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Effect of Rosella (*Hibiscus Sabdariffa* l.) Addition on Antioxidant Activity and Acceptability of Apple Mangrove (*Sonneratia Caseolaris*) Slice Jam

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ABSTRACT

The apple mangrove fruit (*Sonneratia caseolaris*) is highly nutritious, containing vitamins A, B, B2, and C, as well as phytochemicals such as flavonoids, steroids, and alkaloids. This study develops apple mangrove fruit sheet jam to improve conventional jam's practicality. However, the final product's unattractive color requires natural colorants and bioactive compounds to enhance antioxidant activity. Roselle flower extract (*Hibiscus sabdariffa* L.), rich in anthocyanins with antioxidant properties, was used as a natural colorant. The research examines the effect of roselle extract on the antioxidant activity and acceptability of Apple mangrove fruit sheet jam using a Completely Randomized Design (CRD) with four treatments: control (without roselle extract), and additions of 30% (P_1), 40% (P_2), and 50% (P_3) roselle extract, each with five replications. Antioxidant activity, hedonic testing, and proximate analysis (carbohydrates, fats, moisture, ash, and protein) were measured. Data analysis was performed using SPSS with one-way ANOVA and Duncan's Multiple Range Test (DMRT) at a 95% significance level. Results show that adding roselle extract significantly enhances antioxidant activity, with the lowest IC_{50} value at 50% treatment (P_3) at 25.380 ppm compared to the control (P_0) at 55.429 ppm, improves product acceptability.

INTRODUCTION

Apple mangrove fruit (*Sonneratia caseolaris*) is part of the mangrove ecosystem that can be processed into food products because it is non-toxic, can be eaten directly, and has a distinctive aroma and sour taste. According to Manalu (2013), 100 grams of apple mangrove fruit contains vitamin A (11.21 mg), vitamin B (5.04 mg), vitamin B2 (7.65 mg), and vitamin C (56.74 mg). The fruit also contains phytochemical components such as flavonoids, steroids, alkaloids, triterpenoids,

tannins, phenolics, phytosterols, saponins, carbohydrates (77.57%), and proteins (9.21%) with a water content of 84.76% (Dewi and Titik, 2023).

Diversification of food products from apple mangrove fruit includes sheet jam, a modification of conventional jam to make it more practical in serving (Simamora and Evy, 2017). Apple mangrove fruit has a soft texture, making it suitable for processing into sheet jam to produce a good product texture. However, preliminary research shows that the final product's color is less attractive (opaque/pale brown), so natural coloring and bioactive compounds are needed to increase the antioxidant activity of Apple mangrove fruit sheet jam.

Rosella (*Hibiscus sabdariffa* L.) is one of the natural colorants containing anthocyanins, flavonoid compounds with antioxidant properties that can reduce free radicals and support the healing of degenerative diseases (Djaeni *et al*., 2017). This research's novelty lies in using rosella to increase the antioxidant activity and visual appeal of apple mangrove fruit sheet jam. In addition to improving antioxidant activity and consumer acceptability, this study also aims to improve the proximate value of the product. Thus, this research is expected to positively contribute to the development of processed apple mangrove fruit products that are healthy and attractive to consumers and support a sustainable local food industry by utilizing Indonesia's marine biological resources.

LITERATURE REVIEW

According to Prabowo (2015), the apple mangrove fruit is a mangrove fruit that lives in brackish waters. This fruit is wrapped by flower petals, with the following classification: Kingdom Plantae, Division Magnoliophyta, Class Magnoliopsida, Order Myrtales, Family Lythraceae, Genus *Sonneratia*, and Species *Sonneratia caseolaris*. Apple mangrove fruit can be utilized as a food source because it has a distinctive sour taste caused by the high ascorbic acid or vitamin C content, as Rahman (2016) mentioned, reaching 56.75 mg per 100 grams of fruit. Manalu (2013) mentioned that apple mangrove fruit is also rich in vitamin A (11.21 mg), vitamin B (5.04 mg), vitamin B2 (7.65 mg), and has phytochemical components such as flavonoids, steroids, alkaloids, triterpenoids, tannins, phenolics, phytosterols, saponins, carbohydrates (77.57%), and proteins (9.21%). The water content reaches 84.76% (Dewi & Titik, 2023).

Sheet jam is a more compact, elastic, and non-sticky innovation than spreadable jam, as described by Abdillah (2021). The addition of ingredients such as starch, agar, gum Arabic, and others are used to improve the texture of this jam. This product has the advantage of being practical in serving, durable, and easy to produce. Its quality standards are regulated following Indonesian National Standard (SNI) 3746:2008, which includes criteria such as odor, taste, color, fruit fiber, soluble solids, and limits for metals such as tin (Sn) and arsenic (As). Sheet jam is

also assessed through organoleptic tests to assess quality and acceptability by consumers (Sipahelut, 2020).

Rosella (Hibiscus sabdariffa L.) is an ornamental plant known as a medicinal plant. Nurnasari (2017) classifies this plant as part of the Kingdom Plantae, Division Magnoliophyta, Class Magnoliopsida, Order Malvale, Family Malvaceae, Genus Hibiscus L., and Species Hibiscus sabdariffa L. This plant is famous because its petals have healing properties against several diseases, such as hypertension and diabetes (Patel, 2014). Rosella petals are used in various food and beverage products, such as salads, tea, juice, pudding, and jelly, as they contain important active substances such as anthocyanins and hibisci glycosides (Djaeni *et al*., 2017). Studies also show that rosella petals contain high antioxidant compounds, such as anthocyanins that play a role in antioxidant and anti-cancer activities (Mastuti *et al*., 2013). Kustyawati (2015) mentioned that rosella petals are also rich in nutrients such as carbohydrates, protein, vitamin C, and iron.

Proximate analysis is an important method in food chemistry that identifies the main components of a food ingredient. This method includes the measurement of moisture, ash, crude protein, crude fat, and carbohydrates, which are calculated differentially (AOAC, 2005). Food ingredients' Moisture content greatly affects the product's biological and physico-chemical quality. Analysis of protein content by the Kjeldahl method provides information on crude protein content (Sumantri, 2013), while carbohydrates are calculated by the differential method (Qalsum *et al*., 2015; Mukti *et al*., 2018). Despite its advantages as a general method, proximate analysis has limitations, such as not providing specific results for each chemical component and not explaining the texture and digestibility of food ingredients (Suparjo, 2010).

The antioxidant activity of a substance can be measured using the DPPH method, in which antioxidant compounds react with DPPH free radicals to protect cells from damage (Novianti, 2019). DPPH is a free radical compound often used in antioxidant research because of its ease of testing (Alam *et al*., 2013). This assay produces an IC_{50} value indicating the sample concentration required to inhibit 50% DPPH free radical activity. This method is fast, simple, and relatively inexpensive for measuring the antioxidant capacity of samples (Shalaby et al., 2013).

An organoleptic test is a sensory evaluation of the quality and characteristics of food products, beverages, or medicines based on observations about their texture, color, aroma, taste, and shape (Nasiru, 2014). This test can use scoring or ranking methods to assess consumer preference or liking for a product. Organoleptic tests are important in evaluating product changes during storage and to ensure the quality received by consumers (Purbowati *et al*., 2020).

METHOD

This research was conducted in January 2024 at the Food and Chemical Analysis Laboratory, Faculty of Fisheries and Marine Sciences, Airlangga University

Surabaya. The main ingredients included Apple mangrove fruit obtained from the Wonorejo Surabaya area and dried rosella petals. Other supporting materials included DPPH (2,2-diphenyl-1-picrylhydrazyl) powder for antioxidant analysis, methanol as a solvent, and distilled water for laboratory needs. The main tool used was the N4S UV-Vis Spectrophotometer, which measures absorbance in the antioxidant test and is supported by additional equipment such as analytical scales, pipettes, measuring cups, and heating equipment.

This study used a completely randomized design (CRD) with four treatments and five replicates. The treatments consisted of P_0 (control) without the addition of roselle extract, P_1 with the addition of roselle extract as much as 30%, P_2 with the addition of 40%, and P_3 with 50% of apple mangrove fruit pulp. The experimental procedure started with the preparation of rosella extract. The method of making roselle extract follows previous research (Mahfud 2015). First, fresh rosella petals were collected and dried to lose water content. Afterward, the dried petals were pulverized using a blender to form a fine powder. This powder is then mixed with water in the appropriate proportion to obtain the optimal extract. Next, the mixture was filtered using a filter cloth to separate the pulp, resulting in a pure roselle extract with a concentration of 100%.

Once the roselle extract was ready, the process continued with the preparation of mangrove apple pulp following the procedure in the previous study (Visakhadevi *et al*. 2024). Ripe apple mangrove fruits were taken, and the pulp was carefully separated from the skin. The existing seeds were filtered out to get a clean fruit pulp. Then, the apple mangrove fruit pulp is mixed with roselle extract, sugar, margarine, and carrageenan in a clean container and stirred well until all the ingredients are well mixed. The mixture is heated in a saucepan over medium heat while stirring constantly until it thickens and reaches the desired consistency. Finally, the thickened mixture was poured into the prepared molds and allowed to cool to harden before being removed from the molds and ready to serve.

This study used an experimental method to evaluate the effect of rosella extract addition on antioxidant activity, acceptability, and proximate value of apple mangrove fruit sheet jam. The antioxidant activity test was conducted using the DPPH method with a UV-Vis spectrophotometer at a wavelength of 517 nm, measuring the ability of the sample to capture free radicals. Proximate analysis included measurement of protein, carbohydrate, water, fat, and ash content to determine the product's nutritional composition. Product acceptability was assessed through the hedonic test by panelists with parameters including taste, aroma, and color. The data obtained were analyzed using one-factor ANOVA and continued with the Duncan Multiple Range Test (DMRT) test to compare the results of antioxidant activity and proximate between treatments. In contrast, the Kruskal-Wallis Test with Mann-Whitney further test was used to test for significant differences in hedonic test results. All data collected were not only tested

statistically, but also analyzed descriptively by comparing with the Indonesian National Standard (SNI 3746-2008) on Jam.

RESULT AND DISCUSSION

The antioxidant activity value of the Apple mangrove fruit sheet jam product was measured using IC_{50}, which indicates the concentration of sample solution required to inhibit 50% of DPPH free radicals (Table 1). The test results showed that the colorant variation significantly affected the antioxidant activity ($p<0.05$). The DMRT test identified significant differences between treatments, with the highest antioxidant activity in treatment P_3 (50%) with a mean of 25.380±0.19 (very strong), and the lowest in treatment P_0 (control) with a mean of 55.429±0.16 (strong).

Table 1: Antioxidant activity values

Treatments	IC_{50} Value (ppm)
P_0 (0%)	$55{,}429^{a}$±0,16
P_1 (30%)	$40{,}450^{b}$±0,08
P_2 (40%)	$30{,}590^{c}$±0,05
P_3 (50%)	$25{,}380^{d}$±0,19

The addition of rosella extract concentration to apple mangrove fruit sheet jam increased the product's antioxidant activity significantly, with significant differences between the control group and the tested treatments. Based on the IC_{50} value, the antioxidant activity was categorized as strong in treatments P_1, P_2, and P_3, and strong in P_0 (Tristantini, 2016). The increase in antioxidant activity is due to the antioxidant content present in apple mangrove fruit, such as steroids, triterpenoids, flavonoids, carboxyl benzene, and vitamin C (Kurniawan, 2022), as well as rosella flowers containing vitamin C, vitamin E, vitamin A (beta carotene), omega-3, and flavonoids (Zuraida, 2015). This study showed that the more rosella extract was added, the higher the antioxidant activity of the apple mangrove fruit sheet jam product studied. The combination of antioxidant compounds in these two ingredients shows synergistic properties, which means that antioxidant activity will increase as the concentration of ingredients increases, resulting in products with higher antioxidant quality (Ramadhan, 2018).

The hedonic test on apple mangrove fruit sheet jam aims to evaluate panelists' liking for appearance, aroma, taste, and texture and determine the most preferred concentration of rosella extract. The results of the Mann-Whitney test show significant differences in all parameters between the control group and the treatment group with rosella extract concentrations of 30%, 40%, and 50% (Table 2). On the appearance parameter, there was a significant difference between the control group and all treatment groups and between the different treatment concentrations. On the aroma parameter, significant differences were found

between the control and all treatment groups, with the highest aroma value at 50% concentration. For the flavor parameter, the treatment groups with 30%, 40%, and 50% concentrations showed higher values than the control group, although there was no significant difference among the treatment groups. In the texture parameter, there was a significant difference between the control group and all treatment groups, as well as between the 30% and 50% and 40% and 50% concentrations, with the highest value at the 50% concentration. These results indicated that the addition of rosella extract significantly improved the sensory quality of apple mangrove fruit sheet jam.

Table 2. Hedonic test results

Parameters	Mean Value of Hedonic Test Sample			
	Appearance	Aroma	Flavor	Texture
P_0 (0%)	$3,40^{a}$±0,724	$3,13^{a}$±1,008	$3,40^{a}$±0,855	$3,40^{a}$±0,724
P_1 (30%)	$4,13^{b}$±0,629	$3,63^{b}$±0,615	$3,93^{b}$±0,785	$4,13^{b}$±0,629
P_2 (40%)	$4,27^{c}$±0,640	$3,80^{b}$±0,847	$3,97^{b}$±0,809	$4,07^{b}$±0,691
P_3 (50%)	$4,70^{d}$±0,466	$3,87^{b}$±0,819	$4,10^{b}$±0,607	$4,47^{c}$±0,571

The addition of rosella extract to apple mangrove fruit sheet jam significantly changed the appearance, aroma, taste, and texture of the product, and improved its sensory quality so that it was accepted by panelists according to SNI 3746:2008. Anthocyanins in rosella provide an attractive red to purple color and are accepted by panelists, creating a fresh and natural impression that is often associated with product quality (Yuliani, 2011; Nurmasari, 2017). The sour taste of rosella extract, caused by citric acid and malic acid, provided a significant difference between the control and treatments, with all treatments being preferred by panelists (Yuliani, 2011; Agustin, 2020). The sour aroma of rosella, which was also accepted by panelists, reflects the influence of citric acid and malic acid present in rosella flowers (Negara, 2016). The texture test showed that the addition of rosella extract improved the texture of the jam, especially at 50% concentration, which resulted in a more elastic texture due to the interaction of liquid rosella extract with hydrocolloid materials (Iryandi, 2014). The hedonic test results indicated that all sensory parameters (appearance, aroma, taste, and texture) showed significant differences between the control and treatment groups, with the treatment group at 50% concentration showing the highest value and most preferred by the panelists.

Proximate analysis of Apple mangrove fruit sheet jam with the addition of rosella extract showed significant differences in ash, water, fat, and protein content between the control and treatment groups, but there were no significant differences between variants within the treatment groups. The DMRT test in Table 3 revealed that treatment P_1 had the highest ash content (1.824±0.02), while the control P_0 had the lowest (1.648±0.02). The highest moisture content was found in P_3 (48.466±0.04) and the lowest in P_0 (43.446±0.08), indicating an increase in

moisture content as the concentration of rosella extract increased. P_3 treatment also showed the highest fat content (1.842±0.02) compared to P_0 control (1.768±0.01). The highest protein level was found in P_1 (1.594±0.02), with the P_0 control having the lowest level (1.350±0.01). These data indicate that the addition of rosella extract improved the proximate quality of the jam, especially in terms of water, fat, and protein content.

Table 3. Proximate analysis of apple mangrove slice jam

Treatments	Averages of Proximate analysis (%)				
	Carbohydrate	Protein	Fat	Water	Ash
P_0 (0%)	51,788^{a}±0,08	1,350^{b}±0,01	1,768^{a}±0,01	43,446^{d}±0,08	1,648^{a}±0,02
P_1 (30%)	49,300^{b}±0,15	1,594^{a}±0,02	1,816^{b}±0,02	45,466^{c}±0,14	1,824^{b}±0,02
P_2 (40%)	46,844^{c}±0,03	1,576^{a}±0,01	1,832^{b}±0,01	47,932^{b}±0,02	1,816^{b}±0,01
P_3 (50%)	46,328^{d}±0,05	1,584^{a}±0,01	1,842^{b}±0,02	48,466^{a}±0,04	1,800^{b}±0,01

The addition of rosella extract to apple mangrove fruit sheet jam affects the product's proximate value, including moisture, ash, fat, protein, and carbohydrate content. The proximate content of rosella flowers consists of 11.1 grams of carbohydrates, 1.6 grams of protein, 0.1 grams of fat, and 2.5 grams of fiber (Kustyawati, 2015). The addition of rosella extract caused a decrease in the carbohydrate content of jam from 51.788% to 46.328% due to nutrient redistribution (Junianto, 2020; As *et al*., 2023). Protein content increased from 1.350% to 1.584% due to amino acids contained in rosella (Paramita, 2022; Kustyawati, 2015). Fat content also increased from 1.768% to 1.842%, influenced by the fat contained in rosella and the margarine used (Nurhidayanti *et al*., 2017; Crisan *et al*., 2022). In addition, the moisture content increased from 43.446% to 48.466%, in line with the high moisture content of rosella at 87.41% (Rajis *et al*., 2017; Rosalia *et al*., 2016). The addition of rosella extracts also increased the ash content from 1.65% to 1.8%, reflecting the increase of minerals in the jam product (Octavia *et al*., 2015; Komala *et al*., 2013). The assay results of carbohydrate percentage were measured, which involves subtracting other components such as protein, fat, water, and ash from the total composition. Increasing rosella extract increased the water content in apple mangrove fruit jam, indicating that more water was measured. Overall, the addition of rosella extracts improved proximate quality and provided additional benefits to the apple mangrove fruit sheet jam product, reflecting the positive relationship between rosella proximate content and improved jam quality.

CONCLUSION

The results showed that adding rosella flower extract to Apple mangrove fruit sheet jam significantly improved the product's antioxidant activity and hedonic parameters. The highest antioxidant activity was achieved with the addition of 50% rosella extract (P_3), resulting in an IC_{50} value of 25,380 ppm, which showed a significant increase compared to no rosella extract ($p < 0.05$). In addition, adding rosella extracts also improved the consumer acceptability of the product, making it more favorable compared to the product without rosella extract.

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