

## PHYTOCHEMICAL AND ANTIMICROBIAL STUDIES ON *GARCINIA LATTISSIMA* MIQ. FRUIT EXTRACT

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### ABSTRACT

**Objective:** The present investigation was aimed to explore the phytoconstituents present in the fruit part of *Garcinia latisissima* Miq. and their antimicrobial efficacy.

**Methods:** The preliminary phytochemical constituents were qualitatively analyzed using the standard procedures described in *Materia Medica* Indonesia. Antimicrobial screening was performed using disc diffusion and dilution methods.

**Results:** Phytochemical screening of different extracts of *G. latisissima* Miq. fruits revealed the presence of tannins, saponins, flavonoids, and alkaloids, and the results are shown in Table 1. The ethyl acetate and methanolic extracts of *G. latisissima* Miq. fruits showed antimicrobial activity, and the n-hexane extract failed to prove the inhibition against the selected pathogens.

**Conclusion:** The results of the phytochemical and bio-efficacy study revealed most valuable information and also support the continued sustainable use of *G. latisissima* Miq. fruits in the traditional system of medicine.

**Keywords:** *Garcinia latisissima* Miq., Antimicrobial, Phytochemical, Tannins, Saponins, Flavonoids, Alkaloids.

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### INTRODUCTION

This study proposed the existence of active phytochemical compounds and to explore the antimicrobial activity in different solvents of *Garcinia latisissima* Miq. fruit extract.

Plants as living chemical factories provide a vast number of important chemical substances that display a variety of biological actions. About 35,000 (some estimate up to 70,000) plant species are used worldwide for medicinal purposes. Researchers have investigated <0.5% of these for their phytochemical and pharmacological potentials. More latterly there has been a recovery of attracting in the medicinal possibles of therapeutic trees as antimicrobials. Also, others species have been researched to new antimicrobials potential of plant species [1]. The Clusiaceae or Guttiferae family contains 27 genera and 1,090 species, mostly restricted to lowland tropics [2]. Of which, *Garcinia* genus includes about 400 species of evergreen trees or shrubs, occurring from West Africa across tropical Asia to the Fiji Island [3], and most of these contain xanthenes [2].

*Garcinia latisissima* Miq. commonly known as Dolomagota (Maluku, Indonesia) and the gland of the plant used as cure wound [4]. *G. latisissima* Miq. is distributed in East Sepik, Eastern Highlands, West Sepik, Southern Highlands, Western Highlands, Madang, Western Morobe, Milne Bay, Central Gulf, Britain, and Papua Islands [5]. In Indonesia, *G. latisissima* Miq. grows in Seram Island, Maluku, and in Papua, but it has been cultivated in the Bogor Botanic Gardens [6]. Constituents of the stem bark ethanol extract of *G. latisissima* Miq. gathered in Papua New Guinea Central Province were latisxanthone-A, latisxanthone-B, latisxanthone-C, and latisxanthone-D [7]. Latisxanthone is classified as pyranoxanthone. The *G. latisissima* Miq. stem bark ethanol extract collected in Papua New Guinea (Central Province) showed good antibacterial activity (inhibition zone was 8-12 mm) against *Staphylococcus aureus* and *Bacillus subtilis* (Gram-positive bacteria)

and moderate activity (inhibition zone was 4-7 mm) against *Escherichia coli* [8]. With this knowledge, the present investigation deals with the phytochemical analysis and antimicrobial efficacy of *G. latisissima* Miq.

### MATERIALS AND METHODS

#### Plant collection and extraction

*G. latisissima* Miq. fruits were collected and identified from the Center for Plant Conservation Bogor Botanical Gardens, Indonesian Institute of Sciences (LIPI), West Java, Indonesia. The sliced fruits were shade-dried at room temperature and powdered coarsely using a mechanical homogenizer. Powdered plant material was extracted by multilevel maceration using various solvents such as n-hexane, ethyl acetate, and methanolic in a row. The filtrate of the extracts was evaporated to dryness under reduced pressure using a rotary evaporator. The extraction yields were collected, weighted, and stored at 4°C before use [1]. The extraction yield can be calculated by

$$\text{extraction yield (\%)} = \frac{\text{dry weight of extract}}{\text{dry weight of plant powder}} \times 100 [9]$$

#### Phytochemical and antimicrobial activities

The qualitatively analyzed phytochemical constituents used in the standard procedures were described by Fransworth and methods from *Materia Medica* Indonesia Volume VI [10-11]. Antimicrobial screening was performed by disc diffusion method [12]. Two Gram-positive bacteria (*B. subtilis* ATCC 6633 and *S. aureus* ATCC 25923), two Gram-negative bacteria (*Pseudomonas aeruginosa* ATCC 27853 and *E. coli* ATCC 25922), and two fungi (*Candida albicans* and *Trichophyton mentagrophytes*). The zone of inhibition against the selected pathogens was determined and recorded. The standard antibiotics used as positive control were gentamycin for *S. aureus*, erythromycin for *B. subtilis*, ciprofloxacin for *P. aeruginosa*, and amoxicillin for *E. coli*. The first step of the zone of

Table 1. Phytochemical screening of *G. lattissima* Miq. fruits

| Tests          | Reagents used                  | n-hexane extracts | Ethyl acetate extracts | Methanolic extracts |
|----------------|--------------------------------|-------------------|------------------------|---------------------|
| Tannins        | Acidic FeCl <sub>3</sub>       | -                 | -                      | +                   |
|                | Gelatin                        | -                 | -                      | +                   |
| Saponins       | Frothing test                  | +                 | -                      | -                   |
| Flavonoids     | HCl+Mg turnings                | -                 | +                      | +                   |
| Anthraquinones | Borntrager's                   | -                 | -                      | -                   |
| Terpenoids     | H <sub>2</sub> SO <sub>4</sub> | -                 | -                      | -                   |
| Alkaloids      | Dragendorff's                  | -                 | +                      | -                   |
|                | Mayer's                        | -                 | -                      | -                   |
|                | Bouchardat's                   | -                 | +                      | -                   |

Phytochemical screening +: Intensity reaction, -: Non-detected, *G. lattissima*: *Garcinia lattissima*

inhibition used crude extracts (100%) [13]. The positive results from that did the area of inhibition with 2% extracts in DMSO. These methods were in triplicate. The assay of minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) performed for extracts gave a zone of inhibition of 2% extracts. To determine the MIC test used the broth dilution method, and to determine the minimum bactericidal assay by plating out onto each appropriate agar plate [14].

## RESULTS AND DISCUSSION

Fruits from *G. lattissima* Miq. for this research are shown in Fig. 1.

The results of extraction yields from *G. lattissima* Miq. fruits are shown in Table 2.

The screening of phytochemical for different extracts of *G. lattissima* Miq. fruits revealed the existence of different chemical compounds of tannins, saponins, flavonoids, and alkaloids, and Table 1 represents the results. Flavonoids and alkaloids (using Dragendorff's and Bouchardat's reagents) were present in ethyl acetate extracts of *G. lattissima* Miq. The methanolic extracts of *G. lattissima* Miq. demonstrated the presence of tannins and flavonoids. The n-hexane excerpts from *G. lattissima* Miq. illustrated the presence of saponins only.

The antimicrobial efficacy from n-hexane, methanolic, and ethyl acetate extracts (100%) of *G. lattissima* Miq. from this research are shown in Table 3. The inhibition zone diameters  $\geq 10$  mm were shown by the *G. lattissima* Miq. fruits' ethyl acetate extracts opposed to *P. aeruginosa* and *B. subtilis* and the methanolic extracts of *G. lattissima* Miq. fruits opposed to *S. aureus*, *P. aeruginosa*, and *B. subtilis*. The n-hexane extracts of *G. lattissima* Miq. fruits against *B. subtilis* and the ethyl acetate extracts of *G. lattissima* Miq. fruits opposed to *E. coli* and *S. aureus* showed inhibition area diameter  $< 10$  mm. Methanolic extracts of *G. lattissima* Miq. fruits against *E. coli*, *C. albicans*, and *T. mentagrophytes* were resistant to all the extracts.

Table 4 shows the results of the antibacterial activities of 2% *G. lattissima* Miq. fruits' extracts in DMSO. The antibacterial activity has been observed only in the *G. lattissima* Miq. fruits' ethyl acetate and methanolic extracts against the selected bacterial assay. The n-hexane extracts were unsuccessful to give the inhibition against the bacterial assay. This research used the positive control with commercially available standard antibiotic disc (erythromycin for *B. subtilis*, gentamycin for *S. aureus*, and ciprofloxacin for *P. aeruginosa*). The conventional medicine showed positive results against all the tested bacteria. The ethyl acetate extracts (2%) of *G. lattissima* Miq. fruits showed the maximum zone of inhibition against *P. aeruginosa* (9.82 $\pm$ 0.978 mm) and then *B. subtilis* (9.62 $\pm$ 0.431 mm). The methanolic extracts (2%) of *G. lattissima* Miq. fruits showed the highest zone of inhibition against *S. aureus* (9.97 $\pm$ 0.448 mm) and then *B. subtilis* (9.53 $\pm$ 0.416 mm) and *P. aeruginosa* (8.22 $\pm$ 0.506 mm).

Table 5 shows the results of dilution assay of extracts' antimicrobial activities. The results show that MIC and MBC of the methanolic

Table 2: The average of extraction yields from the result of multilevel maceration extraction from *G. lattissima* Miq. fruits

| Solvents      | Yield (%) | Average (%)          |
|---------------|-----------|----------------------|
| n-hexane      | 9.010     | 8.9093 $\pm$ 0.4123  |
|               | 9.262     |                      |
|               | 8.456     |                      |
| Ethyl acetate | 2.414     | 2.7640 $\pm$ 0.7217  |
|               | 2.284     |                      |
|               | 3.594     |                      |
| Methanol      | 18.520    | 16.5640 $\pm$ 1.9201 |
|               | 14.682    |                      |
|               | 16.490    |                      |

*G. lattissima*: *Garcinia lattissima*

Table 3: Antibacterial ability of *G. lattissima* Miq. fruits' extracts (100%)

| Organisms                | Zone of inhibition (mm) |               |          |
|--------------------------|-------------------------|---------------|----------|
|                          | Methanolic              | Ethyl acetate | n-hexane |
| <i>B. subtilis</i>       | ++                      | ++            | +        |
| <i>S. aureus</i>         | ++                      | +             | -        |
| <i>P. aeruginosa</i>     | ++                      | ++            | -        |
| <i>E. coli</i>           | +                       | +             | -        |
| <i>C. albicans</i>       | -                       | -             | -        |
| <i>T. mentagrophytes</i> | -                       | -             | -        |

++: Inhibition zone diameter  $\geq 10$  mm, +: Inhibition zone diameter  $< 10$  mm, -: No inhibition zone, *G. lattissima*: *Garcinia lattissima*, *B. subtilis*: *Bacillus subtilis*, *S. aureus*: *Staphylococcus aureus*, *P. aeruginosa*: *Pseudomonas aeruginosa*, *E. coli*: *Escherichia coli*, *C. albicans*: *Candida albicans*, *T. mentagrophytes*: *Trichophyton mentagrophytes*



Fig. 1. Fruits of *Garcinia lattissima* Miq.

**Table 4: Antibacterial activities from 2% *G. latissima* Miq. fruits' extracts in DMSO using agar diffusion method**

| Bacteria             | The mean±SD of diameter of inhibition zone (mm) |               |            |                     |
|----------------------|---|---------------|------------|---------------------|
|                      | n-hexane  | Ethyl acetate | Methanolic | Antibiotic standard |
| <i>B. subtilis</i>   | 0   | 9.62±0.431    | 9.53±0.416 | 21.08±1.928         |
| <i>S. aureus</i>     | 0   | 0             | 9.97±0.448 | 23.70±1.928         |
| <i>P. aeruginosa</i> | 0   | 9.82±0.978    | 8.22±0.506 | 21.88±0.511         |

Antibiotic standard: Erythromycin 15 µg for *B. subtilis*, gentamycin 10 µg for *S. aureus*, ciprofloxacin 5 µg for *P. aeruginosa*,  
*G. latissima*: *Garcinia latissima*, *B. subtilis*: *Bacillus subtilis*,  
*S. aureus*: *Staphylococcus aureus*, *P. aeruginosa*: *Pseudomonas aeruginosa*,  
DMSO: Dimethyl sulfoxide

**Table 5: MBC and MIC from the *G. latissima* Miq. fruits' methanolic and ethyl acetate extracts (in ppm)**

| Solvents      | <i>B. subtilis</i> |      | <i>S. aureus</i> |       | <i>P. aeruginosa</i> |      |
|---------------|--------------------|------|------------------|-------|----------------------|------|
|               | MIC                | MBC  | MIC              | MBC   | MIC                  | MBC  |
| Ethyl acetate | 2500               | 5000 | -                | -     | 5000                 | 5000 |
| Methanolic    | 1250               | 5000 | 5000             | 10000 | 2500                 | 2500 |

*G. latissima*: *Garcinia latissima*, *B. subtilis*: *Bacillus subtilis*,  
*S. aureus*: *Staphylococcus aureus*, *P. aeruginosa*: *Pseudomonas aeruginosa*,  
MBC: Minimum bactericidal concentration, MIC: Minimum inhibitory concentration

extracts and the ethyl acetate extracts opposed to *P. aeruginosa*, *B. subtilis*, and *S. aureus* were different. Ethyl acetate extracts and methanolic extracts of *G. latissima* Miq. fruits were active against *B. subtilis* (MIC=2,500 ppm and 1,250 ppm, respectively) and against *P. aeruginosa* (MIC=5,000 ppm and 2,500 ppm, respectively). The methanolic extracts of *G. latissima* Miq. fruits were active against *S. aureus* (MIC=5,000 ppm). The ethyl acetate extracts of *G. latissima* Miq. fruits were not tested for Gram-positive bacteria *S. aureus* because they did not give inhibition zone in 2% *G. latissima* Miq. fruits' extracts assay. The methanolic extracts have more powerful activity of antimicrobials as contrast to ethyl acetate extracts. The results of the study indicate that *G. latissima* Miq. fruits' extracts could be useful for antibacterial uses [15].

## CONCLUSION

This study indicated that the methanolic extracts of *G. latissima* Miq. fruits had the highest rendement than the others. The methanolic extracts consist of tannins and flavonoids, qualitatively. The ethyl acetate extracts include flavonoids and alkaloids, and the hexane

extracts consist of saponins only. The antimicrobial activity showed that the methanolic extracts of *G. latissima* Miq. fruits had the best result than the others.

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## REFERENCES

- Johnson M, Kalaiarasi V, Sivaraman A, Janakiraman N, Babu A, Narayani M. Phytochemical and antibacterial studies on *Aristolochia tagala* Cham. World J Pharm Res 2014;3(2):2172-8.
- Stevens PF. The families and genera of fascicular plants. In: Flowering Plants. Vol. 9. Berlin, Heidelberg: Springer-Verlag; 2007. p. 48-66.
- Ramage CM, Sando L, Peace CP, Carroll BJ, Drew RA. Genetic diversity revealed in the apomictic fruit species *Garcinia mangostana* L. (Mangosteen). Euphytica 2004;136(1):1-10.
- Eisai P. Medicinal Herb Index in Indonesia. 2<sup>nd</sup> ed. Indonesia: P.T. Eisai; 1995. p. 71.
- Conn B, Damas K. PNGTreesKey-*Garcinia latissima* Miq. In: Annales Musei Botanici Lugduno-Batavi. Vol. 1. New South Wales, Papua New Guinea: National Herbarium; 1863.
- Sari R, Ruspandi, Ariati SR. An Alphabetical List of Plant Species Cultivated in the Bogor Botanic Gardens. Bogor: LIPI; 2010.
- Ito C, Miyamoto Y, Nakayama M, Kawai Y, Rao KS, Furukawa H. A novel depsidone and some new xanthenes from *Garcinia* species. Chem Pharm Bull 1997;45(9):1403.
- Rao KS. Antibacterial activity of some medicinal plants of Papua New Guinea. Int J Pharmacogn 1996;34(3):223-5.
- Yin NG, Abdullah S, Phin CK. Phytochemical constituents from leaves of *Elaeis guineensis* and their antioxidant and antimicrobial activities. Int J Pharm Pharm Sci 2013;5(4):137-40.
- Fransworth NR. Biological and phytochemical screening of plants. J Pharm Sci 1995;22(3):226-76.
- Depkes RI. Materia Medika Indonesia. Vol. VI. Jakarta: Departemen Kesehatan Republik Indonesia; 1995.
- Bamas SS, Kingsley SJ, Sankaranarayanan S. Antibacterial activity of different phytochemical extracts from the leaves of *T. Procumbens*. Linn.: Identification and Mode of action of the terpenoid compound as antibacterial. Int J Pharm Pharm Sci 2012;4:557-64.
- Mathew S. An evaluation of the antimicrobial activity of various concentrations of *Ocimum sanctum* against various species of bacteria: An *in vitro* study. Int J Adv Appl Sci 2014;3(1):33-6.
- Jeong MR, Kim HY, Cha JD. Antimicrobial activity of methanol extract from *Ficus carica* Leaves against oral bacteria. J Bacteriol Virol 2009;39(2):97-102.
- Sanches NR, Cortez DA, Schiavini MS, Nakamura CV, Dias-Filho BP. An evaluation of antibacterial activities of *Psidium guajava* (L.). Braz Arch Biol Technol 2005;48(3):429-36.