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Phytochemical Profile and Antibacterial Properties of the Fruit and Leaf of the *Momordica dioica* (Roxb.) Willd.

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Summary

This study aims at determination of the antimicrobial activity of the fruit and leaf extracts of *Momordica dioica* on certain pathogenic microbes and screening for substances that may be responsible for these actions. *M. dioica* fruits and leaves were extracted with chloroform, ethanol and distilled water and screened for secondary metabolites. Extracts were found to contain alkaloids, Saponins and cardiac glycolides. The extracts also showed antimicrobial activities against *Escerichia coli, Staphylococcus aureus, Klebsiella pneumoniae* and *Pseudomonas aeruginosa*. The zone of inhibition ranged between 6.5 to 10.33 mm. the inhibitory potentials of the extracts might be ascribed to their content of secondary metabolites. The ability of the extracts to inhibit the pathogens used as indicator organism holds promise for potential application in the pharmaceutical industry.

Key words: Momordica dioica; Phytochemicals; antibacterial; leaf; fruit

Introduction

Momordica dioica (Teasel Gourd) is a dioecious perennial plant with tuberous roots and it is distributed throughout India, Nepal and Bangladesh. The fruit, tuberous roots and whole plant are used in the Indian system of medicine. The leaves are aphrodisiac, anthelmintic; cure 'Tridosha' fever, asthma, bronchitis, hiccups, jaundice eye disease, and injuries. (1, 2). The leaves, fruits and tuberous roots used as folk remedy for diabetes and to cure various illnesses. It is rich in carotene, protein and carbohydrate (3). Fruit contain high amount of vitamin C (4), roots are used for fever (5), piles (6), jaundice, bleeding piles (7). Whole plant is used for injuries (8). In North - Eastern Karnataka young fruits are used as vegetables (personal observation). Various Phytochemicals have been isolated are ascorbic acid, β -sitosterol, triterpenes of ursolic acid, hederagenin oleanolic acid, α -spiranosterol, stearic acid, gypsorenin, momodicaursenol and some aliphatic constituents have been isolated from different part of this plant (9).

Various medicinal plants posses significant antibacterial activity and are used in different alternative systems of medicine successfully *Luffa cylendrica* (10), *Heracleum nepalense* (11), *Leptadenia reticulata* (12) and *Trichosanthes cucumerina* L. var. *cucumerina* (13). Possess significant antibacterial activity. The present investigations were undertaken to find out the antibacterial potentiality of the successive extract of the leaves and fruits against some Gram-positive and Gram-negative bacteria.

Materials and Methods

Collection and identification of plants

The fresh *Momordica dioica* plant and fruits were collected from forest range of Chikli (U) Aurad (B) Taluk, Bidar District. Identified and authenticated with the help of Flora of Presidency of Madras (14), Fascicle of Flora of India (15), Flora of Karnataka (16) and Flora of Gulbarga District (17) and a voucher specimen are deposited in the Herbarium of Botany Department, Gulbarga University, Gulbarga (Vouch. No. HGUG – 803).

Extraction of plant material

50 grams of powdered leaf and fruit material were subjected to the successive soxhlet extraction method using about 500 ml of (Pet. Ether is not considered for test as it is containing oil) chloroform, ethanol (95% v/v) (Qualigens India Ltd.) and Distilled water in order increasing polarity of solvent for a period of 18 hour. The extract obtained were concentrated to dryness *in vacuo* at 40°C and stored at 4°C in the refrigerator until further use.

Preliminary Phytochemical Analysis

The preliminary tests for the detection of secondary metabolites were carried out for all the extracts of *M. dioica* adopting standard methods (18, 19, 20) (Table-1).

Bacterial strains

The indicator bacteria used for this test were obtained from Microbilogy Dept. Gulbarga University Karnataka, India. *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*. All microbial cultivation media were purchased from Qualigens chemicals (India). Streptomycin (Sigma, USA) was checked as positive control.

Media preparation and Antibacterial activity

Successive extraction of plant drug 2mg/ml were screened for their antibacterial activity by agar—well diffusion method (21). The bacteria were inoculated in to nutrient broth and incubated at 37°C in a BOD incubator and the suspensions were checked to approximately to provide 10⁵ cells/ml. From this 200µl of suspension was transferred on to the petriplates containing 20 ml of solidified nutrient agar and then the suspension was spread evenly on the medium with a glass spreader to get a uniform lawn of bacteria. With the help of sterile cork borer wells of 8 mm diameter were made where in, 100µl of the test solution was filled using micropipette. Streptomycin was used as a positive control for antibacterial activity. The plates were incubated at 37±2°C. The zone of inhibition was recorded by measuring the diameter at the end of 24 h.

Results and Discussion

Focus on plant research has increased in recent times, with lot of evidence showing immense potentials of medicinal plants in the field of pharmacology. *M. dioica* has been shown to possess medicinal values (1). In the present study, the results of Phytochemical screening of the leaf and fruit of *M. dioica* is shown in table -1.

Table-1: Phytochemical constituents of the extracts of M. dioica

Phytochemicals	CLE	CFE	ELE	EFE	WLE	WFE
Alkaloids						
Dragendorff's test	+	+	+	+	+	+
Wagner test	+	+	+	+	+	-
Flavonoids						
NaOH test	+	+	+	+	-	-
Shinoda test	-	+	+	+	-	-
Phenols						
Ellagic test	+	+	+	-	+	-
Saponin						
Foam test	+	+	+	+	+	+
Tannin						
Gelatin test	+	+	-	-	+	+
Cardiac glycoside	+	-	+	-	+	+

^{+:} Present, -: Absent, CLE: Chloroform leaf extract, CFE: Chloroform fruit extract, ELE: Ethanol leaf extract, EFE: Ethanol fruit extract, WLE: Water leaf extract, WFE: Water fruit extract,

The presence of secondary metabolites have been proven to be medicinal in nature as they have various protective and therapeutic effects essential to prevent diseases and in the maintenance of a state of well being. Alkaloids have been found to inhibit microbial growth by interfering with cell division (22). Different tannins substances are producing good ant-bacterial activity (23). Saponins are other major phytochemicals present in all the extracts of *M. dioica* Saponins are precursors for the synthesis of steroidal drugs such as sex hormone (24).

The leaf and fruit extracts of *M. dioica* shows appreciable inhibitory effect against most of the indicator bacteria. The chloroform extract of leaf and fruits are more potential against the bacteria like *E. coli* and *P. aeruginosa*. Similarly *S. aureus* and *K. pneumoniae* sps. are more susceptible to water extract of leaf and fruits even as compared to standard streptomycin (Table-2).

Table-2: Zone of inhibition (mm) of indicator bacteria by extracts of *M. dioica* at 200 mg⁻¹ concentration

Indicator bacteria	CLE	CFE	ELE	EFE	WLE	WFE
Escerichia coli	9.50	8.33	8.66	7.33	6.66	8.33
Staphylococcus aureus	7.33	6.50	9.33	7.33	8.33	9.00
Klebsiella pneumoniae	7.66	7.33	8.33	8.00	8.00	10.33
Pseudomonas aeruginosa	10.33	9.00	10.66	6.66	7.33	8.00
Streptomycin Std.	9.73	8.00	11.80	11.00	8.00	9.00

Tests in triplicate.

CLE: Chloroform leaf extract, CFE: Chloroform fruit extract, ELE: Ethanol leaf extract, EFE: Ethanol fruit extract, WLE: Water leaf extract, WFE: Water fruit extract, Std.: standard

The extracts had zones of inhibition ranging from 6.5 to 10.33 mm at concentration of 200 mg⁻¹. It is not surprising that there are differences in the antimicrobial effects of two different parts of the same plant. It is quite possible due to lack of antibiotic properties, or the plant extracts may have insufficient concentration so as to be effective (25). Several authors had reported that plant extracts containing chemicals with antibacterial properties had been useful in the treatment of bacterial and fungal infections (26, 27, 28, 29). The extracts were found to have antibacterial activity against gram positive and gram negative bacteria. This is an indication that the extract has the potential to treat infections as a result of gram positive and gram negative bacteria.

The potential for developing antimicrobials from higher plants appears rewarding as it will lead to the development of a phytomedicine to act against microbes. Plant based antimicrobials have enormous with lesser side effects that are often associated with synthetic antimicrobials (30). Continued further exploration of plant derived antimicrobials is needed today. Further research is necessary to determine the identify of the antimicrobial compounds from within these extracts and also determine their full spectrum of efficacy. However, the present study of *in vitro* antimicrobial evaluation of some plants forms a primary platform for further Phytochemical and pharmacological studies.

In conclusion, *M. dioica* leaf and fruit extracts possess a broad spectrum of activity against a panel of bacteria responsible for the most common bacterial diseases. These promissory extracts open the possibility of finding new clinically effective antibacterial compounds.

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