



Plant Archives

Journal homepage: <http://www.plantarchives.org>
DOI Url : <https://doi.org/10.51470/PLANTARCHIVES.2022.v22.no2.073>

ANTIBACTERIAL ACTIVITY OF LEGUME POD OF *LABLAB PURPUREUS* L. (GUAR) AGAINST THE SELECTED CLINICAL ISOLATES

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(Date of Receiving : 08-08-2022; Date of Acceptance : 09-10-2022)

ABSTRACT

Legumes are one of the most nutritious foods in the world and when mutual with other products are the source of diet for a great part of the world's population, especially in shodder areas where meat, dairy products and fish are economically inaccessible. Legumes belongs to the family Fabaceae or Leguminosae. *Lablab purpureus* is not only an attractive plant, but also has frequent claims on its medicinal values and nutrient content, especially its pods. The antibacterial activity of *Lablab purpureus* by using Petroleum ether, Acetone, Ethanol and Aqueous extracts were examined against gram-positive bacteria's *Bacillus subtilis*, *Staphylococcus aureus* and gram-negative bacteria's *Klebsiella pneumoniae*, *Escherichia coli*. The Antibacterial activity was examined by Disc diffusion method. In *Lablab purpureus*, *Staphylococcus aureus* showed supreme inhibitory concentration zone in acetone extract and the least zone of inhibition was observed in *Bacillus subtilis* of aqueous extract.

Keywords : Legume., *Lablab purpureus* L., Clinical isolates., Antibacterial activity.

Introduction

Legumes are a broad diversity of crops that are often cultured for food and feeds which produce seeds in pods (Lewis *et al.*, 2005). *Lablab purpureus* Linn. is a tall nearly glabrous twining, perennial or annual herb, with smoothy or downy stem. It is cultivated throughout India and tropical and temperate regions of Asia, Africa, and America. (Chopra *et al.*, 1956). The life-threatening infections is one of the most important causes for hospital and community acquired infections and is a common cause of secondary infection in burn patients leading to the mortality (Beheshti, 2011). Hence, there is always a need for search of antimicrobials from natural sources, from plant *Lablab purpureus*. It is distributed all over the world especially in China, Philippines, Bangladesh, Myanmar, India, and Malaysia (Revazishvili *et al.*, 2006). Different parts such as flower, stem, and leaf of the plant have shown to possess various medicinal and pharmacological properties such as anti-inflammatory, antioxidant, antimicrobial, cyto-toxic, insecticidal, anti-obesity, anti-diabetic, immune modulatory, and hypolipidemic (Hoerlle *et al.*, 2009).

Worldwide, infectious diseases emanating from microorganisms such as bacteria, fungi, viruses, and parasites are hazardous to communal health due to the growing resistance of many microorganisms to currently accessible antibiotics. The extensive use of antimicrobial substances in medical, agricultural, and veterinary practices is a topic of great concern to clinical microbiologists all over the world due to the growing emergence of unscrupulous microorganism strains resistant to drugs that cause serious

infections (Marchaim *et al.*, 2012; Yim *et al.*, 2013). Therefore, there is a growing interest in developing new strategies for inhibiting the growth or survival of microorganisms based on the screening of new sources of natural complexes as alternatives to commercially available drugs (De Brito Marques Ramos *et al.*, 2014).

Materials and Methods

The present work was carried out to find the efficiency of *Lablab purpureus* pod against the selected clinical isolates using the solvent extracts Acetone, Ethanol, Aqueous and Petroleum ether. The selected legume Pod of *Lablab purpureus* were collected from the home garden of the senior author at Agasteeswaram, Kanyakumari District, Tamil Nadu. The Pods were washed and dried in shade about a month, then pulverized in a grinder and stored in a sterile container for use.

Test Organisms

For antibacterial study, four strains of clinical isolates, *Escherichia coli* (MTCC 1652), *Staphylococcus aureus* (MTCC 3160), *Klebsiella pneumoniae* (MTCC 7162), *Bacillus subtilis* (MTCC 5981) were selected and obtained from MTCC, Chandigarh. The extraction was done by Soxhlet apparatus techniques. The bacterial strains were maintained on Nutrient Agar (NA).

Nutrient Broth Preparation

Pure culture from the plate were inoculated into Nutrient Agar plate and sub-cultured at 37°C for 24 hours. Inoculum was prepared by aseptically adding the fresh

culture into 2 ml of sterile 0.145 mol/L saline tube and the cell density was adjusted to 0.5 McFarland turbidity standard to yield a bacterial suspension of 1.5×10⁸ cfu/ml. Standardized inoculum was used for Antibacterial test.

Antibacterial Test

Antibacterial test was carried out by using Disc Diffusion Method. (Bauer *et al.*, 1966). The medium was prepared by dissolving 38g of Mueller-Hinton Agar Medium (Hi-Media) in 1000 ml of distilled water. The liquified medium was autoclaved at 15 Lbs pressure at 121°C for 15 minutes (pH 7.3). The autoclaved medium was cooled, mixed well, and poured in to Petri plates (25ml/plate). The plates were swabbed with moribific bacterial culture viz. *Escherichia coli*, *Klebsiella pneumoniae*, *Bacillus subtilis* and *Staphylococcus aureus*. Finally, the Sample loaded disc was then placed on the surface of Mueller-Hinton Agar medium. The standard drug Amikacin 30 mcg concentration

disc was used for positive control and empty sterile disc was used for negative control. The plates were kept for incubation at 37°C for 24 hours. At the end of incubation, inhibition zones were examined around the disc and measured with transparent ruler in millimetres. The size of the zone of inhibition (including disc) was measured in millimetres. The absence of zone inhibition was interpreted as the absence of activity (Kohner *et al.*, 1994; Mathabe *et al.*, 2006). The experimentation was repeated triplicates.

Results

The antibacterial activity of *Lablab purpureus* using Acetone, Ethanol, Petroleum ether and Aqueous extract against bacterial isolates, gram positive *Staphylococcus aureus*, *Bacillus subtilis* and gram-negative *E. coli* and *Klebsiella pneumoniae* were studied. The results obtained were presented in Table-I, Plate-I and Figure-I.

Table I : Antibacterial activity of *Lablab purpureus* Pod against Clinical isolates.

S/N	Clinical Isolates	Zone of Inhibition (mm)				
		Amikacin	Acetone	Ethanol	Petroleum ether	Aqueous
1	<i>Staphylococcus aureus</i>	17.66 ± 0.47	18.00±0.81	16.33±0.94	14.33±0.47	17.33±0.94
2	<i>Bacillus subtilis</i>	25.33±0.47	17.33±0.47	12.33±0.94	15.66±0.94	11.66±0.47
3	<i>Klebsiella pneumoniae</i>	18.33±0.47	13.66±0.47	14.00±0.81	12.66±0.47	15.33±0.47
4.	<i>E. coli</i>	18.00±0.81	15.00±0.81	16.66±0.94	14.66±0.94	14.33±0.47

*Each valve is a mean of three data. *mm- Millimeter

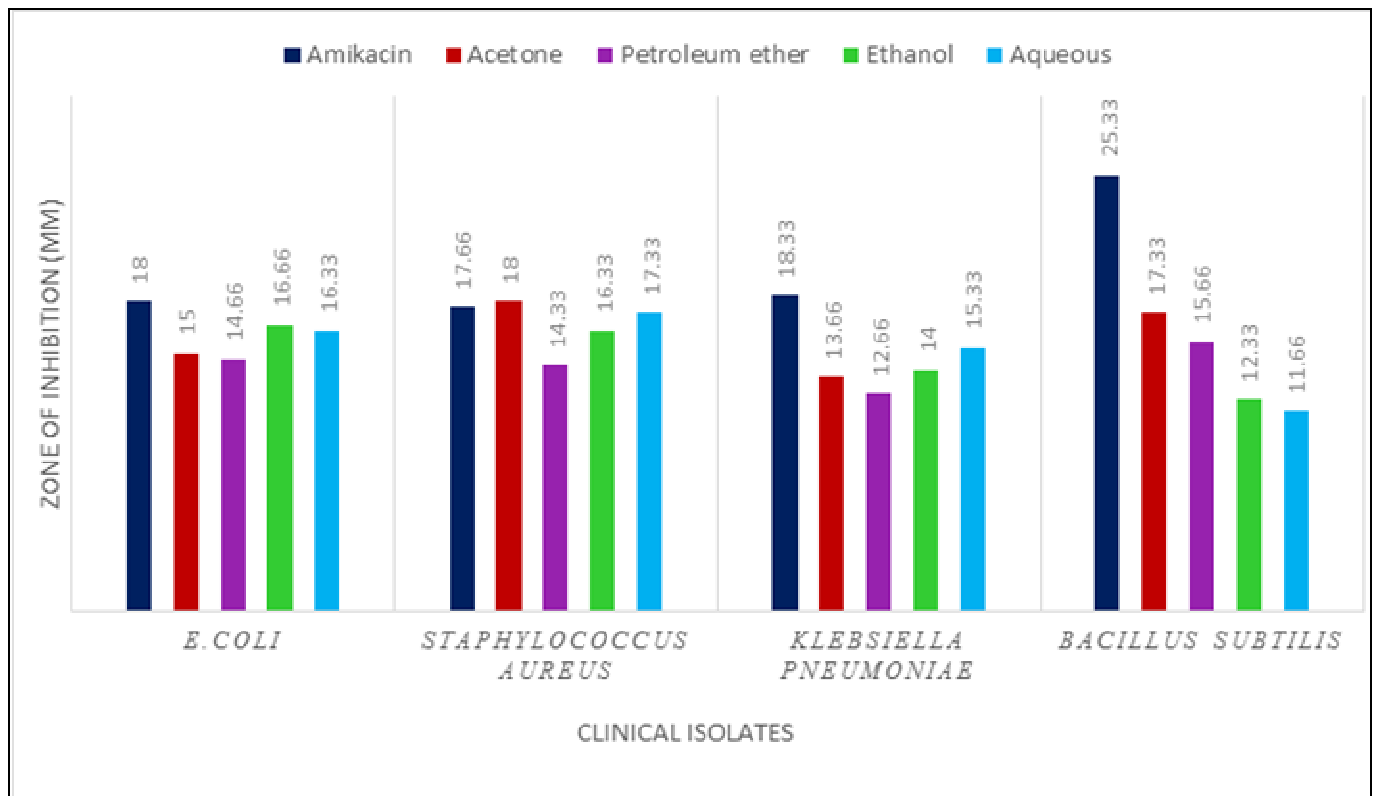
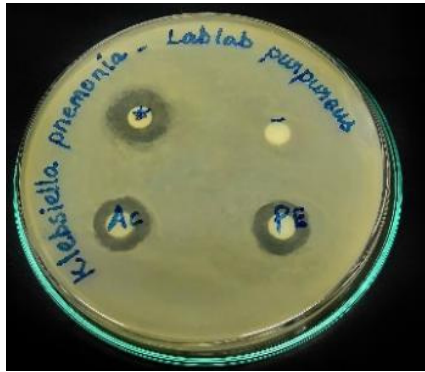
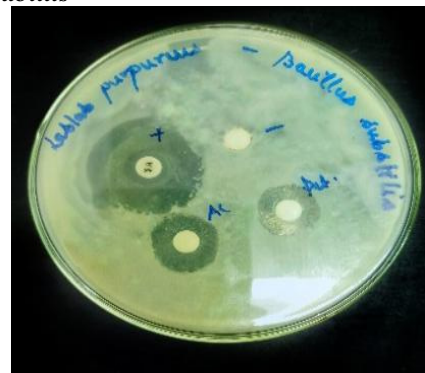


Fig. 1 : Antibacterial activity of *Lablab purpureus* Pod against clinical isolates.

Klebsiella pneumoniae*E. coli**Bacillus subtilis**Staphylococcus aureus*

(+)- Amikacin, P.E - Petroleum ether, Eth- Ethanol, Ac- Acetone, Aq-Aqueous
Plate I : Antibacterial activity of *Lablab purpureus* Pod against clinical isolates.

The Acetone extract of *Lablab purpureus* showed highest zone of inhibition against the isolate *Staphylococcus aureus* (18.00 ± 0.81) followed by *Bacillus subtilis* (17.33 ± 0.47) and lowest zone of inhibition against the

isolates *E. coli* (15.00 ± 0.81) and *Klebsiella pneumoniae* (13.66 ± 0.47). The Ethanol extract of *Lablab purpureus* showed maximum inhibition against the isolates *E. coli* (16.66 ± 0.94) and *Staphylococcus aureus* (16.33 ± 0.94)

followed by *Klebsiella pneumoniae* (14.00±0.81) and lowest zone of inhibition against the isolate *Bacillus subtilis* (12.33±0.94). The Petroleum ether extract of *Lablab purpureus* showed maximum inhibition against the isolate *Bacillus subtilis* (15.66±0.94) followed by *E. coli* (14.66±0.94) and *Staphylococcus aureus* (14.33±0.47) and lowest zone of inhibition against *Klebsiella pneumoniae* (12.66±0.94). The Aqueous extract of *Lablab purpureus* showed maximum zone of inhibition in *Staphylococcus aureus* (17.33±0.94) followed by *Klebsiella pneumoniae* (15.33±0.47) and *E. coli* (14.33±0.47). The lowest inhibition zone in *Bacillus subtilis* (11.66±0.47). In the four solvents, *Lablab purpureus* showed the highest zone of inhibition in *Staphylococcus aureus* (18±1.41) of acetone extract and lowest zone of inhibition in *Bacillus subtilis* (11.66±0.47) in aqueous extract.

Discussion

Plants have been considered as an excellent source of medicinal mediators for thousands of years. Biologically active compounds present in the medicinal plants have always been of great attention. The clinical efficiency of many prevailing antibiotics is being threatened by the emergence of multi drug, resistant pathogens. Mandal *et al.* (2011) showed antibacterial efficiency of aqueous and methanol extract of leaves of *Lablab purpureus*. Abule *et al.* (1995) found inhibitory effect of stem bark extract of *Lablab purpureus* against a panel of gram-positive and gram-negative bacteria. Makembe *et al.* (1996) found inhibitory activity of fresh and dry flower extract of *Lablab purpureus* against methicillin resistant *Staphylococcus aureus*. Therefore, these results can give an indication of feasible antibacterial activity against the clinical isolates of *Lablab purpureus* extracts.

Conclusion

The results obtained from this study considering the bacterial activity using assay of different solvent extracts from the legume *Lablab purpureus* clearly revealed substantial antibacterial activity in contradiction of all the tested human clinical isolates. Additional studies are to be carried out to insulate active principles from the plant materials and to determine their inhibitory activity.

Acknowledgement

I am highly thankful to the Department of Botany and Research Centre, Scott Christian College for providing all facilities for this research work.

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