
ISSN 2622-7258

DOI: 10.31014/aior.1994.06.04.291

The online version of this article can be found at: [https://www.asianinstituteofresearch.org/](https://www.asianinstituteofresearch.org/)

Published by:
The Asian Institute of Research

The *Journal of Health and Medical Sciences* is an Open Access publication. It may be read, copied, and distributed free of charge according to the conditions of the Creative Commons Attribution 4.0 International license.

The Asian Institute of Research *Journal of Health and Medical Sciences* is a peer-reviewed International Journal. The journal covers scholarly articles in the fields of Medicine and Public Health, including medicine, surgery, ophthalmology, gynecology and obstetrics, psychiatry, anesthesia, pediatrics, orthopedics, microbiology, pathology and laboratory medicine, medical education, research methodology, forensic medicine, medical ethics, community medicine, public health, community health, behavioral health, health policy, health service, health education, health economics, medical ethics, health protection, environmental health, and equity in health. As the journal is Open Access, it ensures high visibility and the increase of citations for all research articles published. The *Journal of Health and Medical Sciences* aims to facilitate scholarly work on recent theoretical and practical aspects of Health and Medical Sciences.
Content and Antimicrobial Potential of Cambodia Flower (Plumeria sp.) Extract

I Nyoman Jirna¹, I Gede Sudarmanto², I Nyoman Gede Suyasa³, Ida Ayu Made Sri Arjani⁴, Cok Dewi Widhya Hana Sundari⁵, I Nyoman Purna⁶, Nyoman Mastra⁷

¹,²,³,⁴,⁵,⁶,⁷ Medical Laboratory Technology, Poltekkes Kemenkes Denpasar, Denpasar, Indonesia

Correspondence: I Nyoman Jirna, Medical Laboratory Technology, Poltekkes Kemenkes Denpasar, Denpasar, Indonesia. E-mail: nyomanjirna@ymail.com

Abstract
The case of infectious disease is caused by Streptococcus aureus bacteria. The handling of the disease is performed by antibiotics. However, antibiotics can cause drug resistance to the bacteria. Natural ingredients with antimicrobial plant medicine have potential for the safe, and more effective disease handling. Cambodia flowers (Frangipani P. alba) are expected to contain a variety of chemical compounds with antimicrobial potential. This research may also contribute to the development of new drugs or natural supplements for infection control. The aim of this study is to identify quantitatively antibacterial phytochemical contents and qualitatively test the antimicrobial potential of Cambodia flower extract. The research was conducted using a true-experimental design of a posttest-only-control design with intervention in the treatment group and control for external factors that may potentially affect the research. The data were analyzed using the Kruskal-Wallis test. The results show that Cambodia flower extract contains phenols, tannins, flavonoids, and saponins, and the extract contains 5.95% phenols, 4.85% tannins, and 3.51% flavonoids. There is a difference in antimicrobial potency at various concentrations (55%, 70%, 85%, and 100%) of Cambodia flower extract in inhibiting the growth of S. aureus with a significance value of 0.000. In the in-vitro test, antimicrobial potential is found in the resistant group (with inhibition zone diameter ≤15 mm) in accordance with NCCLS standards. In conclusion, Cambodia flower is expected to provide a beneficial alternative solution for the community in preventing skin infections caused by S. aureus bacteria.

Keywords: Antimicrobial, Cambodia, Phytochemical, Plumeria, Staphylococcus Aureus

1. Introduction

Infectious disease is the second most dangerous cause of human death in the world. Staphylococcus aureus is known as the cause of various clinical disease infections. It normally does not cause infection on healthy skin. Nevertheless, once it enters the internal tissues or bloodstream, the potential serious infection is unavoidable. This bacterium can also cause scale skin syndrome in young children and is highly contagious. S. aureus can cause bacteremia and infective endocarditis, osteoarticular, pleuropulmonary, and device-related infections. It is known
that age is the powerful determinant of *S. Aureus* bacteremia (SAB) in which the occurrence of SAB increases gradually associated with age. Besides, the male gender is associated with SAB incidence with a ratio of ~1.5 compared to females (Laupland et al., 2013; S. Y. C. Tong et al., 2015). Antibiotics have been used against *S. aureus* such as linezolid (100%), trimethoprim-sulfamethoxazole (95%), and tetracyclines (94%). Besides, cephalosporins were agreed appropriate choice for *S. aureus* infections. However, the use of antibiotics and the evolution of bacteria has caused the drug resistance of *S. aureus* to increase gradually (Guo et al., 2020). The development of alternative solutions to prevent *S. aureus* growth is inevitable, and medicine of an antimicrobial nature is needed (Theos et al., 2019).

The use of Indonesian medicinal plants can be used as an alternative to overcome the resistance of microorganisms to antibiotics, considering that Indonesia has various types of plants that have the potential to be used as medicinal ingredients. One of which is Plumeria. Plumeria generally known in Indonesia as frangipani or Cambodia flower, is an ornamental plant popular in Indonesia because of the beauty and distinctive aroma of its flowers. Taxonomically, frangipani belongs to the *Apocynaceae* family which originates from Central America and the Caribbean and has been introduced to Southeast Asia, including Indonesia, especially in Bali where it is used as an ornamental plant (Ayu Fatkhurrohmah Wiyono & Muhtadi, 2020; Gde Sri Adyani Suari et al., 2020). Apart from being an ornamental plant, frangipani is also known to be used as a traditional medicine to treat several diseases, especially the type of *Frangipani P. alba* (Sura et al., 2018). Various bioactive constituents with strong antimicrobial, anti-inflammatory, anthelmintic, antipyretic, and antirheumatic properties have been extracted from the bark, leaves and flowers of *P. alba* (Kaur et al., 2022). The stems and leaves of *P. alba* have been reported to be used to treat skin diseases (R et al., 2014).

One interesting aspect to study is the antimicrobial potential of frangipani flower extract. The ability to inhibit the growth or kill pathogenic microorganisms such as bacteria and fungi is a highly desirable property in the health and medical fields, especially in the context of infection control. In addition, with the increasing resistance of microorganisms to conventional antibiotics (Suganya et al., 2022). Research related to the antimicrobial potential of natural medicinal plants is becoming increasingly important (El-Saadony et al., 2023; Mancuso et al., 2021). By understanding the chemical content of frangipani flower extract and testing its antimicrobial activity, the potential for developing new drugs or herbal supplements to treat infections can be understood. Herbal supplements have been proposed as a potential treatment option for infectious diseases (Mancuso et al., 2021). This study aimed at determining the antibacterial phytochemical content qualitatively and quantitatively as well as determining the antimicrobial potential of frangipani flower extracts in vitro. It is expected that the results of this research can provide an important contribution to the development of natural resources that have the potential new, safer, and more effective antimicrobial agents.

2. Method

This study was carried out by making frangipani flower extract and qualitative testing of phytochemical content. The testing was carried out in the Applied Chemistry Laboratory, Department of Medical Laboratory Technology, Poltekkes Kemenkes Denpasar. Quantitative testing of the phytochemical content of Cambodia flower extract was carried out at the Integrated Analytical Laboratory, Udayana University. The antimicrobial potential test of frangipani flower extract was carried out at the Microbiology Laboratory, Faculty of Medicine, Warmadewa University.

The sample of Cambodia flowers was collected from various areas in 8 districts and 1 city in Bali. The process of extracting Cambodia flowers started from performing air-dried the flowers until 5 kg were obtained as shown in Figure 1. A total of 5 kg of dried Cambodia flowers were oven-dried until dry. This sample was then extracted using the maceration method with a TLC spectrophotodensitometer. The anti-bacterial testing was carried out using a true experimental with a posttest-only-control design. The treatment groups were designed with Cambodia flower extract concentrations of 55%, 70%, 85%, and 100%, with repetition carried out 3 times with 4 multiplications for a total of 48 samples. The positive control used the antibiotic Ciprofloxacin 30 g and the negative control was a sterile 96% ethanol solution. The diffusion (disc) method was used to test anti-microbial potential in vitro using *S. aureus* bacteria grown on Mueller Hinton agar media. The research data were analyzed.
using the Kolmogorov-Smirnov test followed by the Kruskal Wallis test to determine differences in antimicrobial potential for the growth of \textit{S. aureus} with a confidence level of 95\% (α = 0.05) followed by the Least Significant Difference (LSD) test.

3. Results

3.1 Cambodia Flower Water Content

The process of making the test substance was started by preparing a 5 kg Cambodia flower. After being baked, the water content was weighed again to determine the quality of the flowers produced. The results of water content measurements were obtained at 10 \%.

3.2 Content of Phytochemical Compounds in Cambodia Flower Extract

The results of qualitative tests with phytochemical screening on the antibacterial compound content of Cambodia flower extract showed four phytochemical compounds found i.e. phenols, tannins, flavonoids, and saponins as shown in Table 1.

Table 1: The Phytochemical Content of Cambodia Flower Extract

<table>
<thead>
<tr>
<th>No.</th>
<th>Test</th>
<th>Start</th>
<th>End</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Phenol</td>
<td>Clear green</td>
<td>Blue</td>
<td>Positive (+)</td>
</tr>
<tr>
<td>2</td>
<td>Tannins</td>
<td>Clear brown</td>
<td>Blue</td>
<td>Positive (+)</td>
</tr>
<tr>
<td>3</td>
<td>Flavonoids</td>
<td>Clear yellow</td>
<td>Orange-red</td>
<td>Positive (+)</td>
</tr>
<tr>
<td>4</td>
<td>Saponins</td>
<td>Clear green</td>
<td>Light green cloudy, foamy</td>
<td>Positive (+)</td>
</tr>
</tbody>
</table>

Meanwhile, the results of quantitative tests on flavonoid, tannin, and phenol compounds showed varying results from 3.51\% to 5.95\% as shown in Table 2.

Table 2: Quantitative Phytochemical Cambodia Flower Extract

<table>
<thead>
<tr>
<th>No.</th>
<th>Parameter</th>
<th>Method</th>
<th>Result (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Flavonoids</td>
<td>Spectrophotometry</td>
<td>3.51</td>
</tr>
<tr>
<td>2</td>
<td>Tannins</td>
<td>Spectrophotometry</td>
<td>4.85</td>
</tr>
<tr>
<td>3</td>
<td>Phenol</td>
<td>Spectrophotometry</td>
<td>5.95</td>
</tr>
</tbody>
</table>
3.3 Bacterial Inhibition Test

The positive control, discs soaked in the antibiotic Ciprofloxacin 30 g, showed the presence of an inhibition zone starting from repetition groups I to III. The positive control inhibition zone diameter values obtained varied in each repetition group, ranging from 16.250 mm to 16.375 mm as shown in Table 3. Meanwhile, the negative control, discs soaked in 96% ethanol, did not produce an inhibition zone.

Table 3: Inhibition Zone of S. aureus in Positive and Negative Control

<table>
<thead>
<tr>
<th>Repetition</th>
<th>Inhibition Zone Diameter (mm)</th>
<th>Positive Control</th>
<th>Negative Control</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>16.25</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>II</td>
<td>16.375</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>III</td>
<td>16.25</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Average</td>
<td>16.29</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

The growth inhibition zone of S. aureus at various concentrations of Cambodia flower extract (55%, 70%, 85%, and 100%) in replication groups I to III from replications I to IV showed the existence of an inhibition zone that varied from 7.75 mm up to 9.5 mm as shown in Table 4. Compared with the diameter of the inhibition zone for the antibiotic Ciprofloxacin in the National Committee for Clinical Laboratory Standards (NCCLS) table, it is in the resistant category (≤15mm).

Table 4: Inhibition Zone for The Growth of S. aureus at Various Concentrations

<table>
<thead>
<tr>
<th>Extract Concentration</th>
<th>Replication I</th>
<th>Replication II</th>
<th>Replication III</th>
<th>Replication IV</th>
</tr>
</thead>
<tbody>
<tr>
<td>55%</td>
<td>8.5</td>
<td>8.5</td>
<td>7.75</td>
<td>8.375</td>
</tr>
<tr>
<td>70%</td>
<td>8.75</td>
<td>8.625</td>
<td>8.25</td>
<td>8.625</td>
</tr>
<tr>
<td>100%</td>
<td>9.5</td>
<td>9.5</td>
<td>9.375</td>
<td>9.375</td>
</tr>
</tbody>
</table>

3.4 Statistical Analysis

The Kolmogorov Smirnov statistics applied to the data shows that the Asymp sig value obtained is 0.000 < 0.05, which means the data is not normally distributed. Hence, the Kruskal Wallis test was carried out at a confidence level of 95% (\(\alpha = 0.05\)). The results of the Kruskal Wallis test obtained a significance value of (0.000). The value obtained is less than the \(\alpha\) value (0.05), indicating that there is a difference in the antimicrobial potential of
Cambodia flower extract in inhibiting the growth of *S. aureus*. The LSD test results showed that there were differences in antimicrobial potential at various concentrations (55%, 70%, 85% and 100%) of Cambodia flower extract. This can be seen from the significance value of each concentration with control, namely 0.00 < 0.05.

4. Discussion

The Cambodia flower extract in this study had a water content of 10%. Other studies that dried plant leaves reported that the water content of the extracted leaf powder is in the range of 3 to 5% (Wulandari et al., 2020). Based on this, the water content of Cambodia flower extract in this study is still quite high compared to the research results of Wulandari et al. (2020). Lower water content can maintain the stability of the extract powder with a longer shelf life. This is due to extract powder with low water content will prevent microbial growth, minimize oxidation, maintain physical properties, and prevent unwanted chemical reactions.

The results of qualitative tests on several parameters show that there are phenolic, tannin, flavonoid, and saponin compounds in Cambodia flower extract. Quantitatively, Cambodia flower extract shows a phenolic compound content of 5.95% (595 mg/100g), flavonoids 3.51% (351 mg/100g), and tannins 485% (485 mg/100g). Research by Fathoni et al. (2019) also found phenol, tannin, flavonoid, and saponin compounds in yellow Cambodia flowers (*Plumeria alba*), as well as alkaloids, terpenoids, steroids, hydroquinone, and polyphenols (Fathoni et al., 2019). Flavonoids are a secondary metabolite whose production is influenced by the photosynthesis process (Dave Mehta et al., 2023; Kumar et al., 2018; Mipeshwaree Devi et al., 2023). Flavonoids are widely distributed in plants and are responsible for the pigments that color most flowers, fruits, and seeds (Falcone Ferreyra et al., 2012). Flavonoids are natural compounds from the phenolic group and are valuable to human beings for their contribution to impart color and physiologically active members (Mutha et al., 2021).

Flavonoids are reported to have good antioxidant activity and estrogenic, antiviral, antibacterial and anticancer activity (Panche et al., 2016; Ullah et al., 2020). The benefits of flavonoids in the human body are as antioxidants so they are very good for preventing cancer, protecting cell structure, increasing the effectiveness of vitamin C, anti-inflammatory, preventing bone loss, and as antibiotics. In most cases, flavonoids can act directly as antibiotics by interfering with the function of organisms such as bacteria or viruses. The benefits of flavonoids are known to function as phytoalexin, namely as an antimicrobial for bacteria and fungi, thus helping to inhibit the spread of pathogens in the plant body.

Apart from flavonoids, tannin compounds in plants have also been used as medicine. Tannins are a class of polyphenolic compounds that are widely distributed in plant tissues and have various medical and pharmacological applications (Pizzi, 2021). Tannins are astringents, and polyphenols, have a bitter taste, can bind and precipitate proteins, and are soluble in water (especially hot water). Generally, tannin is used to treat skin diseases and as an antibacterial, but tannin is also widely used to treat diarrhea, hemostasis (stops bleeding), and hemorrhoids (Marciaćzyk et al., 2022; Sun et al., 2022). Tannins have been proven to have anti-cancer properties and can inhibit the growth of cancer cells (Pizzi, 2021), antioxidants that help protect cells from oxidative damage (Z. Tong et al., 2022), antimicrobials that inhibit the growth of fungi and bacteria, and antidiabetic properties and helps regulate blood sugar levels (Pizzi, 2021).

With respect to the results of research in the laboratory, the concentration of Cambodia flower extract shows that it has in-vitro antimicrobial potential with a category of resistance to the growth of *S. aureus* compared to the NCCLS table. The diameter of the inhibitory zone formed at various concentrations of Cambodia flower extract tested was classified into the resistant category (≤15mm) with a growth inhibition power of 7.75 mm to 9.5 mm. This shows that the ability to inhibit bacterial growth is not optimal for *S. aureus* bacteria sensitively in the minimum range of 21 mm. Something slightly different was found in the research of (Rupiniasih et al., 2019) who obtained a wider diameter of the inhibition zone of *S. aureus* using Cambodia/frangipani (*P. alba*) flower extract, namely 20.89 mm (versus 7.75–9.5 mm in this study). The size of the inhibition zone formed by an extract determines its antibacterial strength, which is categorized as very strong, strong, medium, or weak. The extract is considered to have a very strong inhibitory effect if the diameter of the resulting inhibition zone is more than 20 mm. If the diameter is between 10 mm and 20 mm, it is classified as having a strong inhibitory effect. A moderate
inhibitory effect was observed when the inhibitory potential of the extract ranged from 5 mm to 10 mm. An extract is considered weak in its inhibitory strength if the diameter of the inhibition zone produced is less than 5 mm (Sinaga & Jaya, 2022). Based on the classification of antibacterial strength, the Cambodia flower extract in this study showed a moderate inhibitory effect on *S. aureus* bacteria. The anti-microbial potential of Cambodia flower extract is not optimal in inhibiting the growth of *S. aureus* bacteria, which is thought to be caused by the antibacterial phytochemical content in the form of phenol (5.95%), tannin (4.85%) and flavonoids (3.51%) which is still relatively low.

**Author Contributions:**
Conceptualization, INJ.; Methodology, INJ.; Software, IGS.; Validation, IAMSA.; Formal Analysis, IAMSA, CDWHS, INP.; Investigation, INP.; Resources, INP, NM.; Data Curation, INJ.; Writing – Original Draft Preparation, INGS.; Writing – Review & Editing, INGS.; Visualization, INGS.; Supervision, INJ.; Project Administration, NM.; Funding Acquisition, INJ.

**Funding:** This research was funded by Poltekkes Kemenkes Denpasar through the Simlitabkes for the research year 2023 and the APC was funded by Poltekkes Kemenkes Denpasar.

**Conflicts of Interest:** The authors declare no conflict of interest.

**Informed Consent Statement/Ethics approval:** All subjects gave their informed consent for inclusion before they participated in the study. The study was conducted in accordance with the Declaration of Helsinki, and the protocol was approved by the Ethics Committee of Poltekkes Kemenkes Denpasar for the research Simlitabkes 2023.

**References**


Asian Institute of Research. Journal of Health and Medical Sciences. Vol.6, No.4, 2023