

ANTIMICROBIAL ACTIVITY OF PETROLEUM ETHER, CHLOROFORM AND ETHANOLIC EXTRACTS OF *CAJANUS SCARABAEOIDES* (L) WHOLE PLANT.

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Summary

Bioassays for antimicrobial activities were carried out using whole plant of *Cajanus Scarabaeoides* (L). Crude petroleum ether, chloroform and ethanolic extracts from whole plant of *Cajanus Scarabaeoides* (L) were prepared and tested against gram positive bacteria i.e. *Bacillus subtilis* and *Staphylococcus aureus*, gram negative bacteria *Pseudomonas aeruginosa* and *Escherichia coli* and fungi *Aspergillus niger*. Both the chloroform and ethanolic extracts showed considerable activity against all the test organisms while petroleum ether extracts showed no activity against any microorganisms (upto 5000 µg/ml). The Minimum Inhibitory Concentration MIC of the plant extracts ranged from 0.01 mg/ml to 100 mg/ml. The antibacterial and antifungal activities of both the ethanolic and chloroform plant extracts were comparable to those of selected antimicrobial chemical suggesting their potential in the treatment of infections caused by these microorganisms. Results showed that the ethanolic have best antimicrobial activities. It can be concluded that antimicrobial activities of the whole plant *Cajanus Scarabaeoides* is due to the phenolic compound present mostly in the ethanolic extract.

Key words: Bioassays, *Cajanus Scarabaeoides* (L), Antimicrobial activity, Agar diffusion, Minimum inhibition Concentration (MIC).

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Introduction

Medicinal plants are gifts of nature to cure limitless number of diseases among human beings ⁽¹⁾. The abundance of plants on the earth's surfaces has led to an increasing interest in the investigation of different extracts obtained from traditional medicinal plants as potential sources of new antimicrobial agents. ⁽²⁾ Researches have shown that all different parts of the plants which include; stem, root, flower, bark, leaves, e.t.c. possess antimicrobial property. In recent years, drug resistance to human pathogenic bacteria has been commonly and widely reported in literature ⁽³⁾. Because of the side effects and the resistance that pathogenic microorganisms build against antibiotics, many scientists have recently paid attention to extracts and biologically active compounds isolated from plant species used in herbal medicines ⁽⁴⁾. Antimicrobial properties of medicinal plants are being increasingly reported from different parts of the world ⁽⁵⁾. The antimicrobial compounds from plants may inhibit bacterial growth by different mechanisms than those presently used. Antimicrobials therefore, may have a significant clinical value in treatment of resistant microbial strains ⁽⁶⁾. In particular, the antimicrobial activities of plant oils and extracts have formed the basis of many applications including raw and processed food preservation, pharmaceuticals, alternative medicine, and natural therapies ⁽⁷⁾. *Cajanus Scarabaeoide* belongs to the Fabaceae family. Locally known as Rantur, Banurkali or Thitkalai. It is traditionally claimed as antidiarrhoeal and antimicrobial drug ^(8,9). The plant is found in the Indian states of Maharashtra and West Bengal as a weed. The whole plant closely resemble like small variety of *Cajanus Cajan* (L). However the work on pharmacological profile of the plant is very less. The photochemical reported in this plant are terpenoids and flavonoids ^(10,11). No known economic use has been reported for this plant. The present investigation was carried out on whole plant of *Cajanus Scarabaeoides* in order to determine the antimicrobial activity of their petroleum ether, chloroform and ethanolic extracts against four bacterial and one fungal isolates.

Materials and methods

Plant materials

The Whole plant of *Cajanus Scarabaeoides*(L) was collected in September 2008 from Vimasankar hill, Maharashtra, India. The whole plant material was taxonomically identified by Dr.S.C.Majumdar, Taxonomist, Botanical Survey of India, Koregaon Road, Pune 411001. The whole plant were dried under shade with occasional shifting and then powdered with a mechanical grinder and stored in an airtight container.

Drugs and chemicals

The following drugs and chemicals were used. Drugs: Norfloxacin(Mankind Pharma Ltd., New Delhi), Chemicals:Petroleum ether (60-80oC) A.R. (SD FINE, Mumbai), Chlorform A.R. (SD FINE, Mumbai), Methanol A.R (SD FINE, Mumbai).

Preparation of extracts

The powder obtained was subjected to successive soxhlet extraction with the solvents with increasing order of polarity i.e. Pet. Ether (50°), Chloroform (50°), methanol (60°). Yield respectively 3.5, 1.19, and 8.94%.The crude extracts were used for bioassay against both gram negative and gram positive bacteria and fungi by agar diffusion method.

Antimicrobial assay:

The antimicrobial activity of *Cajanus Scarabaeoides*(L) extracts was tested against four pathogenic bacteria, viz, gram positive, *Bacillus subtilis* and *Staphylococcus aureus* and gram negative *Pseudomonas aeruginosa* and *Escherichia coli*, while the fungal isolates include *Aspergillus niger*. The agar diffusion method of Garg and Jain⁽¹²⁾ was followed for the antimicrobial assay. Inoculum was prepared from the 24 hours old culture of bacterial isolates in nutrient broth while mycelia plug was cut off from the 48 hours culture of fungal isolates.

Nutrient agar plates were prepared and the inocula were seeded by spread plate method, for the fungal isolates, Potato Dextrose agar were prepared and the mycelia plugs were put at the centre of the prepared plates. The extracts were applied to sterile Whatman No. 1 filter paper discs. All the samples were done in duplicate. Both positive and negative controls were determined, for negative control the two solvents (distilled water and ethanol) were also used to determine their effect on test organisms. Norfloxacin was used to compare the effectiveness of the extracts against bacteria and fungi. After 24 hours of 37⁰c and 48 hours of 25⁰c for bacteria and fungi inoculation, the inhibition zone surrounding the discs by the diffusion of compounds was measured in mm diameter. The minimum inhibitory concentration (MIC) of the extracts against bacteria was also determined. This was done by soaking the paper disc in different concentration of the plant extracts using the same method of agar diffusion. Zones of inhibition in mm were also measured.

Results and discussion

The data revealed that petroleum ether extract of *C.Scarabaeoides* showed no activities against all microbes up to 5000µg/ml concentration. The chloroform extract shows some extant of antimicrobial activity due to the semipolar phyto chemical constituent. Methanolic extract shows good antimicrobial activity due to the presence of phenolic compound in it. Results of preliminary phytochemical tests suggest that petroleum ether extract shows the presence of alkaloids and glycosides, chloroform extract shows the presence of glycosides and steroids and methanol extract shows the presence of glycosides, flavonoids and steroids .This indicates that the antimicrobial principles are polar and semi compounds present in *Cajanus Scarabaeoides*(L).

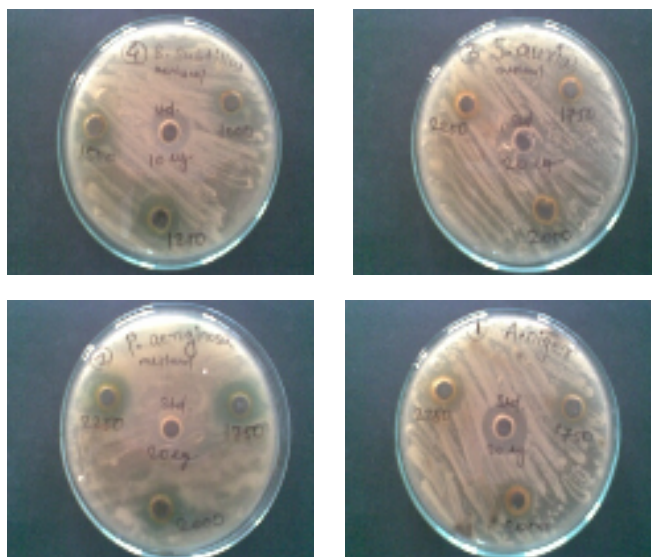


Table 1: Antimicrobial Activity of *Cajanus Scarabaeoides*

A)Pet ether extract: No antimicrobial activity observe up to 5000µg/ml concentration.

B) Chloroform extract:

Microorganism	Zone of inhibition(mm) of extract in µg/ml concentration						Zone of inhibition (mm)of Norfloxacin in µg/ml concentration	
	1000	1250	1500	1750	2000	2250	10	20
<i>Bacillus subtilis</i>	1	2	2	2	3	4	4	6
<i>Staphylococcus aureus</i>	1	1	1	2	2	3	4	5
<i>Pseudomonas aeruginosa</i>	1	1	2	2	3	3	1	2
<i>Escherichia coli</i>	1	1	2	2	3	3	2	3
<i>Aspergillus niger</i>	1	1	2	2	3	4	4	6

C) Methanol extract:

Microorganism	Zone of inhibition(mm) of extract in µg/ml concentration						Zone of inhibition (mm)of Norfloxacin in µg/ml concentration	
	1000	1250	1500	1750	2000	2250	10	20
<i>Bacillus subtilis</i>	3	4	4	4	4	5	4	6
<i>Staphylococcus aureus</i>	1	2	2	3	4	4	4	5
<i>Pseudomonas aeruginosa</i>	2	4	4	4	5	6	1	2
<i>Escherichia coli</i>	2	4	4	5	5	6	2	3
<i>Aspergillus niger</i>	3	3	3	3	4	4	4	6

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