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Brief communication

Phytochemical Screening and *In-vitro* Antibacterial activity Evaluation of *Lantana camara* Linn. Leaf Extract

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ABSTRACT

With both antibiotic-susceptible and -resistant bacteria, the antimicrobial activity of plant extracts and phytochemicals was assessed. The activity of leaves of *Lantana camara* was found by using two different solvents i.e. petroleum ether, and ethanol. *Lantana camara* is a traditional medicinal plant that was determined against two bacterial strains both the solvent extracted by using the Soxhlet apparatus. Its activity tested against two organisms they are *Staphylococcus aureus* and *Escherichia coli*. The activity was determined by inhibition zone by using a standard drug [Amoxicillin]. The extract used inhibited the growth of two organisms to a maximum level. An analysis of the phytochemistry reveals the presence of terpenoids, tannins, and saponins. These substances are thought to be in charge of the leaf extracts' broad-spectrum activity.

Keywords: *Lantana camara*, Antibacterial activity, Petroleum ether extract, Ethanol extract, Soxhlet extraction.

INTRODUCTION

Nowadays the rate of infections is increasing drastically due to various climatic conditions. All these infections are due to different microorganisms such as bacteria, viruses, fungi, parasites, etc. To overcome these microbial infections, other techniques are followed like using synthetic chemicals or natural compounds. Natural compounds are mostly obtained from plants with medicinal properties[1].

A significant source of chemicals with essential medical use is medicinal plants. Medicinal herbs have been used to treat various illnesses since the dawn of time. A range of bioactive compounds are produced through systemic analysis of these plants, which can be used to create novel medicinal medicines[2]. The pharmacological analysis of diverse plants utilized in various traditional systems of treatment has recently attracted increasing attention. Many traditionally used plants have undergone

extensive research using cutting-edge scientific methods in the past few decades, and Numerous findings have been documented regarding a range of therapeutic characteristics[3]. These encompass but are not limited to effects against cancer, inflammation, diabetes, parasitic infections, bacterial and fungal growth, liver protection, oxidative stress, mosquito larvae elimination, and additional benefits[4-6].

One of the plants with many medicinal properties is *Lantana camara* Linn. *Lantana camara* is a flowering plant belonging to the Verbenaceae family (Fig. 1). It is also known as wild sage or red sage and contains flowers of different colors i.e., red, yellow, white, and violet. It is an evergreen shrub with a strong scent, and its opposite, simple leaves have thick petioles. The oval blades are rough and hairy, and the margins are bluntly serrated. *Lantana camara* produces spherical, meaty, two-seeded beans as berries. *Lantana camara* seeds start green, then turn purple, and finally turn a blue-black tint. A native plant called *Lantana camara* can be found in tropical areas. It is regularly used as a medicinal herb to treat many health problems. All parts of this plant i.e., flowers, roots, shoots, leaves, bark, and berries have medicinal properties due to the presence of several biological active constituents[1,4].

Fig. 1. *Lantana camara* plant

The most prevalent and clinically significant species is *E. coli*, which also accounts for a large number of nosocomial infections, gastroenteritis, and more than 80% of all community-acquired urinary tract infections in developing nations. Children, the elderly, and those with compromised immune systems are frequently affected by the purulent infections and abscesses caused by the *S. aureus* bacteria[6].

The primary objective of this study was to conduct a phytochemical analysis and assess the *In-vitro* antibacterial activity of leaf extracts from *Lantana camara* Linn.

MATERIALS AND METHODS

Plant Collection

In June 2023, recently harvested leaves of *Lantana camara* Linn. were gathered from mallepally [vil], kondapur [mdl], and sangareddy [dist]. After collection, leaves that showed signs of damage were sorted out, and the remaining leaves underwent a thorough washing process to eliminate any dust or contaminants. Following this, the cleaned leaves were air-dried in the shade and subsequently ground into a powdered form.

Preparation of plant extract

Lantana camara leaf powder was extracted using soxhlet apparatus with petroleum ether and ethanol solvents for 2-3 days. 50 g each of crushed *Lantana camara* were kept for extraction by using 500 mL of petroleum ether and 500 mL of ethanol separately. The Soxhlet apparatus is maintained at a temperature of 70 degrees centigrade[7]. The extract was collected from the Soxhlet apparatus and the amount of extract was measured. It is further used for phytochemical analysis and activity testing.

Qualitative phytochemical analysis

A small amount of extracts were utilized for the determination of phytochemical constituents such as carbohydrates, steroids, proteins, triterpenoids, flavonoids, resins, tannins, and fixed oils[8].

Carbohydrates

Add 1 mL each of the Molisch reagent to both the extracts taken in test tubes and slowly introduce concentrated sulfuric acid with a gentle addition along the inner walls of the test tubes. Detection of a violet ring forming at the juncture between the two liquids signifies the existence of carbohydrates within the extracts.

Steroids

Add 10 mL of chloroform to each of the extracts, followed by the introduction of 10 mL of sulfuric acid. The presence of steroids is indicated if the upper layer displays a red color and the sulfuric acid layer exhibits a yellow-green shade.

Proteins

Introduce 5-6 drops of Millon's reagent to both extracts. The formation of a white precipitate

that turns red upon heating signifies the presence of proteins within the extract.

Triterpenoids

Combine 2 mL of chloroform with 1 mL of each extract, and then add a small quantity of sulfuric acid. The emergence of a reddish-brown color at the boundary suggests the presence of triterpenoids.

Flavonoids

Mix 1 mL of each extract separately with 2 mL of 2N sodium hydroxide. The presence of a yellow color indicates the existence of flavonoids.

Resins

Independently add 20 mL of 4% hydrochloric acid to both extracts. The formation of turbidity signifies the presence of resins.

Tannins

In separate test tubes for each extract, introduce 2 drops of 10% lead acetate. The presence of reddish-brown precipitate points to the existence of tannins.

Fixed oils

Place a small quantity of plant extracts between two filter papers. The presence of oil stains on the filter papers indicates the presence of fixed oils.

Evaluation of antibacterial activity

The antimicrobial activity of both extracts was assessed utilizing the disc diffusion technique[9]. The microorganisms employed in this assessment, namely *Staphylococcus aureus* and *Escherichia coli*, were sourced from the microbiology laboratory at VIPER College, Narsapur, Medak, Telangana. For further use, these strains were maintained at 4°C in nutrient agar slants. A loop of each bacterial strain was introduced to a 5 mL sterile nutrient broth in a 10 mL vial the day before the antibacterial activity was discovered. To encourage the growth of bacteria, the vials were then maintained at 37°C for 24 hours. For the preparation of the experimental disc, with the use of a punching machine, Whatman's filter paper No. 1 was cut into 6 mm diameters. The discs were autoclaved at 120°C for 20 min to sterilize them. Drying was accomplished by putting the sterilized moisture discs in a hot air oven set to 50°C. For an antibacterial study, a sterile disc holding 25 µL of plant extracts was created.

For the determination of antibacterial activity, positive and negative controls were also prepared to compare the effectiveness of plant extract. A sterile disc with only extracting solvents was used as a negative control whereas amoxicillin (500 mg), a standard drug was used as a positive control. This method measured the zone of inhibition in millimeters (mm).

RESULTS AND DISCUSSION

After the collection of plant material, it was dried and crushed into powder. Later it was extracted using a soxhlet apparatus with petroleum ether and ethanol as extracting solvents. The extract thus obtained was subjected to qualitative phytochemical analysis for the detection of various phytochemical constituents. The phytochemical analysis for both extracts was presented in Table 1.

Table 1: Phytochemical screening of *Lantana camara* Linn leaf extract

Constituents	Petroleum ether extract	Ethanol extract
Carbohydrates	-	++
Steroids	-	+
Proteins	-	+
Triterpenoids	-	+
Flavonoids	-	+
Resins	-	+
Tannins	-	+
Fixed oils	++	+

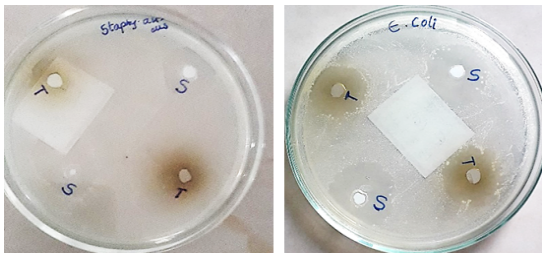
Note: (-) indicates absent, (+) indicates present, (++) indicates heavily present

Subsequent to conducting phytochemical screening on the extracts, the antibacterial efficacy was gauged by employing the disc diffusion technique. The microorganisms selected for examination were *S. aureus* and *E. coli*. Within this process, the diameter of the inhibition zone around the disc was measured in millimeters within the nutrient medium. The zone of inhibition for both extracts was presented in Table 2 and the Petri plates were shown in Figure 2.

Table 2: Antibacterial activity of leaf extract

Plant extract (50 mg/mL)	Inhibition Zone(mm) *Escherichia coli*	Inhibition Zone(mm) *Staphylococcus aureus*
Petroleum ether extract	S: 23; T: 20	S: 25; T: 21
Ethanolic extract	S: 19; T: 15	S: 17; T: 16

S-Standard solution, T-Test solution (Plant extract)



(A) (B)

Fig. 2(A). *Staphylococcus aureus*, (B) *E. coli*

S= Standard, T= Test zone of inhibition provided by two organisms to show the antibacterial activity

The present study showed that petroleum ether extract was potent when compared to ethanolic extraction with more zone of inhibition against *E. coli* (20 cm) and *S. aureus* (21 cm). When compared to previous studies, the zone of inhibition in this study was found to be more than in already existing studies[4,6].

CONCLUSION

Many plants have a variety of physiologically active substances that could be used as sources in herbal medicine. A popular ornamental plant with numerous biologically active ingredients is *Lantana camara*. This plant was chosen as a result of its antibacterial effectiveness against the chosen infections. The type of solvent employed in the extraction process has a significant impact on how well botanical chemicals may be predicted from plant material. According to this study, petroleum ether was the best solvent out of all the ones used since it had the strongest antibacterial effects against particular infections.

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Conflict of interest

The authors declare that there is no conflict of interest.

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