Organoleptic test analysis and effect of liquid smoke concentration on smoked fish

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Abstract. Fish Liquid smoke is one of the food products are preferred by Indonesian society in general and in particular Maluku region. This study aims to analyze organoleptic, TPC, Fat Content, Total Acid, Total Protein, Total Molds, Phenol levels to Concentration of Liquid Smoke Fish. The research method descriptions experimental concentration of 1\% and 3\% and smoked fish. Organoleptic analysis has a high degree of pleasure in the taste of liquid smoke 2430 codes with a value of 2.90, smell on code 2430. For textures in code 2430, 2437, 2438 with a value of 2.70, and the color in code 2438 was received by the panelist. The results of Total Plate Count (PLT) inhibit microbial growth at a concentration of 3\% for 30 minutes soaking time, analysis of total acid levels at a concentration of 3\% (57.6\%). The highest total protein level (23.05\%) concentration of 1\%, analysis of total molds at a concentration of 1\%, and analysis of phenol at a concentration of 3\% have the highest phenol content of 4.06\%.

1. Introduction

Indonesia is known as an agricultural and marine country which has a wealth of natural resources, especially of agricultural and fishery products. The agriculture and fisheries sector is an important sector as an input provider for other sectors. So that the agricultural and fisheries sectors are said to influence the structure of the Indonesian economy. Along with the development of the nation's economy, Indonesia began to proclaim the future towards the era of industrialization [1]. One of the potentials of agriculture is the coconut plant (Cocosnucifera) which has an important role in the life of Indonesian society because coconut is cultivated by most people as an annual plant that has economic and social values. Coconut is also often referred to as the tree of life because almost all parts of the plant can be used for life [2-4].

One part of the coconut that can be utilized is the shell. In general, coconut shells are used as active carbon and handicrafts [5]. Coconut shells are categorized as hardwood, but have higher levels of lignin and cellulose content is lower. Coconut shells can also be processed into a preservative that is by pyrolysis produces liquid smoke [6]. Hadanu and Apituley [6] have identified a compound component of liquid smoke (liquid smoke) from coconut shell. The results of the study found no PAH compounds in liquid smoke because the compound is not formed during the process of pyrolysis of coconut shells carried out at temperatures above 350-420\degree C. The factors that influence the formation of PAH compounds are pyrolysis temperature.
Liquid smoke coconut shell contains four types of phenolic compounds are compounds responsible for flavor liquid smoke and act as an anti-bacterial. The four compounds are 2,6-dimethoxyphenol, 1-hydroxy-2-butanol, 1-(acetyloxy)-2-butanol, 2-methoxyphenol, 1,2-benzenediol, 4-ethyl-2-methoxyphenol, 4-hydroxyacetyl-2-methyl phenol, and others [6-8]. The results of the study showed that the highest chemical component of liquid smoke produced by coconut shell pyrolysis at 227-252°C was 9-octadecanoic acid, tetradecylester or oleic acid, tetradecyl ester as much as 71.68%. Other chemical components contained in liquid smoke are groups of acetic acid, esters, phenols, alcohols, and tar [9]. The chemical compound of liquid smoke on pyrolysis temperature 337-428°C is a compound 2-lauro-1,3-didecoino as much as 37.53%. In the results of the research on the identification of chemical components of liquid smoke, PAH compounds were not found because pyrolysis was carried out at a temperature of 300-450°C.

Based on the literature above, it can be said that liquid smoke from pyrolysis above 300-420°C is free of carcinogenic PAH compounds [6], so that liquid smoke can be used as a fish as a substitute for formalin preservative sometimes used in part of the fishermen and fish traders in the Maluku islands and surrounding areas. The model for using fish preservatives from coconut shell liquid smoke needs further investigation. In case of determine product durability, organoleptic tests (taste, odor, colour, texture), analysis of bacteria at various product storage times, analysis of molds, analysis of fat content, analysis of total acid levels, and toxicity tests as indicators of the quality of food products which in turn can improve the economy of coastal communities in the Maluku islands.

2. Materials and Methods

2.1 Equipment
The equipment used in this study were pyrolysis, Erlenmeyer, separating funnels, scales, sample bottles, glassware, vacuum pumps, Gas Chromatography-Mass Spectrometry (GC-MS), and others.

2.2 Material
The materials used in this study are coconut shell, liquid smoke, dichloromethane pa (E. Merck), methanol pa (E. Merck), Na\textsubscript{2}SO\textsubscript{4} anhydrous p.a. (E. Merck), Whatman filter paper No. 42, KLT plate (aluminum plate), silica gel 40 (70-230 mesh), and others.

2.3 Procedures

2.3.1 Organoleptic test
In this oragnoleptik test using 25 panelists with four hedonic scales, Namely No 1. Dislike very much, No. 2 dislike, No. Three like and No. 4 like very much. The method used is a univariate analysis using organoleptic test result data includes an assessment of flavor, odor/smell, texture, and color of smoked fish with a preservative liquid smoke and smoked fish by using the form organoleptic test. The first step is to prepare samples of liquid smoked fish and smoked fish on the table of the presenter who has been given the code. Next, the panelists took each sample to sample each sample smoke. Panelists gave a questionnaire sheet value corresponding preference level in the two samples.

2.3.2 Bacterial analysis
The number of bacteria grown on nutrient media samples are measured using that as many as 10 mL in each petri dish and incubated for two days at 37°C.

2.3.3 Acid resistant dyes
Acid-resistant staining can be done by making topical preparations from the bacteria Mycobacterium sp. and Bacillus subtilis. Topical preparations are covered with suction paper with no excess paper objects. Blotting paper drip with ziehlneelsencarbolfuchsin solution until saturated. Topical preparations are heated over low heat (fixation) for 3-5 minutes, and then successively rinsed with...
running water for 10-30 seconds and alcohol should not be excessive. The next step preparations rinsed again with water flowing, and then pressed with methylene blue solution for 30-45 seconds then rinse again with water then the preparation is dried with suction paper.

2.3.4 Analysis of fat content
A volumetric flask that has been dried in the oven at 105°C for 1 hour cooled in a desiccator for 15 minutes and weighed (W2). Five grams of crushed sample is then weighed (W1) and wrapped in the sleeve-shaped filter paper (Thimble). The sample is inserted into Soxhlet which has been arranged in a set of extract tools. Soxhlet added into n-hexane. The extraction process is done for 6 hours until the solvent drops back into a clear-colored fat, The solvent was removed by evaporating using a rotary evaporator (50 rpm), heated in an oven at a temperature of 105°C for 1 hour. The volumetric flask was cooled in a desiccator for 15 minutes and weighed (W3). Warm up back in the oven for 1 hour, if the difference between the weighing results with the last extraction has not yet reached the previous weighing 0.0002 gram. Fat content is calculated by the formula:
\[ \text{Fat Content} = \frac{W_3 - W_2}{W_1} \times 100\% \]  

2.3.5 Total acid analysis
Substances were then weighed as much as 5 gr, and included in 100 ml of volumetric flask, then added distilled water up to the mark tera, Piped as much as 25 mL and put in Erlenmeyer added 3 indicators, then titrated with 0.1 N NaOH solution to form a pink colour, then note how much volume (mL) NaOH is used. Total acid content calculated using the formula:
\[ \text{Total Acid} = \frac{\text{ml NaOH} \times N \text{ NaOH} \times \text{Grek x fD}}{\text{Weight} \times 1000} \times 100\% \]  

2.3.6 Analysis of molds
Morphological analysis of mold/fungi through macroscopic observation. Among the morphological analysis of mold/fungi was carried out on the genus Aspergillus sp. (BGLB). The initial stage was moved a little culture from a tube containing a pure culture with a sharp needle into a petri dish containing medium BGLB. Then incubated for 4-15 days at room temperature. Observation of mushroom growth is done every day. If there is a color change that occurs in the colony, the surface condition of the colonies (flat, flat, like flour, granules, beledu/like cotton) is still recorded for the absence of kleistotiesia, the presence or absence of drops of esksudat, as well as the color, the presence or absence of a distinctive smell, also the condition of the back of the colony. The final stage of macroscopic analysis of mold/morphology is the depiction of mold/fungal morphological forms as observed.

2.3.7 Phenol rate test
Mashed Liquid smoke fish is inserted into volumetric flask then added 10 ml of dichloromethane and then shaken out briefly. Samples were allowed to stand for 1 hour and then taken into the bottom fraction Erlenmeyer. Added another 10 mL of dichloromethane, shaken, and allowed to stand for 1 hour. Then take the bottom fraction and add it to the first one, and filtered with a filter paper and then added Na₂SO₄. The filter results are ready for GC-MS identification.

3. Results and Discussion

3.1 Organoleptic test
Univariate analysis in this study was used to determine organoleptic criteria. The organoleptic test data includes an assessment of the taste, odor/smell, texture, and color of liquid smoked fish, and smoked fish using an organoleptic test form. The results of organoleptic tests of liquid smoked fish and smoked fish after averaging showed that the panelists had different levels of preference.
Table 1. Organoleptic test result

| Code | Taste | Odor | Texture | Color |
|------|-------|------|---------|-------|
|      | Fish Liquid Smoke | Smoked Fish | Fish Liquid Smoke | Smoked Fish | Fish Liquid Smoke | Smoked Fish |
| 2430 | 2.90 | 2.65 | 2.85 | 2.75 | 2.70 | 2.55 | 2.65 | 2.35 |
| 2431 | 2.55 | 2.60 | 2.70 | 2.85 | 2.50 | 2.60 | 2.45 | 2.50 |
| 2432 | 2.55 | 2.35 | 2.65 | 2.50 | 2.65 | 2.40 | 2.55 | 2.55 |
| 2433 | 2.55 | 2.40 | 2.65 | 2.55 | 2.60 | 2.55 | 2.45 | 2.45 |
| 2434 | 2.60 | 2.35 | 2.60 | 2.40 | 2.60 | 2.35 | 2.65 | 2.35 |
| 2435 | 2.40 | 2.55 | 2.50 | 2.60 | 2.55 | 2.60 | 2.60 | 2.40 |
| 2436 | 2.75 | 2.60 | 2.80 | 2.65 | 2.65 | 2.40 | 2.60 | 2.40 |
| 2437 | 2.80 | 2.60 | 2.80 | 2.55 | 2.70 | 2.35 | 2.65 | 2.45 |
| 2438 | 2.55 | 2.05 | 2.70 | 2.45 | 2.70 | 2.35 | 2.75 | 2.05 |
| 2439 | 2.45 | 2.20 | 2.55 | 2.35 | 2.55 | 2.45 | 2.30 | 2.45 |
| 2440 | 2.50 | 2.30 | 2.45 | 2.55 | 2.60 | 2.40 | 2.30 | 2.30 |
| 2441 | 2.65 | 2.30 | 2.65 | 2.35 | 2.40 | 2.45 | 2.35 | 2.45 |
| Average | 2.60 | 2.41 | 2.66 | 2.54 | 2.60 | 2.45 | 2.52 | 2.39 |
| SD | 0.15 | 0.12 | 0.12 | 0.12 | 0.09 | 0.10 | 0.15 | 0.13 |

3.1.1 Test of the taste

The results of organoleptic tests on the taste of liquid smoked fish (concentration 3%) has the highest value of 2.90 and the lowest value of 2.40 with an average of 2.60 and standard deviation 0.15 while smoked fish has the highest value of 2.65 and the lowest value of 2.05, with an average of 2.41 and standard deviation 0.18. The results of organoleptic tests show that consumer acceptance of the taste of liquid smoked fish (concentration 3%) is very good. From this analysis shows that each tested sample has a real effect, this is evidenced by the results of the taste test panelist ratings are very strong in smoked fish liquid (concentration of 3%), when compared with smoked fish. In this case, the consumer believes that the more the 3% solution of liquid smoke is added, then the taste of liquid smoked fish is more delicious according to the value 2.90. This can be confirmed by Arizona [10] states that the flavor is a psychological suggestion to food that determines the value of satisfaction to those who eat them and have.

3.1.2 Odor/smell

Odor test results show that the odor/smell of liquid smoked fish is preferred at a concentration of 3% has a value of 2.85 with the preference rather likes and dislikes. From the analysis shows that each sample given 3% concentrated liquid smoke has a real effect, because the level of preference of the panelists to the concentration of liquid smoke is 3%, compared to liquid smoke fish with a concentration of 1%, and smoked fish. This was confirmed by Ilmi [11] stated that aroma is one of the important factors in determining the quality of food ingredients. In the food industry, odor testing is considered important because it can provide an assessment of the results of its products, whether the product is liked or disliked by consumers. The taste parameters differ from the aroma and more involve the taste of tasting. Taste is influenced by several factors, including the content of chemical compounds, temperature, concentration, and interactions with other taste components [12-13] confirmed that odors could be detected from near and far because humans can recognize the taste of food that has not been seen only by detecting odors from afar. This serves to assess the smell of a product or commodity in the form of food or non-food.

3.1.3 Texture

The test results of the texture of liquid smoked fish and smoked fish obtained different textures. According to Wibawanti and Rinawidiastuti [14], texture affects the taste of these foods. The texture
test results of liquid smoked fish obtained the highest value of 2.70 and the lowest value of 2.40 with an average of 2.60 and standard deviation 0.09 while smoked fish obtained the highest value of 2.60 and the lowest value of 2.35 with an average of 2.45 and standard deviation 0.1. The texture test results show the texture of liquid smoked fish is preferred at a concentration of 3%. Panelists argued that liquid smoked fish had the best texture with a value of 2.70 compared to the texture of smoked fish, which obtained the lowest value of 2.35. This was confirmed by Amo [15] stating that texture is a thing that can be seen and felt (with fingers) on a surface or object. According to Amo [15], the texture is a group of physical properties caused by structural elements of food which can be felt by the sense of touch, related to deformation, disintegration, and flow of food under pressure which is measured objectively by the function of mass, time, and distance.

3.1.4 Color
Color test results of this study are in the category of light brown to dark brown color. The results of the color test on liquid smoked fish obtained the highest value of 2.75 and the lowest value of 2.30 with an average of 2.52 and standard deviation 0.15 while smoked fish obtained the highest value of 2.55 and the lowest value of 2.05 with an average of 2.39 and standard deviation 0.12. The results of the color test show that the color of the most preferred product by panelists is the color of liquid smoke fish with a value of 2.75, while for the lowest value is the smoked fish with a value of 2.05. Color is one of the visual factors that determine the acceptance of a product. Food is considered nutritious, delicious, and the texture is very good, sometimes it is not liked when it has a bad color [15]. The organoleptic criteria for sensing the color of a material are a form or quality of a commodity from a food ingredient, for example, the green color of fruit indicates that it is immature, still acidic, or tight [14, 16].

3.2 Bacterial analysis

3.2.1 Total TPC method bacteriastest
The measurement results in the number of bacteria in liquid smoked fish and smoked fish after being stored in a certain time. Analysis Result from the concentration of liquid smoke 3% with 30 minutes immersion time has a value of the number of bacteria 1.11 (CFU / ml) x 103/gr. The growth rate of microorganisms is measured by the method of Total Plate Count (TPC). In this study, it was shown that the use of small concentrations of liquid smoke had a faster impact on microbial growth. In addition to liquid smoke concentration, soaking time also gives effect to the growth of microbes. The longer the time of immersion increasingly inhibit microbial growth at the optimum condition in which microbes total as much as 1.11 x 103 CFU/g occurred 3% concentration and immersion time of 30 minutes.

While the sample without liquid smoke, there was a very rapid microbial growth ranges from 10.92 x 10-2 CFU/g. Determination of total bacteria aims to see the number of colonies, the character of growth in each of the smoked fish microorganisms, the form of morphology, structure, and typical characteristics of bacteria. Bacteria that live almost no color and contrast in smoked fish, in which the bacterial cells are suspended. One way to see and observe the shape of bacterial cells in living conditions is very difficult so that it must be identified by the method of painting or staining bacteria cells so that cells can be seen clearly and easily observed. It also functions to determine the physiological properties of knowing the reaction of the bacterial cell wall through a series of stains.

3.3 Analysis of fat content
The results of the analysis of fat content were obtained from a combination of the treatment of the amount of concentration and the time of immersion with liquid smoke have the highest fat content (60%) are found in liquid smoke fish with a concentration of 1% with a soaking time of 10 minutes, followed by a concentration of 2% with a soaking time of 10 minutes and the lowest (28%) at 3% concentration with 30 minutes soaking time.
3.4 Anaylyze total acid content
The results of the analysis of the total acid content obtained from the combination of the amount of concentration and soaking time with liquid smoke. Highest levels of total acid (57.6%) was obtained at a concentration of 3% at the time of soaking 30 minutes, followed by a 2% concentration with soaking time 30 minutes (43.8%) and the lowest (14.2%) at a concentration of 1% and 2% with 10 minutes soaking time.

3.5 Analysis of total protein levels
The highest concentration of liquid fatty fish protein (23.05%) was obtained at 1% concentration, followed by 2% and 3% liquid smoke concentration, i.e. 22.35% and 21.06% respectively. Protein content in liquid smoke concentration 3% lower than protein content in liquid smoke fish concentration 1% and 2%, this is caused by the acidity of liquid smoke which can damage the protein present in liquid smoke fish.

3.6 Total Mold Test
The total mold test is carried out to determine the amount of mold in the food ingredients after being stored for a certain time. Data on total bacterial test results can be shown total levels of mold at the highest liquid smoked fish (0.1400%) obtained at 1% concentration and immersion time 10 minutes, then followed by a concentration of 1% liquid smoke with a soaking time of 20 minutes and total mold with the smallest concentration (0.0224%) in the concentration of liquid smoke 3% with a soaking time of 20 minutes.

3.7 Analysis of Phenolic Content of Liquid Smoke Fish
The highest levels of total phenols (4.06%) obtained at a concentration of 3% smoked fish, followed by successive concentrations of liquid smoke 2% and 1%, which has consecutive phenol content of 2.35% and 2.05%.

4. Conclusions
Based on the result of the research, it can be concluded that liquid smoke fish and smoked fish have a significant effect on the organoleptic test by using a hedonic scale, and from the results of this study can say that the higher the concentration and duration of immersion using liquid smoke, then the smoked fish sample products are more durable and long lasting. The organoleptic test result, total bacterial test dan fat content showed that liquid smoke fish with a concentration of 3% had high quality when compared to 1% liquid smoke fish and smoked fish. The acid test results and the phenol test showed that the smoked fish that have high levels of acid and phenol in fish liquid smoke concentration 3%, otherwise the test result of total protein levels showed that sample which has the highest protein content found in the liquid smoked-fish concentration of 1%. Based on the several conclusions above, the model of liquid smoke administration in the smoked fish production process can increase economic value in the coastal communities of the islands of Maluku Province.

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