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Application of Distilled Liquid Smoke on the Chemical Characteristics of Skipjack Fish Sausage

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ABSTRACT

Smoked sausage is a popular food among people due to its distinctive aroma and taste. Generally, smoked sausages are made using traditional smoking methods. Since the advent of liquid smoke technology, the processing of smoked fish sausages can be done more practically and safely by immersing them in liquid smoke. The aim of this research was to determine the effect of the application of liquid smoke quality and liquid smoke concentration on the chemical characteristics of liquid smoked skipjack tuna sausage. The experiment was executed using a factorial CRD (Completely Randomized Design) with two factors: the quality of liquid smoke, consisting of grade 1, grade 2, and grade 3; and the concentration, consisting of 15%, 25%, and 35% by volume. The results of the study showed that the quality of liquid smoke grades 2, along with a liquid smoke concentration of 25%, can significantly increase the water content, pH, protein content, total acid, and the texture of liquid smoked skipjack tuna fish sausage.

1. INTRODUCTION

One of the processed food products that is widely known and liked by everyone is sausages. Sausage is a ready-to-eat food that has high nutritional content, chewy texture, and has an elongated round shape (Nalendrya *et al.,* 2016). According to Anggraeni & Widjanarko (2014), Indonesian people's consumption of sausages increases annually by of 4.46% in average. In general, sausages are made from beef or chicken. The obstacle in making beef or chicken sausages is the relatively expensive price of meat as a raw material. For this reason, it is essential to diversify fish-based products. The choice of fish is due to its high content of omega-3 and unsaturated fatty acids which are not found in beef or chicken as well as its large potential and more affordable price (Nalendrya *et al.,* 2016).

Nowadays, processed products made from fish meat are often found. *Cakalang* fish or skipjack tuna (*Katsuwonus pelamis*) is one of the fisheries commodities with great demand in Indonesian and has become an important export commodity. The high nutritional content of skipjack tuna and the relatively cheap selling price are one of the reasons why people like to consume skipjack tuna. Therefore, skipjack tuna has good potential to be developed into processed meat products, one of which is smoked skipjack tuna sausage. Smoked sausage is a type of sausage whose processing is carried out by smoking. Smoked food products generally have a distinctive taste and aroma that comes from phenolic compounds, organic acids and aldehydes contained in the wood used for smoking. Providing smoke aroma with liquid smoke is a safe smoking method, because in the liquid smoke manufacturing process toxic particles and carcinogenic compounds, which may be present in liquid smoke, have been removed through a purification process (Sulistijowati *et al*., 2011).

The utilization of liquid smoke has been widely applied in the manufacture of food products to provide distinctive aroma, taste, color and texture for several products including cheese, fish, and meat (Soldera *et al*., 2008). The application of liquid smoke to products can be done in several ways, namely by soaking or immersing the product, adding it to the mixture, or adding liquid smoke to water during boiling. According to Abustam & Ali (2010), the addition of liquid smoke with a concentration of 0.75% by volume to meatballs from Bali beef muscle can increase tenderness, elasticity, chewiness, and panelists' preferences. Ernawati (2015) also concluded that African catfish sausages dipped in liquid smoke with a concentration of 20% for 30 min produced sausages with a taste, aroma, color and texture that were more popular with panelists. Sukandar *et al*. (2017) stated that the best treatment of liquid smoke for African catfish sausages was given by concentration of 20% liquid smoke which produced sausages with the sensory characteristics of a distinctive and not overpowering aroma, a distinctive smoky taste and a light brown appearance. The consequences of adding liquid smoke on the sensory characteristics of skipjack tuna sausage has not been widely studied. Therefore, research was conducted to determine the effect of different liquid smoke concentrations and liquid smoke qualities on the quality and sensory attributes of skipjack tuna liquid smoked sausage.

2. MATERIALS AND METHODS

2.1. Research Materials

Research materials included: kusambi wood (*Schleichera oleosa*) for making liquid smoke, fresh skipjack tuna fillets and chemicals for analyzing the quality of liquid smoked sausages, namely; 50% NaOH 0.1N, buffer pH 7-14, 90ml NaCl 0.9%, HCL 0.1N, 30ml H_3BO_3, 3 drops of BCG-MR indicator, and 90ml distilled water. The equipment used includes: pyrolyzer and liquid smoke distillation (purification) equipment to obtain liquid smoke grades, namely grade 1 (G1), grade 2 (G2), and grade 3 (G3).

2.2. Treatment and Experimental Design

This research used an experimental method designed with a Completely Randomized Factorial Design. The factor tested was the liquid smoke quality factor (G), which consisted of three levels, namely grade 1 (G1: 3 times purification stages); grade 2 (G2, 2 times purification stages), and grade 3 (G3, 1 purification stage). The second factor was the concentration of liquid smoke (K), % volume in water, with three levels, namely K1 (liquid smoke 15%); K2 (liquid smoke 25%); and K3 (liquid smoke 35%). In total there were 9 treatment combinations, each repeated 2 times.

2.3. Research Procedure

The research was carried out at the Agricultural Products Technology Laboratory, Kupang State Agricultural Polytechnic, and the Office of Drugs and Food Surveillance (*Balai POM*) Kupang City, which took place from April to September 2023. The research was carried out in three stages, namely:

a. Production and purification (distillation) of liquid smoke. The raw materials for making liquid smoke wer kusambi wood. The liquid smoke was made by pyrolysis the wood based on methods conducted by Bora *et al*. (2022). The liquid smoke obtained from pyrolysis was then purified using the distillation method to remove the tar levels in the liquid smoke (Darmadji, 2002). The distillation process is carried out in 3 stages according to get different grades of liquid smoke, namely for grade 3 the distillation was carried out once at a temperature of 110-120 °C, for grade 2 the distillation process was carried out twice at a temperature of 130-145 °C, and grade 1 was carried out three times at temperature of 170-190 °C. The distillation results of each grade are collected in a flask. Next, liquid smoke analysis was carried out to determine its chemical properties.

b. The stage of making smoked fish by applying treatments. This stage was carried out to determine the concentration of liquid smoke at each grade according to the nine (9) treatment combinations, with two repetitions so that there are 18 experimental units. The concentration of liquid smoke for each grade (grade 3, 2, and 1) was made according to the design treatments. This was performed by adding liquid smoke one liter of water according to the concentration treatment (15% volume, 25% volume and 35% volume). Next, the fresh skipjack fillets were washed and drained, then soaked in each combination of liquid smoke solution treatment for 30 min and then dried in oven at 75 °C for 2 h. After heating, the smoked skipjack fillets were cooled before being processed into sausage.

c. Stages of making sausage. Smoked skipjack tuna was cut into pieces and pureed using a blender. Next, the ground fish meat was weighed according to the composition, namely 80% smoked fish, 10% tapioca flour, and 10% wheat flour, as well as additional ingredients in the form of 100g white egg, 3% powdered sugar, 2g floured pepper, salt 1g, vegetable oil 20g, and ice water 150 ml. All ingredients were ground and mixed until a homogeneous dough was formed. The dough was put into the casing, then the sausage was tied 10 cm long and steamed for 30 min. The cooked sausages were drained and were cooled to room temperature by airing them, then opened the casings. Next, the liquid smoked skipjack tuna sausage was analyzed to evaluate its chemical characteristics.

2.4. Observation variables

Observations were made on the quality of liquid smoke, including the pH of liquid smoke (pH meter), total acid content and phenol content of liquid smoke using the titration method. Whereas, the quality of liquid smoked skipjack sausage included chemical and physical characteristics such as pH (using pH meter), water content (%, using gravimetry), protein (%, using spectrometer of Lawly method), texture (g/mm, using texture analyzer), and total acid (%, using titration method).

2.5. Data analysis method

The data obtained from the measurement was analyzed using ANOVA (analysis of variance) in accordance with the experimental design model. To test differences between treatment variables whose influence was significant, the average value was further tested using the Duncan test at the $\alpha = 5\%$ level.

3. RESEARCH RESULTS AND DISCUSSION

3.1. Chemical Composition of Liquid Smoke

The chemical constituent of the liquid smoke is a decisive element of the quality of the liquid smoke as a preservative medium. Results from the chemical analysis of the liquid smoke composition are presented in Table 1 for pH, acid content, as well as phenol content. It shows that there are differences in the quality of liquid smoke for each grade. The average pH of kusambi wood liquid smoke resulting from distillation once (grade 3) has a pH of 3.18 which is more acidic than the pH of liquid smoke resulting from distillation twice (grade 2) which shows a pH of 2.90 and grade 1 (distillation 3 times) produces a pH of 2.82 which is more acidic than grade 3. Likewise, phenol content and acid content, it can be seen that grade 3 kusambi wood liquid smoke, with 1 distillation, shows lower phenol content and acid content, namely 3.15% and 14 .62% compared to grade 2 liquid smoke which has higher phenol and acid levels, namely 5.12% and 18.57%, followed by grade 1 at 5.76% and 20.65%. The results of the analysis of phenol content in this study show that liquid acid grade 2 and grade 1 are in accordance with the permitted liquid smoke standards, namely a good phenol content of 4-15% as required in SNI number 01-2725-2009 on liquid smoke (BSN, 2009). According to Darmadji (2002) and Bora *et al*. (2022) organic acids and phenol within the liquid smoke play important role as antimicrobial ingredients, and their function will be more escalated given that the two compounds exist together at the same time. The phenol level in the liquid smoke influences the acidity, too. (Akbar, 2013) stated the liquid smoke will become more acidic with the higher phenol content.

3.2. Chemical and Physical Characteristics of Sausages

The chemical characteristics of sausages that were observed included water content, pH, protein and total acid, while the physical characteristics of sausages that were observed were only sausage elasticity. The results of the analysis of variance revealed that there was no interaction between the two factors tested, namely the quality of liquid smoke and the concentration of liquid smoke. Each single factor had a significant influence ($p<0.05$) on the chemical and physical

Table 1. Quality of liquid smoke produced from pyrolysis of kusambi wood

Quality of liquid smoke	pH	Phenol content (%)	Acid content (%)
Grade 3	3.18	3.15	14.62
Grade 2	2.90	5.12	18.57
Grade 1	2.82	5.76	20.65

Table 2. Chemical and physical properties of liquid smoked skipjack sausage on the influence of quality and concentration of liquid smoke

Treatment factor	Water content (%)	pH	Protein (%)	Total acid (%)	Elasticity (g/mm)
Quality of liquid smoke					
Grade 1	58.63^{b} ±2.05	4.06^{b} ±1.05	19.88^{b} ±2.87	20.25^{a} ±8.84	74.97^{a} ±4.13
Grade 2	60.78^{a} ±2.37	4.59^{b} ±1.90	21.35^{a} ±2.67	18.53^{a} ±8.14	73.81^{a} ±7.61
Grade 3	63.58^{a} ±2.15	5.20^{a} ±0.41	21.67^{a} ±2.89	16.64^{b} ±7.07	71.19^{b} ±7.33
Concentration of liquid smoke (% vol.)					
15%	65.43^{a} ±1.95	4.96^{a} ±1.48	23.31^{a} ±0.50	15.68^{c} ±4.45	71.92^{b} ±9.22
25%	63.61^{b} ±2.15	4.66^{b} ±1.62	22.48^{b} ±1.11	18.77^{b} ±5.68	72.70^{b} ±8.94
35%	61.75^{c} ±1.87	4.23^{c} ±2.28	20.88^{c} ±1.27	20.97^{a} ±6.21	75.35^{a} ±6.23

Note: Values tailed by different letters in the identical column designate significant differences according to Duncan's test at the 95% significance level; numbers in the table are average value along with standard deviation of two measurement.

characteristics of liquid smoked skipjack tuna sausage. The results of the analysis of the chemical and physical characteristics of liquid smoked skipjack tuna sausage are presented in Table 2.

3.2.1. Water Content of the Sausage

Water content is a determining indicator of food quality. The single factor of liquid smoke quality and liquid smoke concentration in this study had a significant effect ($p<0.05$) on the water content of liquid smoked skipjack tuna sausage. The grade 3 quality of liquid smoke treatment showed that the water content of skipjack tuna sausage was higher at 63.58% and was significantly different from the Grade 1 (58.63%) and Grade 2 (60.78%) treatments (Table 2). It is suspected that the more repeated the distillation process with hot temperatures, the water content will decrease due to evaporation of water from the liquid smoke. According to Muttakun *et al*. (2017), liquid smoke is able to bind free water in the meatball products produced.

From the aspect of water content, the skipjack tuna sausage produced in this study met the quality standards for meat sausages based on SNI requirements, namely a maximum of 67% (BSN, 2015). The same thing was also seen in the liquid smoke concentration treatment (Table 2). The 35% volume concentration treatment gave a smaller water content of 61.75% by weight, followed by a 25% volume concentration with a water content of 63.61% by weight and the 15% volume concentration produced a higher water content of 65.43% by weight and significantly different with the other two treatments. According to Herwati *et al*. (2017) the increasing concentration of liquid smoke used causes a decrease in the water content of smoked fish because osmosis occurs which causes free water in the fish flesh to be pulled out.

3.2.2. pH of the Sausage

The single factors of liquid smoke quality and liquid smoke concentration had separately significant influence ($p<0.05$) on the pH value of liquid smoked skipjack tuna sausage. The grade 3 liquid smoke (1X purification) shows a higher pH value (5.20) and is different from grade 2 and grade 1 of liquid smoke, each of which has a pH that is not significantly different, namely 4.59 and 4.06. The increase in sausage pH in grade 3 liquid smoke is due to the role of phenol compounds in the liquid acid. The phenol content in grade 3 liquid smoke is lower, 16.64%. The liquid smoke is further acidic at the greater phenol content (Akbar, 2013).

The liquid smoke concentration treatment also showed differences in the pH of liquid smoked skipjack tuna sausage. Increasing the concentration of liquid smoke from 15% to 35% significantly reduced the pH of smoked skipjack tuna sausage from 5.20% to 4.06%. The low pH value of liquid smoked skipjack tuna sausage at a liquid smoke concentration of 35% (pH: 4.23%), suggests the role of high phenolic compounds in producing a more acidic sausage product. According to Ernawati (2015), the presence of phenol compounds in liquid smoke can influence the decrease in product pH.

3.2.3. Protein Content of the Sausage

Protein is an important macromolecular element in food. Apart from that, the protein content also greatly determines the quality of liquid smoked skipjack tuna sausage products. The protein content of liquid smoked tuna sausage in this study ranged from 19.88 – 21.87% (Table 2). Both factors (liquid smoke quality and liquid smoke concentration) resulted in significant difference of protein content of the sausage. Thus, the protein content value of liquid smoked skipjack tuna sausage in this study complies to the standard because it is above the minimum limit for beef sausage protein content required by SNI, which is 13% (BSN, 2015). The quality of liquid smoke tested showed that grade 2 and 3 treatments showed higher protein levels (21.35% and 21.67%) and were significantly different from grade 1 which had a protein content of 19.88%.

The same thing was also seen with the treatment of liquid smoke concentration, where the 15% concentration gave a higher protein content of the skipjack tuna sausage (23.31%) and was significantly different from those of 25% and 35% concentrations, each of which had a concentration of 22. 48 and 20.88%. The protein content of liquid smoked skipjack tuna sausage in this study tended to decrease as the liquid smoke concentration increased. The higher the concentration of liquid smoke, the sausage protein will decrease. This is in accordance with Dwiari *et al*. (2008) and Ernawati (2012) where the presence of phenol compounds tends to react with the sulfur hydrogen of proteins which causes the proteins to denature and form new bonds which results in a decrease in the protein content of the smoked material.

3.2.4. Total Acid of the Sausage

The total acidity of liquid smoked tuna sausages in this study, both in the treatment of liquid smoke quality and liquid smoke concentration, ranged from 16.64% - 20.25% (Table 2). Grade 1 liquid smoke (3X purification) resulted in a higher total acid value (20.25%) and is not different from that grade 2 liquid smoke (18.53%), but is significantly different from that of grade 3 liquid smoke (16.64%). The high total acidity of liquid smoked skipjack tuna sausage in the grade 1 and 2 liquid smoke treatment was due to the role of phenolic compounds in the liquid acid which lowered the pH value. This is in line with other opinion that the high total acid in foodstuffs treated with liquid smoke is closely related to the pH of the liquid smoke (Ernawati, 2012; Akbar, 2013).

The liquid smoke concentration treatment also showed differences in the total acidity of liquid smoked skipjack tuna sausage. Increasing the concentration of liquid smoke from 15% by volume to 35% by volume significantly increased the total acid value of smoked skipjack tuna sausage from 15.68% to 20.97%. The high value of total sausage acid (20.97%) in the 35% liquid smoke concentration treatment was because liquid smoke, apart from having phenolic content, also had acetic, propionic, butyric and valeric acids which increased the sour taste (Hardianto & Yunianta, 2015; Bora & Gasong, 2021).

4.2.5. The Elasticity of the Sausage

Table 2 shows that the elasticity or chewiness of the sausages in this study is significantly affected by liquid smoke grades and liquid smoke concentration treatments. The liquid smoke grade treatment shows that grade 2 and grade 1 have a level of elasticity that is not significantly different and is higher, respectively 74.97g/mm and 73.81 g/mm compared to that of grade 3 which has a lower level of elasticity (71.19 g/mm). The same thing was also shown in the treatment of liquid smoke concentration (Table 2), where increasing liquid smoke concentration tends to increase the elasticity value of liquid skipjack tuna sausage, from 71.94 g/mm to 75.35 g/mm. The high elasticity of liquid smoked skipjack tuna sausage in this study is thought to be due to the role of the water content contained in liquid smoke. In this case, more purification step, there is more water evaporation due to hot temperatures.

Sulistijowati *et al*. (2011) stated that the lower the water content in smoked fish, the greater its structural value. This is because when the water content in the fish body decreases, the fish flesh becomes denser or tougher. According to Purnomo (1995), the water content and water activity of food play a very important role in determining food composition. This is in accordance with Darmaji (2002) stating that fish muscles and cells contract when water is removed from the fish after the osmotic dehydration process. The water that comes out due to the phenolic content acts as a dry structure in the fish. Furthermore, Budijanto *et al*. (2008) stated that liquid smoke is rich in carbonyl compounds but with purification it can increase phenolic derivative compounds which provide textural changes to smoked meat.

4. CONCLUSION

The chemical characteristics of liquid smoked skipjack sausage are largely determined by the quality and concentration of the liquid smoke used. The quality of grade 2 liquid smoke (with 2 purifications) shows better chemical characteristics of sausages, as indicated by water content (60.78%), pH (4.59), protein content (21.35%), total acid (18.53%, and texture (73.81g/mm), and was not significantly different from the characteristics of liquid smoked skipjack sausage in grade 1 treatment (3 times purification). Thus, grade 2 liquid smoke is suitable for preserving liquid smoked sausages because it is more efficient . The correct concentration of liquid smoke was obtained at a concentration of 25% by volume, with better chemical characteristic values for liquid smoked skipjack tuna compared to concentrations of 15% by volume and 35% by volume.

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