# Characteristics of Brownies from Mocaf Flour (Modified Cassava Flour) and Black Soybean Flour (Glycine Soja)

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## ABSTRACT

Brownies are one of the most popular types of cakes. Diversification efforts are made with mocaf flour combined with composite flour from black soybeans which is applied to making brownies. The purpose of this study is to determine the physical characteristics (color and texture) and sensory values, chemical and functional characteristics. The study was conducted using a Randomized Block Design (RBD) with four treatments and three replications. Each treatment is a comparison of the composition of flour in use in the production of brownies. Among the treatments were: A (100% mocaf flour), B (50% mocaf flour: 50% black soybean flour), C (40% mocaf flour: 60% black soybean flour), and D (60% mocaf flour: 40% Black soybean flour). The results showed that the variation of the mixture of mocaf flour and black soybean flour affected the physical characteristics of brownies with hardness values between 31.37gf - 50.97gf, and L color values between 37.85 - 54.22; value a* 42.99 – 32.62; and the value of b* 21.68 – 12.47. The most preferred brownie formulation is in formulation A with variations of 100% mocaf flour and 0% black soybean flour. In the chemical and functional aspects, the brownie formulation in formula C with a variation of 40% mocaf flour and 60% black soybean flour has a protein value of 15.85%, antioxidant activity of 13.813 g/ml, and a glycemic index value of 46.2 and the glycemic load of both samples belong to the high category.

## 1. Introduction

Indonesia as a tropical country has a large potential in the agricultural sector. Various agricultural commodities have reasonably good feasibility to be developed in Indonesia, one of which is tubers. Tubers are food ingredients that have a unique taste and good nutritional content, so they have the potential to be developed as an alternative food source (Ratnawati *et al.,* 2020).

Cassava is a food source that has high nutritional value and has a complete composition as an energy source so that it can be useful for body health. From this potential, cassava can be utilized in various products that are ready for consumption (Zullaikah *et al.,* 2015). Cassava is a non-rice food that has the potential to be made as a substitute or substitute for rice consumption. This tuber can be used as an energy source that is rich in carbohydrates and fiber but the protein content in the tuber is very low.

Mocaf flour is a processed product of cassava with a fermentation technique. The modification process can mean changes in molecular structure, including using physical, chemical, and enzymatic methods (Mustika & Kartika, 2020). Mocaf has characteristics similar to flour, which is white, soft, and does not smell like cassava. With characteristics similar to flour, mocaf flour can be used as a substitute for flour in several food product (Firdaus, 2018). Mocaf flour in its use can be used as raw material for making various pastries, such as nastar, cake, cookies. The production of mocaf flour uses a biochemical method with the addition of enzymes or enzymes producing microbes (Sanjaya *et al.,* 2021). Mocaf flour in its use can be used as raw material for making various pastries, such as nastar, cake, cookies.

Mocaf flour is a processed product of cassava with a fermentation technique. Modification process can mean changes in molecular structure, including using physical, chemical, and enzymatic method (Lubis *et al.,* 2021). Mocaf has characteristics similar to flour, which is white, soft, and does not smell like cassava. With characteristics similar to flour, mocaf flour can be used as a substitute for flour in several food product (Yaqin *et al.,* 2019). Mocaf flour in its use can be used as raw material for making various pastries, such as nastar, cake, cookies. The production of mocaf flour uses a biochemical method with the addition of enzymes or enzymes producing microbes (Indrianingsih *et al.,* 2019). Mocaf flour in its use can be used as raw material for making various pastries, such as nastar, cake, cookies.

Organic composite flour from soybeans, red beans, and corn has the advantage of containing higher levels of protein, fiber, and fat than wheat flour. The composition of the organic composite flour can bind more water than wheat flour. Wheat flour substituted with organic composite flour-based on nuts such as soybeans and red beans can be used to improve the nutritional value of cakes, especially protein content up to 50% (Krisnan *et al.,* 2018).

Black soybeans *(Glycine soja)* have an advantage because they have a fairly high nutritional content such as protein and carbohydrates. The amino acids possessed by black soybeans include leucine and lysine (Kwon *et al.,* 2007). Processed products from black soybeans such as tofu enriched with fiber/soybean pulp itself were identified to be able to lower blood glucose levels, reduce inflammation, improve insulin action, improve antioxidant status, improve lipid profiles, can also reduce malonaldehyde levels and enzyme activity of SGOT/SGPT diabetic respondents (Li *et al.,* 2018).

Diversification efforts are carried out by utilizing wood tubers that are processed into mocaf flour as a local food for Garut Regency combined with composite flour from black soybeans which can increase the protein value of a processed food product (Dajanta *et al.,* 2013). Utilization of cassava which is processed into mocaf flour can reduce the use of wheat flour to reduce the amount of wheat flour imports.

Brownies are a type of cake that has a brownish-black color and a slightly harder texture than other cakes because brownies do not require gluten or developers (Hansen *et al.,* 2016). The raw materials used in making brownies generally use wheat flour, margarine, eggs, chocolate, and sugar. Diversification based on local resources needs to be developed by the Indonesian state so that the culture of consuming imported food can be improved, therefore there is a need for comprehensive research and development on local food tubers (Ratnawati *et al.,* 2020).

The use of wood tubers which is a local potential of Garut Regency can be used as mocaf flour, of course, it can replace wheat flour, it will certainly reduce the rate of imports of wheat flour. In an effort to increase diversity in a processed product, it is necessary to have innovation by diversifying. This diversification is carried out by using wood tubers that are made into mocaf flour as a local food for Garut Regency combined with composite flour from black soybeans to realize an increase in the protein value of a processed food product.

Based on this background, to optimize the physical characteristics, sensory, chemical, and functional properties of brownies, it is necessary to study and research brownies made from mocaf flour and black soybean flour, to achieve food diversification to reduce the number of wheat flour imports and increase diversity foodstuffs.

## 2. Method
### 2.1. Material
The raw materials used in this study were mocaf flour with the Bandung brand and black soybeans from Garut Regency. Other ingredients used for making brownies are margarine, chocolate bars, cocoa powder, eggs, sugar, and baking powder. Materials for analysis include HCl 25%, aqua dest, solvent N-Hexan, CuSO4, 5H2O, sodium tartrate, NaOH 10%, methanol.

### 2.2. Making Black Soybean Flour
Making black soybean flour is done by soaking clean black soybeans for two hours, after two hours the soybeans are separated from the soaking water and then dried in an oven at 50ºC for 8 hours. Black soybeans that have been roasted are ground using a grinder, then sieved using a sieve.

### 2.3. Making Brownies
The stages of the process in making brownies are melted chocolate bars with butter by steaming using a pan that is placed on top of another pan filled with hot water. The flour mixture according to the treatment is sifted with cocoa powder and baking powder. Beat the sugar and eggs using a mixer on medium speed until they are slightly pale yellow. The mixture of flour, cocoa powder, and baking powder is gradually added to the sugar and egg mixture, beating on low speed. Add the chocolate and butter mixture and stir gently until smooth. The dough is poured into the brownie mold. Bake in the oven at 190ºC for 20-30 minutes.

### 2.4. Physical Characteristics
*Texture (Hardness)*

Texture testing of mocaf flour brownies and black soybean flour was carried out using a texture analyzer (TA-XT Plus) and aimed to test the hardness of brownies with various flour formulations in making brownies. The probe used in texture analysis of brownies mocaf flour and black soybean flour is a cylindrical probe with a diameter of 36 mm. The sample to be measured is placed on top of the test sample, then the load cell will move the probe down to press the sample and then back up. The hardness value is indicated by the absolute (+) peak, which is the maximum force at the first pressure, with units of gram force (gf).

*Color*

Prepare the Minolta Reflectance Chromameter (CR-400) tool for color testing. Put the brownies in the container provided. Sets the Chromameter's initial calibration to default (Y = 93.5; x = 0.3114; y = 0.3190). The result is the value of L, a, and physical characteristics.

### 2.5. Sensory
The panelists used were panelists with an age range of 14 years to 25 years as many as 25 people, with the observed parameters including aroma, taste, color, texture, and total acceptance. Brownies samples are placed in containers that have been assigned a different three-digit code for each treatment, then prepare a questionnaire sheet at each panelist table and invite the panel to enter the testing room. Before testing the panelists were explained the implementation of the organoleptic assessment. After conducting organoleptic testing, the data from the hedonic scale test were collected and analyzed using a variance.

### 2.6. Chemical Characteristics
*Water Content*

The principle of this method is based on the evaporation of water present in the material by heating, then weighed to a constant weight. The weight reduction that occurs is the water content contained in the material. The purpose of this water content analysis is to determine the water content contained in catfish nori snacks. The way these method works is that an empty cup is heated in an oven at a temperature of 1050 C for 30 minutes, cooled in a desiccator for 15 minutes, then weighed ($W_0$). Then a sample of 2 grams was put in a cup whose weight was known, weighed ($W_1$), then dried in an oven at 1050 C for 3 hours, cooled in a desiccator for 15-30 minutes, then the cup and its contents were weighed and dried again for 1 hour and cooled in a desiccator, weighed again ($W_2$) (AOAC, 1991).

*Ash Content*

Carefully weigh 2-3 grams of the sample into a porcelain cup whose weight is known, for a liquid sample, vaporize over a water bath until dry. Charcoal over a burner, then ash in an electric furnace. At a maximum

temperature of 550ºC until complete ashing (occasionally the furnace door opened slightly, so oxygen can enter). Cool in a desiccator, then weigh with a fixed weight (AOAC, 1991).

*Fat Content*

Oven-dried fat pumpkin at 105°C for about 15 minutes. The sample is weighed as much as 5g then inserted into the fat sleeve. Filter paper containing the sample placed in a Soxhlet extraction apparatus coupled with a condenser. Solvent hexane is put into the pumpkin fat. Then the sample was refluxed for 5 hours. The remainder of the solvent in the fat flask is removed by heating in the oven, then weighed (AOAC, 1991).

*Protein Content*

Destruction Stage Weigh 1 gram of the sample that has been blended. Put it into a 100 mL Kjehdahl flask, then pipette 10 mL of concentrated sulfuric acid into the Kjeldahl flask. Add a catalyst (selenium mixture) to accelerate the digestion. Then the Kjehdahl flask is heated starting with a low heat after a while the fire is gradually raised so that the temperature rises. Destruction can be stopped when a clear greenish solution is obtained. The distillation stage The results obtained is then cooled, then diluted with distilled water to 100 mL. After being homogeneous and cold, 5 mL pipettes are pipetted, put into a distillation flask. Add 10 mL of 30% sodium hydroxide solution through the walls in the distillation flask until a layer is formed below the acid solution. The distillate flask is installed and connected to the condenser, then the end of the condenser is immersed in the liquid reservoir. The steam from the boiling liquid will flow through the condenser to the reservoir Erlenmeyer. The reservoir Erlenmeyer is filled with 10 mL of 0.1 N hydrochloric acid solution which has been dripped with methyl red indicator. Check the results of distillation with litmus paper, if the results are no longer alkaline then distillation is stopped. Titration stage. After the distillation process, the next step is titration. The results of the distillation are accommodated in an Erlenmeyer containing 0.1 N hydrochloric acid and 5 drops of methyl red indicator are then titrated with 0.1 N sodium hydroxide solution. The endpoint of the titration is marked with a pink to yellow color (AOAC, 1991).

*Carbohydrate Content*

Carbohydrate analysis in this study was carried out using the carbohydrate by different method which is included in the proximate analysis method. This procedure is an analysis in which the carbohydrate content including crude fiber is known not through analysis but a calculation.

## 2.7. Functional Characteristic's

*Antioxidant Activity*

15.8 mg of DPPH compound was weighed and then dissolved using methanol p.a to 100 ml so that 0.4 mM of DPPH solution was obtained. The solution is then covered with aluminum foil. 2.0 ml of DPPH solution was put into a 10 ml volumetric flask and methanol was added to the limit. Replication was carried out three times (triple). This solution is used as a comparison solution. Frist steps of testing, a calibration curve for the DPPH solution was first made. A total of 5 mg of DPPH was put into a 50 mL volumetric flask and dissolved with methanol solvent to the mark. The DPPH solution made had a concentration of 100 ppm, then a number of the DPPH solution was taken and diluted in a 10 mL volumetric flask to obtain variations in concentrations of 5, 10, 15, 20, and 25 ppm. Furthermore, the absorption was measured at a wavelength of 517 nm. Diluting the sample with a concentration of 10 ppm by weighing 1 mg of the sample and then dissolving it in 100 ml of methanol PA. Total antioxidant was carried out by spectrophotometric method with DPPH (2,2-diphenyl-1-picrylhydrazyl) (Dajanta *et al.,* 2013). 2 ml of the sample was put into a test tube and added 7 ml of methanol p.a then input 2 ml of DPPH solution. Then the solution was vortexed and incubated in the dark at room temperature for 30 minutes. Furthermore, the absorbance measurement at a wavelength of 517 nm

*Glycemix Index*

The combination food product Mocaf flour and black soybeans in the best treatment were tested for their nutritional content so that the carbohydrate content was obtained. The initial screening is looking for prospective normal respondents in the Garut University campus environment, with the following criteria: have an age between 21-25 years, have a body mass index (BMI) between 18.5-24.9, have a blood pressure SIS/DIA <120/<80, have fasting blood glucose levels in capillary blood 100 mg/dL or have temporary blood glucose levels in capillary blood ≤140 mg/dL, do not smoke, do not suffer from gangrene and other chronic diseases, and respondents are not pregnant. The number of respondents used in this study was 8-12 people. The socialization stage is carried out in the Garut University campus environment and respondents are asked to fill out a letter of approval. The day before the treatment the subjects were required to fast (except water) for 10

hours starting at 22.00 until 08.00 the next morning. On the first day, 50 grams of glucose samples were dissolved in 250 ml of water and then consumed by the respondents. Blood glucose was measured for 2 hours with an interval of 0.30, 60, 90, and 120 minutes. On the second day, the product sample equivalent to 50 grams of glucose was consumed by the respondents in less than 15 minutes. Blood glucose was measured for 2 hours using a glucometer with an interval of 0.30, 60, 90, and 120 minutes. During the postprandial glucose measurement process, respondents are expected not to do any activity except for going to the toilet. Capillary blood glucose levels were determined using the Accu check glucometer. The blood required for analysis with this device is as much as 2-5 l per test. Blood was taken from the fingertips that had been cleaned with 70% alcohol using a lancet slowly. Drops of blood come out affixed to the glucometer strip. Blood glucose levels will be measured on the device after 5 seconds and expressed in mg/dl. Then a relationship curve was made between the measurement time (x-axis) and blood glucose levels (y-axis), and the pattern of changes was analyzed based on the average standard glucose value of each respondent (Brand-Miller *et al.,* 2009)**.**

## 2.8. Data Analysis

The study was conducted using a Randomized Block Design (RBD) with four treatments and three replications. Each treatment is a comparison of the composition of flour in use in the production of brownies. Among the treatments were: A (100% mocaf flour), B (50% mocaf flour: 50% black soybean flour), C (40% mocaf flour: 60% black soybean flour), and D (60% mocaf flour: 40% Black soybean flour).

## 3. Result and Discussions
## 3.1. Physical Characteristics
*Texture (Hardness)*

The texture of brownies with mocaf flour and black soybean flour was obtained from the analysis of the texture of brownies with various flour formulations in making brownies, namely the hardness value. The hardness value is indicated by the absolute (+) peak, which is the maximum force at the first pressure, with units of gram force (gf) (Engelen, 2018). The test results in this study are shown in Table 1.

**Table 1.** Results of Texture Test (Hardness) Brownies.

| Brownies Formulation | Texture (gram force) gf |
|:---:|:---:|
| A | $31,37 \pm 0,12^a$ |
| B | $44,16 \pm 0,34^c$ |
| C | $50,97 \pm 0,34^d$ |
| D | $38,36 \pm 0,18^b$ |

Note: The values in the column followed by unequal letters show significantly different results at the 0.05 level according to the Tukey test results.

The addition of black soybean flour composition to the brownie formula can increase the brownie hardness value. The measured brownie hardness value is influenced by the composition, temperature, and baking time (Pratama 2009).

From the results of testing the texture value of mocaf flour brownies and black soybean flour, the texture values in formula A am 31.37 gf, formula B is 44.16 gf, formula C is 50.97 gf, and formula D is 38.36 gf. The highest texture value was in formula C brownies with a ratio of 40% mocaf flour and 60% black soybean flour, and the lowest texture value was in formula A brownies with a ratio of 100% mocaf flour and 0% black soybean flour. The higher the texture value, the harder the brownie texture. The hardness value is influenced by the water absorption index, volume absorption, degree of gelatinization, and water content of flour (Pitriawati, 2008). On the characteristics and functional properties of brownies with raw materials of mocaf flour and black soybean flour. The results of the test on brownies with the addition of more black soybean flour contain less water content. This causes the texture of the brownies to be tougher.

*Color*

The next measure of the physical characteristics of brownies is color. The color of the brownie sample is characterized by an L value, which means it tends to have white, gray, and black chromatic colors. a* value indicates the color tends to be red and the b* value indicates the color tends to be yellow (Purwani, 2006). The appearance of the color on the brownies with mocaf flour and black soybean flour can be seen in Figure 1.

Formula A

Formula B
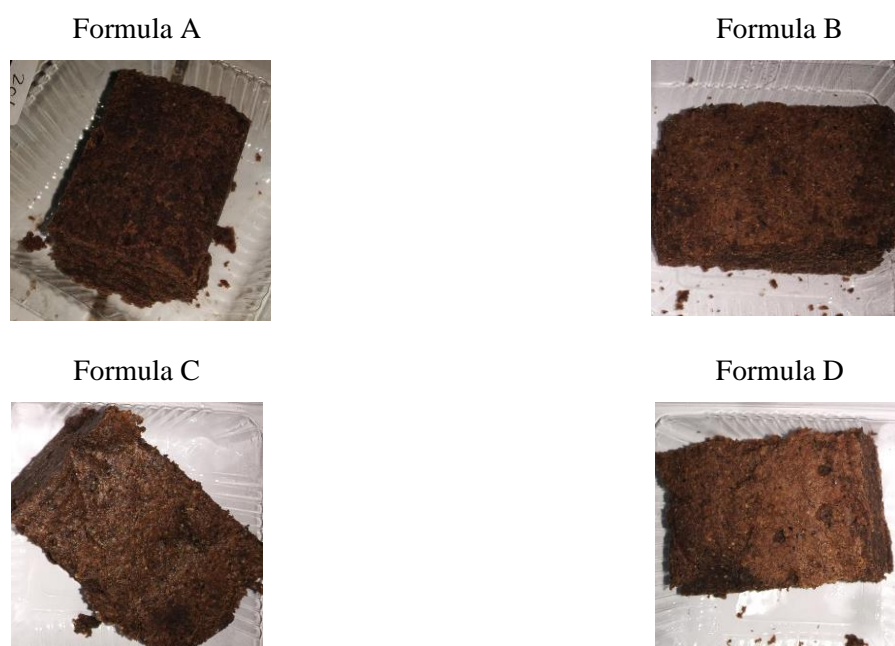
Formula C

Formula D



**Figure 1.** Appearance of brownie color

**Table 2.** Average Value of L, a*, b* Brownies

| Brownies Formulation | Color Attribute | | |
|:---:|:---:|:---:|:---:|
| | **L** | **a*** | **b*** |
| A | $37{,}85 \pm 0{,}39^a$ | $42{,}99 \pm 0{,}68^d$ | $21{,}68 \pm 0{,}54^d$ |
| B | $42{,}51 \pm 0{,}23^b$ | $38{,}39 \pm 0{,}13^c$ | $18{,}64 \pm 0{,}24^c$ |
| C | $51{,}49 \pm 0{,}37^c$ | $35{,}41 \pm 0{,}44^b$ | $15{,}74 \pm 0{,}35^b$ |
| D | $54{,}22 \pm 0{,}56^d$ | $32{,}62 \pm 0{,}11^a$ | $12{,}47 \pm 0{,}40^a$ |

Note: The values in the column followed by unequal letters show significantly different results at the 0.05 level according to the Tukey test results.

The results of the color test using a chromameter tool produce color intensity values that are displayed in the form of L, a*, and b* values. The sample brightness parameter is indicated by the L value in the range $= 0$ (the color is getting darker/darker) to 100 (the color is getting whiter/brighter). a* value as reflected light shows a green-green color if a* value is positive it will show red, and if a* value is negative it will show green. The b* value indicates a blue-yellow mixed chromatic color if a positive b* value indicates yellow and a negative b* value indicates blue (Sinaga, 2019).

Based on the results of the analysis of brownie color in Table 2, it shows that the average value of the L color attribute ranges from 37.85 to 54.22, a* value ranges from 32.62 to 42.99, and the b* value ranges from $12, 47 - 21.68$. The average value of the L a* b* attribute in all brownie treatments has a black color component on the L attribute, a reddish color on a* attribute, and a yellowish color on the b* attribute, resulting in a brown or blackish brown color.

The formation of brown color in brownies is influenced by the raw materials used such as chocolate, sugar, mocaf flour, and black soybean flour. In addition, during the roasting process, a Maillard reaction occurs, namely the browning process of food due to the reaction between carbohydrates, especially reducing sugars with $NH_2$ from protein which produces hydroxyl methyl furfural compounds which continue to become furfural. The furfural formed is then polymerized to form a brown melanoidin compound. Melanoidin is what gives brownies their brown color (Winarno 2004).

### 3.2. Sensory Characteristics
*Hedonic*

The hedonic test is one of the methods in the organoleptic test which is often used to determine the level of liking and consumer acceptance of a product. In this test, the panelists expected to be able to express their

personal opinion about likes or dislikes. The hedonic test can be done with two the way is rating test and the ranking test. Tests are used to measure the level of preference for a product. Levels of pleasure This can be referred to as a hedonic scale, for example very much like, like, somewhat like, somewhat dislike, dislike, and so on. But scale hedonic can be stretched or reduced according to the scale range desired (Rahayu (1998) in Ayustaningwarno (2014).

**Table 3.** Brownies Hedonic Test Results

| Brownies Formulation | Sensory Attributes | | | |
|:---:|:---:|:---:|:---:|:---:|
| | Texture | Aroma | Flavor | Color |
| A | 5,12[b] | 5,28[b] | 5,64[b] | 5,16[a] |
| B | 4,56[ab] | 4,64[ab] | 4,44[a] | 4,92[a] |
| C | 4,16[a] | 4,28[a] | 4,08[a] | 4,72[a] |
| D | 4,60[ab] | 4,84[ab] | 5,24[b] | 5,08[a] |

Note: The values in the column followed by unequal letters show significantly different results at the 0.05 level according to the Tukey test results.

The average preference value for brownie texture is 4.16 – 5.12, which is included in the neutral to moderate category. The highest average texture value is brownies I or brownies made from 100% mocaf flour with a value of 5.12 (somewhat like), while the lowest value is brownies C or brownies made from 40% mocaf flour and 60% black soybean flour with a value 4.16 (neutral). The results of statistical tests show that there is a significant effect between the four brownie samples on the preference value of brownie texture. Based on the results of the analysis, it can be seen that the more addition of black soybean flour, the level of preference for brownie texture decreases.

It is different in the case of making cookies made with the addition of black soybean flour which is preferred by the panelists (Widiawati and Anjani 2017). This is because cookies are preferred with a crunchy texture, while brownies are preferred with a softer texture. This correlates with the brownie hardness test data in Table 10 which shows that the more black soybeans are added, the higher the brownie texture (hardness) value and the lower the preference for brownies in the organoleptic test.

The aroma of brownies has an average preference value of 4.28 – 5.28, which is included in the neutral to moderate category. The highest average value is in the brownie formula I (100% mocaf flour) which is 5.28 (likes), and the lowest is in brownies C (40% mocaf flour, and 60% black soybean flour) with a value of 4.28 (netral). The low level of preference in the formulation of brownies C with the addition of black soybean flour is much as 60% which causes an unpleasant odor that can still be smelled. The unpleasant smell of black soybeans is caused by the activity of the lipoxygenase enzyme (Fizriani *et al.,* 2019). The results of statistical tests, shows that there is a significant effect between the four brownie samples on the preference value of the brownie aroma.

The aroma of brownies has an average preference value of 4.28 – 5.28, Taste is an important parameter in consumer acceptance of a product. The test results on the level of brownie taste preference showed an average value of 4.08 - 5.64 including in the neutral to like category. These results are in line with previous research on the taste acceptance of cookies with black rice flour and black soybeans with a ratio of 65% black rice flour and 35% black soybeans with a value of 2.68 ± 0.98 in the neutral category (Widiawati and Anjani, 2017). The highest level of preference for the taste of brownies is in brownies I with a value of 5.64 (likes), and the lowest value is in brownies C with a value of 4.08 (neutral). the results of statistical tests, it shows that there is a significant effect between the four brownie samples on the value of brownie taste preferences.

The results of the test on the level of preference for brownie color showed that the average value was not much different, namely 4.72 – 5.16, which was included in the moderately like category. The results of statistical tests showed that there was no significant effect between the four brownie samples on the value of brownie color preference.

The overall results of the hedonic test method on the texture, aroma, taste, and color of brownies resulted in the order of brownies in order of the highest to lowest values, namely Brownies A, Brownies B, Brownies C, and Brownies D. From the results of the analysis, it is known that the panelists do not like the addition of too much black soybean flour. However, when compared to the 3 brownies formulas with a mixture of mocaf flour and black soybean flour, the highest level of acceptance is in formula D with a ratio of 60% mocaf flour and 40% black soybean flour). The lowest value is in brownies C with a ratio of 40% mocaf flour and 60% black soybean flour).

### 3.3. Chemical Characteristics
*Water Content*
    Based on the results of the analysis, it can be seen that the treatment of the formulation of mocaf flour and black soybean flour on the water content of brownies is as shown in Table 4.

**Table 4.** Analysis of Brownies water Content

| Treatment | Water content (%wb) |
|---|---|
| A | $16,13 \pm 2,182^a$ |
| B | $15,88 \pm 1,885^a$ |
| C | $15,45 \pm 2,388^a$ |
| D | $15,33 \pm 1,412^a$ |

Note: The numbers in the column followed by unequal letters are significantly different at the 0.05 level according to the Tukey test.

    Water is an important component in food because water can affect the appearance, texture, and taste of food. Water content that is too low will cause the brownie texture to be too dry or less moist (Purnomo *et al.*, 2015). The water content in food ingredients determines the acceptability, freshness, and durability of the product. Therefore, an analysis of the water content was carried out with the aim of knowing the amount of water contained in the resulting brownie product (Winarno, 2008).
    Based on Table 3, it is known that the highest water content is found in treatment A (control) which is 16.13% and the lowest water content is in treatment D (60% Mocaf flour: 40% Black soybean flour) which is 15.33%. This is because the water content of mocaf flour is higher when compared to the water content of black soybean flour, where the water content of black soybean flour is around 8.33% (Putri and Triandita, 2018) while the water content of mocaf flour is 13% (Amanu and Susanto, 2014).

*Ash Content*
**Table 5.** Ash Content Analysis of Brownies

| Treatment | Ash Content (%wb) |
|---|---|
| A | $0,81 \pm 0,080^a$ |
| B | $0,80 \pm 0,1558^a$ |
| C | $0,73 \pm 0,059^a$ |
| D | $0,76 \pm 0,033^a$ |

Description: The numbers in the column followed by the letter which are not the same are significantly different at the 0.05 level according to the Tukey test

    Based on the data in Table 5, it is known that the average ash content of brownies is around 0.73-0.81%. Where the ash content of brownies with substitution treatment of mocaf flour and black soybean flour is lower than the results of research conducted by Purnomo *et al.* (2015) where the highest brownie ash content was 1.65% water content of 75% black soybean flour used.

*Fat Content*
**Table 6.** Fat Content Analysis of Brownies

| Treatment | Fat Content (%wb) |
|---|---|
| A | $11,26 \pm 0,15^a$ |
| B | $11,64 \pm 0,15^a$ |
| C | $11,19 \pm 0,16^a$ |
| D | $10,98 \pm 0,04^a$ |

Description: The numbers in the column followed by the letter which are not the same are significantly different at the 0.05 level according to the Tukey test.

    Based on the results of the analysis of variance, it can be seen that no there is an interaction (p 0.05) namely the significance probability value 0.070 is greater than 0.05 then the hypothesis (h0) is accepted, which means that the treatment of mocaf flour and black soybean flour formulations had no significant effect on brownie

fat. The results of the Tukey test showed that there was no diversity in each treatment, meaning that all treatments were not significantly different (non-significant).

The fat content in brownies is produced from various raw materials such as chocolate, margarine, black soybean flour, and other ingredients. The results of the study of fat content from the four treatments had no effect or were not significantly different at the average fat content level of 11%. brownies with mocaf flour, corn, and black soybeans as raw materials have an average fat content of 13% (Fizriani, et al. 2019).

*Protein Content*

Based on the results of the analysis, it can be seen that the treatment of the formulation of mocaf flour and black soybean flour on the fat content of brownies is as shown in Table 7.

**Table 7.** Protein Content Analysis of Brownies

| Treatment | Protein Content (%wb) |
|---|---|
| A | $7{,}17\pm0{,}16^a$ |
| B | $12{,}13\pm0{,}36^c$ |
| C | $15{,}85\pm0{,}34^d$ |
| D | $9{,}94\pm0{,}25^b$ |

Description: The numbers in the column followed by the letter which are not the same are significantly different at the 0.05 level according to the Tukey test.

Based on the results of the analysis of variance, it can be seen that there is an interaction (p 0.05) namely the significance probability value 0.00 is greater than 0.05 then the hypothesis (h0) is rejected, which means that the treatment of mocaf flour and black soybean flour formulations had a significant effect on brownie protein. The results of the Tukey test showed that there was diversity in each treatment, meaning that all treatments were significantly different (significantly).

In table 12 it can be seen that the protein content of formula C which is made from 60% black soybean flour has the highest protein content of 15.85±0.34d compared to formulas A, B, and C, this indicates that the manufacture of brownies made from Black soybean flour can increase protein content. The use of raw black soybean flour in making brownies will increase the protein content, this is because soybeans contain protein, so the addition of black soybean flour helps enrich the protein nutrition of brownies.

The increase in protein in brownies is strongly influenced by the addition of black soybean flour which has a protein content of 33.13 ± 0.36%. The more soy flour added, the higher the protein content in brownie products (Triandita, 2017). the more content of black soybean flour in cookies causes the protein content to increase. The use of black soybean flour as a basic ingredient for making brownies also affects the fiber content of brownies (Widiawati and Anjani, 2017).

The protein content in this product comes partly from eggs, skimmed milk powder, and composite flour. Each gram of protein contained in the product contributes 4 kcal of energy. According to the Indonesian National Standard (SNI. 01-2973-1992) in 1992, the requirement for brownie protein content is at least 6%, this means that the brownie protein content with the formula in table 12 and the addition of black soybean flour meets the SNI requirements.

*Carbohydrate Content*

Based on the results of the analysis, it can be seen that the treatment of the formulation of mocaf flour and black soybean flour on the carbohydrat content of brownies is as shown in Table 8.

**Table 8.** Carbohydrat Content Analysis of Brownies

| Treatment | Carbohydrat Content (%wb) |
|---|---|
| A | $64{,}62\pm2{,}20^c$ |
| B | $59{,}55\pm2{,}07^b$ |
| C | $56{,}68\pm2{,}15^a$ |
| D | $62{,}97\pm1{,}52^c$ |

Description: The numbers in the column followed by the letter which are not the same are significantly different at the 0.05 level according to the Tukey test.

Based on the results of the analysis of variance, it can be seen that there is an interaction (p 0.05), namely the significance probability value 0.00 is smaller than 0.05 then the hypothesis (h0) is rejected, which means that the treatment of the formulation of mocaf flour and black soybean flour has a significant effect on brownie carbohydrates. The results of the Tukey test showed that there was diversity in each treatment, meaning that all treatments were significantly different (significantly).

Carbohydrates from the brownie formula that had the highest content were Treatment A (64.62±2.20c) which was made from 100% mocaf flour. While the second highest was treatment D (62.97±1.52c) with 60% mocaf flour. Based on the research results of Fiziraini *et al* (2019) Mocaf flour is a very high source of carbohydrates from the manufacture of brownies, mocaf flour which is a modification of starch from cassava certainly greatly affects the carbohydrate content of brownies that mocaf flour (83.22±0.35%) has the highest carbohydrate content compared to corn flour (72.83±0.01%) and black soybean flour (39.61±0.35%), the high carbohydrate content was 60.74% in brownies A (100% Mocaf) because brownies A only uses mocaf flour as raw material for making brownies.

### 3.4. Functional Characteristic's
*Antioxidant Activity*
Based on the results of the analysis, it can be seen that the treatment of the formulation of mocaf flour and black soybean flour on the antioxidant levels of brownies is as shown in Figure 2.
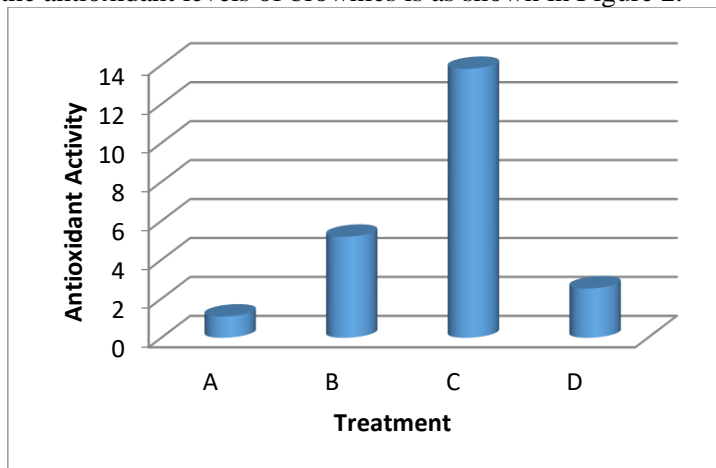


**Figure 2 .** Antioxidant Activity Diagram

Treatment has different antioxidant results, it is known that the highest antioxidant activity is treatment C (40% mocaf flour: 60% black soybean flour) with an antioxidant activity value of 13.81%. This is because the raw material for formula C contains antioxidants from black soybean flour. This is because black soybeans contain a variety of bioactive compounds such as; alkaloids, flavonoids, isoflavones. Black soybeans Besides having a protein content of 40.4g/100 g with complete essential amino acids, it also contains antioxidants, namely anthocyanins and isoflavones. The total content of polyphenols, flavonoids, and anthocyanins was higher than that of yellow soybeans, which were 6.13 mg/g respectively; 2.19 mg/g; 0.65 mg/g. Isoflavones are antioxidants from the flavonoid group found in soybeans and have benefits in people with diabetes mellitus by increasing serum insulin and pancreatic insulin components (Mueller, 2012).

Estimation of antioxidant activity with DPPH radicals aims to determine the ability of brownies to capture radical compounds or their ability as antioxidant compounds. Estimation of antioxidant activity was carried out using the DPPH method. DPPH is a stable free radical and is used to evaluate free radical scavenging in natural materials. The principle of the reaction of this method is that DPPH will be reduced by the hydrogen or electron donation process so that the color will change from violet to yellow with a change in color intensity that is proportional to the number of electron donations followed by a decrease in the absorbance of DPPH (Dris and Jain, 2004). compounds that can cause this can be considered as antioxidants or radical scavengers. The greater the decrease in DPPH absorbance, the stronger the antioxidant activity. In table 9 it can be seen the absorbance of antioxidant activity and %IC50 antioxidant brownies.

**Table 9.** Antioxidant activity and IC50 brownies

| Treatment | Absorbance blank | Solution absorbance | Antioxidant Activity µg/ml | IC 50 | Linear Regression Equation |
|---|---|---|---|---|---|
| A | | 0,464 | 1,090[a] | 2675,73 | y = 0.0187x - 0.0359 |
| B | | 0,445 | 5,188[b] | | |
| C | 0,47 | 0,404 | 13,812[c] | | |
| D | | 0,457 | 2,535[a] | | |

Note: The numbers in the column followed by unequal letters are significantly different at the 0.05 level according to the Tukey test.

Based on the results of the analysis of variance, it can be seen that there is an interaction (p 0.05), namely the significance probability value 0.00 is less than 0.05 then the hypothesis (h0) is rejected, which means that the treatment of mocaf flour and black soybean flour formulations has a significant effect on the antioxidants of brownies. The results of the Tukey test showed that there was diversity in each treatment, meaning that all treatments were significantly different (significantly).

Analysis of antioxidant testing using the DPPH method was carried out by looking at the color changes of each sample after being incubated with DPPH. If all the DPPH electrons are paired with electrons in the extracted sample, there will be a change in the color of the sample starting from dark purple to bright yellow. Then the absorbance value of the sample was measured using a UV-Vis spectrophotometer at a wavelength of 517 nm.

Based on the data from table 9, it is known that the antioxidant activity results have a significant effect on each formulation treatment and has a %IC50 value of 2675.73, this determines that the antioxidants in brownies from the four treatments are included in the weak category. A compound is said to be a very strong antioxidant if the IC50 value is less than 50, strong (50-100), moderate (100-150), and weak (151-200) (Badarinath, 2010). The smaller the IC50 value, the higher the antioxidant activity. This low antioxidant activity occurs due to the heating process that breaks down or opens the network of black soybean flour so that there are active components that can initially be extracted. In the process of frying, the low antioxidant activity is due to the water in the vegetable tissue coming out and being replaced by oil in the vegetable tissue, which will mix with other active components, thereby inhibiting the antioxidant activity of the vegetables. The heating method can affect the texture, nutritional value and antioxidant capacity of black soybean flour (Monreal *et al.,* 2009).

*Glycemix Index*

The glycemic index test was carried out by providing samples of standard food (bread) and test food (brownies A and Brownies C), all test foods containing available carbohydrates were equivalent to 50 grams of glucose. The number of samples given to respondents can be seen in table 10.

**Table 10.** Composition and number of Glycemic Index test samples

| Sample | Karbohidrat by different | Dietary fiber | Karbohdirat avaliabale | Number of samples equivalent to 50 g KH |
|---|---|---|---|---|
| White bread | - | - | - | 106,7g |
| Brownies A | 64,62% | 2,13% | 62,49% | 80 g |
| Brownies C | 56,68% | 2,32% | 54,36% | 92 g |

Based on table 10. that the number of test food samples that must be given to the panelists is equivalent to 50. Food test samples of plain bread were given to respondents 106.7 g, brownies A 80 g, and Brownies C 92 g.

*Blood Glucose Response*

Blood Glucose Response Test Testing samples of white bread (standard), brownies A, and brownies C used 10 respondents. The results of the blood glucose response after consuming the test food can be seen in Figure 3.
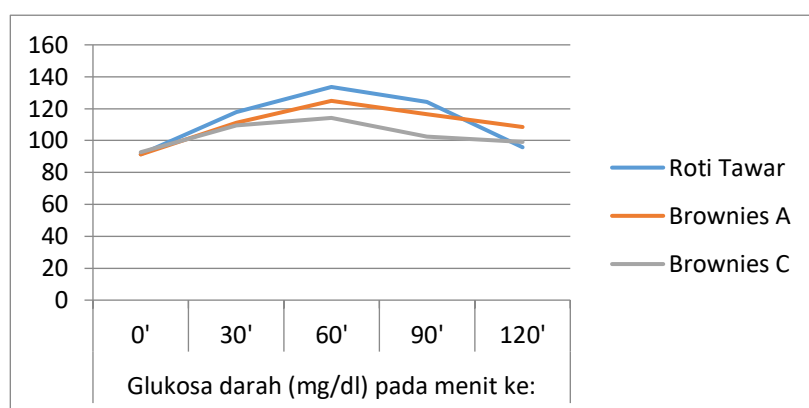
**Figure 3**. Blood Glucose Response

Based on Figure 3, the panelist's blood glucose response after consuming the standard sample and the test sample, it is known that the peak of blood sugar rises at the 60th minute, and decreases in the next minute. The peak of the highest increase in blood sugar levels was found in the provision of plain bread reference food, which was 185 mg/dl. The results of the blood sugar response from the standard sample and the test sample showed that the brownie C sample had a low glycemic index this was due to the formulation of brownies C with 40% mocaf flour and 60% black soybeans, the decrease in blood glucose response was due to black soybeans which had a low glycemic index and Brownies C has fairly high fiber. Table 18 is the result of fiber analysis of the brownie formulation used as a glycemic index sample.

**Table 11.** Fiber content of Brownies formula A and C

| Treatment | Fiber Content (%wb) |
|---|---|
| A = Mocaf Flour 100% : 0%  Black Soybean Flour | 2,13 |
| C = 40% Mocaf Flour : 60% Black Soybean Flour | 2,32 |

Note: Formulations A and C were used as samples of the Glycemic Index Test.

Based on table 11 data, it is known that the fiber content of brownies C is 2.32%, which can affect the decrease in blood sugar response and glycemic index Dietary fiber serves to inhibit the absorption of food in the digestive tract which results in the higher the fiber content of food, the lower the glycemic index of the food (Foresters and Albiner 2004)**.**

Foods without dietary fiber content cause a rapid release of glucose so that it requires a lot of insulin to convert the glucose into energy, dietary fiber can slow down the absorption of glucose in the intestine thereby reducing the need for insulin (Lu *et al.,* 2004)**.**

*Glycemic Index*

The glycemic index of a food is determined by comparing the area of the glycemic response curve for two hours after consuming the test food with the area of the glycemic response curve for two hours after consuming the reference food/standard food. The graph of the blood glucose response of the two ingredients was then calculated for the area of the curve. Then the glycemic index was calculated with these two values (Indrasari and Adnyana, 2007). The results of the calculation of the glycemic index value of the test sample are shown in Figure 4.
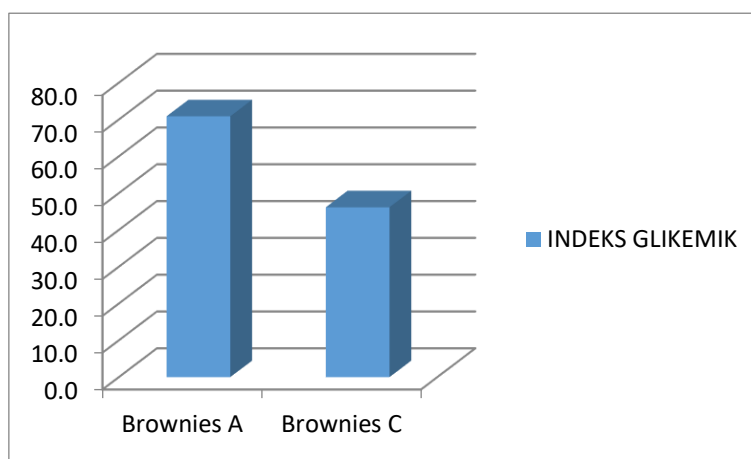
**Figure 4.** Glycemic index value

The glycemic index was determined by comparing the area of the glycemic response curve for two hours after consuming the carbohydrate of the test food with the area of the glycemic response curve for two hours after consuming the standard food. The graph of the blood glucose response of the two ingredients was then calculated for the area of the curve and the glycemic index. After doing the calculations, the glycemic index value of brownies A is 70.9 which is included in the high category, and the glycemic index value of brownies C is 46.2 which is included in the low category. The glycemic index categories are Low GI (<55), medium GI (55-70), high GI (>70).

The bread was determined to be 100 with pure glucose as a standard food for determining the glycemic index of Brownies. The glycemic index value of Brownies C is included in the low category, namely 46.2. So that Brownies C can slow the increase in blood glucose levels. Foods that raise blood sugar levels quickly have a high glycemic index. On the other hand, those that raise blood sugar levels slowly have a low glycemic index. (Forester and albino. 2004). The glycemic index is a way to provide an overview of the relationship between carbohydrates in food and blood glucose response. Foods with a low glycemic index will be digested and converted into glucose gradually and slowly, so the peak sugar content is relatively short. It is very important to control blood sugar levels. Therefore, to control blood glucose levels, it is recommended to consume foods with low GI values. Glycemic index.

*Glycemic Load*

Glycemic Load (BG) is a number that shows the number of carbohydrates in one serving of food. To calculate BG, the following formula can be used: BG = [(GI value x number of available carbohydrates per serving)/100].

**Table 12.** Glycemic Load Formulation of Brownies A and C

| Treatment | Portion (g) | Glycemic Index Value | Carbohydrates available (%) | Glycemic Load |
|---|---|---|---|---|
| Brownies A | 100 | 70.9 | 62.49 | 44.3 |
| Brownies C | 100 | 46.2 | 54.36 | 25.1 |

The purpose of the Glycemic Load is to assess the impact of carbohydrate consumption by taking into account the GI of the food. BG is directly proportional to the carbohydrate content of the food. The lower the carbohydrate content, the lower the BG, the smaller the food served triggers an increase in blood glucose levels. The classification values are low BG (<11), medium BG (11-19), and high BG (>20) (Siagan, 2004). Based on table 11. It was found that the glycemic load of each 100-gram portion of each formulation was 25.1-44.3 so that both brownies formulations were included in the category of high glycemic load.

## 4. Conclusion

The combination of mocaf flour and black soybean flour with different treatment formulations has an effect on texture, aroma, flavor, protein, carbohydrates, and antioxidants. However, it has no effect on color, water content, ash content, fat. Based on the physicochemical analysis of brownies, the best treatment results are brownies with a

combination of 40% mocaf flour and 60% soy flour with a protein value of 15.85%, antioxidant activity of 13.813 g/ml, and a glycemic index of 46.2.

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