Flavor characteristics and quality of mackerel (Scromberomorus commersonii) “otak-otak” affected by various liquid smoke addition methods

D A Susbandi1*, F Swastawati1 and S Suharto1

1Department of Fisheries Processing Technology, Faculty of Fisheries and Marine Science, Diponegoro University, Indonesia

*E-mail: deadessy@gmail.com

Abstract. The efforts to utilize fishery resources by processing fish is needed to fulfill food nutrition, such as “otak-otak” made from mackerel (Scromberomorus commersonii). However, “otak-otak” has a relatively short shelf-life; thus, preservatives are needed. Liquid smoke of coconut shell is used because it contains phenols which has antibacterial activities. This study aimed to determine the effect of liquid smoke addition to sensory and chemical characteristics on the “otak-otak” and to determine antibacterial activities of liquid smoke using Total Plate Count (TPC). Treatments used were adding 3% liquid smoke (control, mixing, boiling, soaking) and two factors addition of liquid smoke and duration of storage time at ±4°C for 0, 8 and 12 days. The results showed that the variation had a significant effect on hedonic testing. The chemical characteristics of the phenol, moisture, and fat levels had a significant effect on the control method with all treatments, but not in the mixing treatment. Treatment without liquid smoke on the 12th day were unacceptable with a TPC value of 4.97±0.02 log CFU/g, while in mixing, boiling, soaking liquid smoke 3% for 12 days were still acceptable with a value of 4.29±0.1 log CFU/g, 4.55±0.04 log CFU/g, 4.39±0.07 log CFU/g.

Keywords: liquid smoke, mackerel, otak-otak

1. Introduction

Mackerel (Scromberomorus commersonii) has the potential to fulfill food nutrition. One of the fisheries diversification products is “otak-otak” (spicy ground fish dough added with flour then boiled and drained) made from mackerel. Based on the National Standards Agency about fish “otak-otak”, it is a processed product supposed to contain fish meat or surimi paste of at least 30% against flour and other ingredients, with or without vegetables and coconut milk who made formation, with or without leaves and cooking [1].

However, “otak-otak” has a relatively short shelf life, so preservative additives are needed to make it last. The effort that can be done is adding natural preservatives like liquid smoke. Liquid smoke contains chemical compounds and has an antimicrobial function and it is quite safe as a natural preservative [2]. There are several options for adding liquid smoke to food products, such as spraying, soaking, boiling, dyeing, or mixing in the ingredients.

The purpose of this study was to determine the effect of liquid smoke addition to sensory characteristics (appearance, aroma, taste, and texture) and chemical characteristics (moisture, protein,
fat, phenol, and pH) on the “otak-otak” and to find out antibacterial activities of liquid smoke using Total Plate Count (TPC).

2. Materials and Methods

The materials used in this study were fresh mackerel, coconut shell liquid smoke, tapioca flour, coconut milk, eggs and kitchen spices. Materials for chemical analysis include aquadest, Follin Denis, Na₂CO₃ solution, fat solvent, selenium, H₂SO₄, 40% NaOH, 2% H₃BO₃, 0.1% bromcresol green indicator, and 0.1% methyl red. The tools used for making “otak-otak” consisted of knives, cutting boards, basins, steaming pans, food processors, and the tools for analysis consisted of ovens, kjeldahl apparatus, soxlet apparatus, and digital scales.

The research was carried out in two phases, namely, preliminary research to determine the critical point of decline in the product quality of “otak-otak” stored at ±4°C and the main research. The first step in the process of making “otak-otak” is to have the fish washed, weeded, washed again, then milled. After that, put it in a food processor and mix it with spices and additional ingredients so that it becomes the mixture of “otak-otak” that has been mixed. The mixture is then separated into 4 parts, transferred into each basin to be treated with the addition of liquid smoke 3%. The treatment given was control (without liquid smoke), mixing liquid smoke 3% into the mixture, adding liquid smoke 3% into boiling water, and soaking in liquid smoke solution 3%. The dough printing process was then boiled with each treatment. The length of the “otak” boiling process ranges between 10-15 minutes. The “otak-otak” is then boiled until cooked, which is characterized by the product floating on the surface of boiled water and soft [3]. The process of making it and sensory testing was carried out at the Processing Laboratory, Fisheries Product Technology, Faculty of Fisheries and Marine Sciences, Diponegoro University.

The research was carried out in an experimental laboratory and the experimental design used was Completely Randomized Design (CRD) for sensory characteristics (appearance, aroma, taste, and texture) and chemical characteristics (water content, protein, fat, and phenol) consisting of 1 factor namely the method of adding liquid smoke 3% (control, mixing, boiling, soaking) in the mackerel “otak-otak” and RAL fish in factorial patterns for pH and Total Plate Count (TPC) which consists of 2 factors, namely variations in the method of adding liquid smoke and variations in storage duration at temperatures of ±4°C (0, 8 and 12 days). Statistical data analysis with ANOVA at α = 5% and continued with BNJ test in the event of a real difference. This research was conducted in April 2018. Testing of water, protein, fat content, and pH were carried out in the Laboratory of Analysis, Fisheries Product Technology, Faculty of Fisheries and Marine Sciences, Diponegoro University. TPC testing was conducted at the Laboratory of Microbiology, Fisheries Product Technology, Faculty of Fisheries and Marine Sciences, Diponegoro University. Testing of phenol levels was carried out in Chem-mix Laboratory, Yogyakarta.

3. Results and Discussion

Preliminary research results obtained from sensory observations of “otak-otak” with different methods of liquid smoke addition showed that the critical point of decline in Otak-Otak product quality occurred on the 11th day in samples of “otak-otak” with control treatment (smokeless liquid).

3.1. Hedonic testing

Hedonic testing is a test of preference which is very important for a product because it is related to consumer acceptance of the product. The purpose of the hedonic test was to determine the level of panelists' preference for fish brain products which added 3% liquid smoke.

As showed in table 1, it can be concluded that the addition of liquid smoke affects the level of panelists' preference. The results of the Kruskal-Wallis non-parametric test analysis showed significant differences between treatments in the hedonic test (p<0.05).
Table 1. Average value of hedonic test “otak-otak” mackerel of 3% liquid smoke.

| Treatment   | Parameter | Appearance | Aroma  | Taste | Texture |
|-------------|-----------|------------|--------|-------|---------|
| Control (K) |           | 7.27±0.77  | 6.77±1.17  | 7.03±1.02  | 7.13±0.72  |
| Mixing (A)  |           | 7.10±0.70  | 6.07±1.19  | 5.40±1.15  | 6.53±0.56  |
| Boiling (B) |           | 7.57±0.76  | 7.00±1.10  | 6.67±1.17  | 7.00±0.58  |
| Soaking (C) |           | 6.40±0.80  | 7.40±1.14  | 7.93±0.77  | 6.93±0.77  |

Information:
This data is the average result of thirty panelists ± standard deviations.
• Data followed by different superscript letters indicate significant differences (P<0.05)

The appearance of the product is the first impression of whether or not the product is attractive and suitable for consumption. The first impression felt by consumers when looking at a product is usually through the appearance of the product and generally consumers prefer products that have an attractive appearance [4]. Then the Mann-Whitney test showed treatment B was significantly different from the treatment of adding liquid smoke in other ways (A and C), but not significantly different from the treatment without liquid smoke. It can be concluded that the addition of 3% liquid smoke in the products of “otak-otak” affects the appearance of the product especially the color appearance. Liquid smoke contains carbonyl compounds which have a role in the coloring and taste of the product [5]. The colour according to the criteria of the meatballs should be whitish or gray, but with the addition of liquid smoke the meatballs turned increasingly brown with the increase of the concentration of liquid smoke used.

Odor testing is considered important in the food industry because it can quickly provide an assessment of whether or not a product is acceptable. Aroma is a smell sensation arising from the stimulation of chemical compounds. The distinct aroma of smoke will be stronger with the increase of the amount of liquid smoke in the product. In addition, liquid smoke can also reduce fishy odor. Based on table 1, panelists preferred the “otak-otak” that does not smell fishy and does not smell too much of liquid smoke. Phenolic compounds contained in liquid smoke consist of various components that have the ability to provide smoky scents that are specific to the product [6].

Then the Mann-Whitney test showed that treatment B was most preferred and significantly different from treatment K and A, but not significantly different from treatment C. The method of adding 3% concentration of liquid smoke that attaches it to the product is also different. The strength of the smell of liquid smoke was also determined by the amount of liquid smoke attached. Phenol compounds in liquid smoke play a role in aroma formation [7]. The higher the concentration of liquid smoke, the more intense the smoke produced will be, even though the results were not significantly different [8].

Taste testing was aimed to determine the level of consumer preference for the taste of mackerel “otak-otak” which was added with 3% liquid smoke in different ways. Sense is a quality attribute of a product which is usually an important factor for consumers in choosing a product [8]. A product can be accepted by consumers if it has the desired taste. Based on table 1, these treatments had a variation of effects on the taste of “otak-otak”. Panelists preferred the immersion treatment in liquid smoke solution 3% and at least like the treatment of 3% liquid smoke mixing into the dough. Differences in panelist values could be caused by each panelist having a different level of preference for taste. Taste gives a strong stimulus to the level of panelist preference [9]. Variety of preference among each treatment may have been caused by different phenol content. The phenol fraction is very important in flavoring smoked fish [7]. The higher the level of phenol in smoked fish, the stronger the taste of smoke.

The assessment of texture was done by assessing the smoothness and elasticity of the product and is one of the parameters of the combination of the physical state of a food product and sensed by the sight and touch. It can be seen in table 1, that those treatments have varied effects on the texture of “otak-otak”, although the results of the Mann-Whitney advanced test analysis showed that they were not significantly different. Differences in the level of preference for “otak-otak” can be influenced by
the processing methods and ingredients added. Different addition methods will affect the compounds in liquid smoke contained in the “otak-otak”, and thus can affect the water content and the texture of the product. Previous study shows that meatball texture is influenced by the quality and quantity of meat used, processing methods used/adopted, and ingredients added [10]. Thus increasing tenderness seemed to be a reflection of higher water content and water holding capacity.

Analysis of variance (ANOVA) test showed the different methods of adding 3% liquid smoke to the “otak-otak” showed significant differences in phenol levels (P<0.05). The highest phenol content was found in the product added 3% of liquid smoke by mixing (556 ppm) and the lowest was in the control (without liquid smoke) (464 ppm) (figure 1). This value is still below the maximum limit of phenol levels in food. Safe Phenol concentration limits in food are 0.02 - 0.1% or 200 - 1000 ppm [8].

3.2. Phenols

![Figure 1. Phenol value of “otak-otak” mackerel that adding 3% liquid smoke.](image)

The addition with mixing method has the highest phenol content because the liquid smoke 3% added was pure with the weight of the dough while the soaked “otak-otak” into liquid smoke 3% in 30 minutes also has no different phenol content. The soaking length will cause the amount of phenol content attached on “otak-otak”. Results of previous research showed that smoked catfish sausage’s phenol content tend to increase with the increase of liquid smoke concentration and the length of soaking increased phenol content [11]. Meanwhile liquid smoke addition with boiling results in the lowest phenol content. This is because liquid smoke solution 3% in boiled water vaporized. Phenol degradation into some phenol short chain resulted in low boiling point [8].

3.3. Proximate composition

The results of analysis of variance (ANOVA) for proximate composition in the “otak-otak” with different methods of adding 3% liquid smoke showed that moisture content, protein and lipid were significantly different (P<0.05) (figure 2). It was indicated that the changes in the proximate composition of “otak-otak” were affected by the chemical compounds of liquid smoke.
Figure 2. Proximate value of “otak-otak” mackerel adding 3% liquid smoke, control, mixing, boiling, soaking.

Based on figure 2, there were different proximate values of “otak-otak” with different methods of liquid smoke adding. It could be influenced by the presence of phenol compounds in liquid smoke contained in each treatment. These values are in accordance with the SNI [1].

The highest moisture content was the control treatment (without liquid smoke) of 59.33% and the treatment of addition of liquid smoke 3% by mixing was the lowest with 54.08% (figure 2). Phenol compounds in liquid smoke attached to “otak-otak” binds the free water in the product, so that the higher the phenol content, the lower the water content of the product. Previous result showed that the greater the concentration of liquid smoke, the smaller the water content value [12]. This is because liquid smoke was able to bind free water that is present in fish during the processing. Decreased levels of smoked skipjack fish fillets were caused by a solution of liquid smoke that seeps into fish meat by osmosis, so that the free water in the flesh was pushed out [13]. Thus, the amount of free water in fish meat was reduced. The amount of liquid smoke that seeps into the flesh of fish increases with the increasing concentration of liquid smoke.

The highest protein content was found in the control treatment (without liquid smoke) with 14.70% and the lowest was in the treatment of addition of liquid smoke 3% by mixing with 10.87% (figure 2). Liquid smoke can reduce protein levels during the manufacturing process. The phenol content of each treatment will react with the protein component, so that the greater the amount of protein that reacts with phenol, the less the amount of protein content. Phenol compounds tend to react with the hydrogen sulphur groups in proteins, because the reaction caused protein denaturation and new bond formation which result in a decrease in protein value from smoked ingredients [14]. The more phenols that stick to the fish react with protein and the protein content decreases. The decreasing in protein content in meatballs was due to the water soluble nature of protein, so it could reduce protein levels after boiling [2]. Significant differences in each treatment were influenced by the concentration of liquid smoke given differently during boiling.

The highest lipid content was found in the treatment of addition of liquid smoke 3% by mixing with 2.85% and the lowest is in the control treatment (without liquid smoke) with 1.36% (figure 2). The difference in the value of the fat content of each treatment in the products of mackerel fish was influenced by the amount of water content and phenol compounds from liquid smoke which can inhibit the damage of fat in the product. The activity of phenolic compounds contained in food can inhibit fat oxidation so as to prevent fat damage. The higher the phenol concentration of the sample, the more the fat damage process can be inhibited. The higher the water content coming out from the material, the greater the amount of fat content measured in the proximate test [12].
3.4. Total plate count (TPC)
Analysis of variance (ANOVA) showed that difference methods of adding liquid smoke 3% in storage times of “otak-otak” significantly affected TPC values (P <0.05). Addition of liquid smoke inhibited bacterial growth. The results of the analysis of the TPC values of “otak-otak” with the addition of 3% liquid smoke treatment showed that “otak-otak” added with liquid smoke experienced bacterial growth during storage slower than the control “otak-otak” (without liquid smoke). Coconut shell liquid smoke contains phenol components, which function as inhibitors of bacterial development [15]. Phenol compounds are one of the main antibacterial chemical compounds.

![Graph showing logarithmic values of “otak-otak” mackerel with added 3% liquid smoke during storage at ±4°C, control, mixing, boiling, soaking.](image)

**Figure 3.** Logarithmic values of “otak-otak” mackerel with added 3% liquid smoke during storage at ±4°C, control, mixing, boiling, soaking.

At day 0 storage, treatments were not significantly affected TPC values. This was because the “otak-otak” has not experienced storage and microbial proliferation has not occurred. On the 8th day, the “otak-otak” with control treatment had the highest number of microbes and was close to the maximum number of microbes in fish brain, namely 4.68 log Cfu/g. According to SNI 7757-2013 concerning fish brain, the maximum limit of TPC value is 4.69 Cfu Log/g, while the number of microbes in each treatment added with 3% liquid smoke had acceptable quality. After having been stored for 12 days, the number of bacteria in the control treatment increased, while the TPC of the “otak-otak” with the treatment of addition of 3% liquid smoke were still acceptable. Of the three treatments, the addition of liquid smoke 3% with the mixing treatment showed the smallest log number of bacteria. This is because in the treatment of mixing liquid smoke allowed for more compounds attached to the “otak-otak” than boiling and soaking.

Liquid smoke is increasingly being used because it has a distinctive flavor and inhibitory effects on pathogens. The preservative effect of liquid smoke in these food occurs because of the presence of antimicrobial and antioxidant compounds, such as aldehydes, carboxylic acids, and phenols [16]. Some common food-borne pathogens such as *Listeria monocytogenes*, *Salmonella*, *Escherichia coli*, and *Staphylococcus* have shown sensitivity to liquid smoke in vitro and in the food system [17]. Therefore, liquid smoke has the potential to be used as a natural antimicrobial in commercial applications with the desired smoky taste.
3.5. Degree of acidity (pH)

Figure 4. The pH value of “otak-otak” mackerel that added 3% liquid smoke during storage at ±4°C, control, mixing, boiling, soaking.

Analysis of variance (ANOVA) on storage times with the different methods of adding liquid smoke 3% in “otak-otak” showed no significant difference in pH values (P> 0.05). The pH values of “otak-otak” showed a decrease in pH values between the control (without liquid smoke) and those that were added with liquid smoke 3%. The lowest pH value was found in the “otak-otak” added with 3% liquid smoke by mixing. This was influenced by the phenol content of liquid smoke absorbed in the mixing treatment of liquid smoke 3% more than the boiling and soaking treatments. The more components of liquid smoke contained, the lower the pH value. The amount of acidity of a product is affected by the acidic composition [2]. This is because liquid smoke contains phenol and acetic acid. The higher the levels of phenol and acetic acid in the liquid smoke of the meal, the higher acidity level as indicated by the decreasing pH value.

The pH value during storage temperatures of ±4°C ranged from 5.03 to 6.67. It can be seen from figure 7, that the result of the analysis of all treatments showed an increase in the pH value of the “otak-otak” during storage. The pH value of meatballs boiled with liquid smoke 2.5% and stored at refrigeration temperature increased until the end of storage. The increase in pH value of the meatballs was aggravated by the development of psychrophilic bacteria which can cause the formation of volatile bases such as ammonia and trimethylamine [18]. The process of protein denaturation will cause the formation of ammonia which is also categorized as a volatile base. The longer the decay process takes place, the higher the ammonia level formed (volatile base), which results in the rise of pH [8]. Protein denaturation causes an increase in pH value by breaking down hydrophobic interactions and increasing the solubility of hydrophobic groups in water. Liquid smoke contains alcohol compounds and phenol compounds that can denaturate proteins [19].

4. Conclusion

The addition of 3% redestilated liquid smoke influenced the flavour characteristics of mackerel “otak-otak”, the most preferred one was “otak-otak” with soaking treatment in liquid smoke solution 3%. The addition of 3% liquid smoke with redestillation affected the proximate value of mackerel “otak-otak”, and it was in accordance with SNI 7757-2013 about “otak-otak”. The treatment of mixing 3% liquid smoke in dough was the best treatment for the proximate quality. Addition of 3% redestillation of liquid smoke also gave an effect on bacterial activity in mackerel “otak-otak” during storage in 4°C temperature. Liquid smoke could provide a longer shelf life, which was based on the number of microbes up to 12 days is still acceptable. Meanwhile “otak-otak” without liquid smoke are still
acceptable for up to 8 days based on the number of microbes. Redestilation of coconut shell liquid smoke contained acidic compounds, phenol, carbonyl which could increase the shelf life in the mackerel “otak-otak”.

References

[1] Standar Nasional Indonesia 2013 Otak-otak Ikan SNI 7757:2013 (Jakarta: Badan Standar Nasional Jakarta)
[2] Muttakun A, Ali R and Sulaeman 2017 Pemanfaatan asap cair dari sabut kelapa muda pada proses pengawetan bakso jamur tiram putih dan ikan patin Jom. Faperta 4 1-15
[3] Putra D A P, Agustini I T W and Wijayanti 2015 Pengaruh penambahan karagenan sebagai stabilizer terhadap karakteristik otak-otak ikan kurisi (Nemipterus Nematophorus) JPBHP. 4 1-10
[4] Karim F A, Swastawati F and Anggo A D 2014 Pengaruh perbedaan bahan baku terhadap kandungan asam glutamat pada terasi. JPBHP. 3 51-58
[5] Hadi A 2014 Pengaruh penggunaan asap cair tempurung kelapa terhadap daya awet bakso Nasuwakes. 7 135-146
[6] Swastawati F, Susanto E, Cahyono B and Trilaksono W A 2012 Sensory evaluation and chemical characteristics of smoked stingray (dasyatis bleekerey) processed by using two different liquid smoke Int. J. Bioscience, Biochemistry and Bioinformatic 2 212-216
[7] Riyadi, N H and Atmaka W 2010 Diversifikasi dan karakterisasi citarasa bakso ikan tenggiri (scomberomus commerson) dengan penambahan asap cair tempurung kelapa. Jurnal Saintek Perikanan 2 29 – 39
[8] Utami RD, Tamrin, and K T Isamu 2016 Efek perendaman vakum asap cair pada bakso ikan tuna (Thunnus Sp.) terhadap penyimpanan. JSTP. 1 193-200
[9] Ernawati 2015 Pengaruh perlakuan asap cair terhadap sifat sensoris dan mikrostruktur sosis asap ikan lele dumbo (Clarias gariepinus). J. Kelautan. 8 52-59
[10] Utami RD, Tamrin, and K T Isamu 2016 Efek perendaman vakum asap cair pada bakso ikan tuna (Thunnus Sp.) terhadap penyimpanan. JSTP. 1 193-200
[11] Sasonoko P, Mushollaeni W and Herman 2014 Aktivitas antibakteri asap cair dari limbah tempurung kelapa terhadap daging kelinci asap Buana Sains 14 193-197
[12] Zuraida I, Sukarno S and Budijanto 2011 Antibacterial activity of coconut shell liquid smoke (cs-ls) and its application on fish ball preservation. Int. Food Res. J. 18 405-410
[13] Lingbeck J M, Cordero P, O’Bryan C A, Johnson M G, Ricke S C, and Crandali P G 2014 Functionality of liquid smoke as all natural antimicrobial in food preservation. Meat Science 97 197-206
[14] Tricahyono A, Widiati A S and Widiyasutti E 2012 Pengaruh penambahan filler komposit (wheat bran and pollard) dan rumput laut terhadap ph, whc, cooking loss dan tekstur nugget kelinci. Jurnal Ternak Tropika 13 19-29