



Short Communication

Evaluation of Antibacterial Activity of *Tecomella undulata* leaves crude Extracts

Sharma Abhishek^{1*}, Patil Ujjwala¹, Kakkar Shivani² and Bhot Meeta¹

¹Department of Botany, Birla College, Kalyan- 421304, MS, INDIA

²Department of Biotechnology, Birla College, Kalyan- 421304, MS, INDIA

Available online at: www.isca.in

Received 25th March 2013, revised 30th April 2013, accepted 3rd May 2013

Abstract

In the present study antibacterial activity of *Tecomella undulata*, leaves were checked on bacterial pathogens of humans. Hexane, chloroform and methanol extracts were used for the study by agar well diffusion method against seven bacteria. The methanol extract showed higher antibacterial activity in comparison to the other two solvents. Methanolic extract showed higher minimum inhibitory concentration against *Klebsiella pneumoniae* and *Micrococcus luteus*; whereas, less inhibitory effect was noted for chloroform and hexane extracts. Gram-negative bacteria were more susceptible than Gram-positive. Present study validates the traditional use of *Tecomella undulata* in medicine.

Keywords: *Tecomella undulata*, Antibacterial activity, Agar-well diffusion.

Introduction

Tecomella undulata (Family, Bignoniaceae) is commonly known as Ammora (in English) or Rohida and is locally known as honey tree, desert teak, Marwar teak or white cedar. It is widely distributed in Arabia, southern Pakistan and northwest India up to an elevation of 1200 meters. In Pakistan, it is found in Baluchistan and Sind¹. Iridoid Glucoside, naphthoquinone, phytosterols, flavonoid glycoside, flavonol, fatty acid and triterpenoids were identified from different parts of *Tecomella undulata* which are pharmacologically active. It is reputed to be of considerable medicinal value in Ayurveda and traditionally, it is extensively used for treatment of several diseases¹. As no data on anti bacterial activity is available on *Tecomella undulata*, present study was proposed to check antibacterial potential of *Tecomella undulata* against *Bacillus subtilis*, *Enterococcus faecalis*, *Escherichia coli*, *Klebsiella pneumonia*, *Micrococcus luteus*, *Proteus vulgaris* and *Pseudomonas aeruginosa* using hexane, chloroform and methanol leaf extracts.

Material and Methods

The present study was done in the Department of Botany, Birla College, Kalyan. Leaves of *Tecomella undulata* were collected from Kalyan during winter season.

Source of Microorganisms: Pure bacterial cultures were obtained from NCIM, Pune, India and were maintained in nutrient agar (Himedia) table 1.

Extraction procedure: Leaf was washed under running tap water, excess water was drained off, shade dried and finally crushed to fine powder in mixture grinder. The dried powder of

the leaves was allowed to Soxhlet for successive extraction with hexane, chloroform and methanol. The liquid extract were collected, filtered and evaporated to dryness. The extracts were dissolved in respective solvents to reach a low and high concentration (100 mg/ml and 300 mg/ml) and were kept in refrigerator till used.

Table-1

List of tested bacterial strains and their catalogue numbers

Bacteria	Catalogue number
<i>Bacillus subtilis</i>	2545
<i>Enterococcus faecalis</i>	2356
<i>Escherichia coli</i>	2641
<i>Klebsiella pneumoniae</i>	2883
<i>Micrococcus luteus</i>	2704
<i>Proteus vulgaris</i>	2813
<i>Pseudomonas aeruginosa</i>	2026

Antibacterial activity: The antibacterial activity was carried out by agar well diffusion method²⁻⁴. For this, the different solvent extracts of leaf of *Tecomella undulata* have been tested against gram positive and gram negative bacteria to find out the antibacterial activity. A nutrient broth was prepared (beef extract-3gm/L; Peptone-5gm/L; NaCl-5gm/L; Agar-16gm/L) and then sterilized in an autoclave at 15lb pressure for 15 minutes. The sterilized medium was poured into petri dishes. The solidified plates were bored with 6mm diameter sterilized cork borer. The plates with wells were used for the antibacterial studies. Various solvent extracts of leaf of *Tecomella undulata* were subjected to individual microbiological tests to ascertain their antibacterial activity against seven species of microorganisms, *Bacillus subtilis*, *Enterococcus faecalis*,

Escherichia coli, *Klebsiella pneumonia*, *Micrococcus luteus*, *Proteus vulgaris* and *Pseudomonas aeruginosa*. The evaluation of antibacterial activity of the extracts was done by observing the clear zone around disks and measuring the diameter of the zone of inhibition (ZI) exhibited by the extracts around disks in mm by sliding digital vernier caliper.

The Minimum inhibitory concentration (MIC) was determined through the dilution method⁵⁻¹⁰. Different concentrations of leaf extract in hexane, chloroform and methanol (ranging from 0.01 to 5.00 mg/ml) was tested separately for each bacterium and inhibition zone of microbial growth in the plates containing test solutions was judged by comparison with blank control plates. Experiments were performed in triplicate. The results of MIC were interpreted as sensitive (<14 mm), intermediate activity (14-17 mm) and susceptible (>17 mm).

Results and Discussion

The results of antibacterial activity and MIC values of solvent extracts of *Tecomella undulata* leaves are summarized in tables 2 and 3, respectively. Methanolic extract was found to be more potent as compare to n-Hexane and chloroform extracts against all tested bacteria. Hexane extract was found to be resistance against *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and *Micrococcus luteus* for low concentration, while *Klebsiella pneumoniae* showed intermediate activity to its high concentration. *Escherichia coli*, *Proteus vulgaris*, *Bacillus subtilis* and *Enterococcus faecalis* did not show any inhibition

zones. Chloroform extract was found effective against *Klebsiella pneumoniae* and *Micrococcus luteus* showed intermediate activity against *Pseudomonas aeruginosa* irrespective of its concentrations whereas *Escherichia coli*, *Proteus vulgaris* and *Enterococcus faecalis* were resistance to low concentration and evinced intermediate activity to high concentration.

Bacillus subtilis did not show inhibition zone. Methanol extract exhibited promising activity against all tested bacteria for both concentrations. It showed fairly high degree of sensitivity against *Klebsiella pneumoniae* followed by *Micrococcus luteus*. Compared with standard antibiotic, methanol extract showed the high inhibition zone values than ciprofloxacin (10µg). Hence it indicated that methanol extract of *Tecomella undulata* leaves has broad spectrum of antibacterial activity against all tested bacteria. Hexane, methanol and chloroform, a negative control, did not show zone of inhibition and indicated that it is not interfering with the formation of zone of inhibition. From the MIC values, it is observed that methanol extract showed significant antibacterial activity followed by chloroform and hexane extracts table 3. The MIC value of methanol extract was 0.01 mg/ml for *Klebsiella pneumoniae* while 0.1 mg/ml against *Micrococcus luteus* and 1.0 mg/ml for *Enterococcus faecalis*. The MIC value of chloroform extract for *K. pneumoniae* was 1.0 mg/ml, whereas for *M. luteus* it was 3.0mg/ml. The hexane extract exhibited the same value against all tested bacteria, it was >5.0 mg/ml.

Table-2
Inhibitory zones of solvent extracts of *Tecomella undulata* against Gram-negative and Gram-positive bacteria

Solvent Extract	Zone of inhibition (mm)							
	Concentrations (mg/ml)	BS	EF	EC	KP	ML	PV	PA
Hexane	100	Nil	Nil	Nil	12	11	Nil	Nil
	300	Nil	Nil	Nil	14	13	Nil	12
Chloroform	100	Nil	Nil	12	13	19	13	15
	300	16	13	14	23	20	15	17
Methanol	100	18	16	19	25	23	19	20
	300	17	20	21	27	25	21	22
Ciprofloxacin	10µg	Nil	23	15	20	22	21	20
Negative Control		Nil	Nil	Nil	Nil	Nil	Nil	Nil

Table-3
MIC value of *Tecomella undulata* leaf extracts

Bacteria	MIC values (mg/ml)		
	Methanol extract	Chloroform extract	Hexane extract
<i>Bacillus subtilis</i>	4.0	>5.0	>5.0
<i>Enterococcus faecalis</i>	2.0	>5.0	>5.0
<i>Escherichia coli</i>	1.0	>5.0	>5.0
<i>Klebsiella pneumonia</i>	0.01	1.0	>5.0
<i>Micrococcus luteus</i>	0.1	3.0	>5.0
<i>Proteus vulgaris</i>	2.0	5.0	>5.0
<i>Pseudomonas aeruginosa</i>	2.0	4.0	>5.0

In the present study hexane, chloroform and methanol extracts of leaves were found to be the most sensitive against *K. pneumoniae* and *Micrococcus luteus* strains. Among the three extracts, the methanol extract exhibited promising results than chloroform and hexane extracts. Methanol proved as the most effective solvent for extracting broad spectrum of antimicrobial compounds from plants.

Conclusion

In conclusion, the present investigation of antibacterial activity of leaf extracts of *Tecomella undulata* reports that the various solvent extracts of leaf showed the antibacterial activity against bacterial pathogens of both gram positive and gram negative. Thus it can be employed as an antibacterial agent against many bacterial diseases. Further its, chemical characterization will disclose the bioactive phytoconstituents responsible for inhibitory activity of *Tecomella undulata*.

References

1. Jain M., Kapadia R., Jadeja R. N., Thounaojam M. C., Devkar R. V. and Mishra S. H., Traditional uses, phytochemistry and pharmacology of *Tecomella undulata*-A review, *Asian Pacific J. of Tropical Biomedicine*, Supplementary 3, 1918-1923 (2012)
2. Rajendran N.K. and Ramakrishnan J., *In vitro* evaluation of antimicrobial activity of crude extracts of medicinal plants against multi drug resistant pathogens, *Biyoloji Bilimleri Araştırma Dergisi*, 2, 97-101 (2009).
3. Penchala S., Talari S., Guguloth V., Azmeera R. and Nanna R. S., Phytochemical Screening and Antimicrobial Activity of Leaf Extract of *Wrightia tomentosa*, *Int. Res. J. Biological Sci.*, 2(3), 23-27 (2013)
4. Yehouenou B., Sessou P., Houinsou R.L., Noudogbessi J.P., Alitonou G.A., Toukourou F. and Sohounhloue D., Chemical composition and Antimicrobial activities of *Cinnamomum zeylanicum* Blume dry Leaves essential oil against Food-borne Pathogens and Adulterated Microorganisms, *I. Res. J. Biological Sci.*, 1(6), 18-25 (2012)
5. Sinha K. and Verma A.K., Evaluation of Antimicrobial and Anticancer activities of Methanol Extract of in vivo and in vitro grown *Bauhinia variegata* L., *I. Res. J. Biological Sci.*, 1(6), 26-30 (2012)
6. Madhavaraj L., Sethumadhavan V.V., Geun H. G., Mathur N. K. and Si W. K., Synthesis, Characterization and Evaluation of Antimicrobial Efficacy of Silver Nanoparticles using *Paederia foetida* L. leaf extract, *I. Res. J. Biological Sci.*, 2(3), 28-34 (2013)
7. Hegde C.R., Madhuri M., Swaroop T. N., Das A., Bhattacharya S. and K.C. Rohit, Evaluation of Antimicrobial Properties, Phytochemical Contents and Antioxidant Capacities of Leaf Extracts of *Punica granatum* L., *ISCA J. Biological Sci.*, 1(2), 32-37 (2012)
8. Verma V., Bhardwaj A., Rathi S. and Raja R.B., A Potential Antimicrobial Agent from *Cocos nucifera* mesocarp extract; Development of a New Generation Antibiotic, *ISCA J. Biological Sci.*, 1(2), 48-54 (2012)
9. National Committee for Clinical Laboratory Standards (NCCLS), Performance standards for antimicrobial susceptibility testing; Eleventh International Supplement, NCCLS, (2001)
10. Hemlal H. and Subban R., GC-MS, HPTLC and Antimicrobial analysis of Root extracts of *Pseudarthria viscida* Wight and Arn and *Desmodium gangeticum* (Linn) DC, *I. Res. J. Biological Sci.*, 1(5), 57-65 (2012)