



EXTRACTION AND PHARMACOLOGICAL EVALUATION OF HIBISCUS AS AN ANTIMICROBIAL AGENT

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ABSTRACT

The rapid emergence of antimicrobial resistance has created an urgent demand for novel and effective therapeutic agents derived from natural sources. Medicinal plants continue to serve as an important source of bioactive compounds with diverse pharmacological activities. The present study focuses on the extraction and evaluation of Hibiscus rosa-sinensis for its antimicrobial potential. Plant materials, including leaves and flowers, were dried, powdered, and extracted using suitable solvents such as ethanol, methanol, and water through standard extraction techniques. The obtained extracts were subjected to preliminary phytochemical screening to determine the presence of secondary metabolites including flavonoids, tannins, alkaloids, saponins, and phenolic compounds. The antimicrobial activity of the extracts was assessed against selected pathogenic microorganisms using the agar well diffusion method. The results demonstrated notable inhibitory

effects, with organic solvent extracts exhibiting greater activity compared to aqueous extracts. The antimicrobial action is likely associated with the presence of phenolic and flavonoid constituents, which are known to interfere with microbial cell wall integrity and metabolic processes. The study highlights the potential of Hibiscus rosa-sinensis as a natural source of antimicrobial agents. Further investigations involving isolation of active compounds, mechanism of action studies, toxicity evaluation, and in vivo experiments are necessary to establish its clinical relevance and pharmaceutical application.

KEYWORDS: Hibiscus rosa-sinensis, medicinal plant, antimicrobial resistance, phytochemical screening, plant extract, natural therapeutics.

1. INTRODUCTION

1.1 Importance of Medicinal Plants

Medicinal plants have been an essential part of human healthcare since ancient times. Long before the development of modern pharmaceuticals, people relied on plants to treat illnesses and maintain health. Even today, many communities around the world depend on traditional medicine systems that use plant-based remedies. Plants are considered valuable because they contain natural chemical substances that can produce therapeutic effects in the human body.



Figure 1: Plant of hibiscus.

In recent years, scientific interest in medicinal plants has increased significantly. Researchers are studying plants to discover new drugs and safer alternatives to synthetic medicines. Many modern medicines are either directly derived from plants or developed based on plant compounds. Natural products from plants are often preferred because they are easily available, relatively affordable, and generally associated with fewer side effects. Therefore, studying medicinal plants plays an important role in the development of new treatments for various diseases.^[1]

1.2 Introduction to Hibiscus

When it comes to medicinal plants, Hibiscus rosa-sinensis stands out as a significant species thanks to its wide range of therapeutic benefits. You might know it better as China rose, shoe flower, or Chinese hibiscus. This lovely plant is part of the Malvaceae family and thrives in tropical and subtropical areas around the world.

The Hibiscus genus boasts a wide variety of species that can be found as herbs, shrubs, or even small trees. These plants are celebrated for their vibrant and eye-catching flowers. Beyond their beauty, many Hibiscus species have a place in traditional medicine. They contain a range of biologically active compounds that can help prevent and treat various health issues. Hibiscus rosa-sinensis, often found in gardens, parks, and homes, is cherished for its stunning flowers and its knack for thriving in warm climates. Beyond its beauty, this plant has caught the eye of researchers due to its impressive pharmacological and medicinal benefits.^[2]

1.3 Botanical Description of the Plant

Hibiscus rosa-sinensis is an evergreen shrub that usually grows between 2 to 5 meters in height. The plant has a strong root system that helps it absorb nutrients and water from the soil.

Young stems are generally green and smooth, while older stems develop a slightly rough texture. The leaves of the plant are simple and arranged alternately along the stem. They are usually ovate in shape with serrated edges and a glossy surface. The green leaves play an important role in photosynthesis and also contain medicinal compounds that are useful in traditional treatments.^[3]

The flowers of Hibiscus rosa-sinensis are large, colorful, and attractive. Although red flowers are the most common, the plant may also produce flowers in pink, yellow, orange, or white shades depending on the variety. The flower contains both male and female reproductive structures, which means it is a complete flower. The central structure of the flower consists of numerous stamens and a long style ending with a stigma. These structures are responsible for pollination and reproduction of the plant.

1.4 Traditional Uses of Hibiscus

Hibiscus rosa-sinensis has been used in traditional medicine for many years in different parts of the world. Various parts of the plant such as flowers, leaves, roots, and stems are used for medicinal purposes. In many traditional healing systems, the plant is believed to possess cooling, soothing, and healing properties.

In Ayurveda, hibiscus is used for promoting hair growth, improving scalp health, and treating menstrual disorders. The flowers are often used in herbal preparations to support skin and

hair care. In Unani medicine, the plant is used to treat respiratory problems such as cough and bronchitis, and it is also considered beneficial for heart health.

In many local communities, hibiscus leaves and flowers are used as home remedies. For example, crushed leaves are sometimes applied to wounds and skin infections, while flower extracts are used as natural hair conditioners. In some cultures, hibiscus drinks or teas are consumed to help manage fever, blood pressure, and digestive problems.^[4]

In the Caribbean, *H. sabdariffa* can be a staple at merry events, particularly known as “Tawny” at Christmas celebrations. This drink, often flavored with ginger and cloves, reflects the social combination of tastes and serves as an image of the neighborhood. The plant is also valued for its high vitamin content including vitamins A, C, and E, which are present in varying but significant amounts. In Middle Eastern countries, Hibiscus tea is known for its sour taste and beneficial effects. It is often served chilled in hot climates as an invigorating refreshment and is often associated with other therapeutic uses including gastric support and antioxidant protection.

These traditional uses suggest that the plant contains important bioactive substances that may provide therapeutic benefits.

Common species of hibiscus

Although there are many notable species in the *Hibiscus* genus, four are particularly noteworthy because of their importance in medicine, culture, and nutrition. *H. sabdariffa* is possibly the most studied species; it is often recognized by its bright red flowers and calyxes. A tart, refreshing tea brewed from it is enjoyed by people from many different cultures. Known as “Bissap” in West Africa and “Sorrel” in the Caribbean, this fruit is valued both for its delicious taste and its high vitamin and mineral content.^[5]

H. rosa-sinensis is often planted as an ornamental plant and is also known as Chinese Hibiscus or shoe flower. Due to its antibacterial and anti-inflammatory properties, it is used in conventional medicine. Studies show that it is good for treating scalp problems and promoting hair development.^[6]

H. acetosella, also known as red sorrel, is valued for its adaptability in the kitchen. Due to their spicy taste, their leaves are often used in salads and drinks, especially in African cuisine.

Additionally, the plant has several health benefits, including acting as an antioxidant and supporting digestive health.^[7]

H. mutabilis, often called the “Confederate rose,” is known for its large, colorful flowers that change from white to pink as they mature. Its flowers are used for mild medicinal purposes in some cultures, but they are primarily grown for their aesthetic value.^[8]

1.5 Phytochemical Constituents of Hibiscus

The medicinal value of *Hibiscus rosa-sinensis* is mainly due to the presence of various phytochemicals. Phytochemicals are natural chemical compounds produced by plants that help them protect against environmental stress and diseases. These compounds can also have beneficial effects on human health.

Some of the major phytochemicals found in *Hibiscus* include flavonoids, tannins, phenolic compounds, alkaloids, saponins, and anthocyanins. Flavonoids and phenolic compounds are well known for their antioxidant activity, which helps protect the body from damage caused by free radicals. Tannins have antimicrobial and astringent properties, making them useful in treating infections and wounds.

Alkaloids and saponins are also important plant compounds that contribute to various pharmacological activities such as anti-inflammatory and antimicrobial effects. Because of these compounds, *Hibiscus rosa-sinensis* is considered a valuable plant for pharmaceutical research.^[9]

1.6 Antimicrobial Potential of Hibiscus

Microbial infections caused by bacteria and fungi remain a major health concern worldwide. In recent years, many microorganisms have developed resistance to commonly used antibiotics. This situation has increased the need to explore new antimicrobial agents, especially from natural sources such as medicinal plants.

Hibiscus rosa-sinensis has shown promising antimicrobial properties in various scientific studies. Extracts prepared from the leaves, flowers, and roots of the plant have been reported to inhibit the growth of different microorganisms. These include both Gram-positive and Gram-negative bacteria as well as certain fungal species.

The antimicrobial effect of the plant is believed to be related to the presence of flavonoids, phenolic compounds, and tannins. These compounds can interfere with microbial cell membranes, inhibit enzyme activity, and prevent the growth and multiplication of microorganisms.^[10]

1.7 Need for the Study

The increasing resistance of pathogens to existing antibiotics has become a serious global challenge. As a result, researchers are exploring natural sources to identify new antimicrobial compounds. Medicinal plants offer a promising alternative because they contain a variety of bioactive substances that may act against harmful microorganisms.

Studying *Hibiscus rosa-sinensis* is important because it is widely available, traditionally used, and contains several biologically active compounds. Scientific evaluation of this plant can help confirm its medicinal properties and support its use in modern healthcare.

1.8 Aim of the Study

The main aim of the present study is to investigate the extraction and pharmacological evaluation of *Hibiscus* as an antimicrobial agent. This study focuses on identifying the phytochemical constituents present in the plant and examining its effectiveness against different microorganisms.

The findings of this research may help in understanding the medicinal importance of *Hibiscus rosa-sinensis* and may contribute to the development of natural antimicrobial drugs in the future.^[11]

2. MATERIALS AND METHODS

2.1 Study Area and Duration

The current study focused on extracting and evaluating the pharmacological properties of *Hibiscus* as an antimicrobial agent, all conducted within the lab facilities of the Department of Pharmacology. This research took place over a set period, during which we collected the plants, carried out the extraction, performed phytochemical analyses, and conducted antimicrobial tests. Every experiment was executed under controlled lab conditions, adhering to standard procedures.

2.2 Collection of Plant Material

We gathered fresh plant material from *Hibiscus rosa-sinensis* at a local garden and nearby areas where this plant is often grown. We carefully chose healthy leaves and flowers to ensure we had high-quality samples for our analysis.

The plant material we collected was thoroughly inspected to get rid of any damaged or infected sections. Once we had everything, we carefully packed the plant material in clean polyethylene bags and transported it to the lab for further processing. We identified and authenticated the plant by looking at its morphological traits, including the structure of the flowers, the arrangement of the leaves, and the characteristics of the stem.^[12]

2.3 Preparation of Plant Material

The plant materials we collected, including leaves and flowers, were first given a good wash with distilled water to get rid of any dust, soil, and other unwanted bits. Once they were clean, we laid them out on tidy trays and let them dry in the shade at room temperature for a few days.

To keep those valuable active compounds intact, shade drying was the way to go instead of letting them get zapped by direct sunlight. The drying process carried on until the plant material was completely dry and had that brittle texture.

After the drying process, we ground the plant materials into a fine powder using a mechanical grinder. To ensure a consistent particle size, the powdered material was sifted through a standard sieve. We then stored the powder in airtight containers, keeping it in a cool, dry place until it was ready for use.

2.4 Chemicals and Reagents Used

In the process of extraction and phytochemical analysis, a variety of chemicals and reagents were utilized. All the chemicals employed in this study were of analytical grade. Some of the most commonly used chemicals include

- Ethanol
- Methanol
- Distilled water
- Hydrochloric acid
- Ferric chloride

- Mayer's reagent
- Wagner's reagent
- Dragendroff's reagent
- Sodium hydroxide
- Sulfuric acid
- Nutrient agar medium

These are all utilized in various extraction processes, phytochemical screenings, and tests for antimicrobial activity.

2.5 Preparation of Plant Extract

Extraction plays a crucial role in separating bioactive compounds from plant materials. In this study, we focused on extracting phytochemicals from Hibiscus powder using various solvent extraction methods.

2.5.1 Maceration Method

In the maceration method, a measured quantity of dried plant powder was placed in a clean conical flask. A suitable solvent such as ethanol or methanol was added to the flask in a specific ratio. The mixture was then sealed and kept at room temperature for several days to allow the solvent to dissolve the active compounds present in the plant material.

During the maceration process, the mixture was shaken occasionally to improve the extraction efficiency. After the extraction period, the mixture was filtered using filter paper to separate the liquid extract from the plant residue.

The liquid that was filtered out was concentrated with a rotary evaporator, keeping the temperature under control. Once concentrated, the extract was carefully collected and stored in airtight containers for future analysis.^[13]

2.5.2 Soxhlet Extraction Method

Soxhlet extraction is an amazing technique for efficiently pulling out plant extracts. In this method, you start by placing dried plant powder into a thimble, which is then set up in the Soxhlet apparatus. A suitable solvent, such as ethanol, is used to carry out the extraction.

The solvent was heated until it hit its boiling point, which allowed the vapor to travel through the apparatus.

Once there, it condensed and mingled with the plant material. This process helped the solvent pull out the bioactive compounds from the plant powder before it flowed back into the flask. This cycle continued until the extraction was completely finished.

After we extracted the solvent, we allowed it to evaporate, which gave us the crude extract. We then stored this extract in the refrigerator, keeping it ready for any upcoming experiments.

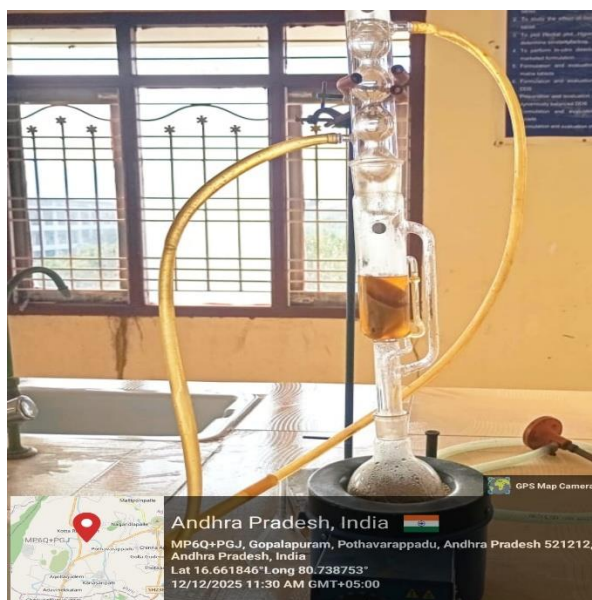


Figure 2: Soxhlet Apparatus.

2.6 Phytochemical Screening

We conducted a preliminary phytochemical screening to pinpoint the key bioactive compounds present in the plant extract. A series of qualitative tests were carried out following standard procedures.

2.6.1 Test for Alkaloids

A small amount of plant extract was mixed with a few drops of Mayer's reagent or Dragendroff's reagent. If a precipitate forms, it suggests that alkaloids are present in the extract.

2.6.2 Test for Flavonoids

The extract was treated with a sodium hydroxide solution, and then we added some dilute acid. When a yellow color appeared and then faded after the acid was added, it confirmed that flavonoids were present.

2.6.3 Test for Tannins

A tiny bit of extract was combined with a ferric chloride solution. If a dark green or blue-black color appears, it means tannins are present.

2.6.4 Test for Saponins

The extract was shaken up really well with distilled water in a test tube. When stable foam started to form, it showed that saponins were present.

2.6.5 Test for Phenolic Compounds

The sample was treated with a ferric chloride solution, and the emergence of a bluish-green color indicated the presence of phenolic compounds.

These tests were instrumental in pinpointing the phytochemical components found in Hibiscus extract.

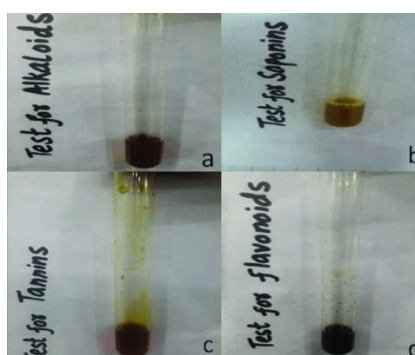


Figure 3: Phytochemical test.

2.7 Preparation of Microbial Culture

For our antimicrobial testing, we sourced specific bacterial strains from a microbiology lab. We cultured these microorganisms using nutrient agar medium, all while maintaining sterile conditions in the lab. To prepare the culture media, we dissolved nutrient agar powder in distilled water and then sterilized it in an autoclave. Once sterilized, we poured the medium into sterile Petri plates and let it solidify.

The bacterial strains were then introduced into the culture media and kept at the right temperature to promote growth.^[14]

2.8 Antimicrobial Activity Test

We looked into the antimicrobial properties of Hibiscus extract using the agar well diffusion method.

PROCEDURE

First, we prepared sterile Petri plates filled with nutrient agar. Then, we spread bacterial cultures evenly across the surface of the agar plates with a sterile swab. Next, we created wells in the agar using a sterile cork borer and added various concentrations of the plant extract into those wells.

The plates were kept at the right temperature for 24 hours. Once the incubation period was over, we took a look at the plates to see if any clear zones had formed around the wells. These clear areas showed that the microbial growth had been inhibited. To assess how effective the plant extract was, we measured the diameter of these inhibition zones using a ruler or caliper.^[15]

Zone of Inhibition

The zone of inhibition is the clear circular area around a well (or disc) on an agar plate where no microbial growth occurs. This happens because the plant extract inhibits the growth of bacteria.

- Larger zone → Stronger antimicrobial activity
- Smaller/no zone → Weak or no activity.

2.9 Statistical Analysis

The experiments were carried out in triplicates to guarantee the accuracy and reliability of the results. We recorded the findings from the antimicrobial testing and presented them as mean values, along with the standard deviation.

We conducted a statistical analysis to compare how effective various extracts and concentrations were. The data we gathered helped us assess the antimicrobial potential of *Hibiscus rosa-sinensis*.

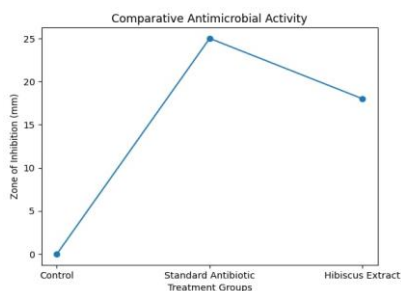


Figure 4: Antimicrobial Activity.

3. RESULTS AND DISCUSSION

The results of this study reveal that extracts from *Hibiscus rosa-sinensis* are packed with various bioactive compounds that could be behind its medicinal properties. In the initial phytochemical analysis, we found key components like flavonoids, tannins, phenolic compounds, alkaloids, and saponins in the plant extract. These phytochemicals are well-known for their positive biological effects. Their presence in the extract hints that this plant might serve as a natural source of compounds that can help protect the body from microbial infections and oxidative damage.

The antimicrobial study conducted with the agar well diffusion method revealed that the plant extract successfully inhibited the growth of certain microorganisms. After the incubation period, we noticed clear zones around the wells where the extract was applied, which indicated its effectiveness in stunting microbial growth. Interestingly, the size of these inhibition zones grew larger with increased concentrations of the extract, suggesting that its antimicrobial properties are indeed dependent on concentration. This phenomenon could be attributed to the action of phytochemicals like flavonoids and tannins, which are known to disrupt the cell structure of microbes and interfere with their normal metabolic processes.

Table 1: zone of inhibition(mm).

Standard drug (mm)	Hibiscus extract (mm)
28	20
27	18
25	15
24	17

Based on what we observed in this study, the findings back up the traditional use of hibiscus for managing infections and related health issues. The antimicrobial properties we've seen really showcase the potential of this plant in creating herbal-based treatments. That said, we

still need to dive deeper with more research, like detailed phytochemical analyses, isolating the active compounds, and conducting advanced biological studies. This will help us get a clearer picture of how the plant works and confirm its effectiveness for use in pharmaceuticals.

4. CONCLUSION

The present study demonstrates that *Hibiscus rosa-sinensis* possesses notable antimicrobial potential, thereby supporting its traditional use in herbal medicine. Extraction techniques such as maceration and Soxhlet effectively yielded bioactive compounds, particularly when organic solvents were used. Preliminary phytochemical screening confirmed the presence of important constituents including flavonoids, tannins, phenolic compounds, alkaloids, and saponins, which are known for their therapeutic properties. The antimicrobial activity assessed through the agar well diffusion method showed clear inhibition of microbial growth, indicating the effectiveness of the plant extracts against selected microorganisms. The increase in inhibition zones with higher concentrations further suggests a dose-dependent activity. Overall, these findings highlight the potential of *Hibiscus rosa-sinensis* as a natural source of antimicrobial agents, and further research involving isolation of active compounds and advanced pharmacological studies is recommended to support its application in modern medicine.

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