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(Research Article)

Premna microphylla Turcz. (Potential as Food Ingredients and Its Bioactivity)

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Abstract

Premna microphylla Turcz is an introduced plant in Indonesia that is used as an ornamental plant, whereas in its country of origin (China) it has long been used as a food ingredient and traditional medicine. This study aims to explain its botany, nutrition and bioactivity as well as its secondary metabolites. The research method was carried out by studying literature obtained online, especially from Google Scholar using keywords such as *P. microphylla*, pectin of *P. microphylla* and bioactivity of *P. microphylla*. The information obtained was synthesized again so that it could explain the botany, bioactivity, and secondary metabolite content of *P. microphylla*. *P. microphylla* has a shrubby habitus with many branches, opposite leaves with serrated edges and berries. In traditional Chinese medicine, *P. microphylla* is used to treat infectious diseases such as dysentery, appendicitis, treating hemostasis, rheumatism, and dysentery. The use of *P. microphylla* as a traditional medicine is related to its bioactivity, including: antioxidants, antimicrobials, cytotoxics and sunscreens. The leaves of *P. microphylla* are rich in nutrients, including pectin (17.33 g/100 g), crude protein (14.85 g/100 g), vitamin C (8.14 mg/100 g). The bioactivity of *P. microphylla* to protect skin from ultraviolet rays is very potential to be developed in the beauty/cosmetics industry while its pectin is developed as in the gel-making industry.

Keywords: *Premna microphylla*; Sunscreen; Pectin; Infectious diseases

1. Introduction

Premna microphylla Turcz is a species belonging Verbenaceae that is mostly spread in the mountains of Southeast China [1], but has now spread to various other countries including Indonesia as an introduced plant. In Indonesia, this plant is widely used as an ornamental plant because it has a flowering and fruit structure. Unlike in Indonesia, in its country of origin (China), *P. microphylla* has long been used as a traditional medicine, especially to treat various infectious diseases [2,3].

The traditional Chinese medicine, *P. microphylla* is used to treat infectious diseases such as dysentery [2,4], appendicitis [2], treat hemostasis, rheumatism, and dysentery [4]. The roots, stems, and leaves can be used for detoxification, swelling, and hemostasis [1]. The stem extract of *P. microphylla* for strong therapeutic potential against oxidation-related diseases [2]. In addition to being used as a traditional medicine, *P. microphylla* leaves have been used to prepare "green tofu" by local people in China [5]. The leaf juiceof *P. microphylla* is also processed into a traditional Chinese food resembling jelly known as "Guanyin Tofu" [6]. Empirically, Guanyin Tofu is used to relieve symptoms related to inflammation [6].

The use of *P. microphylla* as a food ingredient is associated with its nutritional content, especially polysaccharides in the form of pectin compounds [5]. The content of ascorbic acid and β-carotene in leaves and stems is quite high [2]. Design of bioactive polysaccharide composite gels for the field of food engineering [7]. The *P. microphylla* can be a non-toxic and promising natural food supplement to improve diseases caused by chronic inflammation [6]. Flavonoids and

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terpenoids are the main phytochemicals in *P. microphylla*, and methylation, sulfonation, and dihydroxylation are the main metabolic pathways in vivo [4].

Pectin is a complex polysaccharide that is very potential to be used in the pharmaceutical and food industries as a gel or food preservative. The water extract of *P. microphylla* leaves and glycerol are very potential as plasticizers [8]. Alcohol-insoluble solids from *P. microphylla* leaves contain high amounts of pectin [5]. The leaf pectin *P. microphylla* can be used as a potential additive in the food industry [9]. The essential oil has the potential to be used in the cosmetic and pharmaceutical industries [3]. The extract *P. microphylla* will be a potential candidate for application in food preservation to improve food safety [10]. This study aims to explain the botany, potential utilization of pectin and bioactivity of *P. microphylla*.

2. Methods

This study is based on literature studies obtained online, especially from Google Scholar. Some keywords used are *P. microphylla*, pectin of *P. microphylla* and bioactivity of *P. microphylla*. The information obtained is re-synthesized so that it can explain the botany, potential uses of pectin, and its bioactivity of *P. microphylla*.

3. Results and Discussion

3.1. Botany of *Premna microphylla* Turcz.

Premna microphylla belongs to Verbenaceae of the Genus *Premna*. This genus consists of more than 200 species worldwide. Description *P. microphylla*: is a perennial shrub with a height of 2–6 m. The stem has many branches. The leaves, the blades are oblong-lanceolate, elliptical, ovate, or inverted ovate, measuring 3–13 cm long and 1.5–6 cm wide. The tip is long tapering to sharp, and the base is narrow and pointed with an entire or notched to sometimes serrated edge. The leaf stalk ranges from 0.5 to 2 cm. The inflorescence is in the form of a conical panicle. The fruit is purple with a round to inverted ovate shape (Figure 1). The seeds, weighing about 1000 grains, are difficult to breed even though the formation rate is high (Tang et al 2018). This plant is native to South-Central China, Southeast China, Japan, Nansei-shoto, Taiwan [11].

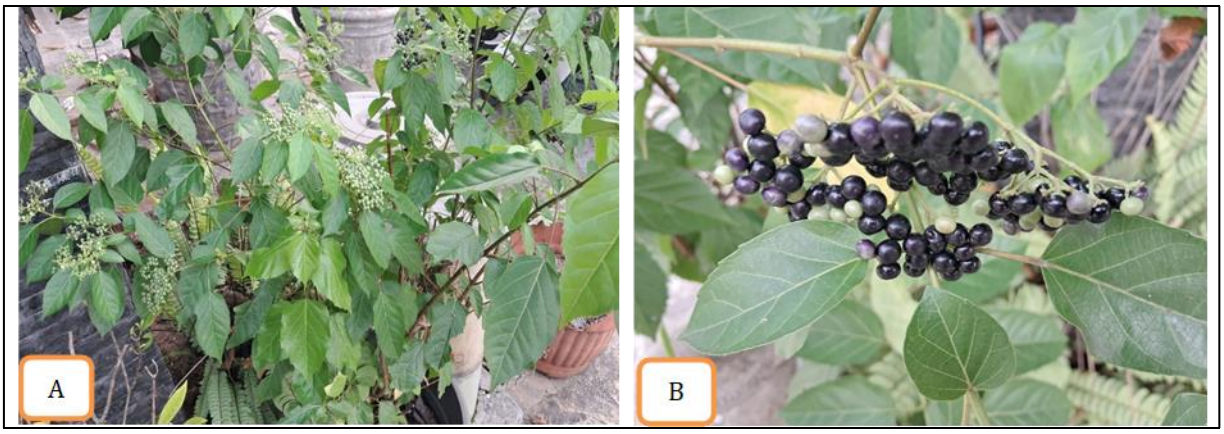

Figure 1 *Premna microphylla* Turcz. A. Habitus; B. Branches supporting ripe fruit and leaves

Premna microphylla Turcz has several synonymous names with *Gumira japonica* (Miq.) Kuntze in Revis. Gen. Pl. 2: 507 (1891); *Premna formosana* Maxim. in Bull. Acad. Imp. Sci. Saint-Pétersbourg, sér. 3, 31: 80 (1886); *Premna japonica* Miq. in Ann. Mus. Bot. Lugduno-Batavi 2: 97 (1865); *Premna luxurians* Nakai in Rep. Veg. Isl. Shikashima: 37 (1927); *Premna microphylla* var. glabra Nakai in Bot. Mag. (Tokyo) 40: 487 (1926); *Premna microphylla* var. luxurians (Nakai) Moldenke in Phytologia 3: 172 (1949) [11].

3.2. Nutritional Content and Pectin

Based on its function, food ingredients are grouped into sources of carbohydrates, proteins, fats and sources of fiber. Although the need for fiber is less than other ingredients (carbohydrates), the health of the digestive system is greatly influenced by fiber. One source of plant types as a source of fiber for food ingredients is *P. microphylla* leaves because they contain high pectin [5]. The *P. microphylla* leaf pectin can be used as a potential additive in the food industry [9], because of its excellent gelation properties [12].

P. microphylla has long been used as a food ingredient by local people in China as Chinese “green tofu” [5]. The leaves of *P. microphylla* are rich in nutrients, vitamins and mineral salts such as pectin (17.33 g/100 g), crude protein (14.85 g/100 g), vitamin C (8.14 mg/100 g), total amino acids (14.24 g/100 g), potassium (22,683.68 mg/kg) and calcium (2,835.15 mg/kg [13]. Alcohol-insoluble *P. microphylla* leaf solids contain high amounts of pectin [5]. The pectin of *P. microphylla* has several characteristics including (1) good thermal stability and pectin degrades at a temperature of 259.3°C [14]; (2) has a high molecular weight and poor solubility, thus limiting its application [15]; (3) the ability to form gels and texture is influenced by the solubility of pectin and the degree of methoxylation (methoxyl content) [12].

The facts show that fiber as a food ingredient is sometimes less preferred because of its hard texture, so further processing is needed to obtain the appropriate structure and texture. Plant fibers are generally in the form of polysaccharides stored in cell walls, including pectin. Pectin from *P. microphylla* leaves consists of rhamnose, glucose, galactose [9,12,14], arabinose, mannose [9], D-galacturonate [14]. The polysaccharides of *P. microphylla* are negatively charged acidic polysaccharides consisting mainly of glucose, galactose and rhamnose [12], so they are known as heteropolysaccharides [7]. The polysaccharide *P. microphylla* is an acidic heteropolysaccharide with →4)-α-D-GalpA-(1→ and →4)-α-D-Galpa-6-Ome-(1→ as the backbone structure) [7].

The pectin content of *P. microphylla* leaves varies greatly depending on the processing. The yield of pectin extracted by the ultrasonic enzyme method (35.53%) was higher than that of the traditional acid extraction method (17.28%) [14]. Ultrasonic destroys the regular structure in pectin *P. microphylla* resulting in a looser microscopic morphology and higher thermal stability [15]. The gel structure of *P. microphylla* pectin can change with the intervention of various factors such as ultrasonic [15], the addition of Ca^{2+} ions [12], the drying process [16] and the compounds used [17]. The resulting ammonium oxalate-soluble pectin showed high galacturonic acid content (76.15%), low neutral sugar content (6.41%) and methoxylation level (14.90%) [5]. The Ammonium oxalate was found to be the most effective *P. microphylla* pectin extracting agent, compared to water, hydrochloric acid and sodium hydroxide [5].

Research shows that the addition of *P. microphylla* to various foods will improve food quality. For example, packaged tofu added with 0.8% (w/w) *P. microphylla* leaf pectin extract has 1.43–1.49 times higher hardness, elasticity, and elasticity than the control so it is preferred [18]. Ultrasonic treatment of pectic will significantly reduce its molecular weight [15]. The gel strength, hardness, and thermal stability of pectin of *P. microphylla* gels are enhanced with increasing Ca^{2+} concentration. The microstructure of pectin of *P. microphylla* gels changes significantly after the addition of Ca^{2+}, resulting in lamellar and agglomerate-like structures due to pectin of *P. microphylla* aggregation [12]. The optimal Na^{+} concentration (0.6 M) for *P. microphylla* to form gels with the highest solid-like properties, water holding capacity, and crystallinity. With further increase in Na^{+} concentration from 0.6 M to 0.7 M, the gel properties of pectin gels will weaken [17]. The addition of low Na^{+} content (10 mM and 20 mM) can increase gel hardness, water holding capacity, thermal stability and crystallinity of the composite gel [19].

3.3. Uses and Bioactivity

Traditional Chinese medicine, *P. microphylla* is used to treat infectious diseases such as dysentery [2,4], appendicitis [2], treat hemostasis, rheumatism, and dysentery [4]. The roots, stems, and leaves can be used for detoxification, swelling, and hemostasis [1]. The use of *P. microphylla* as a traditional medicine is related to its bioactivity such as antioxidant, antimicrobial, cytotoxic and sunscreen.

3.3.1. Antioxidants

Antioxidants are compounds that inhibit free radicals. Empirical facts show that Premna microphylla leaf juice relieves symptoms associated with inflammation [6]. The bioactivity of *P. microphylla* as an antioxidant in vitro has been reported by various researchers such as Xu et al [20], Huang et al [10], Li et al [6], and in vivo by Song et al [1].

The bioactivity of *P. microphylla* extracts is influenced by the pH of the solvent and the compounds used for extraction [6,10], the organs used, and the process. Huang et al [10] stated that the ethanol extract of *P. microphylla* leaves showed better antioxidant properties compared to the water extract. The antioxidant activity of *P. microphylla* stem extract had significantly higher iron-reducing antioxidant properties than the leaves [20]. The pectin *P. microphylla* showed excellent radical scavenging ability, and especially those extracted using basic compounds [6]. The yield of *P. microphylla* pectin extracted by ultrasonic enzyme method had IC50 of 2,2-diphenyl-1-picrylhydrazyl (DPPH) and 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid)/ ABTS free radicals of 0.37 and 0.34 g/L, respectively, in the antioxidant activity test [14]. The *Drosophyla* that underwent inflammation (treated with sodium lauryl sulfate) then fed *P. microphylla* polysaccharide extract significantly extended the life span from 5 days to 6 days [1].

The bioactivity of *P. microphylla* pectin as an antioxidant is related to the increase in the enzymatic activities of endogenous superoxide dismutase, glutathione peroxidase, and catalase, thereby reducing inflammatory cytokines [6]. The increase in the lifespan of *Drosophyla* fed with *P. microphylla* polysaccharides reduced the rupture of microvilli in the midgut and restored the nuclear structure and increased the expression levels of immune-related genes [1]. The root *P. microphylla* extraction increased ConA to induce T lymphocyte proliferation reactions [21]. On the other hand, the leaf extract of the plant had higher levels of proanthocyanidins [2]. In addition, the leaf extract also had higher radical scavenging activity than the stem as shown in the DPPH [20].

3.3.2. Anti-Microbial

Antimicrobial compounds are compounds that can inhibit growth or cause death of microbes. Antimicrobial bioactivity can be applied to improve food preservation, thereby increasing food safety [10,20]. Methanol extracts from leaves and stems have significant activity against Gram-positive and Gram-negative bacteria, namely *Escherichia coli, Bacillus subtilis* and *Staphylococcus aureus* [20].

The bioactivity of *P. microphylla* as an antimicrobial varies depending on various factors such as the polarity of the solvent and the organ used. Ethanol extract of *P. microphylla* leaves showed satisfactory antimicrobial performance compared to water extract [10]. The activity of leaf extract was more prominent in several bacterial species, compared to stem extract [20]. Antimicrobial bioactivity is thought to be related to the content of essential oils such as blumenol C, β-cedrene, limonene, α-guaiene, cryptone, and α-cyperone [3]. The activity of *P. microphylla* essential oil showed a fairly large antibacterial capacity against *Escherichia coli* (MIC/Minimum Inhibitory Concentration value of 0.15 mg/mL) compared to *Bacillus subtilis* and *Staphylococcu*s aureus (MIC value of 0.27 mg/mL) [3] (Zhang et al 2017).

3.3.3. Cytotoxic

Plants that produce cytotoxic compounds have the potential as anticancer because they can inhibit cell growth. The study of food ingredients that have the potential as anticancer including *P. microphylla* has an interesting prospect because it is not toxic. The essential oil of *P. microphylla* has cytotoxic activity against HepG2 (hepatocellular liver cells) and MCF-7 cells (human breast adenocarcinoma cell lines) which through the MTT test (3-(4,5-dimethyl-2-thiazolyl)-2,5-diphenyl-2-H-tetrazolium bromide), and showed cytotoxic activity with IC50 of 0.072 and 0.188 mg/mL for 72 hours [3].

3.3.4. Sunscreen

Excessive exposure to ultraviolet B (UVB) radiation is one of the major risk factors for premature skin aging [22]. A cream containing *P. microphylla* pectin effectively inhibited water loss, epidermal hyperplasia, matrix metalloproteinase-1, and collagen damage in mice subjected to UVB-induced skin injury [22]. In addition, topical application of *P. microphylla* pectin cream increased the protein levels of nuclear factor erythroid 2-related factor 2 (Nrf2), Kelch-like ECH-associated protein 1, macrophage-activating factor, and heme oxygenase 1 (HO-1), and the expression of antioxidant enzymes including superoxide dismutase, catalase, and glutathione peroxidase. In contrast, application of cream containing *P. microphylla* pectin to the skin of mice decreased the levels of nuclear factor-kappa B (NF-κB), inhibitor of kappa B kinase β, and cyclooxygenase-2 proteins, and pro-inflammatory cytokines. *P. microphylla* pectin can protect skin aging caused by UVB through activation of the Nrf2 pathway and suppression of the NF-κB pathway [22]. The bioactivity of *P. microphylla* to protect the skin from UV rays has great potential to be developed in the beauty/cosmetic industry.

Secondary Metabolites

The potential use of *P. microphylla* in the health sector is related to its secondary metabolite content. The secondary metabolite content of plants varies between organs and between growth phases, which are directly or indirectly related to their bioactivity. The bioactivity of *P. microphylla* as an antimicrobial is thought to be related to the content of essential oils such as blumenol C, β-cedrene, limonene, α-guaiene, cryptone, and α-cyperone [3]. The content of phenolic compounds is often associated with bioactivity as an antioxidant. Studies on the content of secondary metabolites of *P. microphylla* are also still limited, even though it has good potential in medicine, especially as an antioxidant. The following is the content of secondary metabolites of *P. microphylla*.

Roots: contain compounds β-sitosterol, palmitic acid, trans-p-methoxycinnamic acid, eugenin, luteolin, luteolin-7-O-β-D-glucopyranoside, pedalitin [23], xanthones (1-hydroxy-2,3-methylenedioxy-6-methoxycarbonyl-7-acetylxanthone; 1,3-dihydroxy-2-methoxycarbonyl-6-methoxycarbonyl-7-acetylxanthone) [24].

Stem: contains the compounds vitexin, schaftoside, vicenin-2, apigenin-6, 8-di-c-arabinoside, apigenin-7-o-β-d-glucoside, carnosic acid, apigenin-8-c-β-d-xylopyranoside, prostratin, aurantio-obtusin-β-d-glucoside, royleanone, 5-hydroxy-7,3′,4′-trimethoxy flavonols, 6-hydroxy-5,6-dehydrosugiol, 14-deoxycoleon, arucadiol, obtusinone-b, trehalose, citric acid and betaine [2].

Leaves: contain compounds including diosmetin, blumenol A, (3S,5R,6S,7E,9R) 5,6epoxy3,9dihydroxy7megastigmene, ixerol B, (−)dehydrovomifoliol, 3βhydroxy5α,-6αepoxy7megastigmen9one, 3S,5Rdihydroxy6S,7megastigmadien9one, loliotide, (+)dehydro-loliolide, (+)medioresinol, 4oxopinoresinol, torturet acid, and indole3carboxylic acid [25], triterpene glycosides (tormic acid ester 28-O-α-L-rhamnopyranosyl (1→2)-β-D-glucopyranoside), triterpenes (arjunolic acid, hiptatic acid A) [26], (2S,3S,4R,11E)-2-[(2R)-2-hydroxytetracosa-noylamino]-11-octadecene-1,3,4-triol; 1-O(9Z,12Z, 15Z-octadecatrienoyl)-3-O-[β-d-galactopyra-nosyl-(1→6)-O-β-d-galactopyranosyl-(1→6)-α-d-galactopyranosyl] glycerol [27].

4. Conclusion

- The *Premna microphylla* has a shrubby habitus with many branches, opposite leaves with serrated edges and berries.
- In traditional Chinese medicine, *P. microphylla* is used to treat infectious diseases such as dysentery, appendicitis, treating hemostasis, rheumatism, and dysentery.
- The leaves of *P. microphylla* are rich in nutrients, including pectin (17.33 g/100 g), crude protein (14.85 g/100 g), vitamin C (8.14 mg/100 g).
- The bioactivity of *P. microphylla* to protect skin from ultraviolet rays is very potential to be developed in the beauty/cosmetics industry while its pectin is developed as in the gel-making industry.

Compliance with ethical standards

Disclosure of conflict of interest

No conflict of interest to be disclosed.

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