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Antibacterial activity of *Momordica cymbalaria* extracts against some common pathogens

S Jamila Jasmin[1], S Gajendhini[2], Dr. Joys Selva Mary Albert[3], Dr. M Sugumari[1]
[1] Assistant Professor, Department of Microbiology, Sardar Raja Arts & Science College, Vadakkangulam, Tamil Nadu, India
[2] Assistant Professor, Department of Biochemistry, Pushkaram College of Agricultural Science, Pudukkottai, Tamil Nadu, India
[3] Assistant Professor, Department of Microbiology, St. Mary's College, Thoothukudi, Tamil Nadu, India

Abstract
Antibacterial activity of the of *Momordica cymbalaria (*Cucurbitaceae) were tested against different bacteria by agar diffusion method in comparison with standard antibiotics, Ampicillin, Tetracycline, Streptomycin and Gentamycin. The antibacterial activity of petroleum ether, chloroform, methanol and aqueous extract of aerial parts of the plant were studied using *Staphylococcus aureus, Klebsiella pneumoniae*, *Escherichia coli, Pseudomonas aeruginosa* as test organisms. All the extracts were effective against all the four microorganisms. The result reveals that the plant extract has very good inhibitory activity against Gram negative organism when compared to standard antibiotics. The methanol and aqueous extracts of plant has shown significant activity against *K. pneumoniae, E.coli, P.aeruginosa* and *S. aureus*. While all standard antibiotics had a zone of inhibition less than the extracts of *M. cymbalaria* indicating that the plant can fight these organisms effectively and it could be a better alternative to the modern medicine.

Keywords: *Momordicacymbalaria*, Antibacterial, Bacteria, Clinical isolate, *Escherichia coli, Klebsiella pneumoniae, Pseudomonas aeruginosa, Staphylococcus aureus*

Introduction
Infectious diseases represent a critical problem to health and they are one of the main causes of morbidity and mortality worldwide. The increasing prevalence of multidrug resistant strains of bacteria and the recent appearance of strains with reduced susceptibility to antibiotics raises the spectra of untreatable bacterial infections and adds urgency to the search for new infection fighting strategies. Plants provide a valuable material base for the discovery and development of new drugs of natural origin. Contrary to the synthetic drugs, antimicrobials of plant origin are not associated with many side effects and have an enormous therapeutic potential to heal many infectious diseases (Marjorie MC 1999) [1]. In this regard, one such plant which has number of traditional uses is *Momordica cymbalaria*.

It is a perennial herb distributed over tropical parts of Western peninsular, India and well known as *Karchikayee*. It is also found in the states of Karnataka and Andhra Pradesh in India. It is traditionally used as abortifacient (Nadkarni AK and Nadkarni KM 1982) [2]. Methanol extract is reported to have anti-ovulatory, abortifacient and anti-implantation activity (Koneri R *et al.*, 2006) [3]. The extracts and the dried form of fruit and leaves were shown to have antidiabetic, hypolipidemic and anti- hyperglycemic activities (Kameswara RB *et al.*, 2003) [5].

Since there is no report on the antibacterial activity of *M. cymbalaria*, an attempt was made to evaluate its petroleum ether, chloroform, methanol and aqueous extracts by agar diffusion method using *Staphylococcus aureus, Klebsiella pneumoniae, Escherichia coli, Pseudomonas aeruginosa* (Clinical isolate, Bacteria) as test organism.

Materials and Methods
Collection of plant material
The fresh Plant of *M. cymbalaria* (stem, leaves and fruits; Plate 1) were collected from Tirunelveli district. The fresh plant material was dried under shade. Dried plant material was powdered using mechanical grinder and passed through sieve no.60 to get the powder o desired coarseness. Powdered material was preserved in an air tight container.

Extraction procedure
Shade dried plant parts such as stem, leaves and fruits (470 g) were coarsely powdered and subjected to successive solvent extraction using petroleum ether, chloroform, methanol and water by continuous hot extraction (Soxhlet). Each time, the marc (exhausted plant material) was air dried and later extracted with other solvents. All the extracts were concentrated by distilling the solvent in a rotary flash evaporator.

Plate 1: Momordica cymbalaria

Microorganisms and media

Various microorganisms used in the study were: Grampositive bacteria: *Staphylococcus aureus*; Gram negative bacteria: *Escherichia coli, Pseudomonas aeruginosa, Klebisella pneumonia.* Bacterial isolates were the clinical isolates obtained from samples collected from the Rajas Medical Institutions, Kavalkinaru, Thirunelveli. The bacterial stock cultures were maintained on Muller Hinton agar and stored at 4ºC.

Antimicrobial activity

The extracts obtained above were screened for their antimicrobial activity in comparison with standard antibiotics, viz. Ampicillin, Tetracycline, Streptomycin and Gentamycin by disc diffusion method (Greenwood *et al.*, 2002) [8] using *S.aureus, K.pneumoniae, E.coli* and *P.aeruginosa* (Clinical isolate, Bacteria) as test organism. Muller Hinton agar were prepared, sterilized and poured into petriplates up to a depth of 3mm.The organisms were suspended in saline and 0.1ml of organisms (10^{10}colony forming units per ml) were spread on these plates on which wells were made using an 8 mm cork borer. To each well, 100µl of each extracts were added and plates were incubated at 37ºC for 24h for bacteria. After incubating for 24h, the results were recorded by measuring the diameter of zone of inhibition surrounding the well. For standard, antibiotic test-disc method was employed. Standard antibiotic discs of 6 mm diameter (Hi-Media) for different antibiotics such as Ampicillin, Tetracycline, Streptomycin and Gentamycin were used. The experiments were done in triplicate.

Result and discussion

The results of antibacterial activities are given in Tables 1 and 2. From Table 1, it is very clear that all the extracts have shown antimicrobial activity against all tested organisms. The methanol and aqueous extracts of plant has shown significant activity against *K.pneumoniae* (18mm; 19.5mm), *E.coli*(17mm; 17mm*), P.aeruginosa* (19.5mm; 19 mm) and *S. aureus* (15 mm; 17mm). The antibacterial activities of different extracts against test organisms are given below:

Escherichia coli: methanol and aqueous extracts showed maximum zone of inhibition (17 mm) followed by chloroform extract (15 mm), while petroleum ether extract showed less zone of inhibition (13 mm).

Pseudomonas aeruginosa: Methanol and aqueous extracts showed maximum zone of inhibition (19 mm). While petroleum ether and chloroform extracts showed less zone of inhibition (16mm) but equal to that of standard antibiotics (Tetracycline:16 mm).

Klebsiella pneumoniae: Aqueous (19.5 mm) and chloroform extracts (19 mm) showed maximum zone of inhibition followed by ethanol extract (18 mm), while petroleum ether extract showed less zone of inhibition (14 mm).

Staphylococcus aureus: Aqueous extract showed maximum zone of inhibition (17 mm) followed by ethanol extract (15 mm), while chloroform(14.5 mm) and petroleum ether extracts (12.5 mm) showed less zone of inhibition.

Table 1: Antibacterial activity of different extracts of *Momordica cymbalaria* against microorganisms

Test Organisms / Extract	Zone of inhibition in mm			
	P.E	C.E	M.E	A.E
Escherichia coli	13	15	17	17
Pseudomonas aeruginosa	16	16	19.5	19
Klebsiella pneumoniae	14	19	18	19.5
Staphylococcus aureus	12.5	14.5	15	17
P.E= Petroleum ether extract; C.E= Chloroform extract; M.E= Methanol Extract; A.E=Aqueous Extract				

Susceptibility test of these test organisms to traditional antibiotics was done using standard antibiotics such as Ampicillin, Tetracycline, Streptomycin and Gentamycin. The zone of inhibition of the standard antibiotics against the test organism was measured and the results are given in Table 2.

Escherichia coli: Standard antibiotic Ampicillin showed maximum zone of inhibition (17 mm) followed by Streptomycin (13 mm), while Tetracycline (9 mm) and Gentamycin (7 mm) showed less zone of inhibition.

Pseudomonas aeruginosa: Standard antibiotic Tetracycline showed maximum zone of inhibition (16 mm) followed by Ampicillin (13 mm) and Gentamycin (12 mm), while Streptomycin showed less zone of inhibition (11 mm).

Klebsiella pneumoniae: Standard antibiotic Tetracycline showed maximum zone of inhibition (17 mm) followed by Gentamycin (16 mm) and Streptomycin (14 mm), while Ampicillin showed less zone of inhibition (11 mm).

Staphylococcus aureus: Standard antibiotic Streptomycin showed maximum zone of inhibition(13 mm) followed by Ampicillin (12 mm) and Tetracycline (11 mm), while Gentamycin showed less zone of inhibition (9 mm).

Table 2: Antibacterial effect of standard antibiotics on tested microorganisms

Test Organisms / Antibiotics	Zone of inhibition in mm			
	Ampicillin	Tetracycline	Streptomycin	Gentamycin
Escherichia coli	17	09	13	07
Pseudomonas aeruginosa	13	16	11	12
Klebsiella pneumoniae	11	17	14	16
Staphylococcus aureus	12	11	13	09

Summary and conclusion

It can be concluded from the results that *M. cymbalaria* plant extracts possess antimicrobial activity against various test organisms used. Some of the extracts (methanol and aqueous) were more effective than traditional antibiotics to combat the pathogenic microorganisms studied. This possibly means that the compound responsible for the antimicrobial activity is present in each extract at different

concentrations. The chance to find antimicrobial activity was more apparent in methanol and aqueous extracts than in petroleum ether and chloroform extracts. The extracts were found to be effective against Gram negative (*E.coli, K.pneumoniae, P. aeruginosa*) pathogens when compared to Gram positive (*S.aureus*) pathogen. The phytoconstituents present in the extracts may be responsible for the antimicrobial activity. The mechanism is yet to be identified.

The study of antibacterial activity of *M. cymbalaria* on various test organisms may help to discover new class of antibiotic substances that could serve as selective agents for infectious chemotherapy and control. This approach has opened up the possibility of the use of this plant in drug development for human consumption for future use.

Further work is needed to isolate the secondary metabolites from the extracts studied in order to test specific antibacterial activity. This study demonstrated that folk medicine can be as effective as modern medicine to combat pathogenic microorganisms. The millenarian use of these plants in folk medicine suggests that they represent an economic and safe alternative to treat diseases.

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