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A Preliminary Study on the Phytochemical Analysis, Antioxidant Activity and Antibacterial Activity of Abutilon Indicum

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Abstract: ***Medicinal plants are used to create, or serve as the basis for, modern pharmaceutical drugs. Roughly 50% of new drugs over the last 60 years originated from natural sources. Abutilon indicum (L.), commonly referred to as Indian mallow or atibala, is a medicinal shrub belonging to the family Malvaceae. It has long been utilized in traditional ayurvedic medicine due to its wide range of pharmacological activities, including anti-inflammatory, wound-healing, diuretic, and antimicrobial effects. The present study aims to screen the phytochemicals, antioxidant activity and the antibacterial potential of Abutilon indicum in hexane and ethyl acetate extract. Phytochemical screening showed the presence of important compounds such as phenolic compounds, flavonoids, alkaloids, and quinones, especially in the ethyl acetate extract, while some compounds like reducing sugars, proteins, saponins, and tannins were mostly absent. The antioxidant activity was evaluated using the DPPH radical scavenging assay at different concentrations, with ascorbic acid used as the standard. The results showed that the percentage of inhibition increased with concentration, indicating moderate antioxidant activity of the plant extract. The antibacterial activity was tested using the agar well diffusion method against Klebsiella pneumoniae on Mueller–Hinton agar medium, using Meropenem as the positive control. The formation of inhibition zones around the wells indicated that the plant extract exhibited antibacterial activity, which increased with higher concentrations. Overall, the results suggest that Abutilon indicum possesses significant phytochemical compounds and shows potential antioxidant and antibacterial properties.***

Keywords: Abutilon indicum, Phytochemicals, Antioxidant, Antibacterial, phenol & Flavonoids

1. Introduction

Plants that contain active compoundssuch as alkaloids, flavonoids, and terpenoidswhich have therapeutic effects on the human body. Beyond traditional use, medicinal plants are used to create, or serve as the basis for, modern pharmaceutical drugs. Roughly 50% of new drugs over the last 60 years originated from natural sources.

“Abutilon indicum (L.), commonly referred to as Indian mallow or Atibala, is a medicinal shrub belonging to the family Malvaceae. The plant is characterized by its soft, velvety, cordate leaves and yellow flowers. It has long been utilized in traditional Ayurvedic medicine due to its wide range of pharmacological activities, including anti-inflammatory, wound-healing, diuretic, and antimicrobial effects. In addition to its therapeutic value, A. indicum is also recognized for its fiber content and ornamental potential, although it has exhibited invasive tendencies in certain geographical regions.Native to tropical and subtropical areas, A. indicum has been traditionally employed in the management of ailments such as ulcers, diabetes, piles, jaundice, gonorrhea, and leprosy. Recent investigations have highlighted the antidepressant activity of the crude methanolic extract of A. indicum (Ai.Cr). **(Zhou, X., Hassan et al.,(2021).** The anti-obesity potential of A. indicum leaf extract was assessed by examining its effects on adipogenesis, lipolysis, and cholesterol esterase activity using 3T3-L1 adipocyte cell lines. suggesting its possible role as an adjunct therapeutic agent in the management of type 2 diabetes. **(Lakshminarayana, L., et al., 2023).** It is commonly known as “Country mallow” or “Thuthi,” A. indicum has also been traditionally recommended for respiratory disorders such as bronchitis and for metabolic diseases including diabetes.

Figure 1: Abutilon indicum plant

The plant thrives in warm, sunny conditions and is capable of growing in dry, nutrient-deficient soils. In India, it is frequently found along roadsides and in wastelands, where ittypically emerges after the monsoon season and blooms during winter. In Poland, the species is additionally grown for ornamental purposes.

Medicinal uses:
Various parts of Abutilon indicum have been reported to exhibit a wide range of pharmacological properties. These include antimicrobial, antifertility, anticonvulsant, anthelmintic, antidiarrhoeal, wound healing, hepatoprotective, antihypertensive, antitumor, antidiabetic,

anti-inflammatory, and free-radical scavenging activities. Hence the present study aims to screen the phytochemicals, determine the antioxidant activity and the antibacterial potential of Abutilon indicum in hexane and ethyl acetate extract.

2. Materials & Methods

Abutilon indicum plants were collected from roadsides nearby Vyasrarpadi, Chennai.

Figure 2: Sample preparation of Abutilon indicum

Preparation of plant extract: The collected plant material was thoroughly washed with water and shade-dried at room temperature. After drying, the sample was finely powdered. About 10 g of the powdered plant material was extracted using 200 mL of hexane and ethyl acetate. The mixture was then filtered through Whatman No. 1 filter paper, and the obtained extract was stored for further analysis.

Preliminary Phytochemical Screening: The therapeutic potential of a plant is largely determined by the bioactive compounds it contains. Therefore, identifying the biochemical constituents of medicinal plants is essential, as it provides insight into their medicinal value and supports the validation of traditional remedies. In the present investigation, the extracts and their fractions were qualitatively analyzed for the presence of phytochemical constituents following standard procedures.

Preparation of Sample: Approximately 100 mg of each extract was accurately weighed and dissolved in 25 mL of the corresponding solvent. The resulting solutions were subsequently used for qualitative phytochemical screening.

1) **Test for Phenolic Compounds: Ferric Chloride Test:** To the sample, few drops of alcoholic $FeCl_2$ solution was added. Bluish green or Bluish black indicated the presence of phenol.
2) **Test for Reducing Sugar:** Fehling's Test: The sample was mixed with Fehling's solution I and II and formation of red coloration indicated the presence of sugars
3) **Test for Flavones: Alkaline Reagent Test:** The sample was mixed with 10% sodium hydroxide solution or ammonia. Dark yellow color indicated the presence of flavones.
4) **4.Tests for Saponins: Frothing Test:** The samples were shaken well with water and formation of copious lather indicated the presence of saponins.
5) **Test for Alkaloids: Mayer's reagent**: To the sample few drops of acetic acid and Mayer's reagent was added and shaken well. A creamy white colored precipitation indicated the presence of alkaloids.
6) **Wagner's reagent:** To the sample, few drops of acetic acid and Wagner's reagent was added and shaken well. A reddish brown precipitation indicated the presence of alkaloid.
7) **T est for Quinones:** To the sample, sodium hydroxide was added. Blue and green or red color indicated the presence of quinones.
8) **Test for Proteins: Biuret Test:** To the sample few drops of Biuret reagent was added and formation of blue colour indicated the presence of protein.
9) **Test for Tannins: Lead Acetate Test:** The samples were mixed with basic lead acetate solution. Formation of orange red precipitate indicated the presence of tannins.

Antioxidant Assay:

In-vitro DPPH Radical Scavenging Activity: This method is a simple, rapid and sensitive method to measure the antioxidant activity of any compound (Koleva et al., 2002). DPPH is a stable free radical with purple in colour, readily accepts the hydrogen from the antioxidants and the colour change from purple to yellow. 2,2-Diphenyl-1-picrylhydrazyl will get converted to 2,2-Diphenyl-1-picrylhydrazine when combined with the antioxidant.

Antibacterial Activity by Agar Well Diffusion Method

Medium: Mueller–Hinton Agar (MHA) was used for evaluating antibacterial activity.

Test Organism: Klebsiella pneumoniae

Procedure: A sterile cuvette containing nutrient broth was prepared. A small amount of the test organism (Klebsiella pneumoniae) was transferred into the broth using a sterile inoculation loop. The inoculated broth was allowed to incubate to obtain uniform bacterial suspension. After incubation, the bacterial culture was evenly spread over the surface of Mueller–Hinton agar plates using a sterile cotton swab to obtain a lawn culture. Wells were then aseptically created on the agar surface using a sterile well borer. The prepared plant extract was carefully added into the respective wells. A standard antibiotic (meropenem) was used as a positive control and placed on the same plate for comparison. The plates were incubated under suitable conditions to allow bacterial growth and diffusion of the extract and antibiotic.

Observation: After incubation, clear zones around the wells indicated inhibition of bacterial growth. The presence of a

zone of inhibition around the extract well demonstrated antibacterial activity, while the antibiotic disc showed sensitivity or resistance based on the size of the inhibition zone.

3. Results

Phytochemical Analysis :

Table 1: Phytochemical Screening of Abutilon indicum in Hexane and Ethyl acetate extract

S. No	Phytochemical Parameters	Hexane	Ethyl Acetate
1	Test for Phenolic Compounds	-	++
2	Test for Reducing Sugar	-	-
3	Test for Flavones	+	+++
4	Test for Saponins	-	-
5	Test for Alkaloids (Wagner's Test)	++	+++
6	Test for Alkaloids (Mayer's Test)	-	-
7	Test for Quinones	++	+++
8	Test for Proteins	-	-
9	Test for Tannins	-	-

As the results showed the strong presence of phenolic compounds, alkaloids, flavonoids and quinones in ethyl acetate extract, the antioxidant and antibacterial studies were carried out in ethyl acetate extract of the plant.

Antioxidant Assay – DPPH Assay

DPPH assay activity of Ethyl acetate extract of Abutilon indicum

% of inhibition = (Abs of control – Abs of test) / Abs of control x100

Control –0.6255

Concentration µg/ml	% inhibition AS	% inhibition AI
50	80	25
100	79	35
150	81	37
200	84	55
250	90	63

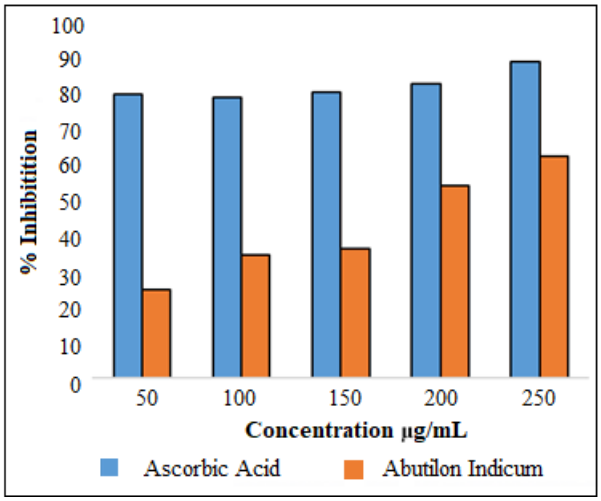

Figure 3: DPPH Radical Scavenging Assay of Ascorbic acid (standard) and Ethyl acetate extract of Abutilon indicum

Table 3: Antibacterial activity of ethyl acetate extract of A.indicum

Concentration of Standard (Meropenem) (µg/ ml)	Zone of inhibition of Standard (Meropenem) (mm)	Concentration of AI (µg/ ml)	Zone of inhibition of AI (mm)
10	35	50	-
10	35	100	-
10	35	150	8
10	35	200	11

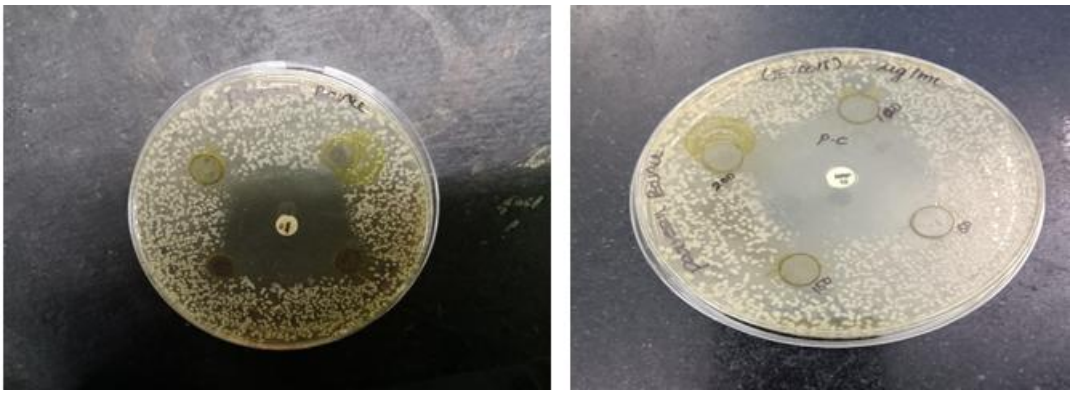

Figure 4: Antibiotic sensitivity test for determining zone of inhibition

4. Discussion

Abutilon indicum (L.) Sweet is a medicinal plant widely used in traditional Asian systems of medicine for the management of various ailments, including diabetes mellitus. Despite its long-standing ethnomedicinal use, the molecular basis underlying the antidiabetic activity of A. indicum has not yet been clearly established. The findings indicate that the A. indicum L. extract could play a role in improving insulin sensitivity. **(Krisanapun et al., (2011).**

The hierarchical CuO NPs was synthesized via green chemistry method by using of Abutilon indicum (A. indicum) leaf extract. The synthesized CuO NPs was performed antibacterial activity against human pathogenic organisms of both Gram negative (Escherichia coli and Salmonella typhi) and Gram positive (Bacillus subtilis and Staphylococcus aureus) bacteria by using agar well diffusion method. It is observed that Abutilon indicum leaf extract can reduce silver ions into silver nanoparticles within 15 min of reaction time. The AgNPs thus obtained showed highly

potent antibacterial activity toward Gram-positive (Staphyloccocus aureus and Bacillus subtilis) and Gram-negative (Salmonella typhi and Escherichia coli) microorganisms. **(Ashok Kumar et al., (2015).** Preliminary analyses indicated that ELEAI was rich in both primary and secondary metabolites. Luteolin, a known phytochemical, was quantitatively confirmed in the extract using HPTLC. Impressively, the DPPH assay highlighted ELEAI's remarkable antioxidant capabilities. **(Sunil et al., (2023).**

The presence of such bioactive compounds in medicinal plants like abutilon indicum often makes them attractive for therapeutic applications. These compounds have been reported to contribute to hepatoprotection and may offer protection against various chronic diseases, including cancer, diabetes, and cardiovascular disorders, as documented in a study by **(Choi et al. in 2015).**The findings of the study showed the phytochemical constituents, antioxidant activity, and antibacterial properties of Abutilon indicum was strongly positive in ethyl acetate extract. Phytochemical screening showed the presence of important compounds such as phenolic compounds, flavonoids, alkaloids, and quinones, especially in the ethyl acetate extract, while some compounds like reducing sugars, proteins, saponins, and tannins were mostly absent.

The antioxidant activity was evaluated using the DPPH radical scavenging assay at different concentrations, with ascorbic acid used as the standard. The results showed that the percentage of inhibition increased with concentration, indicating moderate antioxidant activity of the plant extract.

The antibacterial activity was tested using the agar well diffusion method against Klebsiella pneumoniae on Mueller–Hinton agar medium, using meropenem as the positive control. The formation of inhibition zones around the wells indicated that the plant extract exhibited antibacterial activity, which increased with higher concentrations. Overall, the results suggest that Abutilon indicum possesses significant phytochemical compounds and shows potential antioxidant and antibacterial properties.

5. Conclusion

The current investigation aimed to evaluate the phytochemical profile and antioxidant potential of Abutilon indicum, a medicinal plant widely used in traditional medicine. The findings indicate that A. indicum exhibits notable antioxidant activity, which also shows the presence of diverse bioactive compounds such as phenolic compounds, flavones, alkaloids, and quinones. Abutilon indicum demonstrates considerable medicinal value due to its abundant phytochemical constituents and potent antioxidant properties, indicating its potential usefulness for advanced pharmacological investigations and the development of plant-based therapeutic agents. The antibiotic sensitivity test performed on Klebsiella pneumoniae using the agar well diffusion method demonstrated clear zones of inhibition around the antibiotic and extract wells. The formation of inhibition zones indicates that the test substance possesses antibacterial activity against the selected organism. Meropenem showed a distinct zone of inhibition, confirming its effectiveness and serving as a positive control. Based on the zone diameter, the organism was categorized as sensitive or resistant to the tested agents. Overall, the study confirms that the tested extract exhibits antibacterial potential against Klebsiella pneumoniae, and further studies can be conducted to determine its minimum inhibitory concentration and therapeutic significance.

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