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PRELIMINARY PHYTOCHEMICAL SCREENING OF ANTIBACTERIAL ACTIVITY OF PALMYRA PALM (*BORASSUS FLABELLIFER)* ROOT EXTRACT

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ABSTRACT

The phytochemical constituents of *Borassus flabellifer* was identified and evaluated the antibacterial activity of the acetone, benzene, chloroform, ethanol and methanol extracts. A phytoconstituent analysis of alkaloids, glycosides, terpenoids, steroids, flavonoids, tannins, phenols and saponins were performed on the various solvent extracts. Antibacterial activity was analyzed by agar well diffusion method against five pathogenic microorganisms, *E. coli, S. aureus, K. pneumonia, P. aeruginosa* and *B. subtilis.* The maximum zone of inhibition was exhibited for *E.coli* (14mm); *P. aeruginosa* (13mm) and *B. subtilis* (11mm)against compared to tested micro organisms. The methanolic extract of the palm root has showed consistently significant inhibitory activity on different bacterial species tested. Furthermore, identification of bioactive compounds will be analyzed.

INTRODUCTION: *Borassus flabellifer* L, belongs to family Arecaceae, commonly known as Palmyra palm is a native of tropical Africa but cultivated and naturalized throughout India [1]. The different parts of the *B. flabellifer* are being used for medicinal properties viz. Male flowers are used for anti-inflammatory activity [2], the juice from flowering stalks used for diabetes [3].

Other than these pharmacological uses the juice of the plant is used in preparation of health drinks, jellies etc. The leaves are used to make baskets, hats and many other useful items [4]. The midrib of the leaves and the fibers from the stalks are used in making brushes and base of young leaf stalks is used for straining the Toddy and for making torches [5].

B. flabellifer contains gums, albuminoids, fats and the fresh pulp is reportedly rich in vitamins A and C [6]. The fresh sap is a good source of vitamin B-complex [7]. Studies have indicated that diets rich in fruits and vegetables and those of selected natural antioxidants such as plant poly-phenols, vitamin C and flavonoids are correlated with reduced incidence of cardiovascular and chronic diseases and certain cancer [8]. Hence, the present study was carried out on phytochemical screening and the antibacterial activity of *Borassus flabellifer* in Krishnagiri district.

MATERIALS AND METHODS

Collection of Roots: Palmyra palm roots were collected from Krishnagiri District, Tamil Nadu. It was peeled and washed with water. The shoot was removed and then dried under the shade. It was coarsely powdered using a mixer. The powdered materials were packed in aluminum pouch and stored in atmospheric condition.

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Solvent Extraction: Solvent systems used for the extractions were acetone, chloroform, benzene and ethanol, n-butanol and petroleum ether. 10g of the powered samples were packed in thimble and used for extraction by soxhlet apparatus at a temperature below the boiling temperature of each solvent. A portion of the powdered plant samples was soaked in the conical flask containing solvent, wrapped with aluminum foil and placed in rotary shaker at 120-130 rpm for 48 h. The extracts were filtered using Whatman filter paper No. 1. The solvent was evaporated and the residue was dissolved in sterile dimethylsulfoxide (DMSO-9:1) in 50 mg/ml concentration. The extract was filtered using 0.22 micro filters (Type GV- Millipore) and stored at 4°C for further study.

Phytochemical Screening: Phytochemical screening was performed in all extracts. Alkaloids test was performed by Meyer's tests, amino acids by ninhydrin, carbohydrates by Barfoed's and Fehling tests, flavonoids by $FeCl_3$, glycosides by Legal test, saponin by alcoholic vanillin test, tannins by $FeCl_3$ and lead acetate & triterpenoids by Libermanan-Burchard's test. The test for tannins, phenol, was also carried out as in literature [9, 10].

Test Microorganisms: The five bacterial cultures of both gram-positive and gram-negative bacterial strains used for screening *Escherichia coli, Klebsiella pneumonia, Bacillus subtilis, Staphylococcus aureus* and *Pseudomonas aeruginosa* **.**The stock cultures were maintained in nutrient agar (NA) slant at 4°C and sub-cultured monthly. Working cultures were prepared by inoculating a loopful of each test microorganism in 3 ml of nutrient broth (NB) from NA slants. Broths were incubated at 37°C for 12 hours. The suspension was diluted with sterile distilled water to obtain approximately 10^6 CFU/ml.

Screening for Antibacterial Activity: Disk diffusion: 5 mm of sterile disks were incorporated in 100 µl of root extracts (5 mg/disc). The disk was completely saturated with the extract and allowed to dry. Mueller Hinton (MH) agar plates were swabbed with test bacteria and six extract disks with one of the standard positive control disks (Penicillin) was placed on the MH agar plate. DMSO was taken as the negative control. Plates were incubated overnight at 37°C.

RESULTS AND DISCUSSION

From the phytocontent screening of *B. flabellifer* root extracts, alkaloids, glycosides, steroids, flavanoids, saponins, tannins, phenols and triterpenoids were found to be present study (**Table 1**).

Table 1: Phytochemical constituents of *Borassus flabellifer* Root

Phytochemical Constituents	Acetone	Benzene	Chloroform	Ethanol	Methanol
Alkaloids	-	-	-	-	-
Glycosides	-	-	-	-	-
Carbohydrates	+	+	+	-	-
Tannins and phenols	-	-	-	+	+
Steroids	+	+	-	+	+
Flavanoids	-	-	-	+	+
Saponins	+	-	+	+	+
Triterpenoids	-	-	+	-	-

(+) Present; (-) Absence

Among all the solvents, acetone and benzene were evaluated in carbohydrates, steroids and saponins. The chloroform extract showed the presence of carbohydrates, saponins and triterpenoids. The ethanol root extract showed the presence of steroids, saponins, tannins and phenolic compounds. Methanolic extract exhibited saponins, steroids, flavonoids, tannins and phenolic compounds, which may involve in showing antibacterial activity.

Phytochemical screening of *Borassus aethiopum* extracts revealed the presence of terpenoids, steroids & saponins. All of these compounds have been shown to be potent antioxidants [11, 12, 13 & 14]. The antibacterial activity of methanol extract of *B. flabellifer* root extract against *E. coli, S. aureus,* K. *pneumoniae, P.aeruginosa and B. subtilis* were represented (**Table 2**).

TABLE 2: THE ANTIBACTERIAL ACTIVITY OF *BORASSUS FLABELLIFER* ROOT BY DISC DIFFUSION METHOD

Test microorganism	Zone of inhibition(mm diameter)						
	Positive control	Solvent control	Acetone	Benzene	Chloroform	Ethanol	Methanol
E. coli	13	-	8	5	9	10	14
S. aureus	14	-	10	6	8	11	11
K. pneumoniae	12	-	11	5	8	14	9
P. aeruginosa	11	-	-	7	7	12	13
B. subtilis	13	-	8	8	6	-	11

Concentration of extract- 5 mg/disc, (-) - No zone of inhibition observed, Positive controls –Penicillin (10 µg/ml), Solvent control - 10% DMSO

Penicillin, Standard antibiotic was used for comparing with the root extracts. In comparison with other solvents used in the present study, methanol and ethanol extracts of roots were given better efficiency and showed significant antibacterial activity against all the studied micro-organisms. The root extracts have showed different degrees of antibacterial activity ranging from 8 to 14mm against studied micro-organisms. The antibacterial activity of the root extracts showed magnitude of inhibition patterns in comparison with standard positive control.

Overall, 5 microorganisms, Similarly all the extracts were found to determine the zone of inhibition against, Wherein the methanol and ethanol extracts of palmyra palm root extract exhibited maximum zone of inhibition against *E. coli* (i.e., 14mm); *P. aeruginosa* (13mm) and *B. subtilis* (11mm). The zone of inhibition is in accordance with increase in the concentration of the tested extract. Therefore, a systematic approach should be made to find out the efficacy of bioactive compounds from plants against pathogenic organisms. Some drugs are showed potential against general laboratory used test micro-organisms but less effective against pathogenic organisms.

CONCLUSION: The methanol extracts of root *Borassus flabellifer* exhibited significant antibacterial activity. Further, to isolate bioactive compounds which is present in *Borassus flabellifer* antibacterial activities and therapeutically potential.

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