

The effect of Baking on Antioxidant Activity, Phenolic Compounds, and Hedonic Value of *Snack Bar* Supplemented with *Moringa* Seeds

Eny Idayati¹, Kartiwan¹

¹*Food Engineering Technology Study Programme, Department of Food Crops and Horticulture, State Agricultural Polytechnic of Kupang, Jalan Prof. Dr. Herman Yohanes Lasiana Kupang 85111*

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ABSTRACT

The snack bar is an alternative to local food development with a combination of nuts and fruits with a dense texture unified by a binding agent. Moringa seeds are known to have high nutritional content and antioxidant activity. This study aimed to analyze the effect of the baking process on the content of antioxidant compounds and the sensory quality of the snack bar supplemented with Moringa seeds. The snack bar treatment used the percentage of Moringa seeds, temperature, and time in the baking process. Determination of the phenolic content of the product using the Folin Ciocalteu method and antioxidant activity using the free radical scavenger DPPH and followed by a preference test (hedonic analysis). The experimental design used was a completely randomized design using ANOVA. If the result obtained showed a significant difference at the 5% ($p < 0.05$), it would then be continued using Duncan's Analysis. The results showed that there were significant increases in the antioxidant activity, phenolic compounds, and hedonic analyses, the higher the percentage treatment of 100% Moringa seeds with a baking time of 40 minutes at a temperature of 130 °C.

Key words: Moringa seeds, Temperature, Time, Antioxidant, Snack bar

Introduction

According to several published studies (Singk *et al.*, 2013 and Gu *et al.*, 2020), Moringa is a source of functional compounds rich in nutrients and bioactive compounds including alkaloid, flavonoid, phenolic, triterpenoid, and steroid (Ikalinus *et al.*, 2015). The compounds have the potential to be developed as medicinal plants with medicinal properties for health. The development of Moringa as an industrial raw material is starting to increase, especially in the fields of pharmacy, cosmetics, and various other benefits related to the environment such as coagulant in wastewater treatment (Paixao *et al.*, 2020). By having several bioactive compounds,

Moringa has high antioxidant and antimicrobial properties (Prasad and Elumalai, 2011), and therefore, Moringa be used as a natural preservative to prolong the shelf life of processed meat-based products that are stored at 4°C without any changing in color (Qwele *et al.*, 2013).

The functional compounds in moringa seed oil consist of 82% unsaturated fatty acids, 70% oleic acid (Kurniaty *et al.*, 2018), resembling the profile of olive oil except for linoleic acid (Tsaknis *et al.*, 1998). In addition, it also contains ascorbic acid, sterols, tocopherols, and flavonoids (Tsaknis *et al.*, 1998; Lalas *et al.*, 2002). Furthermore, the protein content of Moringa seeds could reach 35.97% (Olagbemide *et al.*, 2014) which is higher than the other parts such as

leaves and flowers and thus, has the potential to be an alternative source of protein that is feasible to be developed to meet the protein needs. However, there are several phytochemical compounds such as phenolic, flavonoids, saponins, phytates, cyanogenic glycosides, and glucosinolates which are thought to contribute to the bitter taste of Moringa seeds (Foidl *et al.*, 2001). With the right processing method, it can create products that can be received by the sensory but with a high percentage of antioxidant compounds.

Snack bars are small casual food establishments serving light and easy foods with complete nutritional components such as carbohydrates, proteins, fats, vitamins, and minerals. The application of diversified snack bar products with a combination of local food ingredients, namely dried fruit and nuts fortified with Moringa seeds is expected to become a healthy snack with a complete nutritional composition.

The processing of Moringa seed snack bars goes through several stages, namely washing, soaking, and peeling Moringa seeds, as well as the process of baking the dough using high temperatures which aims to obtain better sensory values, and kill and inactivate pathogenic microorganisms and enzymes and reduce anti-nutritional compounds. However, because of the processing stages during processing, the product experienced a significant reduction in nutritional value such as vitamins, minerals, and protein as well as several antioxidant compounds. Consequently, the levels of functional compounds became decreased.

These study aims are to find the right snack bar processing steps by formulating the percentage of moringa seeds, temperature and baking time in order to maintain the retention of functional compounds based on moringa seeds, by analyzing antioxidant activity, phenolic compounds, and vitamin C as well as hedonic analysis in obtaining a panelist's preferred *snack bar* product.

Materials and Methods

Ingredients

The raw material formulation of functional snack bar products with supplemented moringa seeds consists of *crispy rice*, cornflakes, honey, dried tomatoes, peanuts, and moringa seeds. Moringa seeds were taken from Kupang district at an altitude of

150-500 m above sea level, local tomatoes were dried to a moisture content of 9%, and Natural Honey brand honey was obtained from the "Gunung Mutis" honey center, Fatumnasi district of South Central Timor, East Nusa Tenggara province with total dissolved solids of ± 78 °Brix, peanuts from the local market at Kupang city than went through baking and peeling process with a moisture content of $\pm 3\%$. On the other hand, *crispy rice* and cornflakes were obtained from local supermarkets around the city of Kupang.

Moringa Seed Preparation

The preparation of Moringa seeds to reduce its bitterness undergone several modified stages by Gunawan *et al.* (2020), namely sorting and washing Moringa seeds, then soaking in a 0.5% w/v sodium bicarbonate (NaHCO_3) solution with a ratio of 1:30 w/v (1 g of moringa seeds in 30 ml of solution NaHCO_3) for 24 hours. The process of draining and re-washing the Moringa seeds was followed by a pressurized boiling process with a temperature of $\pm 115^\circ\text{C}$ using a pressure cooker for 10 minutes which was calculated after making a hissing sound. Moringa seeds were then drained and dried at 60°C for 12 hours and stripped to produce white seeds without their skin.

The process of making Moringa Snack bars

The production of snack bars referred to Ibrahim *et al.* (2021) in their research on healthy *snack bars* that have been modified, namely:

1. Mixing all raw materials according to the formulation in Table 1, until evenly mixed.

Table 1. Formulation of Ingredients for *Snack bar Moringa Seeds and Dried Tomatoes*

No	Ingredient	Weight (g)
1	Moringa Seeds	Treatment-dependent
2	Corn Titi	100
3	Rice Crispy	100
4	Honey	100
5	Dried Tomatoes	100

2. Baking of the *Snack bar*

The dough mixture was prepared into 12 (twelve) according to the percentage treatment of Moringa seed weight (0% = control/K0, 100%=K1, 60%=K2), temperature (110=S1, and 130=S2)°C and time (20=W1, and 40=W2) minutes during the baking process. For the next step, an 18 cm x 18 cm tin was

prepared, and put parchment paper in it so that the dough did not stick to the tin at the time of molding. Before baking the dough, the oven (Cosmos type CO-958) was preheated until it reached the treatment temperature. The dough that had been mixed, was then poured into the pan until the dough was evenly distributed on each side of the pan. The baking pan was then put inside the oven, during the treatment period. After the bar snack was cooked, it was let to cool down for 30 minutes, removed from the pan, and then cut into 2 x 2 x 10 cm sizes to get a similar size. The packaging of the moringa seed fortification snack bar product used food-grade plastic with a size of 5.5 x 13 cm.

The statistical method used in this study was Completely Randomized Design (CRD) with 3 repetitions and 2 samples without addition as controls. Data-obtained were analyzed statistically by analysis of variance (ANOVA) at a significant level of 5%. A significantly different result would be proceeded by Duncan's Multiple Range Analysis (DMRT).

Determination of antioxidant activity using the DPPH method (Yen and Cheng, 1995)

The determining antioxidant activity in the meso-carp of lontar fruit was:

$$\% \text{ antioxidant activity} = \frac{\text{absorption of control} - \text{absorption of sample} \times 100\%}{\text{absorption of control}}$$

Absorption of sample: the absorption of the reaction results between 5.0 ml of DPPH with a concentration of 0.4 mM with 1 ml of sample extract.

Determination of Total Phenolic Compounds Folin-Ciocalteu Method (Orak, 2006)

Determination was conducted by weighing 100 mg of extract and dissolved it in 10 ml of distilled water to obtain a concentration of 10 mg/ml, where then 1 ml was pipetted and diluted with distilled water to 10 ml, to have an extract concentration of 1 mg/ml. As much as 0.2 ml of extract was pipetted, added with 15.8 ml of distilled water and 1 ml of Folin-Ciocalteu reagent, then mixed by shaking. It was then let stand for 8 minutes then added 3 ml of 10% Na₂CO₃ to the mixture. The solution was left for 120 minutes at room temperature. The absorbance was measured with a UV-Vis spectrophotometer at a wavelength with an absorbance peak of 765 nm. The repetition was carried out thrice to get the volume of phenol content produced equivalent to gallic acid/g of fresh sample.

Determination of Hedonic Analysis

The hedonic Analysis was carried out by 25 semi-trained panelists with a rating scale of strongly dislike (score 1), dislike (score 2), neutral/normal (score 3), like (value 4) and strongly like (score 5) on the color, aroma, texture and taste attributes (Shobha *et al.*, 2015), as well as unpleasant taste in snack bars due to the *after taste* of Moringa seeds. To achieve similar product conditions in the Analysis, samples were pre-stored at room temperature for at least 2 hours (Soukoulis *et al.*, 2008). Each of the samples was placed in a plastic bowl with 6 samples in the packaging, coded with a three-digit random number and presented sequentially to the panelists in separate booths, to assess the level of preference for product attributes as a result of the treatment of the percentage of *Moringa* seeds and the effect of baking. Test between the average treatments carried out by the BNJ test with 5% significance level.

Results and Discussion

Antioxidant Activity Analysis

The results of the antioxidant activity Analysis (%) on *the snack bar* supplemented with Moringa seeds could be seen in Figure 1.

The data in Figure 1 shows a significant increase in the antioxidant activity for the treatment of the percentage of supplemented Moringa seeds in the *snack bar* that was higher even though there was a treatment temperature and length of baking time, namely 100% treatment at 130 °C for 40 minutes. The increased antioxidant activity is due to the presence of components of bioactive compounds such as tocopherols, phenolic compounds, and flavonoids in *Moringa* seeds (Adebayo *et al.*, 2018; Sharma *et al.*, 2020). Meanwhile, the effect of temperature and baking time also affected the percentage of antioxidant activity in the product, which increased along with the higher temperature and length of baking time, presumably due to the reduced water content resulting in the percentage of antioxidant activity becoming higher. Ozcan *et al.* (2020) report an increase in gallic acid, 3,4-dihydroxybenzoate, trans-cinnamic, coumarin acid, and quercetin on drying kiwi in the oven. It is associated with the concentration of compounds due to evaporation moisture so that phenolic compounds are retained in the material and its content increases. The oven drying process by Slatnar (2011) can increase chlorogenic acid,

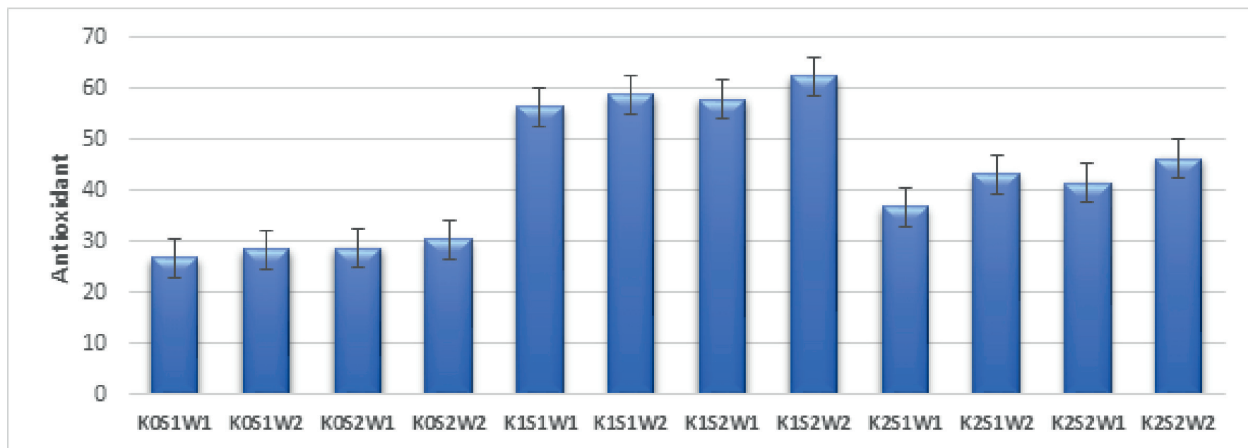


Fig. 1. Antioxidant activity levels (%) of Moringa Seed *Snack bar* samples

catechins, epicatechin, kaempferol-3-O-glucoside, rutin, and Quercetin-3-O-glucoside in dried fruit than in fresh fruit. Improvements that occur due to damage to cell walls by heat or effects easy release macrowave and extraction of phenolic compounds during the process of rehydration. It is also suspected that this treatment damaged plant tissues so that the active ingredients released would increase. In line with the statement of Khatun *et al.* (2006) that the increase in antioxidant activity occurred continuously at an optimal temperature until it decreased due to the loss of antioxidant compounds due to prolonged high temperatures.

Total Phenolic Compound Analysis

The analysis results of total phenolic compounds (%) on *snack bars* fortified with Moringa seeds can be seen in Figure 2.

Figure 2 shows the tendency for the highest total content of phenolic compounds to be found in 100% moringa seed fortification samples compared to 60%. Moreover, the control sample was without for-

tification treatment, while the sample with the highest temperature treatment and the longest time during the baking process showed a higher total phenolic value. These results were almost in agreement with Wang *et al.* (2012) who stated that the long extraction time allowed exposure to active bubbles to disrupt cells, thereby increasing the total amount of phenol. However, prolonging the extraction time can expose phenolic compounds to light and oxygen, enabling it to be oxidized, resulting in a decrease in total phenolic (Bazykina *et al.*, 2002). Sari *et al.* (2012) in the research of *Kappahycus alvarezzi* stated that the total phenol will increase by the time of extraction and increase the temperature until it reached the optimal limit and will decrease by as much as if it exceeds the optimal limit.

Hedonic Analysis

The average value of the hedonic Analysis results on the Moringa seed fortified *snack bar* sample can be seen in Figure 3. Statistically, the percentage of moringa seed fortification treatment, temperature

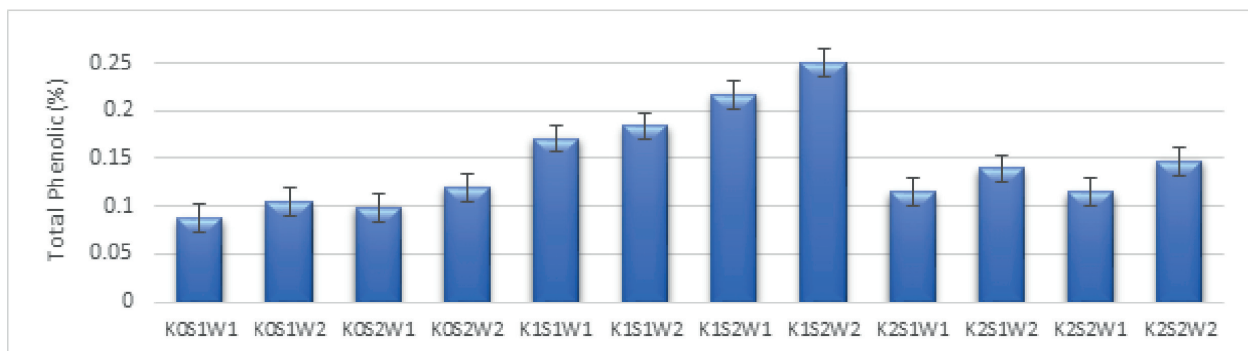


Fig. 2 . Total Phenolic Compound Concentration (%) of Moringa Seed *Snack bar* sample

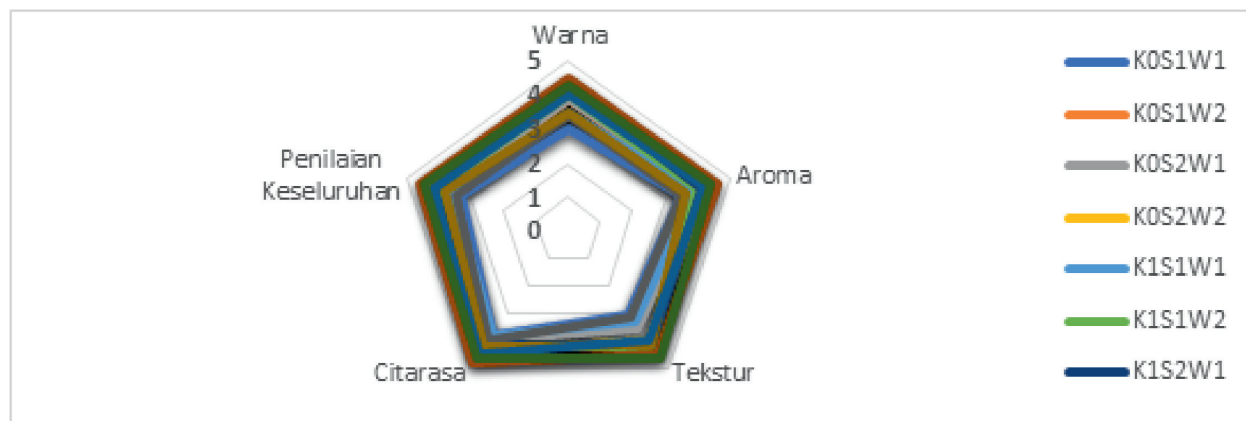


Fig. 3. Hedonic Analysis of Moringa Seed *Snack Bar* Sample

and baking time had an effect on the color, texture, and taste attributes of the *snack bar*.

Based on the results of the analysis, the panelists' level of preference for an overall assessment of the sample, the highest score was in the baking process with 10 0% fortification treatment of Moringa seeds at a temperature of 130 °C for 40 minutes.

From the graph presented, the average hedonic Analysis value for the taste attribute was between 3.8 (neutral) to 4.6 (like). The panelists' preference for the *snack bar* was still acceptable with the percentage of Moringa seed supplemented reaching 100% although the panelists could feel the sensation of typical Moringa seeds aftertaste from the moringa supplemented *snack bar*, compared to samples without supplement. The pre-treatment process on Moringa seeds greatly helped reduce the unpleasant and bitter taste in the final product (Gunawan *et al.*, 2020). On the other hand, the higher temperature and baking time resulted in a sweeter taste due to the caramelization process in honey and moringa seeds during *snack bar* baking. The caramelization process generally occurred due to the influence of high temperature treatment, causing the Maillard reaction in honey. Research by Antony *et al.* (2000) showed that the Maillard reaction to honey produces an attractive taste and smell.

In the color attribute, the percentage treatment of the amount of Moringa had a significant effect between 0% and 100% and 60%, while it did not have a significant effect between 100% and 60%. Panelists liked the *snack bar* color attribute from a score of 3 (neutral) to 4.5 (like), while for the treatment, the higher baking time and the temperature were in line with the reaction to the taste attribute, the color attribute of the product became browner. The brown

color formed in food products is usually the result of the Maillard reaction. Maillard reaction is a reaction that occurs between carbohydrates, especially reducing sugars, and primary amine groups (Winarno, 2004). This is also supported by Nagai *et al.* (2018) that the Maillard reaction occurs by condensation of reducing sugars such as fructose and glucose containing a carbonyl group (aldehyde or ketone) with an amino group that does not contain amino acids, peptides, or proteins. Tamanna *et al.* (2015) and Lund *et al.* (2017) also reported that the higher the temperature and heating time, the more Maillard reactions occurred, which in turn, affected changes in food color, and organoleptic properties and a decrease in the value of some nutrients.

A higher percentage of Moringa seeds would also increasingly form a denser texture of the product but was still within the limits of the panelists' favor with a score between 3 (neutral) to 4.6 (like). For the treatment of solid and sturdy texture, it was in line with the addition of time and temperature during the *snack bar* baking process.

The average hedonic Analysis value for *snack bar* aroma with 0%, 100%, and 60% percentage addition of Moringa seeds ranged from 3.5 (neutral) to 4.6 (like). Based on the results of panelists' acceptance, all samples with the addition of Moringa seeds had an aroma that was in accordance with the assessment standard, which was normal, especially in the 100% sample which had a better score than the sample without the addition of Moringa seeds. Effects due to treatment time and higher temperatures during the baking process also gave the *snack bar* an aromatic effect, this was due to Moringa seeds containing volatile compounds of which if based on the research by Zhang *et al.* (2021) using HS-SPME-GC/

MS showed that there were 52 volatile compounds identified in Moringa seed oil, including 12 hydrocarbons (20.66%), 3 acids (23.99%), 8 alcohols (8.28%), 2 esters (2.61%), 8 aldehydes (14.58%), 2 ketones (0.82%), 7 phenols (6.25%) and 10 heterocyclics (8.74%). The high content of acids, hydrocarbons, and aldehydes covered 59.23% of the total volatile compounds which served as an important factor that contributes to the overall aroma in the final product.

Conclusion

Based on the study results, the treatment samples with the best panelist acceptance were 100% of Moringa seed supplementation, with a baking temperature of 130°C for 40 minutes. There was a significant change in antioxidant activity during the processing of Moringa seed *snack bars* from fresh raw materials to the final product, in which the antioxidant activity decreased by 25-30%, while the total phenolic compounds decreased by 50%. However, based on the hedonic analysis, treatment with 100% Moringa seeds still gave the best result as the most preferred by the panelists compared to other treatments with lower concentrations of Moringa seeds.

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