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THE EFFECT OF SOLVENT CONCENTRATION ON BIOACTIVE COMPOUNDS AND ANTIOXIDANT ACTIVITY OF MANGROVE (*Rhizophora* sp.) LEAF EXTRACT

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Abstract

Mangrove forests contain diverse bioactive compounds with considerable antioxidant activities. However, limited information is available on how variations in solvent concentration influence the extraction efficiency of phenolic and flavonoid compounds in Rhizophora species. This study aimed to evaluate the effects of different methanol concentrations (70, 80, and 90%) on the bioactive compound content and antioxidant activity of *R. mucronata* and *R. apiculata* leaves. Leaf samples were extracted by maceration, and the resulting crude extracts were analyzed for total phenolic content (TPC), total flavonoid content (TFC), and DPPH radical-scavenging activity (IC_{50}). A completely randomized factorial design was applied, followed by Tukey's test for mean separation. Increasing methanol concentration led to higher TPC and lower IC_{50} values, indicating enhanced antioxidant capacity, whereas TFC showed no significant differences across treatments. The 90% methanolic extract of *R. mucronata* exhibited the highest TPC (101.24±46.18 mg GAE/g) and lowest IC_{50} (74.33±4.93 ppm). A similar trend was observed for *R. apiculata*, with the 90% extract exhibiting the strongest antioxidant activity (72.55±1.75 ppm). These findings demonstrate that 90% methanol is the most effective solvent concentration for extracting antioxidant compounds from *Rhizophora* leaves and highlight the potential of these extracts as natural antioxidant sources for future food-and health-related applications.

Keywords: flavonoid, IC_{50}, phenol, *R. apiculata*, *R. mucronata*

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Pengaruh Konsentrasi Pelarut terhadap Senyawa Bioaktif dan Aktivitas Antioksidan Ekstrak Daun Mangrove (*Rhizophora* sp.)

Abstrak

Hutan mangrove mengandung beragam senyawa bioaktif dengan potensi antioksidan yang signifikan. Namun, informasi mengenai bagaimana variasi konsentrasi pelarut memengaruhi efisiensi ekstraksi senyawa fenolik dan flavonoid pada spesies *Rhizophora* masih terbatas. Penelitian ini bertujuan mengevaluasi pengaruh berbagai konsentrasi metanol (70, 80, dan 90%) terhadap kandungan senyawa bioaktif dan aktivitas antioksidan daun *R. mucronata* dan *R. apiculata*. Sampel daun diekstraksi menggunakan metode maserasi, dan ekstrak kasar yang dihasilkan dianalisis untuk menentukan total fenolik (TPC), total flavonoid (TFC), serta aktivitas penangkal radikal DPPH (nilai IC_{50}). Rancangan acak lengkap faktorial digunakan dalam penelitian ini, dilanjutkan dengan uji Tukey. Hasil penelitian ditemukan bahwa peningkatan konsentrasi metanol menghasilkan TPC yang lebih tinggi dan nilai IC_{50} yang lebih rendah, yang menunjukkan peningkatan kapasitas antioksidan, sementara TFC tidak menunjukkan perbedaan signifikan antarperlakuan. Ekstrak metanol 90% dari *R. mucronata* menunjukkan TPC tertinggi (101,24±46,18 mg GAE/g) dan nilai IC_{50} terendah (74,33±4,93 ppm). Pola serupa juga diamati pada *R. apiculata*, di mana ekstrak 90% menghasilkan aktivitas antioksidan terkuat (72,55±1,75 ppm). Temuan ini menunjukkan bahwa metanol 90% merupakan konsentrasi pelarut yang paling efektif untuk mengekstraksi senyawa antioksidan dari daun *Rhizophora*, serta menegaskan potensi ekstrak tersebut sebagai sumber antioksidan alami untuk aplikasi pangan dan kesehatan pada masa mendatang.

Kata kunci: fenol, flavonoid, IC_{50}, *R. apiculata*, *R. mucronata*

INTRODUCTION

Mangrove forests play an essential role in supporting sustainability and human livelihoods, as they are utilized as sources of food, timber, fuel, and medicine. In addition, mangroves serve as natural barriers against disasters such as tsunamis, tropical storms, and tidal surges, and help reduce the impacts of coastal erosion (Carugati *et al*., 2018). Mangroves are halophytic plant species distributed across approximately 112 countries. In Indonesia, mangrove forest ecosystems cover an estimated area of 3,364,076 hectares (Mitra *et al*., 2023). The mangrove species *Rhizophora mucronata* and *Rhizophora apiculata* are important components of coastal ecosystems, particularly in tropical regions (Arifanti *et al*., 2022).

Mangroves are recognized for their high ecological value and serve as sources of various bioactive compounds, including steroids, triterpenes, saponins, flavonoids, alkaloids, and tannins, which have potential therapeutic benefits (Syahidah & Subekti, 2019). Extracts from the Rhizophoraceae family show strong potential as antioxidant sources that promote human health by counteracting the effects of free radicals (Indriaty *et al*., 2023). The selection of solvents depends on the plant species, plant parts to be extracted, characteristics of the bioactive compounds, and availability of the solvents. Polar solvents, such as water, methanol, and ethanol, are generally used to extract polar compounds (Abubakar & Haque, 2020; Pandey *et al*., 2014).

The use of mature mangrove leaves is advantageous because of their availability and higher phytochemical content, such as flavonoids, alkaloids, tannins, triterpenoids, and phenols, compared to young and senescent leaves (Zamani *et al*., 2019). The antioxidant compounds in mature mangrove leaves are classified as very high, reaching 21.12 µg/mL (Sari *et al*., 2024). Another study by Mokhtar *et al*. (2022) reported that, compared with stems and roots, the ethanol extract of *R. mucronata* mangrove leaves exhibited the highest antioxidant activity, with an IC_{50} value of 9.84 µg/mL. According to

Podungge *et al.* (2015), the methanolic extract of *R. mucronata* fruit contains flavonoids, hydroquinones, triterpenoids, tannins, and saponins. Qualitative phytochemical screening revealed that the fruit extract of *R. mucronata* contained saponins and steroids, whereas the fruit extracts of *R. apiculata* and *A. marina* contained tannins, saponins, and steroids (Ramli *et al.*, 2020; Yuniarti *et al.*, 2020). Egra *et al.* (2023) demonstrate that the methanol extracts of *R. mucronata* and *R. apiculata* mangrove leaves show IC_{50} values of 37.58 µg/mL and 63.30 µg/mL, respectively, whereas extracts from the wood and bark present IC_{50} values exceeding 65 µg/mL.

Akasia *et al.* (2021) reported that, based on the phytochemical analysis of mangroves from Tuban (Bali) extracted using methanol, *R. apiculata* tests showed positive results for phenols, alkaloids, flavonoids, tannins, saponins, and steroids, but negative for terpenoids. In contrast, *R. mucronata* tests showed positive results for phenols, flavonoids, tannins, saponins, and terpenoids but negative for alkaloids and steroids. Usman *et al.* (2022) found that the dichloromethane and ethyl acetate extracts of *R. mucronata* leaves tested positive for secondary metabolites, including alkaloids, flavonoids, tannins, and phenolics. Another study by Kasitowati *et al.* (2017) showed that *R. mucronata* leaf extract contains alkaloids, flavonoids, and tannins. Quantitative phytochemical testing demonstrated that the methanol extract contained the highest levels of these compounds (1,895.47 ppm), followed by ethyl acetate (129.75 ppm) and n-hexane (108.79 ppm).

Issusilaningtyas *et al.* (2023) found that methanol extraction of mangrove leaves resulted in *R. mucronata* leaf extract containing alkaloids, flavonoids, tannins, saponins, steroids, and triterpenoids. Maulana and Sasmito (2021) reported that the ethanol extract of *R. apiculata* leaves exhibits the highest antioxidant activity, with an IC_{50} value of 49.45 ppm. Methanol is commonly used to isolate organic compounds because it can dissolve almost all classes of secondary metabolites (Akasia *et al.*, 2021). Variations in the composition of bioactive compounds indicate that each part of the mangrove plant provides distinct benefits for humans (Rozirwan *et al.*, 2022).

Plant extracts rich in antioxidants not only enhance food safety but are also influenced by various environmental factors that determine the activity of their bioactive compounds. One of the key factors that affects the composition and effectiveness of phenolic compounds is the habitat in which the plants grow (Mutia, 2022).

Despite the growing number of studies evaluating the antioxidant properties of *Rhizophora* species, most investigations have focused on comparing different solvent types rather than examining variations in solvent concentration within the same extraction system. In addition, comparative data between *R. mucronata* and *R. apiculata* extracted under standardized conditions are scarce. The limited understanding of species-solvent concentration interactions highlights the need for a more systematic approach to determine how methanol concentration influences the extraction of phenolic and flavonoid compounds, as well as antioxidant activity.

Optimizing the solvent concentration is crucial for improving the efficiency of phenolic recovery, which directly affects the antioxidant performance of plant extracts. Such optimization is particularly important for developing natural antioxidant sources that can be applied to food preservation and functional food ingredients. Therefore, understanding the relationship between solvent concentration and bioactive compound extraction from mangrove leaves will strengthen the scientific foundation for the utilization of *Rhizophora* species in food and health-related applications. Based on this background and considering the potential of *R. mucronata* and *R. apiculata*, this study aimed to identify the effects of varying solvent concentrations on the bioactive compound content of mangrove leaves and determine the optimal concentration of methanol that maximizes TPC and antioxidant activity (lowest IC_{50}) for each species.

MATERIALS AND METHOD

Preparation and Extraction

Leaf samples of *R. mucronata* and *R. apiculata* were collected from the BAPPL Serang mangrove conservation area in

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Indonesia. The samples consisted of fresh, intact, mature leaves attached to the branches. The leaves were transported by land in polybags to the laboratory. The collected mangrove leaves were then re-sorted to check for any damage, followed by thorough washing of each leaf under running water. The leaves were wiped dry, and the moisture content of the fresh leaves was measured. The samples were subsequently dried at room temperature (air-drying) (Arifin *et al*., 2023) for approximately 20 days or until a moisture content of <15% was achieved. The dried samples were ground using a grinder and extracted with methanol at concentrations of 70%, 80%, and 90% (v/v) to determine the optimal concentration. Extraction was performed by soaking the samples in the solvent at a ratio of 1:5 (w:v). Maceration was performed on an orbital shaker (SCILOGEX-SKo330-Pro) for 48 h at room temperature at a speed of 180 rpm. The macerate was filtered using Whatman No. 42 filter paper and evaporated with a rotary vacuum evaporator (BUCHI-Heating bath B-100) at 40°C under reduced pressure until most of the solvent was removed, obtaining a viscous crude extract rather than a completely dry extract. The crude extract was subsequently analyzed for antioxidant activity (DPPH), phytochemical composition, total phenolic content, and total flavonoid content (TFC). The extract yield was weighed and calculated using the following formula:

$$\text{Yield (\%)} = \frac{\text{extract weight (g)}}{\text{initial sample weight (dry) (g)}} 100$$

Phytochemical Screening

This analysis consisted of several components and followed the procedure described by Tiwari *et al*. (2011).

Alkaloid test: A total of 0.05 g of the extract was dissolved in 10 drops of 2N H_2SO_4, filtered, and divided into three test tubes. Each tube was then treated with Wagner's reagent (brown precipitate), Dragendorff's reagent (orange–red precipitate), or Mayer's reagent (yellowish-white precipitate) as indicators of a positive result. Flavonoid test: A total of 0.05 g of extract was mixed with 0.1 mL of Mg powder, 0.4 mL of amyl alcohol, and 4 mL of ethanol. A positive reaction was indicated by the formation of red, yellow, or orange color in the amyl alcohol layer.

Saponin test: A total of 0.05 g of extract was mixed with hot water, shaken for 1 min, and then 1 drop of 2N HCl was added. The formation of stable foam indicated a positive result.

Tannin test: A total of 0.05 g of extract was infused with hot water for 3 min, filtered, and then treated with 1% $FeCl_3$. A dark blue or greenish-black color indicated a positive reaction. Phenol test: A total of 0.05 g of extract was mixed with 2.5 mL of ethanol and two drops of 5% $FeCl_3$. The appearance of green, red, purple, or blue colorations indicated a positive result. Steroid and triterpenoid test: A total of 0.05 g of extract was mixed with 2 mL of chloroform, 10 drops of acetic anhydride, and 3 drops of H_2SO_4. Blue coloration indicates the presence of steroids, whereas reddish-brown or purple coloration indicates the presence of triterpenoids.

Antioxidant Activity (DPPH)

The antioxidant activity of the mangrove leaf extract was evaluated using the DPPH method (Boeing *et al*., 2014). The assay was conducted by mixing 300µL of the sample extract with 600 µL of 0.1 mM DPPH solution, followed by vortexing for 30 s. All mixtures were incubated for 30 min in the dark at room temperature, and the absorbance was measured at 517 nm using a spectrophotometer. The samples were tested at concentrations of 50, 100, 150, 200, 250, and 300 ppm, while ascorbic acid was used as a positive control at concentrations ranging from 1–6 ppm. The percentage inhibition and IC_{50} were calculated using the following formula:

$$\%\text{DPPH} = \frac{\text{blank absorbance-sample absorbance}}{\text{blank absorbance}} \times 100$$

$$IC_{50} = \frac{50 - \text{bx (intercept)}}{\text{a (slope)}}$$

Total Phenolic Content (TPC)

This analysis was conducted to determine the total phenolic content in the mangrove leaf extract, which contributed to its antioxidant properties, following the method described by Alara *et al*. (2020),

with modifications. In this assay, 50% Folin–Ciocalteu reagent was prepared by mixing 3 mL of Folin reagent with 3 mL of distilled water. A 5% Na_2CO_3 solution was prepared by dissolving 0.5 g of Na_2CO_3 in 10 mL distilled water. A gallic acid standard solution (1:1, w:v) was prepared at concentrations of 0, 20, 40, 60, 80, and 100 ppm.

A total of 0.125 mL of the standard solution was mixed with 0.125 mL of ethanol, 0.625 mL of distilled water, and 62.5 µL of 5% Folin reagent and incubated for 5 min. After incubation, 0.125 mL of the 5% Na_2CO_3 solution was added, followed by a 60-minute incubation in the dark. After incubation, 180–200 µL of the mixture was transferred into a microtube and measured using a spectrophotometer at 725 nm wavelength.

Total Flavonoid Content (TFC)

This analysis began with the preparation of a quercetin standard solution using ethanol and quercetin (1:1) at a concentration of 1000 ppm, followed by the preparation of a 2% $AlCl_3$ solution. A series of quercetin standard concentrations (0, 20, 40, 60, 80, and 100 ppm) was prepared. A total of 500 µL of the sample was mixed with 500 µL of 2% $AlCl_3$, vortexed, and incubated for 10 min. The absorbance was measured at 415 nm using a spectrophotometer. The same procedure was performed for the blank and all quercetin standard concentrations Alara *et al.* (2020).

Data Analysis

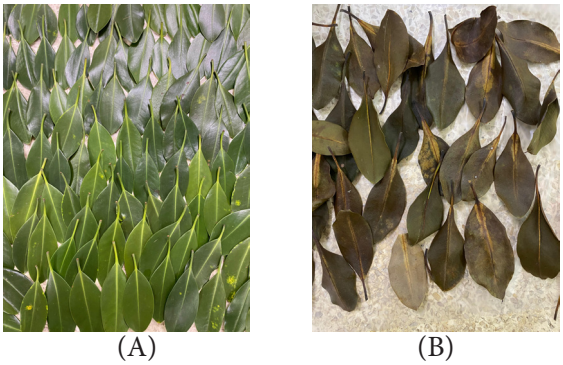
The data obtained in this study were analyzed using statistical testing tools. The analysis was performed using a factorial completely randomized design (CRD) ANOVA with two mangrove species and three solvent concentrations. Variables showing significant effects ($p<0.05$) were further evaluated using Tukey multiple range test.

RESULT AND DISCUSSION

Mangrove plants grow in tropical and subtropical regions. In this study, two types of mangrove leaves, R. mucronata and R. apiculata, were collected from the BAPPL Serang Conservation Area, Banten. Fresh and dried mangrove leaves are shown in Figure 1. The moisture content of fresh mangrove leaves ranged from 62% to 65%, whereas after 20 days drying period, the moisture content of the dried leaves decreased to 11–13%. According to Arifin et al. (2023), drying mangrove leaves at room temperature for 288 h resulted in a moisture content of 11.13%. The longer the drying duration, the lower the moisture content of the mangrove leaves.

The results showed that the extract yields of mangrove leaves varied according to the species and solvent concentrations used (Table 1). The yield of *R. mucronata* decreased with increasing methanol concentration, from 14.63% at 70% methanol to 10.49% at 90% methanol.

A different pattern was observed for *R. apiculata*, whose yields remained relatively stable, ranging from 14.25% to 15.12%, with no clear decrease at higher methanol concentrations. In a related study, Do *et al.* (2014) reported yields of 26.06% for 100% methanol extract and 32.92% for 75% methanol extract, respectively. The methanol, ethyl acetate, and n-hexane extract of *R. mucronata* macerated for 4×24 h yielded 35.30%, 13.60%, and 1.20%, respectively (Kasitowati *et al.*, 2017).



(A) (B)

Figure 1 Fresh (A) and dried (B) mangroves leaves

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Table 1 Mangrove leaves yield extract

Methanolic extraction	Concentration (%)	Yield (%)
Rhizophora mucronata extract (MME)	70	14.63
	80	13.75
	90	10.49
Rhizophora apiculata extract (MAE)	70	15.11
	80	14.89
	90	14.25

The efficiency of extraction is determined by the chemical characteristics of the phytochemical compounds, extraction method, sample particle size, type of solvent used, and possible presence of interfering compounds (Stalikas, 2007). The extraction yield is influenced by various factors, including solvent polarity, pH, temperature, extraction duration, and composition of the extracted material. Under the same temperature and time conditions, the type of solvent and sample composition are the most determining factors (Do *et al*., 2014). The decrease in the extract yield in the MME treatment is presumed to be associated with the polarity characteristics of the solvent. Methanol at a lower concentration (70%) contains a higher proportion of water, enabling the more efficient dissolution of both polar and semi-polar compounds. In contrast, 90% methanol, which is relatively less polar, can solubilize only a portion of the bioactive components, resulting in a lower extraction yield. This finding is consistent with that of Harborne and Williams (2000), who reported that extraction efficiency is strongly influenced by the compatibility between solvent polarity and the polarity of the target compounds. The differing responses observed between the two mangrove species further suggest variations in their leaf chemical composition, which may affect the solubility of compounds in the solvent (Do *et al*., 2014).

The findings of this study indicate that selecting an appropriate solvent concentration is essential for obtaining an optimal extraction yield without compromising the quality of the target compounds. Although the highest MME yield was achieved using 70% methanol, increasing the methanol concentration to 90% was more effective for extracting phenolic compounds and enhancing antioxidant activity.

Phytochemical Screening

Mangrove plants have been reported to contain important bioactive compounds with potential applications in various industries. The distribution of phytochemicals varies among different plant parts, including leaves, fruits, flowers, stems, and roots.

Phytochemical analysis (Table 2) revealed that the leaf extracts of *R. mucronata* and *R. apiculata* contained steroids, tannins, phenols, and flavonoids. These findings are consistent with previous studies reporting that mangrove leaf extracts generally contain various bioactive compounds, including alkaloids, phenols, flavonoids, tannins, saponins, steroids, and terpenoids. (Priyanto & Rimba, 2023; Akasia *et al*., 2021; Dahibhate *et al*., 2018; Eswaraiah *et al*., 2020; Krisnafi *et al*., 2024; Rahmawati *et al*., 2025; Syahidah & Subekti, 2019; Thao *et al*., 2022). Sachithanandam *et al*. (2022) reported that the mangrove leaf extract of *R. mucronata* contains a diverse range of bioactive compounds, including phytochemicals or secondary metabolites such as alkaloids, flavonoids, steroids, saponins, tannins, and phenolic compounds. Steroids are derived from various plants, animals, and microorganisms and are characterized by 17 carbon atoms arranged in four fused rings. Plant-derived steroids possess a wide range of medically, pharmaceutically, and agrochemically relevant properties (B. Gunaherath & Gunatilaka, 2014). The leaf extract of *R. mucronata* was found to contain high levels of steroids, whereas the levels of alkaloids, saponins, tannins, phenols, and flavonoids were relatively low. Steroids are known to enhance stamina (aphrodisiac effects) and exhibit anti-inflammatory activities (Egra *et al*., 2023; Rahmawati *et al*., 2025). Steroids also play important roles as

Table 2 Phytochemical compounds of methanolic mangrove leaves extract

Sample	Steroid	Triterpenoid	Saponin	Tannin	Fenol	Flavonoid	Alkaloid		
							Mayer	Dragendorff	Wagner
MME 70%	+	-	-	+	+	+	-	-	-
MME 80%	+	-	-	+	+	+	-	-	-
MME 90%	+	-	-	+	+	+	-	-	-
MAE 70%	+	-	-	+	+	+	-	-	-
MAE 80%	+	-	-	+	+	+	-	-	-
MAE 90%	+	-	-	+	+	+	-	-	-

(+): detected; (-): not detected; MME: Methanolic *R. mucronata* extract; MAE: Methanolic *R. apiculata* extract

antioxidants, anticancer, anticholesterol, and antiviral agents (Lein *et al*., 2025).

Phenols are characterized by an aromatic ring structure attached to one or more hydroxyl groups. Phenolic compounds are generally found in various plant parts, such as leaves, fruits, and bark (Sachithanandam *et al*., 2022). Flavonoids consist of two benzene rings (A and B) connected through a heterocyclic pyran ring (C). Flavonoids are an important group of polyphenolic compounds that possess a benzo-γ-pyrone structure and are primarily found in plants. Flavonoids exhibit strong antioxidant and fungicidal activities and are used as therapeutic agents for menstrual and skin disorders, as well as for wounds, including leprosy and dermatitis (Liu *et al*., 2021; Parthiban *et al*., 2022). Alkaloids are a group of secondary metabolites and the most diverse nitrogen-based bioactive compounds, typically characterized by heterocyclic rings. Alkaloids are commonly found in flowering plants (Maldoni, 1991). Mangrove plants that contain alkaloids are known to be toxic or medicinal (Parthiban *et al*., 2022).

The results showed that the extract did not contain alkaloid compounds. This outcome is presumed to be influenced by several factors. The polarity of the solvent significantly affects the number of chemical components present, the efficiency of the extraction process, and the resulting biological activity (Ali *et al*., 2021; Truong *et al*., 2019). This finding is consistent with the study of Akasia *et al*. (2021), which reported that phytochemical tests of mangroves from Tuban (Bali) extracted using methanol revealed that *R. mucronata* was positive for phenols, flavonoids, tannins, saponins, and terpenoids, but negative for alkaloids and steroids. Similarly, Hardiningtyas *et al*. (2024) reported that alkaloid compounds were not detected in the leaf, flower, and fruit extracts of *R. mucronata*, as indicated by the absence of white precipitates following the addition of Mayer's reagent.

The compounds detected in the phytochemical analysis were associated with the antioxidant capacity of the sample. Several previous studies have reported that high levels of phenolic compounds, alkaloids, and terpenoids are linked to strong antioxidant activity. In addition, phytochemical compounds such as alkaloids, tannins, saponins, and terpenoids are known to act as free radical scavengers and chain-breaking agents in oxidation reactions (Bulan *et al*., 2022; Miranti *et al*., 2018; Nguyen *et al*., 2021).

The solvent concentration range used in this study (70–90%) was not sufficiently wide to alter the solubility threshold required for these metabolites to yield different qualitative outcomes. In other words, although the concentrations of phenolic, flavonoid, tannin, or other compounds may vary quantitatively, as reflected in the TPC, TFC, and IC_{50} values, their levels remain above the detection limit of the qualitative assays across all treatments, resulting in similar positive reactions. This phenomenon is consistent with previous reports stating that qualitative phytochemical tests are not sensitive enough to distinguish moderate variations in compound concentrations within polar solvent systems (Tiwari *et al*., 2011).

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Antioxidant Activity

DPPH is a stable free radical compound widely used to evaluate the free radical-scavenging potential of antioxidants (Jha *et al*., 2022). The antioxidant activity of the extract was indicated by its IC_{50} value. The IC_{50} value of DPPH scavenging activity is inversely proportional to the percentage of DPPH scavenging activity, indicating that the highest antioxidant activity is represented by the lowest IC_{50} value (Priyanto & Rimba, 2023). Analysis of variance (ANOVA) showed that mangrove leaf type, solvent concentration, and their interaction ($p<0.05$) had a highly significant effect on the IC_{50} value of the extract (Table 3).

This indicates that the effectiveness of the extract in scavenging free radicals is influenced by variations in leaf type and the solvent concentration used. Tukey's post-hoc test showed that the IC_{50} values differed significantly among the treatments. The 90% concentration yielded the lowest IC_{50} value and was categorized as having strong antioxidant activity, which was significantly different from the 80% and 70% concentrations.

The IC_{50} values showed significant variation among the treatments, which was attributed to the differences in the levels of bioactive compounds present in the samples. In the *R. mucronata* extract, the IC_{50} value decreased with increasing solvent concentration, from 141.58 ppm (70%) to 89.00 ppm (80%) and 74.33 ppm (90%). A similar trend was observed for the *R. apiculata* extract, with the IC_{50} values decreasing from 100.79 ppm (70%) to 88.49 ppm (80%) and 72.55 ppm (90%). These findings are consistent with previous studies reporting that methanolic extracts of *R. mucronata* have an IC_{50} value of 117.498 µg/mL (Kasitowati *et al*., 2017), methanolic extracts of *R. apiculata* have an IC_{50} value of 92.75 µg/mL and *R. mucronata* 93.21 µg/mL (Vittaya *et al*., 2022), and methanolic extracts of *R. apiculata* have an IC_{50} value of 68.00 µg/mL (Chelliah *et al*., 2023). According to Ramli *et al*. (2025), increasing the methanol concentration enhanced the antioxidant activity of the extract, with the 90% methanol extract showing the lowest IC_{50} value (62.42±3.23 ppm), whereas the 80% and 70% methanol extracts showed lower antioxidant capacities. The study also demonstrated that the highest total phenolic content (TPC) was obtained from the 80% methanol extract, while the 90% methanol extract contained the highest total flavonoid content (TFC). The decrease in the IC_{50} values indicates that the use of solvents at higher concentrations enhances the antioxidant activity of the extract. Among the ten different methanolic extracts of mangrove plants, *R. mucronata* was shown to possess efficient phytochemicals that exhibit strong antimicrobial and antioxidant activities. The methanolic extract of *R. mucronata* has an IC_{50} value of 70.93 µg/mL, whereas *R. apiculata* has an IC_{50} value of 127.79 µg/mL (Sachithanandam *et al*., 2022).

Table 3 IC_{50}, total phenol, and total flavonoid content of *R. mucronata* and *R. apiculata*

Sample	IC_{50} (ppm)	TPC (mgGAE/g)	TFC (mgQE/g)
MME 70%	141.58±1.82^{aa}	79.71±21.80^{aa}	5.41±5.93^{aa}
MME 80%	89.00±1.13^{ab}	94.32±33.73^{aa}	5.52±5.87^{aa}
MME 90%	74.33±4.93^{ac}	101.24±46.18^{aa}	8.47±4.07^{aa}
MAE 70%	100.79±3.55^{aa}	32.96±4.46^{aa}	3.31±3.42^{aa}
MAE 80%	88.49±5.17^{ab}	31.21±7.97^{aa}	5.42±4.21^{aa}
MAE 90%	72.55±1.75^{ac}	36.04±6.85^{aa}	3.49±4.05^{aa}
Ascorbic acid	0.79±0.00	-	-

IC_{50}: <50 (very strong); 50-100 ppm (strong); 100-150 (moderate); 150-200 (weak) (Molyneux, 2004);

MME: Methanolic *R. mucronata* extract; MAE: Methanolic *R. apiculata* extract;

Distinct letter superscripts denote statistically significant differences according to Tukey's HSD test ($p<0.05$).

The results show a relationship between the total phenolic content and the IC_{50} values of the mangrove leaf extracts (Table 3). This relationship indicates that a higher phenolic content results in a lower IC_{50} value, reflecting stronger antioxidant activity. The antioxidant activity in the samples is presumed to originate from compounds capable of donating protons to neutralize the free radicals. Phenolic and flavonoid compounds are known to possess these capabilities (Fidrianny *et al*., 2014, 2015).

The total phenolic content is closely related to the antioxidant activity of black mangrove leaves (Arifin *et al*., 2023). Phenolic compounds are known to donate hydrogen atoms to neutralize free radicals, thereby contributing to the reduction of IC_{50} values as their concentration increases in the extract. These findings are consistent with previous studies reporting strong negative correlations between total phenolic, flavonoid, and carotenoid contents and IC_{50} values in DPPH radical scavenging assays. This indicates that the higher the levels of total phenols, flavonoids, and carotenoids, the lower the IC_{50} value, reflecting stronger DPPH radical-scavenging activity (Heim *et al*., 2002, Fitriansyah *et al*., 2018). The higher the total phenolic content, the stronger the antioxidant activity (Sobuj *et al*., 2021).

Total Phenolic Content

The phenolic content of the material was expressed in gallic acid equivalents (GAE). Analysis of variance (ANOVA) showed that mangrove leaf type significantly affected total phenolic content, whereas solvent concentration did not. The interaction between leaf type and solvent concentration also did not have a significant influence. Tukey's HSD post-hoc test indicated that variations in solvent concentration (70%, 80%, and 90%) did not result in significant differences in phenolic content, as all concentration groups fell within the same homogeneous subset. The analysis further showed that the total phenolic content in the leaf extracts of *R. mucronata* and *R. apiculata* increased with the use of higher methanol concentrations during the extraction process. Although minor fluctuations in the mean values were observed across concentrations, no consistent directional trend was evident. The absence of significance is likely attributable to the relatively large within-group variability, as indicated by substantial standard deviations, which reduced the ability to detect differences among treatments. The overlapping variability suggests that the solvent concentration did not meaningfully influence the total phenolic extraction under the tested conditions. The total phenolic content of the *R. mucronata* extract at 90% solvent concentration was 101.24±46.18 mg GAE/g, and no significant differences were observed among the tested solvent concentrations. This increase indicates that methanol 90% is more effective in dissolving phenolic compounds. Phenols are key bioactive compounds that contribute to antioxidant capacity; therefore, optimal extraction enhances the levels of these components (Banerjee *et al*., 2008; Mokhtar *et al*., 2022).

The study conducted by Hardiningtyas *et al*. (2024) showed that the leaves, flowers, and fruits of *R. mucronata* are rich in phenolic compounds. A positive result in the phenol test was indicated by a color change to blue. According to Haryati *et al*. (2015), this color change occurs due to a reaction between $FeCl_3$ and the –OH groups of phenolic compounds. Another study by Rahmawati *et al*. (2025) reported that the total phenolic content in the ethanol extract of *R. mucronata* leaves from Surabaya waters was 6.70%. In addition, Kaur *et al*. (2019) showed that the total phenolic content in the methanolic extract of *R. mucronata* leaves reached 21.25 mg/g. Black mangrove leaves contain phenolic compounds, including flavonoids. Nearly all mangrove species are capable of producing flavonoids because they inhabit relatively extreme environments. These compounds play an essential role in protecting mangroves from various environmental stresses (Arifin *et al*., 2023; Mierziak *et al*., 2014). The use of solvents with different polarities results in varying total phenolic contents (Hardiningtyas *et al*., 2024).

Total Flavonoid Content

Flavonoids are secondary phenolic compounds widely found in plants, known for their antioxidant properties and strong chelating activity (Heim *et al*., 2002).

Analysis of variance (ANOVA) showed that neither mangrove leaf type nor solvent concentration had a significant effect on total flavonoid content. Their interaction also had no significant effect. The average flavonoid content was 5.00±0.85 mg QE/g. This indicates that within the solvent concentration range used (70–90%), the ability of methanol to extract flavonoids remains relatively stable, and variations in leaf type do not result in meaningful differences in total flavonoid levels. Previous studies have reported that the amount of flavonoids extracted is strongly influenced by the extraction method applied and the conditions or characteristics of the material during the extraction process (Zhu *et al*., 2021). In fresh mangrove leaves, moisture content may affect the stability of flavonoid compounds because water contributes to the vulnerability of flavonoids, which are sensitive to temperature and humidity (Prayitno *et al*., 2025).

In plant tissues, flavonoids generally occur as glycosylated derivatives with diverse functions. Their distinctive colors, as seen in flavones, flavonols, and anthocyanidins, may serve as visual signals for pollinators. Compounds with astringent tastes, such as catechins and other flavanols, are presumed to function as defense mechanisms against herbivorous insects (Mazza & Miniati, 2018). In addition, flavonoids may act as catalysts in the light phase of photosynthesis or as regulators of ion channels involved in phosphorylation (Harborne, 2017), protecting plant cells from stress by scavenging reactive oxygen species generated during photosynthesis (Alara *et al*., 2020). Phenolic compounds, such as flavonoids, function as antioxidants because they contain hydroxyl groups attached to aromatic carbon rings, enabling them to scavenge free radicals (Phillipson, 2001).

CONCLUSION

This study demonstrated that increasing methanol concentration enhanced the extraction of phenolic compounds and improved the antioxidant activity of *R. mucronata* and *R. apiculata* leaf extracts. Extraction using 90% methanol resulted in a relatively high total phenolic content and strong DPPH free-radical scavenging activity, indicating its effectiveness as a solvent for recovering antioxidant compounds from mangrove leaves. These findings suggest that extracts obtained using 90% methanol have the potential for further development as natural antioxidant ingredients applicable in food preservation, functional foods, and health-related products.

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REFERENCES

Abubakar, A. R., & Haque, M. (2020). Preparation of medicinal plants: Basic extraction and fractionation procedures for experimental purposes. In *Journal of Pharmacy and Bioallied Sciences* (Vol. 12, Issue 1). https://doi.org/10.4103/jpbs.JPBS_175_19

Adi Prayitno, S., Utami, D. R., & Putri, S. N. A. (2025). Phytochemicals analysis and antioxidant (IC_{50}) value of dried and fresh mangrove (*Rhizopora racemosa*) leaves extract for herbal drink base. *Journal of Tropical Food and Agroindustrial Technology*, *6*(01), 1–7. https://doi.org/10.21070/jtfat.v6i01.1643

Adi Priyanto, R., & Rimba, F. (2023). Antioxidant activity and bioactive compound in mangrove fruit (*Rhizophora mucronata Lamk.*). *Jurnal Ilmiah PLATAX*, *11*(2). https://doi.org/10.35800/jip.v11i2.48758

Akasia, A. I., Nurweda Putra, I. D. N., & Giri Putra, I. N. (2021). Skrining fitokimia ekstrak daun mangrove *Rhizophora mucronata* dan *Rhizophora apiculata* yang dikoleksi dari kawasan mangrove Desa Tuban, Bali. *Journal of Marine*

Research and Technology, *4*(1). https://doi.org/10.24843/jmrt.2021.v04.i01.p03

Alara, O. R., Abdurahman, N. H., & Olalere, O. A. (2020). Ethanolic extraction of flavonoids, phenolics and antioxidants from *Vernonia amygdalina* leaf using two-level factorial design. *Journal of King Saud University - Science*, *32*(1). https://doi.org/10.1016/j.jksus.2017.08.001

Annisa Mutia. (2022, November 7). Kondisi Mangrove Indonesia *2021*. Databoks. Katadata.Co.Id.

Arifanti, V. B., Sidik, F., Mulyanto, B., Susilowati, A., Wahyuni, T., Subarno, Yulianti, Yuniarti, N., Aminah, A., Suita, E., Karlina, E., Suharti, S., Pratiwi, Turjaman, M., Hidayat, A., Rachmat, H. H., Imanuddin, R., Yeny, I., Darwiati, W., & Novita, N. (2022). Challenges and strategies for sustainable mangrove management in Indonesia: A Review. In *Forests* (Vol. 13, Issue 5). https://doi.org/10.3390/f13050695

Arifin, M. H., Jacoeb, A. M., Abdullah, A., & Riyadi, P. H. (2023). Drying method comparison of black mangrove leaves (*Rhizophora mucronata*) for an antioxidant activity assay. *Food Research*, *7*, 67–75. https://doi.org/10.26656/fr.2017.7(S3).10

B. Gunaherath, G. M. K., & Gunatilaka, A. A. L. (2014). Plant Steroids: Occurrence, biological significance and their analysis. In *Encyclopedia of Analytical Chemistry*. https://doi.org/10.1002/9780470027318.a9931

Banerjee, D., Chakrabarti, S., Hazra, A. K., Banerjee, S., Ray, J., & Mukherjee, B. (2008). Antioxidant activity and total phenolics of some mangroves in Sundarbans. *African Journal of Biotechnology*, *7*(6).

Bulan, D. E., Nurfadilah, N., Syahrir, M. R., Mismawati, A., Torambung, A. K., & Rachmawati, M. (2022). Phytochemical composition and antioxidant activity of leaf extracts from three *rhizophora* species from Bontang Waters, Indonesia. *Tropical Journal of Natural Product Research*, *6*(8). https://doi.org/10.26538/tjnpr/v6i8.2

Carugati, L., Gatto, B., Rastelli, E., Lo Martire, M., Coral, C., Greco, S., & Danovaro, R. (2018). Impact of mangrove forests degradation on biodiversity and ecosystem functioning. *Scientific Reports*, *8*(1). https://doi.org/10.1038/s41598-018-31683-0

Chelliah, C. K., Murugan, M., Rajivgandhi, G., Gnanasekaran, C., Govindan, R., Maruthupandy, M., Quero, F., Arulraj, A., Viswanathan, M. R., Alharbi, N. S., & Alshammary, N. H. (2023). Phytochemical derivatives and secondary metabolites rich *Rhizophora mucronata* as an active anti-oxidant and anti-bacterial agent against multi drug resistant bacteria. *Journal of King Saud University - Science*, *35*(8). https://doi.org/10.1016/j.jksus.2023.102912

Costa, D. Da. (2017). Pasang Surut Pantai Kampus Serang (Sekolah Tinggi Perikanan).

Dahibhate, N. L., Saddhe, A. A., & Kumar, K. (2018). Mangrove plants as a source of bioactive compounds: A Review. *The Natural Products Journal*, *9*(2), 86–97. https://doi.org/10.2174/2210315508666180910125328

Do, Q. D., Angkawijaya, A. E., Tran-Nguyen, P. L., Huynh, L. H., Soetaredjo, F. E., Ismadji, S., & Ju, Y. H. (2014). Effect of extraction solvent on total phenol content, total flavonoid content, and antioxidant activity of *Limnophila aromatica*. *Journal of Food and Drug Analysis*, *22*(3), 296–302. https://doi.org/10.1016/J.JFDA.2013.11.001

Dyah Kasitowati, R., Yamindago, A., & Safitri, M. (2017). Potensi antioksidan dan skrining fitokimia ekstrak daun mangrove *Rhizophora mucronata*, Pilang Probolinggo. *Journal of Fisheries and Marine Science*, *1*(1), 72–77. http://jfmr.ub.ac.id

Egra, S., Kuspradini, H., Kusuma, I. W., Batubara, I., Imra, Nurjannah, Wahyuni, E., Yamauchi, K., & Mitsunaga, T. (2023). Potential of prospective medicinal plants of *Rhizophoraceae* from North Kalimantan, Indonesia. *Biodiversitas*, *24*(3). https://doi.org/10.13057/biodiv/d240303

Eswaraiah, G., Peele, K. A., Krupanidhi, S.,

Kumar, R. B., & Venkateswarulu, T. C. (2020). Studies on phytochemical, antioxidant, antimicrobial analysis and separation of bioactive leads of leaf extract from the selected mangroves. *Journal of King Saud University - Science*, *32*(1). https://doi.org/10.1016/j.jksus.2019.03.002

Fidrianny, I., Aristya, T., & Hartati, R. (2015). Antioxidant capacities of various leaves extracts from three species of legumes and correlation with total flavonoid, phenolic, carotenoid content. *International Journal of Pharmacognosy and Phytochemical Research*, *7*(3).

Fidrianny, I., Darmawati, A., & Sukrasno. (2014). Antioxidant capacities from different polarities extracts of cucurbitaceae leaves using FRAP, DPPH assays and correlation with phenolic, flavonoid, carotenoid content. *International Journal of Pharmacy and Pharmaceutical Sciences*, *6*(SUPPL. 2).

Fitriansyah, S. N., Aulifa, D. L., Febriani, Y., & Sapitri, E. (2018). Correlation of total phenolic, flavonoid and carotenoid content of *phyllanthus emblica* extract from bandung with DPPH scavenging activities. *Pharmacognosy Journal*, *10*(3). https://doi.org/10.5530/pj.2018.3.73

Harborne, J. B. (2017). The flavonoids: Advances in research since 1986. In *The Flavonoids: Advances in Research since 1986*. https://doi.org/10.1201/9780203736692

Harborne, J. B., & Williams, C. A. (2000). Advances in flavonoid research since 1992. In *Phytochemistry* (Vol. 55, Issue 6). https://doi.org/10.1016/S0031-9422(00)00235-1

Hardiningtyas, S. D., Purwaningsih, S., Sastra, M., Dan, A., & Sinulingga, F. (2024). Potential of mangrove *Rhizophora mucronata* as tisane rich in phenols and antioxidants. *Jurnal Akuatika Indonesia*, *9*(2), 94–110.

Heim, K. E., Tagliaferro, A. R., & Bobilya, D. J. (2002). Flavonoid antioxidants: Chemistry, metabolism and structure-activity relationships. In *Journal of Nutritional Biochemistry* (Vol. 13, Issue 10). https://doi.org/10.1016/S0955-2863(02)00208-5

Idris Ali, A., Paul, V., Chattree, A., Prasad, R., Paul, A., & Amiteye, D. (2021). Evaluation of the use of different solvents for phytochemical constituents and antioxidants activity of the leaves of *murraya koenigii* (linn.) *Spreng*. (rutaceae). *Plant archives*, *21*(no 1). https://doi.org/10.51470/plantarchives.2021.v21.no1.137

Indriaty, Djufri, Ginting, B., & Hasballah, K. (2023). Phytochemical screening, phenolic and flavonoid content, and antioxidant activity of *Rhizophoraceae* methanol extract from Langsa, Aceh, Indonesia. *Biodiversitas*, *24*(5). https://doi.org/10.13057/biodiv/d240541

Issusilaningtyas, E., Aji, A. P., & Fauziyah, A. R. (2023). Pengaruh metode ekstraksi terhadap kandungan metabolit sekunder ekstrak daun bakau hitam (*Rhizopora mucronata*). *Jurnal Ilmiah Nusantara*, *1*(April).

Jha, N., Esakkiraj, P., Annamalai, A., Lakra, A. K., Naik, S., & Arul, V. (2022). Synthesis, optimization, and physicochemical characterization of selenium nanoparticles from polysaccharide of mangrove *Rhizophora mucronata* with potential bioactivities. *Journal of Trace Elements and Minerals*, *2*. https://doi.org/10.1016/j.jtemin.2022.100019

Krisnafi, Y., Sumartini, S., & Mardiah, R. S. (2024). Aplikasi serbuk daun mangrove (*Rhizopora* sp.) sebagai pengawet alami tali rami pada alat tangkap jaring ikan. *Jurnal Pengolahan Hasil Perikanan Indonesia*, *27*(1), 62–74. https://doi.org/10.17844/jphpi.v27i1.47046

Lein, M. A., Desniar, D., & Hardiningtyas, S. D. (2025). Fermentasi daun mangrove *Rhizophora mucronata* sebagai teh herbal. *Jurnal Pengolahan Hasil Perikanan Indonesia*, *28*(2), 170–186. https://doi.org/10.17844/jphpi.v28i2.61684

Liu, W., Feng, Y., Yu, S., Fan, Z., Li, X., Li, J., & Yin, H. (2021). The flavonoid biosynthesis network in plants. In *International Journal of Molecular Sciences* (Vol. 22, Issue 23). https://doi.org/10.3390/ijms222312824

Maldoni, B. (1991). Alkaloids: Isolation and purification. *Journal of Chemical*

Education, *68*(8). https://doi.org/10.1021/ed068p700

Maulana, D. M., & Sasmito, B. B. (2021). The dose effect of mangrove leaf extract (*Rhizophora apiculata*) on anticancer activity in hela cells. *Journal of Stem Cell Research and Tissue Engineering*, *5*(1). https://doi.org/10.20473/jscrte.v5i1.29380

Mazza, G., & Miniati, E. (2018). Anthocyanins in fruits, vegetables, and grains. In *Anthocyanins in Fruits, Vegetables, and Grains*. https://doi.org/10.1201/9781351069700

Mierziak, J., Kostyn, K., & Kulma, A. (2014). Flavonoids as important molecules of plant interactions with the environment. In *Molecules* (Vol. 19, Issue 10). https://doi.org/10.3390/molecules191016240

Miranti, D. I., Ichiura, H., & Ohtani, Y. (2018). The bioactive compounds and antioxidant activity of food products of *Rhizophora stylosa* fruit (coffee and tea mangrove). *International Journal of Forestry Research*, *2018*. https://doi.org/10.1155/2018/2315329

Mitra, S., Naskar, N., Lahiri, S., & Chaudhuri, P. (2023). A study on phytochemical profiling of Avicennia marina mangrove leaves collected from Indian Sundarbans. *Sustainable Chemistry for the Environment*, *4*, 100041. https://doi.org/10.1016/j.scenv.2023.100041

Mohd Mokhtar, R. A., Misrah, M. F., Kansil, T., Amin, Z., Yusof, N. A., & Budiman, C. (2022). Evaluation of antioxidant activity and total phenolics of selected mangrove plants in Sabah. *Borneo International Journal of Biotechnology (BIJB)*, *2*. https://doi.org/10.51200/bijb.v2i.3330

Molyneux, P. (2004). The use of the stable free radical diphenylpicryl-hydrazyl (dpph) for estimating antioxidant activity. *Songklanakarin Journal of Science and Technology*, *26*(December 2003). https://doi.org/10.1287/isre.6.2.144

Nguyen, K. Q., Scarlett, C. J., & Vuong, Q. V. (2021). Assessment and comparison of phytochemicals and antioxidant properties from various parts of the Australian maroon bush (*Scaevola spinescens*). *Heliyon*, *7*(4). https://doi.org/10.1016/j.heliyon.2021.e06810

Oktavia, S., Halim Ismail, M., Ulfa Sa, F., Lavanya Wati, K., Sephiana, L., Oktaviani, L., Muzahidi, M., Guruh Gumelar, P., Nurasiyah, S., Munandar, E., & Ali Khalifa, M. (2025). Mangrove ecosystem dynamics in Kasemen district, Serang city: a spatial and ecological approach. *Jurnal Maiyah*, *4*(1), 40–48. https://doi.org/10.20884/1.maiyah.2025.4.1.14306

Pandey, A., Tripathi, S., & Pandey, C. A. (2014). Concept of standardization, extraction and pre phytochemical screening strategies for herbal drug. *Journal of Pharmacognosy and Phytochemistry JPP*, *115*(25).

Parthiban, A., Sivasankar, R., Sachithanandam, V., Khan, S. A., Jayshree, A., Murugan, K., & Sridhar, R. (2022). An integrative review on bioactive compounds from Indian mangroves for future drug discovery. *South African Journal of Botany*, *149*. https://doi.org/10.1016/j.sajb.2021.10.004

Phillipson, J. D. (2001). Phytochemistry and medicinal plants. *Phytochemistry*, *56*(3). https://doi.org/10.1016/S0031-9422(00)00456-8

Podungge, F., Purwaningsih, S., & Nurhayati, T. (2015). Karakteristik buah bakau hitam sebagai sediaan ekstrak sumber antioksidan. *Jurnal Pengolahan Hasil Perikanan Indonesia*, 18(2).

Rahmawati, R., Sanjaya, Y. A., Pratiwi, Y. S., Hendrawan, E. P. N., Salsabila, Z. N., & Amalia, T. (2025). Antioxidant activity and phytochemical screening of *Rhizopora mucronata* mangrove leaf extract from mangrove botanical garden, Surabaya City, Esat Java. *Jurnal Perikanan Universitas Gadjah Mada*, *27*(1), 1. https://doi.org/10.22146/jfs.90770

Ramli, H. K., Nurhayati, T., Abdullah, A., Yuniarti, T., & Primahana, G. (2025). Bioactive Compounds of Methanolic Leaf Extracts of Rhizophora mucronata from Pantai Indah Kapuk. Food Security Symposium.

Ramli, H. K., Yuniarti, T., Lita, N. P. S. N., & Sipahutar, Y. H. (2020). Uji fitokimia

secara kualitatif pada buah dan ekstrak air buah mangrove. *Jurnal Penyuluhan Perikanan Dan Kelautan*, 14(1). https://doi.org/10.33378/jppik.v14i1.198

Rozirwan, Nugroho, R. Y., Hendri, M., Fauziyah, Putri, W. A. E., & Agussalim, A. (2022). Phytochemical profile and toxicity of extracts from the leaf of *Avicennia marina* (Forssk.) Vierh. collected in mangrove areas affected by port activities. *South African Journal of Botany*, *150*. https://doi.org/10.1016/j.sajb.2022.08.037

Sachithanandam, V., Lalitha, P., Parthiban, A., Muthukumaran, J., Jain, M., Misra, R., Mageswaran, T., Sridhar, R., Purvaja, R., & Ramesh, R. (2022). A comprehensive in silico and in vitro studies on quinizarin: a promising phytochemical derived from *Rhizophora mucronata* Lam. *Journal of Biomolecular Structure and Dynamics*, *40*(16). https://doi.org/10.1080/07391102.2021.1894983

Sari, N. I., Sidauruk, S. W., Dewita, Ananda, N. T., & Leksono, T. (2024). Valorization of mangrove leaves (*Sonneratia alba*) based on leaf age as a functional salt preparation. *Kuwait Journal of Science*, *51*(1). https://doi.org/10.1016/j.kjs.2023.06.003

Sobuj, M. K. A., Islam, M. A., Haque, M. A., Islam, M. M., Alam, M. J., & Rafiquzzaman, S. M. (2021). Evaluation of bioactive chemical composition, phenolic, and antioxidant profiling of different crude extracts of *Sargassum coriifolium* and *Hypnea pannosa* seaweeds. *Journal of Food Measurement and Characterization*, *15*(2). https://doi.org/10.1007/s11694-020-00758-w

Stalikas, C. D. (2007). Extraction, separation, and detection methods for phenolic acids and flavonoids. In *Journal of Separation Science* (Vol. 30, Issue 18). https://doi.org/10.1002/jssc.200700261

Syahidah, & Subekti, N. (2019). Phytochemical analysis of mangrove leaves (*Rhizophora* sp.). *IOP Conference Series: Materials Science and Engineering*, *593*(1). https://doi.org/10.1088/1757-899X/593/1/012007

Thao, N. P., Linh, K. T. P., Quan, N. H., Trung, V. T., Binh, P. T., Cuong, N. T., Nam, N. H., & Van Thanh, N. (2022). Cytotoxic metabolites from the leaves of the mangrove *Rhizophora apiculata*. *Phytochemistry Letters*, *47*. https://doi.org/10.1016/j.phytol.2021.10.014

Tiwari, P., Kumar, B., Mandeep, K., Kaur, G., & Kaur, H. (2011). Phytochemical screening and extraction: A Review. *Internationale Pharmaceutica Sciencia*, *1*(1).

Truong, D. H., Nguyen, D. H., Ta, N. T. A., Bui, A. V., Do, T. H., & Nguyen, H. C. (2019). Evaluation of the use of different solvents for phytochemical constituents, antioxidants, and in vitro anti-inflammatory activities of *severinia buxifolia*. *Journal of Food Quality*, *2019*. https://doi.org/10.1155/2019/8178294

Usman, U., Fildzania, D., & Fauzi, I. (2022). Uji aktivitas antioksidan dan antidiabetes ekstrak daun mangrove *Rhizopora mucronata*. *Jurnal Sains Dan Kesehatan*, *4*(1). https://doi.org/10.25026/jsk.v4i1.724

Vittaya, L., Charoendat, U., Janyong, S., Ui-eng, J., & Leesakul, N. (2022). Comparative analyses of saponin, phenolic, and flavonoid contents in various parts of *Rhizophora mucronata* and *Rhizophora apiculata* and their growth inhibition of aquatic pathogenic bacteria. *Journal of Applied Pharmaceutical Science*, *12*(11). https://doi.org/10.7324/JAPS.2022.121113

Yuniarti, T., Sipahutar, Y., Ramli, H. K., & Lita, N. P. S. N. (2020). Application of mangrove extract for the melanosis inhibition in whiteleg shrimp (*Litopenaeus vannamei*). *Jurnal Pengolahan Hasil Perikanan Indonesia*, 23(1). https://doi.org/10.17844/jphpi.v23i1.30862

Zaky Zamani, M., Prajitno, A., & Fadjar, M. (2019). Morphological characteristics of bioactive compounds on api-api mangrove leaves extract (*Avicennia marina*) based on leaves age. *Research Journal of Life Science*, *6*(3). https://doi.org/10.21776/ub.rjls.2019.006.03.4

Zhu, Z., Zhao, Y., Zhang, Y., Wu, X., Liu, J., Shi, Q., & Fang, Z. (2021). Effects of ultrasound pretreatment on the drying kinetics, water status and distribution in scallop adductors during heat pump drying. *Journal of the Science of Food and Agriculture*, *101*(15), 6239–6247. https://doi.org/10.1002/jsfa.11290