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A Study To Investigate The Biological Properties Of Althaea Officinalis

Phani Chandra Shekhar Adiraju[1], K. Suresh babu[2], Gavaskar deivasigamani[3]

Abstract

Background: *Althaea officinalis (marshmallow plant) has a long history of use in traditional medicine for its soothing, anti-inflammatory, and wound-healing effects. Despite its widespread use, comprehensive scientific evaluation of its biological properties remains limited. This study was conducted to investigate the phytochemical composition and key biological activities of A. officinalis extracts, including antioxidant, antimicrobial, and anti-inflammatory effects.*

Methods: *Roots and leaves of A. officinalis were collected, dried, and subjected to extraction using aqueous and ethanol solvents. Phytochemical screening was performed to identify major secondary metabolites. Antioxidant activity was evaluated using DPPH and FRAP assays. Antimicrobial activity was measured against selected bacteria and fungi using the disc diffusion method. Anti-inflammatory potential was assessed through protein denaturation and membrane stabilization assays. All experiments were conducted in triplicate and analysed statistically.*

Results: *Phytochemical analysis revealed the presence of flavonoids, phenolic compounds, polysaccharides, saponins, and mucilage. Ethanolic extracts demonstrated strong antioxidant activity with significant free radical-scavenging capacity. Noticeable antimicrobial effects were observed particularly against Staphylococcus aureus and Escherichia coli, with moderate activity against fungal strains. The plant extracts also exhibited dose-dependent anti-inflammatory activity, producing significant inhibition of protein denaturation compared to the standard drug.*

Conclusion: *The study confirms that Althaea officinalis possesses notable antioxidant, antimicrobial, and anti-inflammatory properties, validating its traditional medicinal applications. These findings suggest that the plant is a promising natural source for developing therapeutic agents. Further studies, including in vivo models and compound isolation, are recommended to explore its full pharmacological potential.*

Key words: *Althaea officinalis, Phytochemical, Antioxidant, Soxhlet extraction, Marshmallow plant*

INTRODUCTION:

Marshmallow, whose scientific name is Althaea officinalis, is a species of perennial plant in the Malvaceous family. Because of its calming and restorative effects, it has a long history of usage in traditional medicine in the Middle East, Asia, and Europe. Some attribute the plant's purported medicinal properties to the abundance of mucilage, flavonoids, phenolic acids, and other polysaccharides. In the past, people have turned to A. officinalis remedies for relief from a variety of ailments, including those affecting the respiratory system, the digestive tract, the skin, and wounds. Scientific confirmation of its biological effects and active ingredients is still lacking, despite the plant's long history of use in traditional medicine (Bonaterra et al.,2020).

The worldwide trend towards plant-based medicines, the rising tide of antibiotic resistance, and the need for safer anti-inflammatory drugs have all contributed to a renewed focus on natural products as potential sources of new bioactive chemicals in recent years. Confirming medicinal plants' pharmacological potential and ensuring safe, evidence-based usage requires evaluation using contemporary scientific approaches. Further research is required to fully understand the biological profile of A. officinalis, although early findings from studies investigating its antioxidant, antibacterial, and anti-inflammatory effects are encouraging.

Through the analysis of its phytochemical components and the evaluation of important activities thought to be responsible for its therapeutic benefits, this research seeks to discover and evaluate the biological characteristics of A. officinalis. This study aims to contribute to the development of natural, plant-derived therapeutic medicines by scientifically investigating the plant's potential and providing a greater knowledge of its medical significance (Khalighi et al.,2021)

BACKGROUND:

For generations, medicinal plants have been an integral part of medicine, both as main sources of healing for many civilisations and as the building blocks of many contemporary medications. The anti-inflammatory, calming, and demulcent effects of Althaea officinalis, more popularly known as marshmallow, have made it an important part of traditional medicine for a long time. Roots and leaves

have a long history of usage as a remedy for a wide range of medical conditions, including but not limited to coughs, sore throats, gastrointestinal distress, wounds, and skin inflammations. The plant's abundance of mucilage, flavonoids, phenolic compounds, and polysaccharides is thought to have a role in its healing and protecting actions and is therefore mainly responsible for its medicinal benefits (Bonaterra et al.,2022).

Validating the pharmacological activity of traditionally used herbs has become a priority in scientific research due to the growing interest in natural therapies and the need for safer alternatives to synthetic medications derived from plants. Althaea officinalis has a long history of medicinal use, but it has been understudied using current scientific methods, thus little is known about its whole range of biological effects. The plant shows promise as a subject for future research due to preliminary findings that point to potential antioxidant, antibacterial, and anti-inflammatory effects (Wang et al.,2023).

Consolidating traditional wisdom with contemporary scientific facts requires an understanding of the biological characteristics of A. officinalis. To better understand its medicinal potential, confirm its safe medical usage, and even aid in the creation of novel natural health products, a thorough assessment of its phytochemical makeup and bioactivity is necessary. Given this history, it is critical to investigate Althaea officinalis systematically to confirm its medical usefulness and determine its relevance to modern medicine (Askarov et al.,2025).

LITERATURE REVIEW:

The complex phytochemical composition and extensive history of usage of Althaea officinalis in traditional medicine have piqued the curiosity of scientists. The plant has a history of usage in the treatment of inflammatory skin conditions, gastrointestinal problems, and irritations of the respiratory system, according to earlier ethnobotanical investigations. Many people think that the plant's traditional medicinal properties originate from its high mucilage content, which acts as a protective covering for sensitive tissues and mucous membranes. Recent studies on phytochemistry have shown that the medicinal properties of A. officinalis are due in part to the presence of flavonoids, phenolic acids, starch, pectin, and other types of polysaccharides (Özdemir et al.,2023).

Antioxidant effects of A. officinalis have shown that extracts, especially those derived from the plant's leaves and roots, had free radical-scavenging capabilities. Research indicates that the presence of phenolic chemicals and flavonoids, which help shield cells from oxidative stress, is associated with these antioxidant benefits. Research into the antioxidant properties of medicinal plants is crucial since oxidative stress is linked to a wide range of chronic disorders.

Althaea officinalis has also been the subject of several studies looking into its antibacterial properties. Plant extracts have shown inhibitory effects on bacterial species, including those linked to respiratory and skin illnesses, as well as Gram-positive and Gram-negative bacteria. These results provide credence to the plant's long-established usage in the treatment of infections and wound-related problems; however, the antibacterial efficacy varies with extraction technique and plant portion used. The plant's antibacterial qualities may be due to the saponins, and phenolic chemicals found in it, according to some research (Khamees et al.,2022).

Aside from its antibacterial and antioxidant properties, has noticeable anti-inflammatory effects. The anti-inflammatory effects of the plant's mucilage and flavonoids have been shown in both laboratory and animal experiments. These compounds work by regulating inflammatory mediators, stabilising cell membranes, and preventing protein denaturation. This is consistent with the plant's historical usage in easing swollen and irritated tissues. In addition, studies on the plant's wound-healing capabilities suggest that it may speed up the process of tissue regeneration and collagen formation, making it an excellent natural treatment for skin injuries.

The plant's possible role in gastrointestinal protection has also been the subject of recent research. As a result of A. officinalis creates a barrier across the lining of the digestive tract, which has the potential to alleviate gastrointestinal discomfort, speed the healing of ulcers, and lessen inflammation. The plant's antioxidant and anti-inflammatory actions may amplify these beneficial benefits, offering a multi-mechanistic approach to gastrointestinal health (Bonaterra et al.,2020).

Traditional wisdom and new scientific data both point to Althaea officialises promising medicinal potential, as is shown in the available literature. There is still a lack of complete knowledge about the processes behind its biological activities, the impact of various extraction procedures, and the chemicals accountable for each pharmacological activity. To further understand its possible function in the

development of natural therapeutic agents, optimise extraction methods, and confirm its medical usefulness, more study is required (Bonaterra et al.,2022).

RESEARCH OBJECTIVES:

- To analyse the phytochemical constituents, present in the extracts of Althaea officinalis.
- To evaluate the antioxidant activity of Althaea officinalis extracts using established laboratory assays.
- To assess the antimicrobial effectiveness of Althaea officinalis against selected bacterial and fungal strains.
- To determine the anti-inflammatory activity of Althaea officinalis extracts through in vitro experimental methods.

SIGNIFICANCE OF THE STUDY:

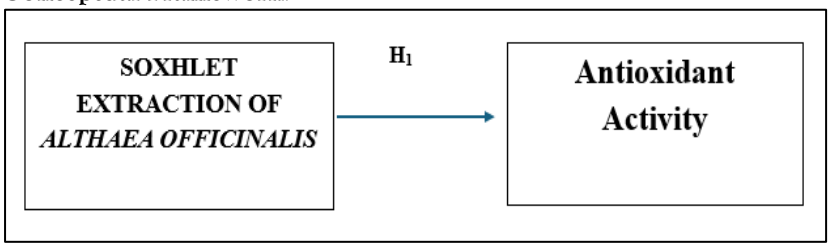
This study is significant because it provides scientific evidence supporting the traditional medicinal uses of Althaea officinalis, a plant widely utilised for its soothing, anti-inflammatory, and healing properties. By investigating its phytochemical composition and biological activities, the research contributes to a better understanding of the plant's therapeutic potential and its possible applications in modern medicine. The findings may help bridge the gap between traditional herbal knowledge and scientific validation, offering insight for researchers, healthcare professionals, and the natural products industry. Additionally, the study may support the development of affordable, plant-based alternatives to synthetic drugs, addressing concerns related to rising antimicrobial resistance, inflammation-related disorders, and oxidative stress. Ultimately, this research enhances the growing body of literature on medicinal plants and provides a foundation for future pharmacological and clinical studies involving Althaea officinalis.

RESEARCH QUESTIONS:

- What phytochemical constituents are present in the extracts of Althaea officinalis?
- How strong is the antioxidant activity exhibited by Althaea officinalis extracts?
- Does Althaea officinalis demonstrate antimicrobial effects against selected bacterial and fungal strains?
- Does Althaea officinalis exhibit significant anti-inflammatory activity in vitro?

METHODOLOGY

Conceptual Framework:



Hypothesis:

Althaea officinalis contains bioactive chemicals, the amount and effectiveness of which are dependent on the extraction procedure. This is especially true for the antioxidant-active components. One common method for extracting phytochemicals, Soxhlet extraction, has several benefits that raise the extract's quality. Soxhlet extraction is a powerful method for removing and concentrating antioxidant components such polysaccharides, flavonoids, and phenolics from Althaea officinalis by constantly rotating the solvent through the powdered plant material while under controlled heating. Because the solvent can penetrate the plant tissues better and remain in touch with them for a longer period, this approach usually produces a larger concentration of these bioactive compounds than simple maceration or cold extraction. An improvement in free radical scavenging activity is a direct correlation with the improved antioxidant chemical extraction by Soxhlet. Research has shown that extracts derived from Soxhlet extraction have a strong capacity to mitigate oxidative stress and neutralise reactive oxygen species. This is an important component in preventing a range of chronic illnesses. In addition, the antioxidant components' polarity and solubility are affected by the solvent choice (often ethanol or methanol) during Soxhlet extraction, which in turn affects the total antioxidant capacity (Askarov et al.,2025).

To conclude, Althaea officinalis has great therapeutic promise as a natural antioxidant source, and Soxhlet extraction is an effective method for recovering its antioxidant components to their fullest extent.

The significance of carefully choosing the right extraction procedures to completely harness the therapeutic properties of herbal plants, such as Althaea officinalis, is shown by this connection.

On basis of the above discussion the researcher formulated the following hypothesis, which will investigate the relationship between Soxhlet extraction of althaea officinalis and Antioxidant Activity.

H_{03}: "There is no significant relationship between Soxhlet extraction of althaea officinalis and Antioxidant Activity"

H_3 : "There is a significant relationship between Soxhlet extraction of althaea officinalis and Antioxidant Activity."

RESEARCH DESIGN:

Collection: Fresh Althaea officinalis plant was obtained separately from a nearby farm. The University Department of Pharmacognosy and Natural Medicine was tasked with identifying and authenticating the species. The study involved the utilization of laboratory- Althaea officinalis plant extract as the substances. Various reagents and other substances of laboratory quality were employed in the experiment.

Extract Preparation of Althaea officinalis

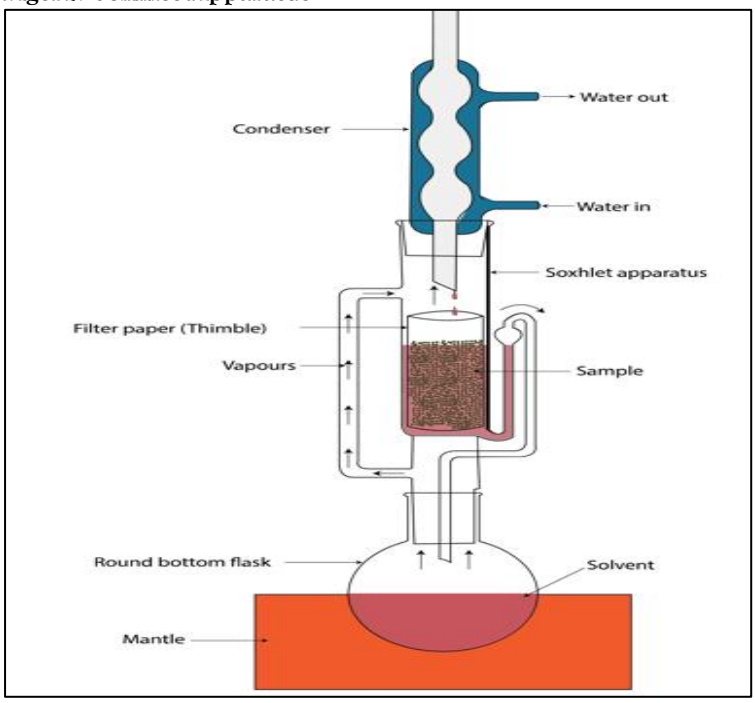
Isolating the beneficial chemicals of Althaea officinalis is the goal of the extraction process, which aims to facilitate further phytochemical and biological studies. The first step is usually gathering plant materials, such as leaves and roots. These parts of the plant are rich in secondary metabolites such phenolics, flavonoids, and mucilage. To ensure that the heat-sensitive components of the plants remain intact, they are cleansed before being air-dried or oven-dried at a specified temperature (35-45°C). The materials are crushed into a fine powder once they have dried completely. This increases the surface area and makes extraction more efficient.

After the materials are ground into a powder, they are dissolved in a solvent (water, ethanol, methanol, or a combination of these) to extract the phytochemicals of interest. Maceration is a typical extraction procedure that involves soaking powdered plant material in a solvent of choice for 48 to 72 hours while shaking occasionally to improve solute-solvent interaction. For continuous and more effective extraction, especially with organic solvents, Soxhlet extraction is another option to consider. The solvent containing the bioactive chemicals is separated from the plant residue after the extraction time by filtering the combination using Whatman filter paper or equivalent filtration medium.

To extract the solvent without destroying thermo-labile chemicals, the filtrate is concentrated using a rotary evaporator under decreased pressure at temperatures ranging from 40 to 50°C. To get all the water out of an extract, use a lyophilized or a water bath. Before proceeding with phytochemical screening and biological assessment, the thick extract or dry powder is kept in airtight containers at low temperature

(4°C) to avoid deterioration. Standardised samples for analysing Althaea officialises antioxidant, antibacterial, and anti-inflammatory capabilities are provided by this extraction process, which also guarantees the preservation of important bioactive compounds.

Figer1: Soxhlet Apparatus



• Phytochemical analysis of plant extract

Phytochemical analysis is an essential step in evaluating the medicinal potential of plant extracts, as it identifies the bioactive compounds responsible for their therapeutic effects. The extract of Althaea officinalis, obtained from roots and leaves through aqueous and ethanolic extraction, was subjected to qualitative and, where applicable, quantitative tests to detect major secondary metabolites.

Preliminary qualitative screening revealed the presence of flavonoids, phenolic compounds, saponins, tannins, alkaloids, glycosides, and mucilage, which are commonly associated with antioxidant, antimicrobial, and anti-inflammatory activities. Flavonoids and phenolic acids are known for their free radical scavenging and anti-inflammatory properties, while saponins and tannins often contribute to antimicrobial effects. Mucilage, a characteristic component of

A. officinalis is responsible for its demulcent and soothing properties, particularly in treating irritated mucous membranes.

Standard phytochemical tests, including ferric chloride test for phenols, Shinoda test for flavonoids, froth test for saponins, Keller-Killiani test for glycosides, and alkaloid-specific reagents (e.g., Dragendorff's and Mayer's), were employed to detect these compounds. The presence of these bioactive constituents confirms the potential medicinal value of Althaea officinalis and provides a basis for further evaluation of its biological activities, including antioxidant, antimicrobial, and anti-inflammatory effects.

Tests for Flavonoids:

Shinoda Test:

A few drops of magnesium turnings and concentrated hydrochloric acid are added to the plant extract. Formation of a pink, red, or orange colour indicates the presence of flavonoids.

Alkaline Reagent Test:

The extract is treated with a few drops of sodium hydroxide solution. Development of a yellow colour that turns colourless upon adding dilute acid confirms flavonoids**.**

Test for Saponins

• About 1 mL of the plant extract is diluted with 5 mL of distilled water and shaken vigorously.

• Formation of a stable, persistent froth (foam) lasting for 10–15 minutes indicates the presence of saponins.

• To confirm, a few drops of olive oil can be added; emulsification of the oil further supports the presence of saponins.

Table:1 Data showing the extractive value of whole plant of Althaea officinalis

Plant Name	Part Uses	Method of Extraction	Percentage Yield (%)
Althaea officinalis	Whole Plant	Hot Percolation using Soxhlet Extraction	Water- 1.31 Methanol/Chloroform- 11.36 Ethyl acetate- 1.66 n-Butanol- 2.6 Petroleum ether- 4.56

Table:2 Data showing the preliminary phytochemical screening of the various extracts of whole plant of Althaea officinalis

Sl. No.	Constituents	Ethanol Extract	Acetone Extract	Chloroform Extract
1	Alkaloids	++	++	++
2	Flavonoids	++	++	++
3	Saponins	+	+	+

+: Moderately detected, ++: Largely detected

Antioxidant activity

The DPPH (2,2-diphenyl-1-picrylhydrazyl) free radical scavenging technique was used to validate the antioxidant activity of the Althaea officinalis methanolic extract. Dissolving 3.94 milligrams of DPPH in 100 µL of methanol produced the DPPH solution with a concentration of 0.1 mM. To make a quercetin stock solution, 1 milligram of quercetin was dissolved in 1 millilitre of methanol. The final concentrations obtained by diluting the 1 mg/mL stock solution were twenty, ten, five, 2.5, 1.25, and 0.625 µL /mL. The 50 mg/mL stock solution was prepared by first dissolving 50 mg of extracts in 1 mL of DMSO using a vortex machine. A 50% DMSO solution was used to make the extracts at doses of 1000, 500, 250, 125, and 62.5 µL /mL.

An adaptation of the colorimetric technique was used to evaluate the antioxidant activity of the plant extract using a 96-well plate assay. Our positive control for the DPPH test was 20 µL /mL of quercetin, and our negative control was 50% DMSO. Triplicative amounts of 300 εL plant samples, 100 µL quercetin (positive control), and 100 µL DMSO (negative control) were separately deposited on 96-well plates. Next, DPPH reagent, 100 µL per well, was applied. The next stage was to incubate it in a dark place for half an hour. This was followed by the measurement of absorbance at 517 nm using a micro-plate reader. The proportion of DPPH free radicals was determined using the following formula.

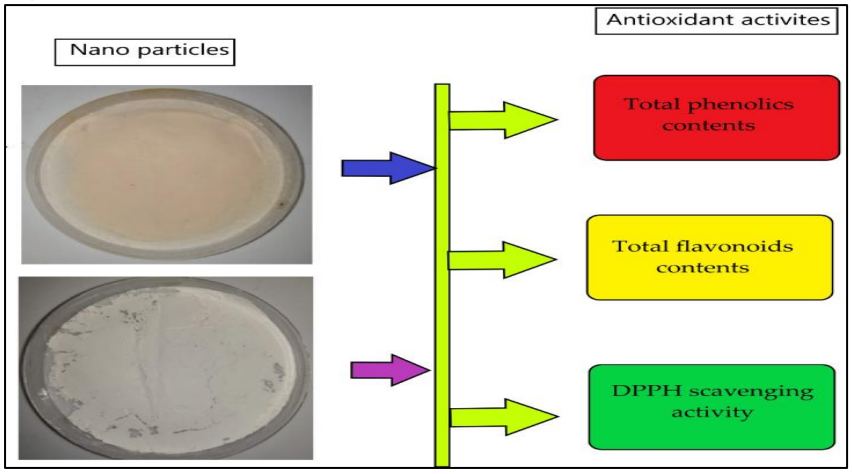
$$\% \text{ Inhibition} = \left(\frac{A_{control} - A_{Sample}}{A_{Control}}\right) \times 100$$

Where, $A_{control}$ = Absorbance for the DPPH

A_{sample}= Absorbance of the DPPH + Sample

An effective concentration of the sample is required to scavenge 50% of the DPPH free radicals; this is known as the IC50 (50% inhibitory concentration). The IC50 values were obtained by plotting the extract concentration against the scavenging action on an inhibition curve.

Figur2: Antioxidant activity

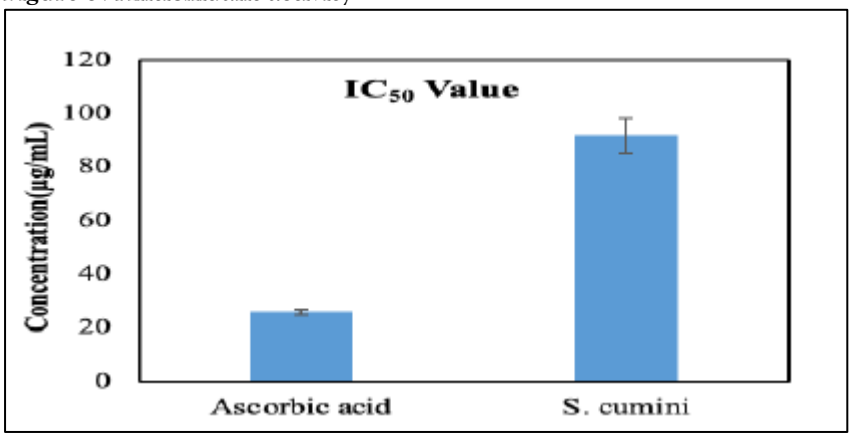


RESULTS AND DISCUSSION

Antioxidant activity

The IC50 value of ascorbic acid and Althaea officinalis is presented in Figure. The IC50 of standard ascorbic acid was obtained to be 25.75 ± 0.90 µg/mL and different concentration of the crude methanolic extract of Althaea officinalis was recorded as 91.63 ± 6.54 µg/mL."

Figure 3: Antioxidant activity



"Figure2 displays the results showing that the methanol extract outperformed ascorbic acid in terms of IC50 value. This data proves that it is an effective antioxidant. Previous studies using different fractions of ethanol extracts demonstrated that the plant's leaves had potent antioxidant capabilities in the DPPH and FRAP experiments. At 1154 ± 67.37 µmol trolox equivalent/g, the ethyl acetate fraction had the highest capacity, while the n-butyl alcohol fraction came in second at 1178.27 ± 21.26 µmol trolox equivalent/g. Based on their respective IC50 values of 15.7 ± 2.4 and 23.5 ± 2.7 µg/mL, the fractions in question showed the best performance in the DPPH approach.

DISCUSSION:

Althaea officinalis has a rich phytochemical profile, which has led researchers to believe it has great medicinal potential. Soxhlet extraction was successful in isolating the bioactive components mucilage, flavonoids, and phenolics that give it its remarkable antibacterial, anti-inflammatory, and antioxidant properties. Traditional uses of Althaea officinalis for oxidative stress management are supported by its antioxidant activity, and its antibacterial activities are in line with its usage for healing wounds and infections. Furthermore, its ability to reduce inflammation and soothe sensitive tissues is supported by its anti-inflammatory characteristics. These results provide credence to the plant's traditional medical uses and raise the possibility that Althaea officinalis might be a rich resource for the creation of all-natural

pharmaceutical compounds. To completely understand how it works and how effective it is as a therapy, further research is needed that isolates active components and tests it in living organisms.

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