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Evaluation of Antimicrobial Activity of *Balanites Roxburghii* Planch Fruit Extracts

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Abstract
Evaluation of antimicrobial activity was performed by cup-plate method. The test microorganisms used for the antimicrobial activity were four bacterial species (two Gram positive and two Gram negative) – *Bacillus subtilis, Staphylococcus aureus, Pseudomonas aeuroginosa,Escherichia coli*. The test microorganisms used for the antimicrobial activity were four bacterial species (two Gram positive and two Gram negative) – *Bacillus subtilis, Staphylococcus aureus, Pseudomonas aeuroginosa, Escherichia coli*.

Keywords: ***Balanites roxburghii* Planch.,** Antioxidant, DPPH, Reducing power, β-carotene

Introduction[1,2,3,4,5,6,7,8,9]

Synonyms : Balanites aegyptiaca (Linn.) Delile
Biological Source: It contains whole plant of *Balanites roxburghii* Planch.
Family: Balanitaceae, Simaroubaceae.
Part used: Leaves, Stem, Fruits, Seeds
Scientific Classification
Division : Magnoliophyta
Class : Magnoiiatae or Dicotyledons
Subclass: Rosidae
Superorder: Rutanae
Order : Gcraniales
Family : Balanitaceae
Vernacular names
Sanskrit : Angavriksha
Hindi : Hingan, Hingoli.
Kannada : Ingalore, Ingalkai
Marathi : Hingonbet
Malyanam: Nanjunta
Gujarati : Ingoriyo
Telugu : Gara
Oriya : Ingudihala
Bengali : Hingon
Literature survey reveals that Balanites roxburghii Planch. is shrub or small evergreen tree rarely reaching 9 m. Young parts pubescent Twings armed with stout auxiliary or supra auxiliary spines, 1-6cm long, which often bears leaves or flowers. Fruits are an ovoid drupe, 2.5-6 cm long on a short thick, stalk, faintly 5-grooved, pale yellow when ripe, pulp 5mm thick, with an offensive greasy smell; stone hard, fibrous. It produces date-like fruits. The fruit is an ovoid drupe with a sweet pulp possessing an unpleasant odour. The stone encloses a single oily seed. Fruit Pulp contains five steroidal saponins, designated as balanitism A, B, C, D and E. Diosgenin content of the fruits varies from 0.3 to 3.8 %. Two furostanol glycosides and 6-methyl diosgenin were also obtained from the fruits.

Ethnomedical uses of Fruits:

Unripe ones are Anthelmintic and cathartic, useful curing skin diseases and whooping cough when ripe; massage on chest with the pulp mixed with goat-milk is known to be highly beneficial in the treatment of pneumonia in children. The pulp of the fruit is edible and is reported to be used for cleaning silk and cotton. It contains some Saponin and is not astringent. In Indian indigenous medicine the fruit is also considered useful for boils, leucoderma and other skin diseases. The African Arabs use the pulp as a fish poison. In Uganda the oil is used as a remedy for sleeping sickness and in Spain as a purgative. The seed kernels have long been known to contain a water-soluble Saponin which is toxic to cold-blooded animals. The fruits are reported to be excellent corrosion-inhibitors for brass, and aluminum alloy in acidic solution. Protracted administration of fruit pulp extract produced hyperglycemia-induced testicular dysfunction in dogs.

Materials and Method:[10,11,12,13]

Preparation of extract:

Methanolic extracts of fruits prepared by successive solvent extraction method in a Soxhlet extractor were used for the screening of antimicrobial activity.

Microorganisms used:

The test microorganisms used for the antimicrobial activity were four bacterial species (two Gram positive and two Gram negative) – *Bacillus subtilis,Staphylococcus aureus, Pseudomonas aeuroginosa,Escherichia coli*. The stock cultures were maintained on nutrient agar medium at 4ºC. The microorganisms were activated by inoculating a loopful of the strain in the nutrient broth (25ml).

Antimicrobial activity:

Evaluation of antimicrobial activity was performed by cup-plate method. Sterile Muller Hinton agar media was poured in sterile Petri plates under aseptic conditions. The test organisms 0.1 ml was spread on agar plates. Cups were made at the size of 6 mm diameter, in the agar plates using the sterile borer. Streptomycin (10 µg/disc) was served as reference standard. The disc (6 mm in diameter) was impregnated with 10 µl of each of 125 mg/ml (1.25 mg/disc), 250 mg/ml (2.5 mg/disc) and 500 mg/ml (5 mg/disc) methanol extracts of fruits of the *Balanites roxburghii Planch***.**. The plates containing bacterial strains and standard were maintained at 37±0.5ºCfor 1 h to allow the diffusion of solution into the medium. All the plates containing bacterial strains were incubated at 37°C±0.5ºCfor 48 h. The zone of inhibition (mm) was calculated by measuring the diameter of zone of bacterial growth around the cup. The average of three independent determinations was recorded.

Statistical analysis:

The values are represented as Mean ± S.E.M. for triplicate sets of experiments and the statistical significance was evaluated by One-way analysis of variance (ANOVA) followed by Dunnett's t-test.

Result and Discussion:

The results of antibacterial activity are shown in Table 1 and Figure 1

Table 1: Antimicrobial activity of methanol extracts of fruits of *Balanites roxburghii Planch*.

Sample	***Conc.***	***Mean ± SEM of diameter of zone of inhibition(in mm)***			
		Gram positive bacteria		***Gram negative bacteria***	
		B. subtilis	***S.aureus***	***E.coli***	***P.aeuroginosa***
Methanolic extract (a)	1.25 mg/disc	6.5±0.12	7.2±0.24	6.9±0.12	7.5±0.18
Methanol extract (b)	2.5 mg/disc	16.3±0.26	17.0±0.23	16.8±0.19	17.4±0.15
Methanol	5	20.0±0.38	19.3±0.33	20.6±0.17	20.8±0.14

extract (c)	mg/disc				
Streptomycin (d)	10 µg/disc	24.7±0.11	23.7±0.18	25.2±0.18	25.6±0.20

Values are expressed as Mean ± S.E.M. of triplicate measurements. A value of P<0.05 was considered statistically significant (By one way ANOVA, followed by Dunnett's t-test).

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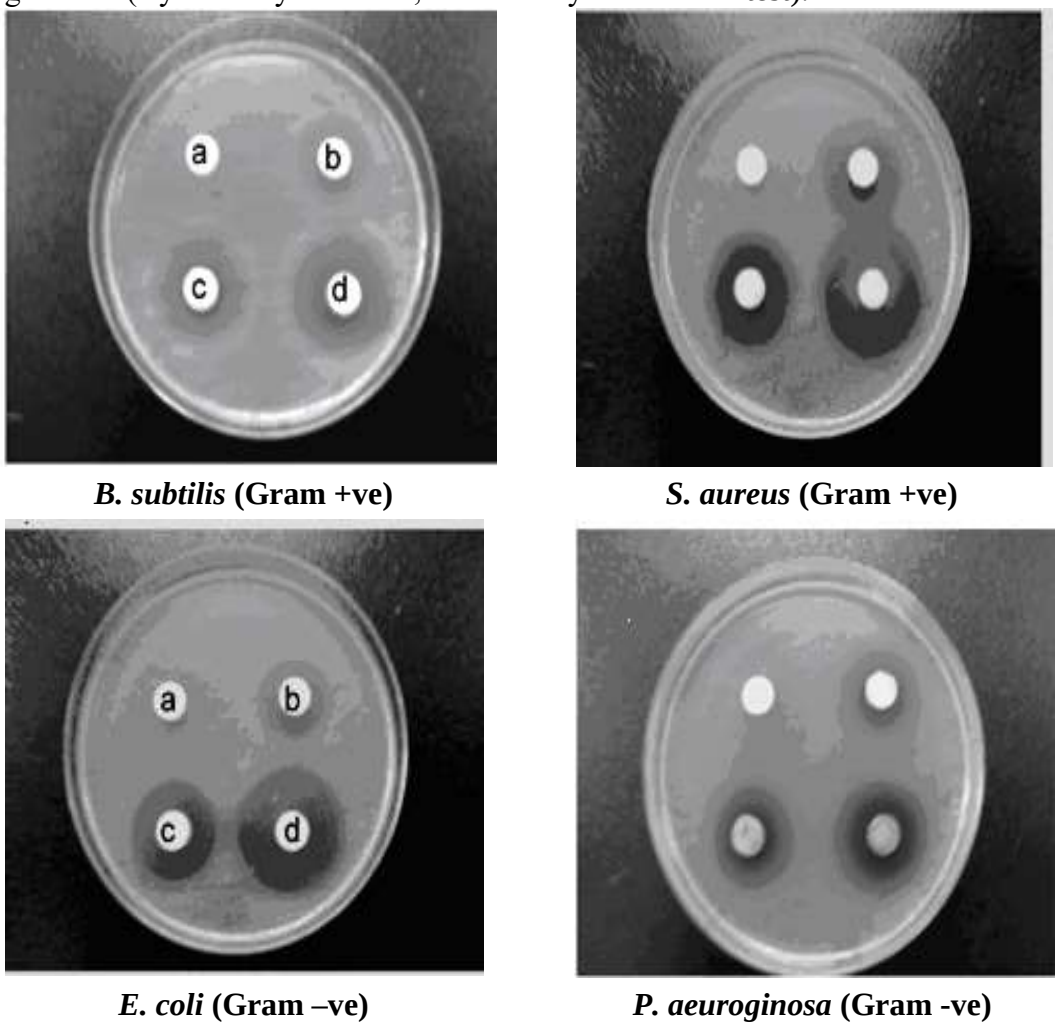

***B. subtilis* (Gram +ve)** ***S. aureus* (Gram +ve)**

***E. coli* (Gram –ve)** ***P. aeuroginosa* (Gram -ve)**

Figure 1: Antibacterial activity of methanol extracts of fruits of *Balanites roxburghii Planch*. with Gram positive bacteria and Gram negative bacteria

Methanolic extract of plant shows antimicrobial activity against multiple gram positive and gram negative strains.

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