



JURNAL RONA TEKNIK PERTANIAN
ISSN: 2085-2614; e ISSN : 2528-2654
Journal homepage: <https://jurnal.usk.ac.id/RTP>



Phytochemicals - Bioactivity of *Avicennia marina* Leaves Extract, and Its Application in Food Products: A Brief Literature Review

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Abstract

Avicennia marina (*A. marina*) is tropical and non-tropical biodiversity that grows along the coast worldwide, including Indonesia. *A. marina*, also known as the "grey mangrove" tree, is a mangrove plant of the *Avicenniaceae* family. A widely used plant part is *A. marina* leaves, which are further processed into *A. marina* leaves extracts. Various studies on *A. marina* leaves, extract, and their application to food products have been extensively researched and communicated in various scientific papers. So, this study aims to provide an overview of the current knowledge on *A. marina* leaves extract, focusing on its phytochemical-bioactivity and application in food products. Various studies have reported that *A. marina* leaves extract contains polyphenols, flavonoids, steroids, terpenoids, alkaloids, cardiac glycosides, saponins, tannins, anthraquinones, and other phytochemical compounds based on phytochemical screening. Several recent studies also reported that *A. marina* leaves extracts have many benefits due to their biological activities, including antioxidant, anti-bacterial, anti-fungal, and anti-cancer activities. The application of *A. marina* leaves in food products, such as chocolate bars and noodles, is reported to increase the specific nutritional value. In addition, the water extract of *A. marina* leaves can also maintain the quality of fishery products, especially fresh fish, and acts as a natural bio-formalin or bio-preservation. In future research, the application and supplementation of water extract of *A. marina* leaves in other food products need to be studied further. In addition, other food process engineering must be applied and evaluated to produce *A. marina* leaves extract with maintained bioactive compounds and their bioactivity.

Keywords: *A. marina* leaves, bioactivity, food application, natural products, phytochemical

Fitokimia - Bioaktivitas Ekstrak Daun *Avicennia marina* dan Aplikasinya pada Produk Pangan: Tinjauan Literatur Singkat

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Abstrak

Avicennia marina (*A. marina*) merupakan keanekaragaman hayati tropis dan non-tropis yang tumbuh di sepanjang pantai di seluruh dunia, termasuk Indonesia. *A. marina*, yang juga dikenal sebagai pohon "bakau abu-abu", adalah tanaman bakau dari keluarga *Avicenniaceae*. Bagian tanaman yang banyak dimanfaatkan adalah daun *A. marina*, yang selanjutnya diolah menjadi ekstrak daun *A. marina*. Berbagai penelitian mengenai daun *A. marina*, ekstrak, dan aplikasinya pada produk pangan telah banyak diteliti dan dikomunikasikan dalam berbagai karya ilmiah. Oleh karena itu, penelitian ini bertujuan untuk memberikan gambaran umum tentang pengetahuan terkini tentang ekstrak daun *A. marina*, dengan fokus pada fitokimia-bioaktivitas dan aplikasinya pada produk pangan. Berbagai penelitian telah melaporkan bahwa ekstrak daun *A. marina* mengandung polifenol, flavonoid, steroid, terpenoid, alkaloid, *cardiac glycosides*, saponin, tanin, antrakuinon, dan senyawa fitokimia lain berdasarkan skrining fitokimia. Beberapa penelitian terbaru juga melaporkan bahwa ekstrak daun *A. marina* memiliki banyak manfaat karena aktivitas biologisnya, seperti aktivitas antioksidan, anti-bakteri, anti-jamur, dan anti-kanker. Aplikasi daun *A. marina* pada produk makanan, seperti pada *chocolate bars* dan mie, dilaporkan dapat meningkatkan nilai gizinya secara spesifik. Selain itu, ekstrak air daun *A. marina* dapat menjaga kualitas produk perikanan, terutama ikan segar, dan berperan sebagai bio-formalin atau pengawet alami. Pada penelitian selanjutnya, aplikasi dan suplementasi ekstrak air daun *A. marina* pada produk pangan lain perlu dikaji lebih lanjut. Selain itu, rekayasa proses pangan lain perlu diterapkan dan dievaluasi untuk menghasilkan ekstrak daun *A. marina* dengan senyawa bioaktif dan bioaktivitas yang terjaga.

Kata kunci: Aplikasi pada makanan, bioaktivitas, daun *A. marina*, fitokimia, produk alam

INTRODUCTION

Phytochemicals are naturally occurring compounds found in plants that offer a range of potential health benefits. Among these, the *Avicennia marina* (*A. marina*), known as the "grey mangrove", has gained attention for its bioactive properties. *A. marina* is a mangrove plant that grows in tropical and subtropical coastal and swampy areas (Kathiresan & Bingham, 2001). This plant is typically found in areas inundated by seawater during high tide and is resilient to extreme environmental conditions such as drought, high humidity, and low soil acidity (Budiadi et al., 2022; Chen & Ye, 2014;

Purnobasuki & Utami, 2016). The growth of *A. marina* is influenced by various factors such as temperature, humidity, and water salinity (Kodikara et al., 2018; López-Hoffman et al., 2007; Nguyen et al., 2015; Yanti et al., 2021). The plant can grow up to 3-10 meters, depending on the environmental conditions in which it grows. *A. marina* has an intense and complex root system that allows it to survive in challenging environments.

One commonly used plant part is *A. marina* leaves, which are further processed into *A. marina* leaves extracts. Over the years, numerous studies have been conducted on this plant species due to its rich phytochemical composition and potential bioactivities. In particular, the leaves of *A. marina* have been a subject of interest in many of these studies (Andriani et al., 2021; Febriani et al., 2020; Ibrahim et al., 2022; Sivalingam, 2021; Sohaib et al., 2022; Vasanthakumar et al., 2019), as they are known to contain a wide range of bioactive compounds such as alkaloids, flavonoids, tannins, and phenolic compounds. Furthermore, the extracts from *A. marina* leaves have also been shown to possess various pharmacological activities such as antioxidant (Iranawati et al., 2018; Yassien et al., 2021), anti-microbial (anti-bacterial and anti-fungal) (Ananthavalli & Karpagam, 2017; Sohaib et al., 2022; Vasanthakumar et al., 2019), and anti-cancer (Andriani et al., 2021; Huang et al., 2016) activity, and so on. These properties make *A. marina* leaves extract a promising candidate for developing new drugs and nutraceuticals. Given the growing interest in *A. marina* leaves extract, it is not surprising that many studies have been conducted to investigate its bioactivity and potential applications. One of the critical applications of *A. marina* leaves extract is in the food industry, where it has been utilized as a natural food preservative (Iswadi et al., 2015; Rofik & Ratnani, 2012; Safitri et al., 2020; Sumartini et al., 2021), or improving specific nutrition value or sensories acceptance in the food product (Ratrinia & Sumartini, 2021; Sumartini et al., 2022; Zahara & Zuraidah, 2018). So, this brief literature review aims to provide an overview of the current knowledge on *A. marina* leaves extract, focusing on its phytochemical-bioactivity and application in food products.

MATERIALS AND METHODS

This research uses a literature study (Paré & Kitsiou, 2017). The research papers used in this study are related to the topics discussed, which include the phytochemical-bioactivity of *A. marina* extracts and the use of *A. marina* leaves water extract in food products and its effect, as reported by some prior researchers from around the world, and were found in open-access sources such as Google Scholar (scholar.google.com) and Crossref (<https://www.crossref.org/>).

RESULTS AND DISCUSSION

Physical Appearance, Chemical, and Bioactive Compositions of *A. marina* Leaves

Avicennia marina leaves have been known as traditional medicine for treating skin disease, rheumatism, smallpox, and many other diseases long ago (Bandaranayake, 2002; Fauvel et al., 1993). Physically, the *A. marina* leaves have a different colour on the upper and lower surfaces, the upper surface is green, and the lower surface has a yellowish-green colour and becomes white when the increasing of leaves age. The shape of the leaves was oval or ovoid with a tapered end, with the length, width, and thickness of the leaves being 69.36 mm, 36.29 mm, and 0.77 mm, respectively.

Moreover, the upper surface of *A. marina* leaves smooth, but the lower surface has a rough texture (Jacoeb et al., 2011). The appearance of *A. marina* leaves is shown in Figure 1.

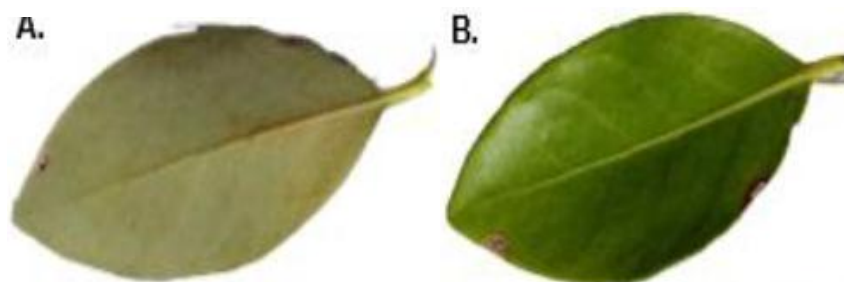


Figure 1. The appearance of the lower surface (A) and upper surface (B) of *A. marina* leaves (Mariano et al., 2019)

Chemically, *A. marina* fresh leaves contain a water content of 68.16%, protein content of 3.67%, fat content of 0.72%, ash content of 4.45%, carbohydrate content of 23.00, and the rest was crude fiber (Jacoeb et al., 2011). In addition, the dried *A. marina* leaves have moisture content of 15.6%, ash content of 7.17-15.3%, the fiber content of 12.18-18.7%, total protein of 6.8 – 13.37%, total lipid of 3.18 -7.3%, total carbohydrate of 23.4%, with total organic matter and other organic matter in respectively was 59.6 - 92.83 and 7.17 -11.5 (Al-Mur, 2021; Jamarun et al., 2020). Moreover, Jamarun et al. (2020) reported that neutral detergent fiber, acid detergent fiber, cellulose, hemicellulose, and lignin content was 45.99%, 35.95%, 35.95%, 10.03, and 7.34% in dried *A. marina* leaves in sequence. Furthermore, dried *A. marina* leaves also have a macro-content and microcontent of minerals. The microcontent mineral dominates by Cl (1.03%), followed by P (0.51%), K (0.48%), Ca (0.38%), Na (0.20%), Mg (0.20%), and S (0.01%). In contrast, the microcontent mineral was dominated by Fe (388 ppm), Mn (211 ppm), Zn (164 ppm), and Cu (128 ppm) (Jamarun et al., 2020). (Huang et al., 2016) revealed that *A. marina* leaves contain the trace elements of Fe (128.7 mg/kg), Ni (2.7 mg/kg), In (2.6 mg/kg), Rb (10.3 mg/kg), Zn (12.7 mg/kg), but Ag, Al, As, Cu, Cd, and Cr were not detected. In other reports, Yanti et al. (2021) stated that micromineral in *A. marina* leaves in several cost areas of West Sumatra consists of N (3.08 to 3.49%), K (1.83 to 2.04%), Mg (0.15 to 0.87%), Ca (0.38 to 0.69%), P (0.14 to 0.17%), and S (0.005 to 0.009%). Meanwhile, the micronutrient consisted of Zn (2.02 to 2.52 ppm), Mn (2.02 to 2.52 ppm), Fe (1.00 to 1.19 ppm), Cl (0.57 to 0.75 ppm), Cu (0.49 to 0.89 ppm), and Mo (0.41 to 0.47 ppm). Moreover, heavy metals, Hg (0.04 to 0.08 ppm) and Pb (0.23 to 0.25 ppm) were also detected in *A. marina* leaves.

The dried *A. marina* leaves contain total phenolic content (TPC) of 1.06 to 1.50 mg GAE/L (Aprilliana et al., 2021) or 13.02 to 24.51 mg GAE /g (Febriani et al., 2020; Su'aidah et al., 2021; Yanti et al., 2021). Hastuti et al. (2020) also reported that the TPC of *A. marina* leaves an average of 1.28%, with internal in 0.88 to 1.62%. In another study, Al-Mur (2021) also reported that the TPC and total flavonoid content (TFC) of dried *A. marina* leaves were 9.5% and 10.9%, respectively. Sediment nutrients, water quality, environmental conditions, level maturity of leaves, and location growth of plants influenced TPC and TPF in *A. marina* leaves (Aprilliana et al., 2021; Hastuti et al., 2020; Rahman & Sasmito, 2021; Su'aidah et al., 2021). Aprilliana et al. (2021) also reported that *A. marina* leaves contain chlorophyll a of 7.86 to 10.86 mg/L, chlorophyll b of 9.25 to 13.29 mg/L, and carotenoid of 608.37 to 1723.22 mg/L. The *A. marina*

leaves also reported having a total coumarin content of 8.418×10^{-3} mg CE/g (Febriani et al., 2020) and a tannin content of 1.40 to 4.24% (Yanti et al., 2021).

Extraction Methods of Bioactive Compounds from *A. marina* Leaves

Extracting bioactive compounds from *A. marina* leaves commonly uses maceration methods with various solvents, like water, methanol, ethanol, n-hexane, and ethyl acetate. However, any studies used the infusion method using water as a solvent. Maceration is regarded as the most straightforward and popular extraction technique, with the disadvantages of lengthy extraction durations and low extraction effectiveness. However, raw materials with thermolabile properties can be extracted using this technique (Zhang et al., 2018). For preparation, the *A. marina* leaves were dried to obtain dried *A. marina* leaves, followed by milling and sieving to obtain the uniform size of *A. marina* leaves powder. For the extraction procedure, *A. marina* leaves powder or chopped fresh leaves with a specific weight were combined with the solvent with a specific ratio at a specific time, with or without continuous stirring. Commonly, after extraction, the extracts were filtered using filter paper or centrifuge. Any two ways for filtered *A. marina* leaves extracts. First ways, the filtered extract of *A. marina* leaves is directly stored in liquid form at 4°C in the refrigerator (Rozirwan et al., 2022; Sumartini et al., 2021; Safitri et al., 2020; Iswadi et al., 2015; Afzal et al., 2011) or redissolved in DMSO (Ananthavalli and Karpagam, 2017). In other ways, the filtered extracts of *A. marina* leaves were evaporated using a rotary evaporator at a specific temperature (37-55 °C) to obtain a paste crude extract of *A. marina* leaves and further stored until used (Sohaib et al., 2022; Ibrahim et al., 2022; Andriyani et al., 2021; Yassien et al., 2021; Rahman & Sasmito, 2021; Zamani et al., 2019; Ananthavalli & Karpagam, 2017; Huang et al., 2016; Hardiningtyas et al., 2014). The summarized extraction conditions using the maceration method of *A. marina* leaves are tabulated in Table 1.

Infusions are often diluted solutions of easily soluble chemical components in the raw plant material. Previously, a brief maceration of the raw plant in either cold or scalding hot water might result in additional infusions of the phytochemicals present in the plant sample. The fundamental steps in making fresh infusions are moisturizing the powdered raw material plant, treating it with cold water for approximately 15 min, following that treating it with boiling water for 30 min, and then filtering it (Manousi et al., 2019) for further application or after filtering, the water extracts were evaporated using a vacuum rotary evaporator to obtain paste water extract (Huang et al., 2016). The main disadvantage of infusions is that water extracts contain much water and must be consumed quickly after preparation, so they are susceptible to bacterial and fungal growth (Manousi et al., 2019). The extraction of water extract of *A. marina* leaves using this method was performed by Huang et al. (2016). Briefly, the dried *A. marina* leaves were dissolved in water with a ratio material to solvent 1:10 w/v for 30 min at room temperature, then boiled at 95 °C for the next 30 min. After that, the water extracts were filtered using filter paper and followed by evaporation using a vacuum rotary evaporator at 80°C to obtain paste water extract of *A. marina* leaves.

Table 1. The extraction conditions using the maceration method of *A. marina* leaves

Raw material types	Extraction conditions	Reference
Fresh leaves	• Solvent: ethyl acetate • Ratio material to solvent: 1:10 w/v • Extraction time: 24 h • Extraction stages: 4 times	Rozirwan et al. (2022)
Dried leaves powder	• Solvent: absolute ethanol • Ratio material to solvent: 1:5 w/v • Extraction time: 5 days •	Sohaib et al. (2022)
Dried leaves powder	• Solvent: ethanol, acetone, or ethyl acetate • Ratio material to solvent: no information • Extraction time: 4 days	Ibrahim et al. (2022)
Dried leaves powder	• Solvent: methanol • Ratio material to solvent: 1:5 w/v • Extraction time: 24 h	Andriyani et al. (2021)
Dried leaves powder	• Solvent: ethanol • Ratio material to solvent: 1:5 w/v • Extraction time: 24 h	Sivalingam (2021)
Fresh leaves	• Solvent: water • Ratio material to solvent: 1:3 w/v • Extraction time: 24 h	Sumartini et al (2021); Safitri et al. (2020); Iswadi et al. (2015)
Dried leaves powder	Stage 1: • Solvent: n-hexane • Ratio material to solvent: 1:2 w/v • Extraction time: 48 h Stage 2: • Solvent: ethyl acetate • Ratio material to solvent: 1:2 w/v • Extraction time: 48 h Stage 3: • Solvent: ethanol • Ratio material to solvent: 1:2 w/v • Extraction time: 48 h	Rahman & Sasmito (2021)
Dried leaves powder	• Solvent: 80% ethanol and methanol • Ratio material to solvent: 1:20 w/v • Extraction time: 24 h	Yassien et al. (2021)

Table 1. *continued*

Raw material types	Extraction conditions	Reference
Dried leaves powder	• Solvent: ethanol • Ratio material to solvent: 1:5 w/v • Extraction time: 24 h	Zamani et al. (2019)
Dried leaves powder	• Solvent: ethanol or water • Ratio material to solvent: 1:3 w/v • Extraction time: 24 h	Ananthavalli & Karpagam (2017)
Dried leaves powder	• Solvent: ethanol and methanol • Ratio material to solvent: 1:10 w/v • Extraction time: 24 h	Huang et al. (2016)
Dried leaves powder	• Solvent: methanol, n-hexane, or ethyl acetate • Ratio material to solvent: 1:5 w/v • Extraction time: 24 h	Hardiningtyas et al. (2014)
Dried leaves powder	• Solvent: ethanol • Ratio material to solvent: no information • Extraction time: 7 days	Afrizal et al. (2011)

Phytochemical and Bioactive Compounds of *A. marina* Leaves extracts

The phytochemical composition of *A. marina* leaves extract is similar to the several leaves extracts reported by several researchers previously (Utoro et al., 2022a; Utoro et al., 2022b), which consists of polyphenols, flavonoids, steroids, terpenoids, alkaloids, cardiac glycosides, saponins, tannins, and anthraquinones based on the phytochemical screening, and depending on the extracts used for the analysis (Ananthavalli & Karpagam, 2017; Jacob et al., 2011; Rozirwan et al., 2022; Sivalingam, 2021; Vasanthakumar et al., 2019). The qualitative phytochemical screening comparison of various *A. marina* leaves extracts is presented in Table 2.

Besides the *A. marina* leaves, the various *A. marina* leaves extracts also contain the same bioactive compounds. The quantitative bioactive compounds were reported in various *A. marina* leaves extracts. The ethanolic extract of *A. marina* leaves contains total phenolic content (TPC) in the range of 1915.92 -24.236 mg GAE/g (Rahman & Sasmito, 2021; Yassien et al., 2021) and total flavonoid content (TFC) of 10.022 mg rutin/ g, and IC₅₀ of 193.9 µg/mL (Yassien et al., 2021). In contrast, the ethyl acetate and n-hexane of *A. marina* leaves extracts have a TPC of 1.3205 to 752.97 mg GAE/g (Rahman & Sasmito, 2021) and 113.22 mg GAE/g (Rahman & Sasmito, 2021; Rozirwan et al., 2022), respectively. Moreover, the methanolic Extract of *A. marina* leaves also contains TFC of 21.7 mg/L and TPC of 1908.8 mg/L (Al-Mur, 2021). Huang et al. (2016) reported that the TPC of methanolic, ethanolic, water, and ethyl acetate of *A. marina* leaves extracts was 46.96 mg/g, 22.82 mg/g, 47.06 mg/g, and 80.96 mg/g, respectively. In addition, the TFC was 9.26 mg/g, 11.96 mg/g, 6.83 mg/g, and 18.69 mg/g for methanolic, ethanolic, water, and ethyl acetate of *A. marina* leaves extract in sequence.

Table 2. The phytochemical screening qualitative of various *A. marina* leaves extracts

Extract Types	Compounds identified
Water or aqueous	Cardiac glycosides, leuco-anthocyanin, xanthoprotein, coumarin, quinones, glycosides, alkaloids, saponin, phenolics, flavonoids, diterpenes, tannin, and terpenoids (Ananthavalli & Karpagam, 2017).
Ethanol	Phenolics (Ananthavalli & Karpagam, 2017; Sivalingam, 2021; Zamani et al., 2019), flavonoids, tannins (Ananthavalli & Karpagam, 2017; Rahman & Sasmito, 2021; Sivalingam, 2021; Vasanthakumar et al., 2019; Zamani et al., 2019), steroids (Rahman & Sasmito, 2021; Sivalingam, 2021; Vasanthakumar et al., 2019), terpenoids (Ananthavalli & Karpagam, 2017; Rahman & Sasmito, 2021; Vasanthakumar et al., 2019; Zamani et al., 2019), alkaloids (Ananthavalli & Karpagam, 2017; Vasanthakumar et al., 2019; Zamani et al., 2019), cardiac glycosides, anthraquinones (Vasanthakumar et al., 2019), saponins (Ananthavalli & Karpagam, 2017; Rahman & Sasmito, 2021), diterpenes, oxalate, anthocyanin, leucoanthocyanin, carboxylic acid, xanthoprotein, coumarin glycosides, and quinones (Ananthavalli & Karpagam, 2017)
Ethyl Acetate	Flavonoids (Hardiningtyas et al., 2014; Jacob et al., 2011; Rozirwan et al., 2022), steroids (Hardiningtyas et al., 2014; Jacob et al., 2011; Rozirwan et al., 2022), and saponin (Rozirwan et al., 2022)
Methanol	Phenolics, tannin (Jamarun et al., 2020; Sivalingam, 2021), steroids, terpenoids, alkaloids (Jamarun et al., 2020), saponin (Sivalingam, 2021), terpenoids (Hardiningtyas et al., 2014; Rahman & Sasmito, 2021), and tannins (Rahman & Sasmito, 2021)
n-hexane	Flavonoids, steroids, terpenoids, and alkaloids (Rahman & Sasmito, 2021)

Biological Activities of *A. marina* Leaves Extracts

Several studies reported that *A. marina* leaves extracts have biological activities, including antioxidant, anti-bacterial, anti-fungal, and anti-cancer effects. The biological activities of the *A. marina* leave extracts, both *in vitro* and *in vivo*, were summarized in this section.

Antioxidant: Methanolic extract of *A. marina* leaves has lower antioxidant activity than *A. alba* leaves. The IC₅₀ of methanolic Extract of *A. marina* leaves was 123.23 ppm, classified as a moderate antioxidant with the range of inhibition by DPPH was 7.05 to 88.41% at the concentration of 31.25 to 250 ppm. Moreover, the antioxidant activity of the methanolic Extract of *A. marina* leaves was higher than that of the bark (Iranawati et al., 2018). Hardiningtyas et al. (2020) and Jacob et al. (2011) reported that crude-ethyl acetate extract has a lower IC₅₀ than other extracts, such as methanolic and hexane extracts. The range of IC₅₀ in crude-ethyl acetate, methanolic, and hexane extracts was 181.73 to 182.33 ppm, 254.77 to 257.58 ppm, and 1033.66 to 1013.43 ppm,

in sequence. The lowest IC_{50} in crude ethyl acetate extract indicated the highest antioxidant activity. Ethanolic Extract of *A. marina* leaves also had an antioxidant activity with DPPH IC_{50} of 193.9 $\mu\text{g/mL}$, ABTS of 326.8 $\mu\text{M TE/mg}$, and FRAP of 340.29 $\mu\text{M TE/mg}$ (Yassien et al., 2021). The antioxidant activity of *A. marina* leaves extract is caused by the presence of phenolic and flavonoid compounds composition in their Extract (Sivalingam, 2021; Yassien et al., 2021).

Anti-bacterial: Several researchers reported various *A. marina* leaves extracts' anti-bacterial activity in gram-positive and gram-negative bacteria. The ethanolic Extract of *A. marina* leaves at a low concentration of 2.5 to 10% inhibited the growth of *Streptococcus mutans*, but could not inhibit the growth of *Candida albicans* (Dharmautama et al., 2017). *In vitro* studies performed by Yassien et al. (2021) reported the 50 μL of ethanolic Extract of *A. marina* leaves revealed the anti-microbial in the various pathogenic bacteria using the disk diffusion methods *Bacillus subtilis*, *Candida albicans*, *Salmonella typhimurium*, *Vibrio damsela* and *Staphylococcus aureus*, with the diameter zone inhibition of 22 mm, 25 mm, 16 mm, 16 mm, and 14 mm, in sequence, but cannot inhibit the *Escherichia coli*, and *Pseudomonas aeruginosa*. Ananthavalli & Karpagam (2017) revealed that the ethanolic and aqueous of *A. marina* leaves at concentrations of 25 to 100 μL effectively inhibit the growth of *Salmonella typhi*, *Klebsiella pneumoniae*, *Escherichia coli*, and *Staphylococcus aureus*. However, the effectivity was lower inhibition than the Amoxicillin as the positive control. Ibrahim et al. (2022) also reported that the ethyl acetate crude extract of *A. marina* leaves was the most effective in inhibiting *P. fluorescens*. However, the *B. subtilis* was inhibited effectively by crude acetone extract. Moreover, different pathogenic bacteria like *S. epidermis*, *E. coli*, *K. pneumonia*, and *B. pertussis* are also inhibited by either acetone, ethanol, or ethyl acetate crude extract with different inhibition degrees.

Anti-Fungal: Distilled water and dimethyl sulphoxide (DMSO) of *A. marina* leaves extracts at all concentrations of 2000, 4000, and 6000 ppm were found to be the most effective solvents preventing the growth of seven allergenic fungi (*Alternaria alternata*, *Aspergillus flavus*, *Aspergillus fumigatus*, *Aspergillus niger*, *Cladosporium herbarum*, *Penicillium notatum*, and *Saccharomyces cerevisiae*, with inhibition percentage of 72.91%, 80.33%, 63.25%, 83.00%, 65.25%, 78.58%, and 49.5%, respectively, and highly significantly different compared with the negative control. Moreover, the chloroform, ethanolic, and acetone of *A. marina* leaves extract also had an anti-fungal activity lower than the two. A dose-dependent tendency was discovered during the investigation, and the highest inhibition was discovered at 6000 ppm. The *A. marina* leaves are potent and effective against fungi because they contain phenols and flavonoids (Afzal et al., 2011).

Anti-Cancer: The ethyl acetate extract and fractions had higher anti-cancer activity and cytotoxic effects than the other extracts (methanolic, ethanolic, and water), was it caused by contained avicennones D and E based on $^1\text{H-NMR}$ and $^{13}\text{C-NMR}$. Furthermore, xeno-transplanted human MDAMB-231 breast cancer cells into the back skin of BALB/c nude mice were shown in *in-vivo* investigations to be suppressed by ethyl acetate extract (Huang et al., 2016). In another research, Andriani et al. (2021) reported the methanolic extract of *A. marina* leaves to have cytotoxic activity and anti-proliferative on HeLa cervical cancer cells through *in vitro* studies, and more effectively than taurine, free amino acids, in the same purposes studies based IC_{50} and cell viability parameters at the same concentration used. The phenolic compound contained the A.

marina leaves extracts, which were predicted as the primary contributor to its anti-cancer activity (Sohaib et al., 2022).

Application in Food Products

The community has widely used the *A. marina* leaves powder or water extract of *A. marina* leaves for various purposes, such as for preservation or improved food quality. Sumartini et al. (2022) added the 10% *A. marina* leaves powder during chocolate bars manufacturing and observed it during storage at room temperature for 0, 7, and 14 days. The addition of 10% *A. marina* leaves powder effectively decreases the ash content of chocolate bars during 14 days of storage. Ratrinia & Sumartini (2021) reported that adding marina leaves powder during chocolate bar manufacturing increases water, protein, ash, carbohydrate, dietary fibre, vitamin C, and decreased fat content in chocolate bars compared to controls. Zahara & Zuraidah (2018) reported that the enrichment of 30 g of *A. marina* leaves powder had a better proportion likely by panellists than another proportion in wet noodles.

Another study reported *A. marina* leaves as a preservation agent in fisheries products. Sumartini et al. (2021) evaluated the effect of the concentration of water extract of *A. marina* leaves (15%, 20%, 25%) under 3 different conditions storage (room temperature (23°-27°C), cold temperature (10°C), and freezing temperatures (0°C) on the quality of the *Layang Benggol* fish at 24 h of storage time. The findings indicated that storage temperature considerably impacted the quality of the *Layang Benggol* fish. However, the concentration of water extract from *A. marina* leaves had a negligible impact on water content. With a water extract of *A. marina* leaves of 25%, which showed a water content value of 76.73%, the optimum freshness was discovered in freezing temperature storage. In contrast, the *E. coli* test revealed that *Layang Benggol* fish was negative after exposure to cold and freezing temperatures at all water extracts of *A. marina* leaves concentrations. Safitri et al. (2020) conducted an experiment application of water extract *A. marina* leaves for the preservation of fresh cob fish, with different concentrations of 0% to 30%, with intervals of 5% during 12 h and 24 h, and the microbial growth (ALT and MPN *E. coli*) as the response. The experimental research revealed that increased water extract of *A. marina* leaves applied in fresh cob fish decreased the ALT and MPN *E. coli* during storage. Moreover, the fulfilment standard in ALT parameters was achieved by applying water extract of *A. marina* leaves in a concentration of 15% in 12 h and 30% in 24 h. Iswadi et al. (2015) evaluated the application of various concentrations of water extract from *A. marina* leaves in a range of 5 to 20%, with intervals of 5% on the microbial growth in fresh cob fish during 12 and 24 h storage at room temperature. The best concentration to inhibit microbial growth in fresh cob fish during storage was 20% of concentrations extracts. The physical observed at 12 h storage time with the application of 20% of concentrations showed clear eyes, tense skin, red gills, supple flesh, and solid consistency. The organoleptic testing by panellists showed fresh cob fish treated with 20% extract concentrations still favoured because of the fish's aroma, taste, colour, and texture, like fresh fish.

Rofik & Ratnani (2012) also reported the *A. marina* leaves with various proportions (1:5, 1:7.5, and 1:10 w of *A. marina* leaves powder/ v of water) as a bio-formalin solution for preserving milkfish. The milkfish was immersed in the bio-formalin solution for 15 min before storage at room temperature for 20 h and observed each for 2 h. The best proportion was revealed at 1:5 w of *A. marina* leaves powder/ v of water because it can be preserved longer than 20 h compared with other proportions

and controls. The visual characteristics of milkfish were white skin, flat eyes, slightly open mouth, slightly chewy flesh, strong scales, and fishy aroma.

CONCLUSION

Avicennia marina (*A. marina*) leaves are a part of the *A. marina* plant, found along the coasts worldwide in tropical and subtropical regions, including Indonesia, with high biological activities. Based on phytochemical screening, *A. marina* leaves extracts to dominated contain polyphenols, flavonoids, steroids, terpenoids, alkaloids, cardiac glycosides, saponins, tannins, and anthraquinones. According to some studies, *A. marina* leaves extracts have various biological activities, such as antioxidant, anti-bacterial, anti-fungal, anti-cancer, etc. It has also been reported that using *A. marina* leaves water extract in food products has an impact on increasing the nutritional value of food products or the maintenance of the quality of fisheries products (especially fish), which act as bio-preservation or bio-formalin. Further research is needed regarding applying and supplementing water extract from *A. marina* leaves in other food products. Other process techniques needed more to extract *A. marina* leaves with maintained bioactive compounds and their bioactivity.

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