Moisture Content, Protein, Crude Fiber and Antioxidant Activity of Cookies with Boiled Papaya Leaf

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Abstract. This research aims to determine the effect of the addition of papaya leaves after boiling to moisture, protein, crude fiber and antioxidant activity and in cookies. The cookies were made by the addition of papaya leaf in to the dough. The treatments conducted were T0: 0%; T1: 10%; T2: 20%; T3: 30% which has been boiled, the formation of dough and roasting. The results of testing of moisture, protein, crude fiber and antioxidant activity have increased. Moisture content of 4.64 – 7.38%, protein 6.74 – 7.96%, crude fiber 5.08 – 10.96% and antioxidant activity 46.73 – 68.42%. The best treatment for increasing the concentration of papaya leaves in cookies is T1: 10%.

Keywords: moisture content, protein, crude fiber, boiled papaya leaf

1. Introduction
Papaya leaves are part of the papaya plant which contains many good nutrients such as bioactive components. Bioactive components in papaya leaves are very abundant such as alkaloids, flavonoids, saponins and papain [18]. Some of these components are beneficial to the body because they have a role as antioxidants. These antioxidant compounds can prevent their heaps of various degenerative diseases such as heart attacks, leukemia and premature aging.

The abundance of beneficial components in papaya leaves is apparently not enough to attract the attention of the public to consume them. The main reason for the low interest of the community in consuming papaya leaves is the presence of a very strong bitter taste in this leaf. This bitter taste is caused by a compound called karpain alkaloid (C₁₄H₂₅NO₂) found in papaya leaves [4].

Boiling is one of the easy steps to eliminate bitter taste in papaya leaves. During the boiling process, karpain alkaloids in papaya leaves will decompose by heat so that the bitter taste can be minimized [12]. The nature of the alkaloid karpain which can be decomposed by heat makes papaya leaves to be a substance that is very suitable when combined with food which in the processing requires heating and minimal nutrient content such as cookies.

The dominant nutritional component in cookies is carbohydrate, protein and fat [20]. As a product with a lot of consumer interest and a high level of consumption, improving the nutrient component in cookies should need to be improved. Addition of nutrients such as fiber and bioactive compounds is...
deemed necessary to balance the nutritional components in cookies. This study aims to determine the effect of adding papaya leaves after boiling to moisture, protein, crude fiber, antioxidant activity and sensory properties in cookies. The benefit of this research is that it can provide information regarding the use of papaya leaves added in cookies as an effort to increase nutrition.

2. Materials and methods

2.1. Materials

The materials used in this study include papaya leaves from papaya plants (Carica papaya L.) orange papaya and obtained from the area of Undip Agrotechnopark, flour, refined sugar, margarine, egg yolk, skim milk, baking soda, salt, aquades, selenium, H₂SO₄, H₃BO₃, NaOH, HCl, MB and MR indicators, 70% alcohol, hot water, DPPH solution, and methanol. The equipment used includes mixers, basins, blenders, Whatman 41 paper, cup glass, oven, porcelain saucer, desiccator, analytical scales, heater extract, furnace, destilator, pumpkin Kjeldahl, acid cupboard, pipette, burette, erlenmeyer, and spectrophotometer UV-Vis (Shimadzu, UV-1280, 220-240 (CE)).

2.2. Methods

The research methods included making cookies, testing moisture content (AOAC, 2007), protein content (AOAC, 2007), crude fiber, antioxidant activity (UV-Vis spectrophotometer), and sensory properties.

2.2.1. Process of Cookies. The first stage in making papaya leaf cookies is boiling papaya leaves in water at a temperature of 60° C for 10 and doing 2 repetitions of boiling. The next stage is making dough with the composition of flour, papaya leaves after boiling (T0: 0%; T1: 10%; T2: 20%; T3: 30%), refined sugar, margarine, egg yolk, skim milk, baking soda and salt mixed with mixer until blended. The mixture is then weighed 15 grams and flat circles are formed. Then the mixture is baked in an oven at 130 ° C for 35 minutes.

2.2.2. Water Content Analysis. The percentage of water content is determined by the oven method. This method is carried out by means of a porcelain dish dried in an oven at 105° C for 1 hour then put in a desiccator for 15 minutes until it is cool then weighed (a). The cookie samples were macerated and then weighed as much as 2 grams (b) in a porcelain dish. The sample is then dried in an oven at 105 ° C for 4 hours. Next, the sample was removed from the oven and put in a desiccator for 15 minutes until it cooled. The sample is then weighed. The drying process is repeated for 1 hour to constant weight (c) [1]. Calculation of water content by the formula:

\[
\% \text{water content} = \frac{b - (c - a)}{b} \times 100\%
\]

2.2.3. Protein Analysis. Protein levels were determined by the Kjeldahl method. This method is carried out by means of macerated cookies and then weighed 0.3 grams and inserted into the Kjeldahl flask. Then added 0.3 grams of selenium and 10 ml of concentrated H₂SO₄. Kjeldahl flask is then reconstructed in a smoke cupboard until the color turns clear green. After cooling, add 100 ml of distilled water carefully then add 40 ml of 40% NaOH. Then, the distillation process is carried out by trapping 5 ml of 4% H₃BO₃ mixed with MR and MB indicators. The distillation process is carried out until 50 ml of distillate is obtained. The destillat is then titrated with 0.1 N HCl until a light purple color is formed. The process of destruction, distillation and titration is also carried out on blank solutions containing only aqueous and NaOH solutions [1]. Calculation of protein content by the formula:
\[
\% \text{ Protein} = \left( \frac{ml \ HCl \ sampel - ml \ HCl \ blanko}{berat \ sampel \times 1000} \right) \times 14.01 \times 6.25 \times 100 \%
\]

2.2.4. Rough Fiber Analysis. Crude fiber testing using the gravimetric method was done by means of macerated cookies and then weighed as much as 1 gram (x). The sample was added 50 ml of H\textsubscript{2}SO\textsubscript{4} 0.3 N and then added to the heater extract and extracted for 30 minutes. Then added 25 ml of NaOH 1.5 N and extracted again for 30 minutes. Filter paper of Whatman 41 is heated in an oven at 105°C for 1 hour then weighed (a). The extract that was extracted was then filtered with heated Whatman 41 paper. The filtered sample is then washed successively with 50 ml of hot water, 50 ml of H\textsubscript{2}SO\textsubscript{4} 0.3 N, 50 ml of hot water and 25 ml of 70% alcohol. Filter paper and the contents of the sample are put in a porcelain dish and then dried in an oven at 105°C for 6 hours. The sample is lifted and cooled in the desiccatior for 15 minutes then weighed (Y). After weighing, the sample was put in a furnace temperature of 550°C for 6 hours then lifted and cooled in a desiccator and then weighed (Z) [19].

Calculation of crude fiber by the formula:

\[
\% \text{ Crude Fiber} = \frac{Y - Z - a}{x} \times 100 \%
\]

2.2.5. Antioxidant Activity Analysis. The percentage of antioxidant activity was determined by 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radical capture method. This method is done by sampling macerated cookies and weighing 0.1 gram. The sample was then added with 1 ml of ethanol solution. The solution is centrifuged until separate deposits and solutions are formed. The sample solution was then taken as much as 200 µl and added 1 ml of DPPH solution. Then the solution is left at room temperature with darkness for 30 minutes. The solution was then transferred in cuvette for absorbance measurements with a UV-Vis spectrophotometer wavelength of 517 nm. The control solution was made with the same procedure where the solution contained only ethanol and DPPH solution [10].

Antioxidant activity is then calculated by the formula:

\[
\% \text{ Antioxidant activity} = \frac{\text{absorbance} \text{ control} - \text{absorbance} \text{ sample}}{\text{absorbance} \text{ control}} \times 100\%
\]

2.2.6. Statistic Analysis. Data of the result analysis obtained from water, protein and crude fiber test results were analyzed by SPSS 22.0 using the Analysis of Variant (ANOVA) method to determine the effect of treatment at the 95% confidence level. Data from antioxidant test results were interpreted descriptively.

3. Results and Discussions

Table 1. The Result of Water Content, Protein, Crude Fiber Analysis

| Treatment | Water Content | Protein | Crude Fiber |
|-----------|---------------|---------|-------------|
| T0        | 4.64 ± 0.28\textsuperscript{a} | 6.74 ± 0.27\textsuperscript{a} | 5.08 ± 0.37\textsuperscript{a} |
| T1        | 5.63 ± 0.25\textsuperscript{c} | 7.17 ± 0.12\textsuperscript{c} | 6.36 ± 0.61\textsuperscript{c} |
| T2        | 6.52 ± 0.53\textsuperscript{b} | 7.53 ± 0.12\textsuperscript{b} | 7.75 ± 0.25\textsuperscript{b} |
| T3        | 7.38 ± 0.04\textsuperscript{a} | 7.96 ± 0.20\textsuperscript{a} | 10.96 ± 0.57\textsuperscript{a} |

The results showed that in each parameter namely water content, protein content, crude fiber content and antioxidant activity increased with the number of concentrations of papaya leaves added to cookies. Based on the results of the statistical analysis, each parameter had significant differences from each treatment difference in the concentration of adding papaya leaves where P ≤ 0.05. The results of the sensory properties test showed an inverse comparison where the higher the concentration of papaya leaves in cookies the lower the panelist’s acceptance.
3.1. Water Content
Based on Table 1, the water content obtained in cookies with the addition of papaya leaves experienced a significant increase along with the concentration of papaya leaves added in cookies. The results showed that the water content of papaya leaf cookies did not meet the quality standards based on SNI 2973-2011[2] which stated that the maximum moisture content was 5%. This is due to the high water content of the papaya leaves. The percentage of water content per 100 grams of papaya leaves is 81.27% [9].

The water content in cookies is also influenced by the gelatinization process of wheat flour used. That gelatinization is the process of changing starch granules from a crystalline state to expand by heating and water [8]. There are two starch fractions that play an important role in the gelatinization process, amyllose and amylopectin. Amylose functions in strengthening the gel structure, while amylopectin functions in water absorption and development of starch granules [3]. Amylopectin is what absorbs water from papaya leaves, then the water will be trapped in starch granules which affects the percentage of water content of cookies.

3.2. Protein Levels
Based on the testing of protein levels using the Kjeldhal method the results shown in Table 1 that papaya leaf cookies meet the quality requirements based on SNI 2973-2011, which is a minimum of 5% where the higher the concentration of papaya leaves added to cookies, the higher the protein content [2]. Increased levels of protein are caused by papaya leaves contain a fairly high protein component. In 100 grams of papaya leaves contain as much as 9.16% protein [9]. That in papaya leaves there are several protein components including rubisco large subunits, endochitinase, and chymopapain isofom V [13].

Protein in cookies from papaya leaves are also obtained from the ingredients of the dough used, namely flour and skim milk. Wheat flour has a protein content in the range of 8-11%, this value indicates that this flour is low in protein. Making cookies low-protein flour is used because it has a smaller water absorption ability than high-protein wheat flour [11]. Other composition as a protein contributor is skim milk. Skim milk is low-fat milk but high in calcium and protein. Based on Codex Stan 207-1999, there are at least 34% protein in skim milk.

3.3. Crude Fiber Levels
In Table 1, testing of crude fiber levels found results with a straight ratio where the higher the concentration of papaya leaves, the higher the crude fiber content of cookies. The level of crude fiber in 100 grams of papaya leaves was 12.81% [9]. That the fibers in papaya leaves include the most dominant cellulose, hemicellulose and lignin [6].

The benefits of adding papaya leaves to cookies in increasing fiber levels are as a nutritional balancer. The dominant carbohydrate content becomes a problem in product cookies, because excessive carbohydrate consumption can lead to diseases such as obesity and diabetes. That fiber consumption can slow down mastication time accompanied by an increase in saliva production, then fiber in the body will inhibit the absorption of energy and nutrients including glucose from food, inhibited absorption will provide a faster satiety effect due to more gastric emptying time old so it is very good in preventing and treating obesity and diabetes mellitus [15].
3.4. Antioxidant Activity

The percentage result of antioxidant activity in Figure 1. shows that there is an increase along with the high concentration of papaya leaves added in cookies. Papaya leaves have abundant bioactive components and act as antioxidants. That the total antioxidant of papaya leaves in ethanol solvents is 122.47 µg TE / g which consists of bioactive components of polyphenols, flavonoids, saponins and proanthocyanidin [18]. Besides the bioactive components in papaya leaves, antioxidants in cookies are also obtained from maillard reactions that occur in the dough during the roasting process. The sugar component in food will experience a maillard reaction or browning during the roasting process to form melanoidin compounds which have the ability as antioxidants and have a high sensitivity in binding to DPPH free radicals [16].

The percentage of antioxidant activity in papaya leaf cookies treated T0, T1 and T2 did not experience a significant increase, but in the T3 treatment there was a significant increase. These results can be caused due to a long heating process with high temperatures, where bioactive compounds in papaya leaves tend to be damaged by the process so that it does not show a significant level in each treatment. The bioactive component in papaya leaves can survive at 70° C for 20 minutes, the higher the temperature given will reduce the number of bioactive components [18].

4. Conclusions

Based on the results of the study, it can be concluded that the addition of boiled papaya leaves to cookies increases water, protein, crude fiber and antioxidant activity. The best treatment concentration of papaya leaves that have been boiled in cookies is T1: 10%.

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