ABSTRACT

The aim of the study was to evaluate the product fortified sticks with tuna bone meal and kale leaf extract (Brassica oleracea var. Acephala) as dyes. Experimental research was focused on finding the optimal fortification of tuna fish bone meal and kale leaf extract in increasing the nutritional value and sensory properties of the product. The results showed that the stick product with 25% tuna bone and 20% kale leaf extract (P3) had the highest protein content (6.61±0.11%) and calcium (4.133±0.01 mg/100g) compared to other treatments. However, the water content and crispness of the product decreased. The lowest moisture content (0.945±0.12%) and the highest hardness (6.17±0.11 mm/100g/10s) in P3 compared to other treatments. Sensory test results on preference, aroma and color of the product fortified sticks with 20% tuna bone meal and 15% kale leaf extract (P2) were significantly (P<0.05) higher than the control, sticks with 15% tuna bone meal and 10% kale leaf extract (P1) and P3. The highest value of P2 was 43% for texture, 40% for aroma, and 57% for color. However, for taste and texture, P0 was significantly (P<0.05) higher than P1, P2, P3. The more the addition of tuna fish bone meal and kale leaf extract in the sticks, the higher the protein and calcium content, but the water content and crispness of the product decreased. It is recommended to make sticks using the composition of adding 20% tuna fish bone flour and 15% kale leaf extract.

Keyword: Fish bones; Kale Leaves; Sticks; Tuna

INTRODUCTION

Sticks are long flat cakes has a savory and crunchy texture, which are processed by frying or baking. Sticks are generally made from wheat flour, tapioca, and other starch sources with high carbohydrate content, but low in nutrients so they need to be added or fortified with nutrients as other sources of energy such as sources of fat, protein, vitamins, or minerals. The use of by product materials that can be used in the manufacture of food is very limited. Tuna bone meal as a by-product has not been used properly, it is an alternative that should be developed. In relation to increasing the nutritional value of the product, sticks fortified with fish bone meal can produce steak products containing better calcium and protein, which are useful for human health compared to steaks without fortified fish bone meal.

Tuna is one of the fish resource commodities that has a highvalue and as an export commodity in addition to meeting domestic needs. This is related to the high nutritional content and delicious taste of the meat so that it is liked by consumers (Zulfahmi et al., 2014). Fish bones also contain high protein and calcium. In fact, the calcium content in fish bones is almost 40% (Trilaksani et al., 2006). However, tuna bone which is a waste has not been used effectively. In general, fishery waste is up to 35%, while fish meat that has been weeded is an average of 65% (Fishypedia, 2011). One alternative that can be done is processing tuna fish bones into bone meal. The by-product of tuna bones is very large, so far it is just wasted, especially in traditional markets. Tillman et al (1998), stated that fish bones contain 45% water content, 10% fat, 20% protein and 25% ash. Tuna fish bones are usually only used as raw material for making bone meal for animal feed formulas, fish component solutions and others. Utilization of fish bones into fish bone meal as an additive or fortification in making sticks is an...
alternative that needs to be considered. So far, people only use cob meat for further processing, while the bones, head and entrails are disposed as waste.

In relation to increasing the nutritional value of the product, sticks fortified with fish bone meal can produce steak products containing better calcium and protein, which are useful for human health compared to steaks without fortified fish bone meal.

Previous study showed that the sticks added with catfish bone meal in the manufacturing process produced sticks with the highest calcium content compared to sticks that only added catfish meat. The calcium content in the sticks added with catfish meat produces the lowest calcium content. In addition, the contribution needed in stick products with the addition of 20% fish bone meal is the best treatment (Handayani et al., 2017). Food production systems that can increase energy continue to be sought, especially the use of agricultural waste, due to concerns about environmental problems. Good food production is also expected to be able to follow the existing trends without increasing the burden of waste on the environment.

Kale (Brassica Oleracea Var. Sabellica) is a type of vegetable that has high nutritional value, a group of plants such as cabbage, broccoli and kailan. Kale is very high in fiber, vitamin C, iron and magnesium. The use of kale leaf extract is also an alternative as a natural food coloring agent, as well as a medicinal plant. Kale which is mixed in the processing of stick products is expected to increase immunity in humans because it is a source of vitamin C, iron and magnesium (Roni, 2016).

Kale is a medicinal plant that has secondary metabolic compounds that are beneficial to the body, not only as a food additive, but also with biochemical, bioactivity, and other chemical functions in facilitating the body's work functions, especially the digestive system. These compounds include: essential oils, alkaloids, acids, steroids, tannins, saponins, flavonoids and vitamin C as well as other major groups. The use of herbal-based dyes is important and has a great opportunity to be developed because it can help complement essential nutrients, increase food consumption, and optimize product utilization, and have a positive effect on technological characteristics and product quality (Wardah et al., 2017). Currently, kale is also popular as a detox food ingredient for the body because the flavonoid content in kale contains antioxidants for the body (Roni, 2016). The use of kale leaf extract is also an alternative as a natural food coloring agent. Research on the addition of natural dyes in the manufacture of biscuits has been carried out (Ristiana&Suhartatik, 2015). Study uses purple cabbage as a natural dye in the manufacture of biscuits. Previous study showed the best formula, namely the addition of 10% purple cabbage because it produces a fairly crunchy texture and a fairly good color, but a less sweet taste (Ristiana&Suhartatik, 2015).

This study aimed to evaluate changes in the nutritional value of the sticks given the addition of tuna bone meal and kale leaf extract with a certain dose. The results of the research are expected to produce quality sticks and can reduce waste. The potential contribution of food products that utilize waste and the use of plants in the snack food industry is a new breakthrough that deserves attention because the use of fish bone waste can cause ecological problems.

METHODS

The study was carried out at the Agroindustry Laboratory of the Vocational Faculty of Untag Surabaya in June-August 2021. This study focused on evaluating the product fortified sticks with tuna bone meal and kale leaf extract as a natural dye to increase the nutritional value and
sensory properties of the product. The formulation of adding tuna bone meal and kale leaf extract as natural dyes in making sticks is presented in Table 1.

Table 1. Formulation of tuna fish bone sticks and kale leaf natural dyes in research

| No | Materials                  | P0     | P1       | P2       | P3       |
|----|----------------------------|--------|----------|----------|----------|
| 1  | Tuna Fishbones (g)         | -      | 16.9 (15%) | 22.6 (20%) | 28.3 (25%) |
| 2  | Kale leaf extract (ml)     | -      | 11ml (10%) | 16ml (15%) | 22ml (20%) |
| 3  | Wheat Flour (g)            | 113.2  | 113.2    | 113.2    | 113.2    |
| 4  | Tapioca Flour (g)          | 22.6   | 22.6     | 22.6     | 22.6     |
| 5  | Egg (g)                    | 28.2   | 28.2     | 28.2     | 28.2     |
| 6  | Margarine (g)              | 22.6   | 22.6     | 22.6     | 22.6     |
| 7  | Salt (tbsp)                | 0.5    | 0.5      | 0.5      | 0.5      |
| 8  | Onion (g)                  | 1.8    | 1.8      | 1.8      | 1.8      |
| 9  | Garlic (g)                 | 1      | 1        | 1        | 1        |
| 10 | Baking Powder (tsp)        | 0.5    | 0.5      | 0.5      | 0.5      |
| 11 | Water (ml)                 | 35.2   | 23.8     | 18.2     | 12.5     |

Description:  
*P0 = Sticks without treatment (as control);  
P1 = Sticks with 15% tuna bone meal and 10% kale leaf extract  
P2 = Sticks with 20% tuna bone meal and 15% kale leaf extract  
P3 = Sticks with 25% tuna fish bones and 20% kale leaf extract*

Materials  
Wheat Flour, Tuna Bone Flour, Tapioca Flour, Garlic, Onion, Baking Powder, Margarine, Oil, Kale Leaves Extract, Water, and Egg.

Tools  
Weight, Noodle Maker, Spoon, Washbasin, Plastic Bowl, Pan, Spoon for frying, Knife, Pan for Stream, Stove, Kjeldal Tecator 1026/2006, Penetrometer, Direct Spectrophotometer AAS and Drying Oven Memmert UN 55

Procedure of Making Tuna Fishbone Flour  
First, wash the fishbone as much as needed, then boil it in 1-liter water at a temperature of 80˚C for 30 minutes. Second, steam the fishbones at a pressure of 1 atm for 2 hours. Then, cut the fishbone into small pieces and steam it at a temperature of 100˚C for 30 minutes. Next, dry the fish bones using an oven at a temperature of 50 - 60˚C for 24 hours, and grind with a blender and sieve.

Procedure of Making Natural Dye from Kale Leaves Extract  
First, choose the same size and color of kale leaves. Second, clean and wash the leaves using water, then drain them. Third, weigh, cut into small pieces, then grind with a blender until it is slight by adding water with a 1:2 ratio. Next, strain them using plastic filtration to take leaves kale extract.
Procedure of Making Fishbone Stick by Adding Kale Leaves Extract

Procedure of Making Fishbone Stick and Natural Dye from Kale based on instruction (Siswanti, 2017), with the following steps: First mix Tuna fishbone flour each in treatment: 15%, 20% and 25% with wheat flour and tapioca flour until evenly and then add egg, margarine, and mashed seasoning into the admixture, then stir until it is mixed evenly, followed by adding water little by little. After that add natural dye from liquid Kale leaves extract each in treatment: 10%, 15%, and 20%, and mix it evenly into a smooth dough or not sticky. Then shape admixture with dryer and automatic shaper (noodle maker), so shaped it like a sheet and cut into a stick. The last step cut stick, then fry at a oil temperature of 140°C for 3 minutes in every treatment and chill the stick and analyse it for chemistry, physics, organoleptic test.

Chemical and Sensory Analysis

Parameters for the chemical properties sticks included: water content, texture (hardness), protein, calcium and sensory. Parameters of sensory using preference test to predict the level of consumer preference for sticks include aspects of color, texture, aroma, taste and overall.

Moisture Content analysis (AOAC,2002)

Foodstuffs in the form of powder ware weighed as much as 1-2 g in a weighing bottle with a known weight. Then dried in the oven at a temperature of 100-105°C for 3-5 hours. The material was cooled in a desiccator and weighed. The material was heated again in the oven for 30 minutes, cooled in a desiccator and weighed again, the treatment was repeated until a constant weight was reached (the difference in weighing was less than 0.2 mg, respectively). Weight measurement was the amount of water in the material.

Nitrogen Content (N-total Macro-KjeldahlTecator 1026/2006 method) (AOAC,2002)

In the digestion process, the appliance is turned on and the temperature setting was set to 420°C. The material is weighed as much as 1g and put in a Kjeltec flask. Added 15 ml of concentrated sulfuric acid and 2 Kjeldahl tablets. The aspirator faucet is turned on or used in a fume hood with an exhaust pump. The Kjeltec tube was fed into the digestor. Samples were destructed for 45–60 minutes. Destruction is declared complete if the sample turns clear and white smoke was no longer formed. After the digestion ends, remove the Kjeltec flask from the digestor and allow it to cool (15 minutes). In the distillation process, the Kjeltec flask was placed into the automatic distillation apparatus, the AUTO button was pressed (the input has been set for 75 ml aquadest and alkaline - NaOH 40% - 25 ml, and a steaming time of 4 minutes, according to Tecator standards). A total of 25 ml of 4% boric acid (containing indicator methyl red and bromine cresol green in methanol) was measured as a distillate container in an erlenmeyer. Raise the position of the Erlenmeyer until the distillate pipe was immersed and is on the bottom surface of the Erlenmeyer. The distillation tool works automatically, leave it until the process is complete. The sample was titrated with 0.2N titrisol HCl until the end point of the titration. The HCl used was recorded, nitrogen and protein were calculated by the following formula.

\[
N(\%) = 6.25 \times \frac{14.01 \times (\text{Vol sampel} - \text{Vol blank}) \times 0.2}{\text{Sampel weight} \times 10}
\] (1)

Calcium content (Titration method BSN 2004 (Ca as CaCO₃)) (AOAC,2002)
Analysis of Ca levels as follows: The sample solution that was ready to be analysed was then put into a vial (a 10 mL sample holder that looks like a bottle). Analysis of Ca levels in the stick sample was carried out by inserting 10 mL of each sample into the vial, where for the stick sample it was put into 3 vials and the sample was also put into 3 vials by adding the reagent (reagent) Ca, namely methyl ptaline \((\text{C}_{20}\text{H}_{26}\text{NO}_3)\) into each vial, then shaken until all the reagents dissolve to become homogeneous and the solution changes color from clear to pink. In this study, each sample of sticks analyzed for Ca content was measured 3 times. Furthermore, the samples were analyzed using a Direct Spectrophotometer AAS and then the results obtained were averaged.

**Texture Analysis (Sumarmono, 2012)**

The texture of a material was a quality element that can be sensory measured by the touch of a fingertip, using the tongue, mouth and teeth. It can also be measured objectively with special tools, such as tendermeter, texture meter, succulometer, pressuremeter and penetrometer. The penetrometer was a device used to measure the level of hardness or texture of a material with the principle of measuring the depth of the inserting awl.

**Sensory Evaluation**

Sensory evaluation is a scientific measurement in measuring and analysing the characteristics of a food ingredient or product that is received by sight, taste, smell, touch and interpret the reactions and consequences of the sensing process carried out by humans who are also called panelists as measuring instruments. In this study, using the preference test is part of the organoleptic test (Waysima et al., 2010). The likes or hedonic test is a test where the panelists are asked to give personal responses about likes or dislikes and their levels (Wahyuningtias et al., 2014). The parameters observed or tested through organoleptic tests are colour, aroma, colour, and taste in stick. The test was carried out using a preference test and panelists used for this study using 30 panelists trained panellists with the following value categories: 1 = highly dislike, 2 = dislike, 3 = neutral, 4 = like, 5 = highly like.

**Data analysis**

The data from the observation of nutritional values and the results of the panelists' assessment in the study were analysed using a completely randomized design (CRD). If there is a difference, then it is continued with the Honestly Significant Difference Test (BNJ) to find out the location of the difference between treatments if the treatment has a significant effect \((P < 0.05)\).

**RESULT AND DISCUSSIONS**

**Chemical Characteristics**

The results of the chemical analysis of sticks fortified with tuna bone meal and natural dyes kale leaf extract consisting of water content, texture (crunch), protein and calcium content are presented in Table 2.
Table 2. The results of the chemical analysis of sticks

| Treatment | Water content (%) | Hardness (mm/100g/10s) | Protein content (%) | Calcium (mg/100g) |
|-----------|------------------|-----------------------|---------------------|------------------|
| P0        | 2.025±0.11c       | 4.83±0.21b            | 3.46±0.20b          | 0.399±0.31d      |
| P1        | 1.231±0.21b       | 1.50±0.12a            | 5.17±0.44c          | 1.538±0.22c      |
| P2        | 1.135±0.20a       | 4.50±0.44a            | 5.61±0.23c          | 3.520±0.45b      |
| P3        | 0.945±0.12a       | 6.17±0.11c            | 6.61±0.11a          | 4.133±0.01a      |

There was an increase in the chemical content of the stick products fortified with tuna bone meal and natural dyes from kale leaf extract. There was a significant decrease in the average water content (P<0.05) in the sticks fortified with tuna bone meal and kale leaf extract. P3 (25% fortified tuna bone meal and 20% kale leaf extract) contained 0.945±0.12% (lowest) water content compared to P0 (2.025±0.11%), P1 (1.231±0.21%) and P2 (1.135±0.23%). P1 was not significantly different (P>0.05%) with P2. The low water content of the fishbone meal fortified sticks may be due to the presence of calcium which causes water absorption in the product.

Hardness (texture) test using a penetrrometer resulted in sticks P1 (15% fortification of tuna bone meal and 10% kale leaf extract had the best average level of crispness (1.50±0.12 mm/100g/10s) and significant (P<0.05) compared to P0 (4.83±0.21mm/100g/10s), P2 (4.50±0.44mm/100g/10s) and P3 (6.17±0.11mm/100g/10s).This happened because the low amount of fish meal meant that the calcium content was not much in the fish meal. Maulida (2005), stated that the more addition of tuna bone meal, the harder the product produced, this was related to the large calcium and phosphorus content in tuna bone meal. Komar, et.al (2009) stated that the water content is a very important factor in determining the texture. Factors that affect the texture of foodstuffs are the ratio between protein content, fat, processing temperature, water content and water activity (Komar et al., 2009). Morrison (1959), explained that bone contains 85% of minerals in the form of calcium phosphate, 14% calcium carbonate and 1% magnesium.

The average protein content of sticks at P3 (25% tuna bone meal and 20% kale leaf extract) was highest at 6.61±0.11% and significant (P<0.05) compared to P0 (3.46±0.24%), P1 (5.17±0.44%) and P2 (5.61±0.23%). The high protein content in P3 is due to the presence of protein from fish bone meal, because tuna has a very high nutritional content, this is followed by protein content in bones. The protein content in tuna is relatively large, namely 15-25% per 100 grams of fish meat (Fishypedia, 2011).

The average calcium content of sticks at P3 (25% tuna bone meal and 20% kale leaf extract) was highest at 4.133±0.01 mg/100g and significant (P<0.05) compared to P0 (0.399 ±0.31mg/100g), P1 (1.538±0.22 mg/100g) and P2 (3.52±0.45 mg/100g). This is in accordance with the results of Handayani’s research (2015) that sticks added with catfish bone meal produced sticks with the highest calcium content compared to sticks that only added catfish meat with the lowest calcium content. The contribution needed in stick products with the addition of 20% fish bone meal is the best treatment (Handayani et al., 2017).

**Sensory Profiles**

The results of organoleptic tests on sticks fortified with tuna bone meal and the addition of kale leaves as natural dyes included: taste, aroma, texture and color tests conducted on 30 semi-trained panelists.
As many as 4 treatments were coded as follows: P0 (sticks without treatment); P1 (Sticks with the addition of 15% tuna bone meal and 10% natural coloring kale leaf extract); P2 (Sticks with the addition of 20% tuna bone meal and 15% natural coloring kale leaf extract); P3 (Sticks with the addition of 25% tuna fish bones and 20% natural coloring kale leaf extract).

**Taste**

Panelists on average liked the taste of the unfortified sticks (P0) over the fortified sticks with tuna bone meal and kale leaf dye (P1, P2 and P3). This is because the addition of fish bone meal produces a distinctive fish taste, the more additions it causes the fish's distinctive taste to be stronger. Like wise, the addition of kale leaf extract coloring, the more you give it, the stronger the kale flavor in the product (Wardani et al., 2012). The results of the panelists' assessment are presented in Figure 1.

![Figure 1. Histogram of Taste Assessment on Sticks](image)

**Aroma**

Panelists on average liked the sticks with the addition of tuna bone meal and kale leaf dye in P1 treatment. It is possible that the panelists prefer sticks with a slightly rancid tuna aroma and not too green coloring. According to Wardani et al (2012), that the addition of fish bone meal produces a product that has a stronger aroma and is slightly rancid, in accordance with the characteristics of the dominant fish having a distinctive fishy aroma. The results of the panelists' assessments are presented in Figure 2.
Figure 2. Histogram of Aroma Rating on Sticks

Texture
Treatment (P1) produced the highest score with an average value of 43% compared to other treatments (P0, P2 and P3). Maulida (2005), states that the more addition of fish bone meal causes the product to be harder, this is related to the calcium and phosphorus content in fish bone meal. Komar, et.al, (2009) explained that water content is a very important factor in determining the texture of a product. The lower the water content of the product, the harder the product. Factors that affect the texture of foodstuffs are the ratio, protein content, fat, processing temperature, water content and water activity (Fellows, 2009). The results of the panelists’ assessment are presented in Figure 3.

Color
The results of statistical analysis showed that the addition of tuna bone meal and kale leaf extract in 4 treatments obtained significant results ($P<0.05$). The highest score was obtained at P2 of 57% compared to P3 (37%), P1 (31%) and P0 (37%). This happens because some panelists like the golden color like the color of the sticks in general. The results of the panelists’ assessment of the color of the sticks fortified with tuna bone meal and kale leaf natural dyes can be seen in Figure 4.
The results of the organoleptic tests carried out were hedonic tests in the form of color, taste, texture and aroma with a test scale of 1-5. The panelists' preference level on tuna fish bone meal sticks with the addition of natural dyes of kale leaves, the highest panelists' assessment was at P2 (20% Tuna Fishbone Flour + 15% Kale leaf extract) with a value of 43%. For P3 (25% tuna bone meal + 20% kale leaf extract) with a value of 30%, P1 (15% tuna bone meal + 10% kale leaf extract) with a value of 23%. The panelists' assessment of the P1 treatment tends to be smaller because the panelists do not like/accept the product compared to P0 (without treatment) which has a value of 33%. Overall acceptance is a response that includes the results of the panelist's general assessment which includes color, aroma, texture, and taste of the sample (Warsito et al., 2015). The results of the panelists' assessment are presented in Figure 5.

**CONCLUSION**

Fortification of fish bone meal and kale leaf extract had an effect on the chemical content of the sticks. The more addition of fish bone meal and kale leaf extract, the water content and the crispness of the sticks decreased, but the protein and calcium levels in the sticks products actually increase.
In terms of taste, the researcher preferred the sticks that were not fortified with fishbone meal and kale leaf extract. As for the aroma and texture, the researcher liked the sticks fortified with 15% tuna fish bone meal and 10% natural dye from kale leaf extract. But the overall preference and color of the product, the highest score was obtained on the sticks fortified with 20% tuna fish bone meal and 15% natural coloring kale leaf extract.

**RECOMMENDATION**

Based on the research results obtained, it is recommended that in producing sticks with tuna fish bone meal and kale leaf extract natural dyes, the best composition is sticks with the addition of tuna fish bones and 20% + 15% kale leaf extract which is the best formula because it has high value. quite good nutrition and preferred by consumers.

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