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Identification of Antioxidant Compounds using the DPPH Radical Scavenging Method from Ethanol and Methanol Extracts of the Leaves and Fruits of the Renggak Plant (*Ammomum dealbatum* R.)

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ABSTRACT. This study aims to screen antioxidant compounds in the renggak plant (*Amomum dealbatum* R.). Phytochemical screening was carried out on the leaves and peel of the renggak fruit by the extraction method using ethanol and methanol solvents. Extraction was carried out by maceration, and the antioxidant potential test was carried out by measuring the extract's scavenging activity against DPPH radicals. In the fruit peel, both ethanol and methanol extracts tested positive for all phytochemicals, while in the leaves, ethanol extract was positive only for tannins and alkaloids, and methanol extract for flavonoids, saponins, tannins, and alkaloids. Differences in solvent polarity caused the difference in results. The results of the antioxidant test showed that the leaf extract had an IC_{50} value of 44.31 µM, while the fruit peel extract had an IC_{50} value of 144.57 µM. The lower IC_{50} value in the leaf extract indicates that the bioactive compounds contained therein have higher antioxidant potential than the fruit peel, as they can inhibit 50% of free radical activity at lower concentrations. The results of this study provide additional phytochemical and antioxidant candidates from plant sources for use as raw materials in the pharmaceutical and cosmetic industries.

INTRODUCTION

Phytochemical compounds are secondary metabolites synthesized by plants that are not classified as nutrients but serve primarily as a form of protection or defense. These compounds are typically present in different plant parts, including leaves, fruits, stems, and roots (Bachheti *et al*., 2020). Various groups of these substances, such as flavonoids, terpenoids, tannins, saponins, and alkaloids, are widely recognized for their significant roles in health and pharmaceutical applications. For instance, flavonoids, which belong to the polyphenol group, have been reported to exhibit diverse bioactivities, including antiviral, anti-inflammatory, cardioprotective, antidiabetic, anticancer, anti-aging, and antioxidant properties (Arifin and Ibrahim, 2018). Likewise, alkaloids are known for their ability to combat microbial infections, regulate blood pressure, and stimulate the nervous system (Carrie *et al*., 2022).

Renggak (*Amomum dealbatum* R.) is a herb known locally as "Alachengay" and a member of the Zingiberaceae family (Dhakal *et al*., 2023). This common plant on Lombok Island has not been widely used as a medicinal plant. Renggak is often found in forests, has tall leaves, and is known by the Lombok community as a plant with edible fruit that can cure headaches. The benefits of Renggak are not yet known to the public. Therefore, this plant is less popular among the public. Renggak has a high content of primary and secondary metabolites, making it suitable for use as a medicinal plant (Chelleng *et al*., 2024; Muliasari *et al*., 2019).

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Phytochemical compounds in the fruit and leaves of *A. dealbatum* R. are thought to possess various beneficial activities; however, studies on the chemical components of various parts of this plant are still limited. Therefore, further research is needed to identify and characterize the bioactive compounds present in this plant. Phytochemical screening is a qualitative test of the chemical compounds present in plant parts, especially secondary metabolites such as flavonoids, alkaloids, saponins, tannins, and terpenoids (Adekanmi *et al*., 2020). The choice of solvent and extraction technique is an important factor in the phytochemical screening process. The desired active compounds cannot be effectively and completely extracted if the solvent used is inappropriate (Kerton and Marriott, 2013). Ethanol and methanol are organic solvents commonly used in phytochemical research because of their ability to extract polar and semi-polar compounds from plant materials. Methanol can dissolve polar and nonpolar compounds, making it very effective for dissolving chemical compounds (Xu *et al*., 2024). Ethanol has a relatively high solubility and is inert (Arsa and Achmad, 2020).

Antioxidant compounds are molecules that can protect body cells from damage caused by free radicals by inhibiting oxidation reactions (Fakriah *et al*., 2019). Pollution, contamination, radiation (sunlight with a thin ozone layer), fatigue, stress, and various diseases they cause can result in the body releasing excessive oxygen radicals (free radicals). Free radicals cause many health problems. Antioxidants are compounds that have a molecular structure that can donate electrons freely to free radical molecules without being disturbed at all, and can break the chain reaction of free radicals. We find many antioxidants in plant-derived foods (Anggarani *et al*., 2023). Antioxidant activity of the Renggak plant from various regions has different activities. Research by Pintatum and Laphookhieo (2022) showed weak activity (179.8 ± 3.9 mg/L), Azim *et al*. (2025) strong activity (66.515 ± 2.37 mg/L), and Ayu *et al*. (2021) weak activity (150 mg/L). Human demand for new basic ingredients for medicines and skincare continues to increase, in line with rising health standards and human needs (Islam *et al*., 2019; Sovia *et al*., 2020). One way to obtain these preparations is through optimizing local Indonesian plants. The Renggak plant has long been used traditionally as a headache remedy.

Exploration of the phytochemical composition and antioxidant potential of *Amomum dealbatum* Roxb., a plant traditionally used as a headache remedy, but with still very limited scientific studies. Unlike previous studies that generally examined popular medicinal plants, this study is the first to comprehensively qualitatively screen secondary metabolites (flavonoids, alkaloids, saponins, tannins, and terpenoids) in the fruit and leaves of *A. dealbatum* and systematically compare the effectiveness of ethanol and methanol solvents in the extraction process. Integrating traditional knowledge and modern phytochemical analysis, this study not only validates the plant's ethnomedical use but also provides opportunities to discover new bioactive compounds with antioxidant potential for pharmaceutical and skin care applications. Based on its description, research will be conducted to test the antioxidant activity of the Renggak plant (*Ammomum dealbatum* R.) and to provide scientific data on its antioxidant properties.

RESEARCH METHODS

The tools and materials used in this study include instruments and supporting equipment. The tools used were a GC-MS instrument (Shimadzu: QP2010Ms), UV-Vis spectrophotometer (REIGN UV-1900), quartz cuvettes, and supporting glassware. The materials utilized were obtained from Merck Supelco, which included Pro-Analyze ethanol, Supelco Pro-Analyze methanol, 2,2-diphenyl-1-picrylhydrazyl (DPPH), Dragendorff's reagent, Wagner's reagent, Mayer's reagent, 1% $FeCl_3$, acetic acid, and sulfuric acid.

Extraction of Renggak

Samples of Renggak leaves and fruit were collected in Korleko Village, Labuhan Haji District, East Lombok (8°37′22.8″S 116°36′3.6″E). The Renggak fruit was cleaned, and the skin was peeled to separate it from the flesh. The fruit skin was sliced thinly. The fruit skin was air-dried for 3 – 7 days. The dried Renggak fruit skin was blended until a fine powder was obtained. Dry powder of fruit skin and Renggak leaves was extracted with a ratio of sample powder to solvent of 1:10. The sample powder was 50 g, while the solvent used was 500 mL. The maceration process was repeated 3 times in 24 hours (3 × 24 hours). Then, the macerated material was filtered through filter paper and collected in a jar. The macerated liquid extracts were concentrated using a vacuum rotary evaporator at 40 °C for approximately 1 hour to obtain a thick extract from each sample.

Phytochemical Screening

Alkaloid: A total of 1 mL of extract was put into a test tube, and then 1 mL of 2 N HCl was added. Then, 1 mL of each filtrate was transferred to test tubes 1, 2, and 3. A total of 2 drops of the Wagner, Mayer, and Dragendorff reagents were added to test tubes 1, 2, and 3, respectively. All tests show a positive result if a white precipitate forms in test tube 1, a brown precipitate forms in test tube 2, and an orange precipitate forms in test tube 3. For the flavonoid test, 1 mL of extract was placed in a test tube, followed by the addition of 0.1 g of magnesium powder and 1 mL of concentrated HCl. The flavonoid test yields a positive result when the solution color turns yellow or red. For the saponin test, an extract volume of 1 mL was added to 10 mL of distilled water and shaken. A positive result for saponin is obtained if foam forms after being left for 10 – 15 minutes. For phenol and tannin tests: 3 drops of 1% $FeCl_3$ solution were added to 1 mL of extract. A positive result for phenol compounds is indicated by a green, red, yellow, or orange colour, while a dark blue or black colour indicates a positive result for tannin compounds. For the terpenoid test, 1 mL of glacial acetic acid (CH_3COOH) and 1 mL of concentrated sulfuric acid (H_2SO_4) were added to 1 mL of the extract, and a positive result is indicated by the solution changing color to reddish brown.

Antioxidant Activity Test

Antioxidant activity test of leaf and fruit skin extracts was not carried out using the DPPH (2,2-diphenyl-1-picrylhydrazyl) method, which is a spectrophotometric method based on measuring the ability of antioxidant compounds to reduce DPPH free radicals into non-radical forms, which is indicated by a color change from purple to pale yellow (Baliyan *et al*., 2022). In this test, a DPPH solution was prepared in methanol, and absorbance was measured at 517 nm using a spectrophotometer (Molole *et al*., 2022; Thakar *et al*., 2022). Leaf and fruit peel extract samples were diluted to a certain concentration (20 µg/mL, 40 µg/mL, 60 µg/mL, 80 µg/mL, 100 µg/mL), then mixed with DPPH solution and incubated for a specified period to allow the reaction to take place optimally. The decrease in absorbance relative to the control indicates the antioxidant compound's effectiveness in neutralizing free radicals. The percentage of inhibition was calculated from the difference in absorbance between the control ($A_{control}$) and the sample (A_{sample}), while the IC_{50} value was obtained from the relationship curve between sample concentration and percentage of inhibition. This method provides a quantitative picture of the antioxidant capacity of each plant part, using a fast, sensitive approach, and is widely used in the analysis of natural bioactive compounds. The formula commonly used to calculate the % inhibition in the DPPH test is presented in Equation 1.

$$\%\text{inhibition} = \frac{A_{control} - A_{sample}}{A_{control}} \times 100\% \quad (1)$$

RESULTS AND DISCUSSION

Extraction of Renggak Fruit Skin and Leaves

Extraction of Renggak fruit skin and leaves was carried out using the maceration method. Powdered fruit skin and leaves of Renggak, as much as 50 g, were macerated with 500 mL of ethanol and methanol (3 × 24 hours) to produce a concentrated ethanol extract. Several factors affect the maceration process. These include the type of solvent, the size of the simplicia, the duration of the maceration, and the stirring. Due to the concentration difference between the active substance solution inside and outside the cell, during maceration, the concentrated solution is forced out. This event repeats until the concentration between the solution inside and outside the cell is balanced again (Yulandari *et al*., 2023).

Phytochemical Screening

Phytochemical screening was conducted to identify the secondary metabolites present in the plants. The phytochemicals of the plants tested in this study were weak. The results obtained from the phytochemical tests of the Renggak fruit peel and leaf extracts are shown in Table 1. Gas Chromatography-Mass Spectrometry (GC-MS) analysis also identified 20 active compounds with potential biological activities in the methanol extract of Renggak leaves. Only 10 active compounds were detected in the methanol extract of Renggak fruit, although in smaller quantities, indicating the presence of important bioactive components (Figure 1). This difference in the number of active compounds between leaves and fruit indicates variation in phytochemical content across plant parts. These

findings provide important information for selecting the most promising plant parts for further development in phytochemical research and natural product-based industrial applications.

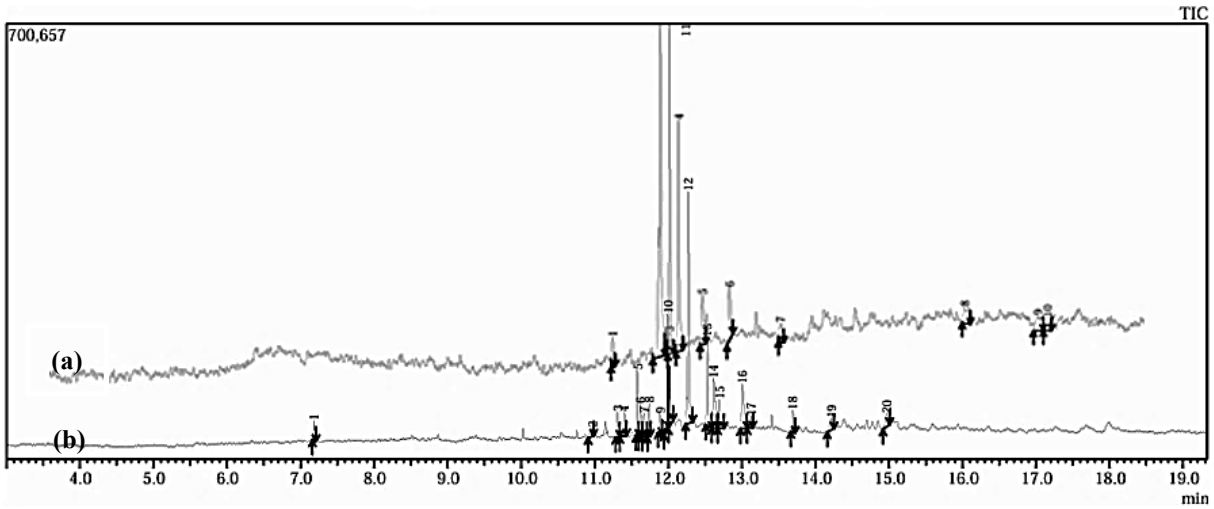

Figure 1. GC-MS test result (a) fruit extract and (b) leaf extract.

Table 1. Phytochemical screening results of the crude extract of Renggak fruit peel.

No.	Compound	Reagent	Identification	Phytochemical Results	
				Ethanol	Methanol
1.	Alkaloid	Dragendroff	Orange sediment	+	+
		Wagner	Reddish solution	+	+
		Mayer	White sediment	+	+
2.	Saponin	water	A persistent foam layer	+	+
3.	Flavonoid	0.1 gram Mg + 1ml concentrated HCl	Red solution	+	+
4.	Terpenoid	1 ml CH_3COOH + 1 ml H_2SO_4	Brownish-red color	+	+
5.	Tannin	$FeCl_3$ 1%	Blue/black solution	+	+

Table 2. Phytochemical screening results of the crude extract of Renggak leaves.

No	Compound	Reagent	Identification	Phytochemical Results	
				Methanol	Ethanol
1.	Alkaloid	Dragendorff	Orange sediment	+	+
		Wagner	Reddish solution	+	+
		Mayer	White sediment	+	+
2.	Saponin	water	Stable white foam	+	-
3.	Flavonoid	$FeCl_3$ 1%	Red solution	+	-
4.	Terpenoid	1 ml CH_3COOH + 1 ml H_2SO_4	Brownish-red color	-	-
5.	Tannin	$FeCl_3$ 1%	Blue/black solution	+	-

Based on the data from Table 1 and Table 2 above in the alkaloid test, the formation of deposits in the Wagner, Mayer, and Dragendorff tests indicates that there are alkaloids in the extract of the fruit skin and leaves of Renggak. Brown deposits in the Wagner test, white deposits in the Mayer test, and orange deposits in the Dragendorff test indicate that there are secondary metabolite compounds of the alkaloid group in the extract of the fruit skin and leaves of Renggak. The purpose of adding HCl is that alkaloids are basic, so they are usually extracted with solvents containing acid. Meanwhile, treatment of the extract with NaCl before adding the reagent aims to remove protein (Hadi and Permatasari, 2019).

A positive result for alkaloids in the Wagner test was observed for both the ethanol and methanol extracts of Renggak fruit peel, as evidenced by a brownish-orange color change and the formation of a brown precipitate. The

precipitate is presumed to be a potassium-alkaloid complex. In the preparation of Wagner's reagent, iodine reacts with I^- ions from potassium iodide to produce I_3^- ions, which are brown in color. During the Wagner test, the metal ion K^+ forms a coordinate covalent bond with the nitrogen atom in the alkaloid, resulting in the formation of an insoluble potassium-alkaloid complex. A positive result for alkaloids in the Mayer test for ethanol and methanol extracts is indicated by a color change in the solution from yellow-orange to slightly cloudy, as if a white precipitate had formed. The presence of protein precipitates after adding a reagent containing heavy metals (Mayer's reagent) can yield a positive reaction for certain compounds.

A positive alkaloid test using the Dragendorff method occurs when a compound contains an alkaloid, which reacts with the Dragendorff reagent to form an orange-brown or reddish-orange precipitate. This is caused by the interaction between the alkaloid compound and the tetraiodobismuthate(III) ion (Sulistyarini *et al*., 2020), which produces a potassium-alkaloid precipitate. In the preparation of the Dragendorff reagent, bismuth nitrate is dissolved in hydrochloric acid to prevent hydrolysis, because bismuth salts easily hydrolyze and form bismuthyl ions (BiO^+). To keep the Bi^{3+} ions in solution, acid is added to shift the equilibrium to the left. The Bi^{3+} ions from the bismuth nitrate then react with potassium iodide to form a black precipitate of bismuth(III) iodide. This precipitate subsequently dissolves in excess potassium iodide to form potassium tetraiodobismuthate. In the Dragendorff test, the alkaloid nitrogen forms a coordinate covalent bond with the K^+ metal ion. Based on the observation results, the Dragendorff method test showed a brownish precipitate/spot. From the three methods, it can be concluded that the skin of the fruit and leaves of the Renggak positively contain alkaloid compounds.

Saponin is an amorphous compound and has at least one glycosidic bond (C–O-sugar bond) at C-3 between the aglycone and one sugar chain. For the saponin test, 10 mL of distilled water was added to 1 mL of the sample, and the mixture was shaken. The purpose of adding distilled water is to facilitate the hydrolysis of glycoside bonds, which can form foam in water and hydrolyze into glucose and other compounds (Deng *et al*., 2023). So that saponin can form a colloidal solution in water and produce foam or suds when shaken (Akasia *et al*., 2021). The purpose of shaking in this test is to expand the field. Based on the observations, it was found that the skin of the Renggak fruit with both solvents produced a lot of foam and remained constant for up to 10 minutes, whereas in the saponin leaf sample, foam was observed only in the methanol extract. This shows that the fruit skin and the leaves of the Renggak contain saponin compounds.

In qualitative testing of phenolic compounds, a 1% Iron(III) chloride solution can be used. The reaction between $FeCl_3$ (3%) and phenolic compounds results in a strong color change from green to red, purple, blue, or black, indicating that the tested sample contains phenol. From the experiments carried out, the ethanol extract of the Renggak fruit skin showed a yellow color change, while the methanol extract of the Renggak fruit skin showed a yellow solution and a dark yellow precipitate. Meanwhile, in the methanol extract of the leaves, the solution turned orange-yellow. The addition of a 3% $FeCl_3$ reagent to this solution reacts with the hydroxyl group on the aromatic ring of the phenol compound. The following is the phenolic test reaction (Candra *et al*., 2021).

Flavonoids are phenolic compounds consisting of two aromatic rings connected by a three-carbon bond. Flavonoids can be tested using the Shinoda test, in which a color change in the sample to red, yellow, or orange in the amyl alcohol layer indicates the presence of flavonoids. This color change is caused by a reduction in the amount of flavonoid compounds in their aglycone form, which then form a complex with magnesium, resulting in a yellow solution (Villela *et al*., 2019). The results of the flavonoid test on both the ethanol and methanol extracts of the Renggak fruit peel showed a dark red precipitate on the surface of the solution, as did the methanol leaf extract. This proves that the Renggak fruit peel positively contains flavonoids. The following is an image of the reaction from the flavonoid test (Lindawati and Ma'ruf, 2020).

Terpenoids are hydrocarbon compounds consisting of large structures derived from isoprene units (C5). Some terpenoid compounds include one or more double bonds, and their structures consist of cyclic allyl. The principle of terpenoid identification is the Lieberman-Bourchardt test using anhydrous acetic acid and concentrated H_2SO_4. This color change begins with the acetylation of the hydroxyl group with anhydrous acetic acid, which forms a double bond. Then, hydrogen bonds are released, which shift the double bond, allowing the compound to undergo resonance and act as an electrophile or carbocation. The presence of a carbocation leads to the release of hydrogen and its electrons, resulting in conjugation elongation and a brownish-orange color (Sulasmi *et al*., 2018). In the terpenoid test, the skin of the Renggak fruit showed positive results with both solvents, while the leaves showed negative results with both solvents.

In the tannin test, an oxidation reaction occurs between $FeCl_3$ and one of the hydroxyl groups in the tannin, leading to flaming. The addition of $FeCl_3$ produces a blackish-blue color, indicating the presence of condensed

tannins. Because tannins form a complex with $FeCl_3$, the extract turns blackish-blue upon addition of $FeCl_3$ (Munadi, 2018). The tannin test conducted by Fajriaty *et al*. (2018) shows the nature of tannins that can precipitate gelatin. The formation of gelatin precipitate showed good results. Tannin forms a copolymer with a higher specific gravity that cannot be dissolved in water, producing a white precipitate (Nurjannah *et al*., 2022). The test results for the ethanol extract of fruit skin samples show that the solution turns blue, and for the methanol extract, the solution surface turns blue. In the leaf extract, the blue color is only observed with methanol as the solvent. Based on the analysis above, the Renggak fruit skin positively contains tannin compounds.

Based on the comprehensive screening results, the fruit peels tested positive for all tests because they often contain high levels of phytochemical compounds. This is because the peels act as a natural barrier against pathogens and the external environment. The fruit peel, which acts as a primary barrier, often contains a concentration of various active compounds that help protect the fruit from microbes, insects, and UV radiation. Therefore, the fruit peel may contain more phytochemicals than other plant parts, making positive results across all tests more likely (Gonzales *et al*., 2005). Compounds such as flavonoids, terpenoids, tannins, saponins, and alkaloids are often found in high concentrations in the fruit peel as a natural defense mechanism (Harborne, 1998). The phytochemical content of the fruit peel may be easier to extract because its cellular structure allows solvents to penetrate cell walls more easily. These compounds may also be more soluble in organic solvents such as ethanol and methanol, as these solvents can dissolve polar and semi-polar compounds. Methanol, being more polar than ethanol, can extract more polar compounds (such as flavonoids and saponins) more efficiently (Hikmawanti *et al*., 2021).

The fruit peel of *Amomum dealbatum* R. may contain a variety of bioactive compounds, including both polar and nonpolar substances, making it suitable for extraction with both polar and nonpolar solvents. Ethanol and methanol can dissolve compounds with a broad spectrum of polarity. Because the fruit peel is rich in diverse compounds, both solvents can effectively extract various phytochemicals (Sultana *et al*., 2009). Meanwhile, the phytochemical screening results of Renggak leaves showed significant differences. The difference in secondary metabolite compound results between methanol and ethanol extracts was caused by the polarity of the solvent. Methanol solvent is slightly more polar than ethanol solvent, making it more effective in extracting polar and nonpolar compounds. Compounds that can be extracted with methanol include flavonoids, saponins, terpenoids, and tannins (Adisti *et al*., 2023). This is consistent with the phytochemical screening results, which showed that saponins, flavonoids, and tannins were detected in the methanol extract, but not in the ethanol extract. The higher dielectric constant possessed by methanol can increase its ability to enter the cell structure and cell walls of plants, which in turn can cause methanol to extract secondary metabolite compounds more effectively than ethanol.

Methanol has one methyl group (CH_3), which is smaller than the ethyl group (C_2H_5), so that the methanol solvent can penetrate the cell wall further and enter the cell cavity containing the active substance. The active substance will dissolve in the organic solvent outside the cell, and the resulting concentrated solution will then diffuse out of the cell. This process will continue until there is a balance between the concentrations of the active substance in the fluid inside and outside the cell (Stein, 2012). The difference in screening results obtained basically occurs due to the use of different solvents at the polarity level and in accordance with the principle like dissolves like, where the saponin, flavonoid, and tannin compound groups tend to have the same level of polarity as methanol, while alkaloids can be extracted by methanol and ethanol solvents. Based on the results of phytochemical screening using methanol solvent, positive results were obtained in the alkaloid, flavonoid, saponin, and tannin tests, while the terpenoid test showed negative results. The screening results with ethanol solvent showed positive results in the alkaloid test and tannin test, while the flavonoid, saponin, and terpenoid tests showed negative results.

Antioxidant Activity

Natural ingredients have been recognized as promising sources of antioxidants, which play a crucial role in combating oxidative stress, a key factor in the development of various chronic diseases, including cancer, diabetes, and neurodegenerative disorders. Phytochemical compounds such as flavonoids, phenolics, and terpenoids found in plants exhibit antioxidant activity through free radical scavenging, transition-metal ion chelation, and lipid oxidation inhibition (Mucha *et al*., 2021). *Moringa oleifera* extract, for example, has been shown to enhance cellular antioxidant capacity by modulating the Nrf2 pathway and reducing reactive oxygen species (ROS) in animal models (Ndlovu *et al*., 2023). Another study showed that polyphenols in pink garnet have a protective effect against oxidative damage in vascular endothelial cells (Said and Ibrahim, 2024). Furthermore, *Rosemary*

officinalis extract and its bioactive compounds, such as rosmarinic acid, have been shown to possess potent antioxidant capacity and hold promise for applications in pharmaceuticals and functional foods (Azlan *et al*., 2023). These findings reinforce the important role of natural products as safe and effective antioxidant-based therapeutic agents for the intervention of oxidative stress-related diseases (Chaudhary *et al*., 2023).

Measurement of antioxidant activity at a constant concentration (80 ppm), monitored periodically from 10 to 60 minutes, illustrates the time evolution of the kinetic reaction type between antioxidant compounds in plant extracts (both leaves and bark) and free radicals, using the DPPH assay. A gradual increase in the percentage of inhibition over time indicates that the radical-scavenging reaction is progressive, and the antioxidant compounds in the extract work effectively but are not immediately depleted at the start of the measurement. Chemically, the percentage of inhibition in the DPPH test is determined by the decrease in the intensity of the purple color of the DPPH solution at 517 nm, due to its interaction with the reducing compound (antioxidant), which causes a decrease in absorbance. The higher percentage of inhibition over time is indicative of a low (or slow) reaction rate between the phenolic or flavonoid compounds present in the extract and the DPPH radical, which is an expected characteristic of high-molecular-weight or complex-structured compounds like tannins and triterpenoids (Muhammad *et al*., 2020).

This increase may also be considered as evidence of the extract's increasing stability and antioxidant capacity. Antioxidant substances that do not react directly with a radical but have a long exposure time to the radical site due to their bulk physical properties (structure, steric hindrance) are known as retarded antioxidants. This is consistent with the findings of Wang *et al*. (2024), who reported that antioxidants derived from natural materials often exhibit kinetic profiles that depend on molecular structure and solvent medium. If the % inhibition continues to increase until the 60th minute and has not reached a plateau, this suggests that the antioxidant compound remains active in the system and has not fully reacted, potentially indicating the potential for gradual release of the active compound from the extract matrix (Lourenço *et al*., 2020). Comparison between leaves and bark can provide deeper insight. Bark contains high levels of lignins and tannins (Kassem *et al*., 2023) and can offer polyvalent antioxidants, which are slower-reactive but stable ones with long-term action. Leaves, however, generally have a higher content of susceptible flavonoids that might activate more rapidly (Table 3 and Table 4).

Table 3. Absorbance in units of time.

No	Extract (80 µM)	Absorbance (nm)						
		0 min	10 min	20 min	30 min	40 min	50 min	60 min
1	Leaf	0.214	0.104	0.093	0.09	0.09	0.09	0.09
2	Fruit peel	0.186	0.115	0.096	0.084	0.077	0.062	0.06

Table 4. Percentage of inhibition.

No	Extract (80 µM)	% Inhibition						
		0 min	10 min	20 min	30 min	40 min	50 min	60 min
1	Leaf	0	51.4	56.5	57.9	57.9	57.9	57.9
2	Fruit peel	0	38.2	48.4	54.8	58.6	66.7	67.7

Table 5. IC_{50} values.

Leaf Extract (µg/mL)	Leaf		Fruit Skin	
	Absorbance (nm)	IC_{50}	Absorbance (nm)	IC_{50}
0	0.802		0.302	
20	0.688		0.268	
40	0.527	44.31	0.243	144.57
60	0.406		0.213	
80	0.214		0.186	
100	0.113		0.168	

Testing the antioxidant activity of leaf and fruit peel extracts at various concentrations revealed significant differences in their free radical-scavenging abilities, as indicated by their respective IC_{50} values. The leaf extract showed an IC_{50} value of 44.31 µg/mL, while the fruit peel extract had an IC_{50} value of 144.57 µg/mL (Table 5). The lower IC_{50} value in the leaf extract indicates that the bioactive compounds contained therein have a higher antioxidant potential than the fruit peel, because they can inhibit 50% of free radical activity at lower

concentrations. The sharp decrease in absorbance in the leaf extract with increasing concentration supports this conclusion, indicating that its antioxidant compounds are more reactive (faster) and more efficient (Figure 2). This finding aligns with previous studies indicating that plant leaves are generally rich in flavonoids, phenolic acids, and simple polyphenols, which are strong free radical scavengers (Pintatum and Laphookhieo, 2022). In contrast, the activity of complex phenolic compounds in fruit skin, such as tannins and lignin, tends to be slower or requires higher concentrations to achieve equivalent inhibitory effects (Mohanty *et al*., 2023). Therefore, in terms of pharmacological and applicative potential, leaf extracts are more recommended as active ingredient candidates for the development of antioxidant-based products across the pharmaceutical, functional food, and cosmetic fields.

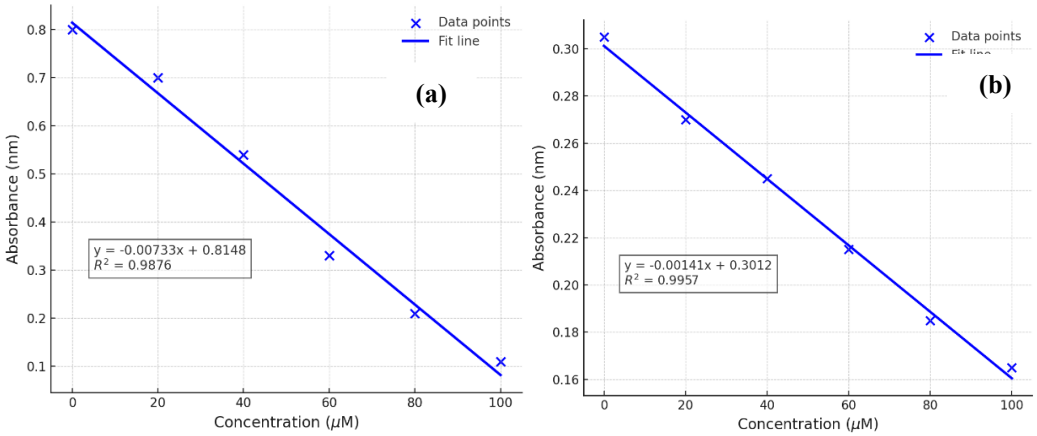

Figure 2. Linear regression of antioxidant activity (a) leaves and (b) fruit skin.

CONCLUSION

The secondary metabolite compounds contained in the ethanol extract and methanol extract of the Renggak fruit skin (Roxb.) are alkaloids, saponins, phenolics, flavonoids, terpenoids, and tannins. The positive results for all tests on the fruit peel are most likely due to the high phytochemical content found in this part. The results of phytochemical screening using methanol solvent were positive for the alkaloid, flavonoid, saponin, and tannin tests, while the terpenoid test was negative. The results of screening using ethanol solvent showed positive results for the alkaloid test and tannin test, while the flavonoid, saponin, and terpenoid tests showed negative results. Variations in the active compound content of the leaves and fruit peel affect the antioxidant activity of the Renggak plant. Renggak leaves have higher antioxidant activity with an IC_{50} value of 44.31 µM compared to the Renggak fruit peel with an IC_{50} value of 144.57 µM. Renggak leaves and fruit have potential as medicinal plants and can be used as materials for environmentally friendly, sustainable development.

CONFLICT OF INTEREST

There is no conflict of interest in this article.

AUTHOR CONTRIBUTION

RR: Conceptualization, Experimental Design, Data Analysis, and Manuscript Preparation; MM, LF, JI: Sample Preparation, Extraction Process, and Antioxidant Testing Experiments; SWAI: Methodology Validation; SPA: Experimental Support and Result Verification.

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DECLARATION OF GENERATIVE AI

The author declares that the generated AI (ChatGPT) was used solely to assist with language refinement, clarity, and style adjustments. After using this tool, the author reviewed and edited the content as needed and takes full responsibility for the content of the published article.

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