



Preliminary Phytochemical Screening and Antimicrobial Activity of Aqueous and Methanolic leaf extracts of *Balanites aegyptiaca* (L.).

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Abstract

In order to scientifically justify the traditional medicinal use of *Balanites aegyptiaca* leaves, the plant was screened for the presence of some phytochemical constituents and the antimicrobial activity of the aqueous and methanolic leaf extracts of *Balanites aegyptiaca* was evaluated. The phytochemicals were analyzed using the standard methods of phytochemical analysis, while the antimicrobial activities against *Escherichia coli*, *Pseudomonas aeruginosa*, *Salmonella typhi*, *Staphylococcus aureus*, *Aspergillus flavus* and *Aspergillus niger* were analysed using agar well diffusion method. The result shows the presence of saponins, flavonoids, cardiac glycosides, alkaloids and tannins in both the aqueous and methanolic leaf extracts of *Balanite aegyptica*. Phenols were only detected in the aqueous leaf extract, while terpenoids were only detected in the methanolic leaf extract of *Balanite aegyptica*. Steroids, and anthraquinones were not detected in both the aqueous and methanolic leaf extracts of *Balanite aegyptica*. The results also revealed that the two extracts possess wider antimicrobial activities against the tested organisms at different concentrations. Therefore, both the aqueous and methanolic leaf extracts of *Balanite aegyptica* may provide a target for drug discovery.

Keywords: Medicinal plant, antibacterial activity, *Balanite aegyptica* and phytochemicals.

INTRODUCTION

Plants have been the basis of many traditional medicine systems throughout the world for thousands of years and continue to provide mankind with new remedies. About three quarter of the world's population relies on plants and plant extracts for their healthcare [1]. The useful products obtained from plants directly or indirectly, demonstrate their importance to man. Plants serve as a source of food, medicinal product, energy and shelter to man and his livestock [2]. Medicinal plant is defined as any plant with one or more of its organs containing substance that can be used for therapeutic purpose or which can be used as precursors for the synthesis of antimicrobial drugs [3]. The modern pharmaceutical industry itself still relies largely on the diversity of secondary metabolites in plants and secondary metabolites of which at least 12,000 have been isolated; a number estimated to be less than 10% of the total [4]. The search for plants with antimicrobial activity or potential has gained increasing importance in recent years due to the development of antimicrobial drug resistance and often the occurrence of undesirable side effect of some antibiotics [5].

Balanites aegyptiaca (L.) Del. belongs to the family Balanitaceae. It is commonly known as desert date. It is a multi-branched, evergreen tree distributed throughout the drier parts of India [6]. It is widely grown in the Sudano-Sahelian region of Africa, the Middle East and South Asia [7].

Balanites aegyptiaca is well known for its medicinal effect. It has been reported to be an antihelminthic, a purgative, emetic and can also cure other types of ailments like skin boils, leucoderma, malaria, wounds, colds, syphilis, liver and spleen disorders and aches. The bark of the plant is useful in curing psychiatric disorders, epilepsy, yellow fever, jaundice and syphilis and can also act as a fumigant to heal circumcision wounds. The boiled root of the plant can be used as a soup against stomach pain, anthrax, and the infusion of root also acts as an antidote to snake bite [8].

This research was carried out to reveal the antimicrobial activity and phytochemical potentiality of *Balanites aegyptiaca* leaves in order to verify the claim of its medicinal properties.

MATERIALS AND METHODS

Collection and preparation of plant materials

The leaves of *Balanites aegyptiaca* linn was collected from Aliero, Kebbi State, Nigeria. It was identified in the Biological Science Department, Kebbi State University of Science and Technology, Aliero, and voucher number (291) was obtained. Disease-free leaves were carefully selected, rinsed under running tap water to eliminate dust and other foreign particles and were air- dried under shade for 2 weeks. The dried leaves were ground into fine powder using mortar and pestle.

Extraction of plant material

100g of the powdered leaves sample were weighed and extracted with 500ml each of distilled water and methanol. The process was run for 72hours after which the extracts were filtered using Whatman filter paper. The filtrate was evaporated to dryness using water baths evaporator and kept in a well labelled sterile specimen bottles and stored in a refrigerator at 4^oc until required.

Microorganisms used

The microorganisms used in this susceptibility assay were *Escherichia coli*, *Pseudomonas aeruginosa*, *Samonella typhi*, *Staphylococcus aureus*, *Aspergillus flavus* and *Aspergillus niger*. They were obtained from the Department of Microbiology Kebbi State University of Science and Technology, Aliero.

Qualitative Phytochemical Screening

The two extracts were screened for the presence of major phytochemicals using standard qualitative methods as described [9, 10, 11]. The plant extracts were screened for the presence of tannins, saponins, flavonoids, alkaloids, steroids, terpenoids, phenols, cardiac glycosides and anthraquinones.

Antimicrobial test

Disc diffusion method for antimicrobial susceptibility testing was carried out according to the standard method [12] of assessing the antibacterial and antifungal activities of the plant extracts. Prepared discs containing the various extracts were carefully placed on the inoculated plates using a sterilized forceps in each case. The plates were then turned upside down and incubate at 37^oc for 24hrs in an incubator. After incubating, the inoculated plates were observed for the zone of inhibition (in mm diameter). Standard antibiotic streptomycin was used as positive control.

RESULT AND DISCUSSION

The Phytochemical constituents present in aqueous and methanolic leaves extracts of *Balanites aegyptiaca* was presented on table 1. It revealed the presence of some phytoconstituents in both aqueous and methanolic leaf extracts of *Balanites aegyptiaca*. The result shows the presence of saponins, flavonoids, cardiac glycosides, alkaloids and tannins in both the aqueous and methanolic leaf extracts of *Balanite aegyptica*. Phenols were only detected in the aqueous leaf extract, while terpenoids were only detected in the methanolic leaf extract of *Balanite aegyptica*. Steroids, and anthraquinones were not detected in both the aqueous and methanolic leaf extracts of *Balanite aegyptica*. This might be as a result of different extraction abilities of varied solvents [13].

Table 1: Phytochemical constituents present in aqueous and methanolic leaf extracts of *Balanites aegyptiaca*.

PHYTOCHEMICALS	EXTRACTS	
	Aqueous	Methanol
Saponins	++	+++
Flavonoids	+	++
Steroids	-	-
Phenols	+	-
Terpenoids	-	+
Anthraquinones	-	-
Cardiac glycosides	++	++
Alkaloids	+	++
Tannins	+++	+

Key: + = slightly present ++ = Moderately present; +++ = Highly present; - = Non detected

Table 2: Antibacterial activity of aqueous and methanolic leaf extracts of *Balanite aegyptica*.

Bacteria	Zones of Inhibition (mm)						
	Aqueous extract			Methanolic extract			Amp. (1mg/ml)
	5mg/ml	10mg/ml	15mg/ml	5mg/ml	10mg/ml	15mg/ml	
<i>E. coli</i>	12.5±0.25	13.7±0.25	15.4±0.25	13.4±0.12	14.7±0.30	15.5±0.16	13.3±0.90
<i>P.aeruginosa</i>	13.1±0.21	14.2±0.25	15.3±0.20	0±0.00	0±0.00	0±0.00	13.6±0.57
<i>S. typhi</i>	12.3±0.25	3.5±0.22	15.2±0.25	12.3±0.15	12.7±0.25	14.7±0.25	12.8±0.29
<i>S. aureus</i>	0±0.00	0±0.00	0±0.00	0±0.00	0±0.00	0±0.00	13.8±1.10

Key: Values are mean and standard deviation (n=3), Amp. : Ampiclox (control), 0±0.00 = No activity.

Table 3: Antifungal activity of aqueous and methanolic leaves extracts of *Balanite aegyptica*.

Bacteria	Zones of Inhibition (mm)						
	Aqueous extract			Methanolic extract			Ket. (1mg/ml)
	5mg/ml	10mg/ml	15mg/ml	5mg/ml	15mg/ml	15mg/ml	
<i>A. niger</i>	6.83±0.34	8.50±1.00	9.17±1.60	0±0.00	8.33±1.10	10.2±0.29	7.00±0.50
<i>A. flavus</i>	0±0.00	0±0.00	0±0.00	0±0.00	0±0.00	7.83±0.29	7.80±0.58

Key: Values are mean and standard deviation (n=3), Ket. : Ketokonazole (control), 0±0.00 = No activity.

The antibacterial activity of aqueous and methanolic leaf extracts of *Balanite aegyptica* was presented in table 2. The result shows zone of inhibition against three (3) tested organisms (*E. coli*, *P.aeruginosa* and *Salmonella typhi*) at a concentration of 5mg/ml, 10mg/ml and 15mg/ml while no zone of inhibition was detected in *Staphylococcus aureus* at the same concentration of *Balanite aegyptica* leaf aqueous extract. While the methanolic extract showed zone of inhibition against *E. coli* and *Salmonella typhi* at all the three (3) concentrations and no zone of inhibition was observed against *Pseudomonas aeruginosa* and *Staphylococcus aureus* at all the three (3) concentrations of *Balanite aegyptica* methanolic leaves extract.

The antifungal activity of aqueous and methanolic leaf extracts of *Balanite aegyptica* was presented on table 3. The result shows zone of inhibition against *Aspergillus niger* at all the three (3) concentrations (5mg/ml, 10mg/ml, 15mg/ml) while there was no zone of inhibition against *Aspergillus flavus* at all the three (3) concentration of *Balanite aegyptica* aqueous leaf extract. However, the methanolic leaf extract demonstrated activity the two fungi.

The presence of alkaloids, saponins, terpenoids and tannins in the leaves extracts of *Balanite aegyptica* has medicinal implications. These phytochemicals are known to be biologically active. Tannins were found to play a role in antifungal, antibacterial, astringent and antibiotic activities [14, 15]. Tannins were also found to form irreversible complexes with proline-rich proteins leading to the inhibition of the cell protein synthesis.

In addition to antimicrobial activity exhibited by tannins, they also react and form complex with proteins to provide the typical tanning effect. This is important medicinally for the treatment of inflamed or ulcerated tissues [16]. Tannins-containing herbs as their main component are astringent in nature and are used in the treatment of intestinal disorders such as diarrhoea and dysentery, thus exhibiting antimicrobial activity. One of the largest groups of chemical produced by plants is the alkaloids and their amazing effect on humans

has led to the development of powerful pain killer medications. Terpenoids also act as antibiotics to protect plants from pathogenic microorganisms [17].

CONCLUSION

The presence of some phytochemical constituents in aqueous and methanolic leave extracts of *Balanite aegyptica* showed that this plant may be used as a basic medicinal agent for curing a number of diseases. The zone of inhibition exhibited by the extract on the tested organisms indicates that the plant can be use as source of antimicrobial agent.

RECOMMENDATIONS

I would recommend the isolation and characterisation of the active components. Also, the mechanisms of action should be investigated.

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