservation techniques. One of these is smoking⁹. Smoking sandfish in North Sulawesi is generally conducted traditionally using direct hot smoking method to preserve the sandfish and to give taste of the smoked product.

Moreover, according to¹⁰ smoke concentration, optimum smoking time, and smoking temperature in traditional practice are not consistent and difficult to control. There is also risk potential to human health in relation with the presence of polycyclic aromatic hydrocarbon (PAH) compounds. These compounds can occur in wood pyrolysis. Highly carcinogenic PAH compounds are benzo(a)pyrene.

Smoking does not only produce color, taste and typical texture, but also can yield carcinogenic components, such as polycyclic aromatic hydrocarbon (PAH) in which benzo(a)pyrene is carcinogenic compounds in meat and fish. Heat produced during smoking process will reduce the water content of the smoked material from evaporation. Water content decline of the food material makes water activity value reduce so that the food material will be more durable since the availability of water for microbial growth is reduced¹¹. In smoking, not a burning fire, but smoke³ works as a preservative via formaldehyde gases arising from imperfect combustion of unburnt materials. Smoking is a method of preserving fish through treatment combination of heat utilization and chemicals produced from wood combustion. Smoking is intended to kill bacteria, ruin the enzymatic activities, reduce water content, and absorb various chemicals of smoke.

Good smoking products are determined by smoke quality and volume produced, temperature, humidity of smoking room, and air circulation in the smoking room. Smoke and quality and volume yielded depends upon types of fuel used¹². Smoke from wood combustion contains important compounds that give flavor or color of the smoked products¹³.

Materials and Method

Sample Preparation

Materials used were fresh sandfish, nutmeg shell, and coconut shell. As many as 40 individuals of sandfish were cleansed and the gut content was discarded, boiled for 10 minutes, and cooled. Twenty individuals were smoked with smoking material of nutmeg shell and the other twenty were smoked with coconut shell. The smoked sandfish were characterized their chemical properties, such as water content, protein, fat, and ash, were determined using¹⁴ method. The microbiological and chemical pollutions were also analyzed.

Statistical analysis

Data analysis used One-Way ANOVA to know the effect of nutmeg shell and coconut shell treatments on the chemical composition of the smoked sandfish. The values were presented as means \pm SD. The significance test was done at $P < 0.05$. The analyses used version 20 SPSS software (Chicago, IL, USA).

Results and Discussion

Nutmeg shell is one of the wastes of nutmeg oil processing that possesses high potential as raw material of active carbon production and smoking material and its availability is very high and continuous. These food wastes have not well used yet. Nutmeg shell can be used as raw material of carbon and active carbon production. Since it has hard texture, this shell is believed to hold high wood materials, such as lignin, cellulose, and hemicellulose¹⁵.

Coconut is one of the raw materials easily found in North Sulawesi. Coconut shell is part of coconut fruit laid beneath the coir biologically functioning as protector of coconut meat. It is hard layer with a thickness of about 3-5 mm. This shell hardness results from silicate (SiO₂) content, in which this part covers 15-19% of total coconut fruit weight. Coconut shell is categorized as hard wood but possessing higher lignin and lower cellulose, and water content of about 6-9% (calculated on dry weight basis), while its metoxyl content is almost the same as that in regular wood, except that number of chemical componets varied with location.

Coconut shell is the outer part of the coconut meat. It has hard texture and can be utilized for various needs. Besides the shell part, coconut plant has many benefits for human, from stem, leaf, meat, water, and coir. Coconut shell is usually left, but it will have benefit after processed in charcoal form.

Chemical composition of sandfish smoked with nutmeg shell and coconut shell is presented in Table 1.

Table 1. Chemical parameter analyses on the sandfish smoked with nutmeg shell and coconut shell.

Smoking material treatment	Chemical Parameters			
	Water (%)	Protein (%)	Fat (%)	Ash(%)
Sandfish smoked with nutmeg shell	25.14 \pm 0.42	60.67 \pm 3.04	2.02 \pm 0.01	16.38 \pm 0.30
Sandfish smoked with coconut shell	24.44 \pm 0.53	64.97 \pm 3.06	2.04 \pm 0.03	16.81 \pm 0.31

Table 1 demonstrates that water content difference in the sandfish smoked with nutmeg shell and that with coconut shell is very small. Heat from smoking process could reduce water content of the product. According to¹⁶, water content is very important to influence the durability of a food material, because water content influences the physical and chemical characteristics and the composition by microorganisms. Water content reduction occurs during smoking. Sandfish smoking will make the sandfish body surface be dry from evaporation. The highest protein content was found in the smoked sandfish using coconut shell, 64.97 %, higher than that reported by¹⁷ 56.62%, and¹⁸, 34.13%. Sandfish is a fishery product containing high protein, while protein is a food component that is very important as energy source, building and controlling material for human body¹⁹. Fat content was almost the same between smoked sandfish using smoking material of nutmeg shell and coconut shell, 2.02% and 2.04%, respectively. Ash content was 16.38 % using nutmeg shell smoking

material and 16.81% using coconut shell smoking material. This finding is similar to²⁰ who found that the ash content of sandfish was 16.8%. The higher the ash content of food material is, the more the mineral content will be. Sandfish have minerals, such as chromium, ferrum, cadmium, mangan, nickel, cobalt, and zinc.²¹ added that minerals in sandfish are phosphorus, magnesium, calcium, iodine, ferrum, and copper.

Chemical contamination of the sandfish smoked with smoking material of nutmeg shell and coconut shell is given in Table 2.

Table 2. Chemical pollution of sandfish smoked with nutmeg shell and coconut shell.

Sample/Smoking Materials	Sample weight (mg)	Sample Volume	Konsentrasi Alat (mg/L)	Mercury (mg/kg)	Cadmium (mg/L)	Plumbum (mg/L)
Smoked sandfish (nutmeg shell)	729.1	0.025	0.05			
	729.1	0.025	0.39	0.042	1.79	3.50
Smoked sandfish (coconut shell)	1,095.7	0.025	0.04			
	1,095.7	0.025	0.34	0.033	0.84	7.71

Chemical contamination analysis found that mercury (Hg) content was 0.042 mg/kg in the smoked sandfish using smoking material of nutmeg shell and 0.033 mg/kg in those smoked with coconut shell. It indicates that Hg, Pb, and Cd content still meet the threshold requirements. Mercury (Hg) has a chemical name "hydrargyrum" meaning liquid silver. In the periodical table, this substance has atomic number of 80 and atomic weight of 200.59 g/mol with melting point of 365.68°C. This metal is produced from sinabar ore (HgS) containing 0.1-4% mercury²². Mercury occurring in the body or waste in the waters is altered by microorganism activity to metal-mercury (Me-Hg) component that has toxic property and high durability in aquatic animals.

Cd content in the sandfish smoked with nutmeg shell is 1.79 mg/L, while those smoked with coconut shell is 0.84 mg/L. Pb content in the smoked sandfish using coconut shell was higher than that in the smoked sandfish using nutmeg shell (Table 2).

Plumbum (Pb) has atomic number of 82 with an atomic weight of 207.2 g/mol at the periodical table and melting point of 1,740°C and functions as protecting layer if contacted with humid air. Plumbum is a very toxic heavy metal and can be practically detected in all dead things in the environment and biological system. Major source of Pb comes from alkyl group, and this component is toxic to all life aspects.

According to²³, marine animal's metabolism, like sandfish, their body will process or transform every entering toxic material (metal) or the metal toxin. It is affected by various factors, such as sampling site and condition at sampling time.

Table 3. Microbiological contamination of sandfish smoked with nutmeg shell and coconut shell.

No	Parameter	Result		Unit	Method
		Sandfish smoked with nutmeg shell	Sandfish smoked with coconut shell		
1.	Total Plate Count	9.09 x 10 ¹	1.36 x 10 ²	colonies/g	SNI 2897:2008 point 4.1
2.	<i>Escherichia coli</i>	< 3	< 3	APM/g	SNI 2897:2008 point 4.3
3.	<i>Staphylococcus aureus</i>	0	0	colonies/g	SNI 2897:2008 point 4.4
4.	<i>Salmonella</i>	Negative	Negative	Nrg/25g	SNI 2897:2008 point 4.5

The ALT analysis on the smoked sandfish using nutmeg shell smoking material found 9.09x10¹ colonies/g, while those using coconut shell smoking material found 1.36x10² colonies/g. *E. coli* analysis gave similar results for both smoking materials used, <3 APM/gr. Analysis of *S. aureus* and *Salmonella* did not give different results between the use of nutmeg shell and coconut shell smoking materials (Table 3). Total Plate Count (TPC) of food material is limited to 10⁶ cfu/g. According to²⁴, colony growth reflects number of

organisms in the sample, such as bacteria, mold. The TPC analysis of the sandfish smoked with nutmeg shell and coconut shell found low number of colonies indicating that the processing has used good and hygienic smoking process. It is indicated with number of samples of detected *E. coli* and *Salmonella* meaning that no contamination occurs in both smoked sandfish products.

Microbiological contamination *Escherichia coli* analysis in sandfish smoked with nutmeg shell and coconut shell found less than 3 APM/g. *E. coli* is gram negative bacteria, unspore, aerobic to facultative aerobic, rod-shaped, and can ferment lactose by producing acid and gas at 35°C for 48 jam. *E. coli* lives in human intestine. Contamination of food material could occur if sanitation and hygiene during processing is not hygienically carried out²⁵. Contamination prevention effort can be done through development of worker's understanding and awareness on benefit of sanitation. *Staphylococcus aureus* is gram positive bacteria that has higher tolerability than other pathogenic bacteria. This bacterium can live in the medium with 0.86 water content and produce toxin at aw of 0.92. *S. aureus* lives at the surface of human's skin, nail, and respiratory tract. Processed product through heating process is easily contaminated by these bacteria on processor's hand. Beside that, storage at optimum tolerable temperature can promote the growth of the bacteria. *Salmonella* is facultative anaerobic gram negative bacterium, unspore, rod-shaped and motile. Food materials from animal and water disposal is known as good media for this bacterial development. *Salmonella* can live in human's and animal's intestines and develop into colonies in excretion. Transmission of *Salmonella* can occur during the transport of raw materials and the processing.

Conclusion

Chemical composition of the sandfish smoked with nutmeg shell contained 25.14%, 60.67% protein, 2.02% fat, and 16.38% ash, while those smoked with coconut shell had 24.44% water, 64.97% protein, 2.04% fat, and 16.81% ash. The lowest water content and the highest protein content were found in the sandfish smoked with coconut shell. The sandfish smoked with nutmeg shell held 0.033 mg/kg mercury, 1.79 mg/L Cd, 3.50 mg/L Pb, while those smoked with coconut shell had 0.042 mg/kg mercury, 0.84 mg/L Cd, and 7.71 mg/L Pb. The lowest mercury (Hg), Cadmium (Cd), and Plumbum (Cd) concentrations were recorded in the sandfish smoked with nutmeg shell.

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