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Antimicrobial study of plant extracts of *Datura metel* L. against some important disease causing pathogens

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ABSTRACT

Objective: In this present study antimicrobial activity of aerial parts of *Datura metel* L were evaluated against the resistant pathogens belong to aquatic, human and plant origin. **Methods:** Soxhlet extraction method was used to get the corresponding extracts of hexane, chloroform and methanol. The antimicrobial activities of the organic solvent extracts on the various test microorganisms, including bacteria and fungi investigated using agar well diffusion technique. The length of inhibition zone was measured in millimeters from the edge of the well to the edge of the inhibition zone. Methanol and chloroform extracts exhibited promising antimicrobial activity than hexane extracts. **Results:** The zone of inhibition of chloroform varies from (9 to 18 mm) where as with methanol (11 to 30 mm) at 100 mg/ml and chloroform (11 to 19 mm) and methanol (12 to 35 mm) with 250 mg/ml DMSO concentrations consequently. Among all microorganisms studied *Erwinia caratovara* and *Pseudomonas syringae* showed the considerable growth inhibition with chloroform and methanolic extracts. **Conclusions:** *D. metel* can be used in the treatment of infectious diseases caused by resistant pathogenic microorganisms. Further studies are being carried out in order to separate the individual components that are present in plant extracts of *D. metel* using column chromatography.

1. Introduction

India has a rich heritage of knowledge on plant based drugs both for use in preventive and curative medicine. A country like India is very much suited for development of drugs from medicinal plant. Historically, plants have provided a source of inspiration for novel drug compounds, as plant derived medicines have made large contributions to human health and well-being. The number of multi-drug resistant microbial strains and the appearance of strains with reduced susceptibility to antibiotics are continuously increasing. Medicinal plants represent a rich source of antimicrobial agents [1-20]. Because of the side effects and the resistance that pathogenic microorganisms build against antibiotics, many scientists have recently paid attention to extracts and biologically active compounds isolated from plant species used in herbal medicines [21]. Plants generally produce many secondary metabolites which constitute an important source of microbicides, pesticides and many pharmaceutical drugs. Plants belonging to family solanaceae are distributed world wide, which includes 85 genera and about 2,800 species in the world. The name Datura comes from the early Sanskrit Dustura [22] or dahatura. *Datura metel* L. classified in the plant family Solanaceae. A perennial herbaceous plant, belonging to the Solanaceae family can reach a height of 1.5m. Leaves are simple, alternate, dark green, broadly ovate, shallowly lobed and glabrous. Flowers are large, solitary, and trumpet-shaped with a sweet fragrance usually appreciated in the mornings and evenings, with a wide range of colors, ranging from white to yellow and light to dark purple. The flowers are hermaphrodite and are pollinated by insects. The fruit is in the form of a capsule covered with short spines. A variety of phytochemicals have been found to occur in *D. metel*. These phytoconstituents comprises alkaloids, flavonoids, phenols, tannins, saponins and sterols. The phytoconstituents of Datura were analyzed from various parts of the plant like the leaf [23, 24] root [25] and shoot [26-28]. The plant finds application in the treatment of diarrhea and skin diseases. It is used in the treatment of catarrh, epilepsy, insanity, hysteria, rheumatic pains, hemorrhoids, painful menstruation skin-ulcers and wounds. It is also used in the treatment of burns. It is used to calm cough and to treat laryngitis and Treacheries [29]. Antibacterial studies were done on *D. metel* [30]. Plant extracts have greater potential as antimicrobial compounds against microorganisms and that they can be used in the treatment of infectious

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caused by disease causing pathogens. In this present study antimicrobial activity of aerial parts of *Datura metel* L were evaluated against the resistant pathogens belong to aquatic, human and plant origin.

2. Methods and Materials

2.1 Plant material and extraction:

Datura metel L was taxonomically identified and the Voucher specimen is stored in the department of botany, Andhra University, Visakhapatnam, INDIA. The aerial plant parts were collected from visakkhapatnam, Andhra Pradesh, India. The plant material were dried under shade with occasional shifting and then powdered with a mechanical grinder and stored in an airtight container. The powder obtained was subjected to successive soxhlet extraction with organic solvents with increasing order of polarity i.e. Hexane, Chloroform and Methanol respectively.

2.2 Test microorganisms

Microbial strains of clinical, plant and aquatic origin i.e. Aeromonas hydraophylla (MTCC 646), Alternaria alternate (MTCC 1362), Ustilago maydis (MTCC 1474), Asperigellus niger (MTCC 2723), Acremonium strictum (MTCC 3072), Pencillium expansum (MTCC 2006), Fusarium oxysporum (MTCC 1755), Xanthomonas compestries(MTCC 2286), Erwina caratovara (MTCC 3609), Lactobacillus acidophilus (MTCC 447), Pseudomonas marginalis (MTCC), *Pseudomonas syringae* (MTCC 1604), Pseudomonas aeruginosa (MTCC 1688), Streptococcus mutans (MTCC 890), Steptococcus salivarious (MTCC 1938) and Staphylococcus aureus (MTCC 96) including both fungi and bacteria were procured from Microbial Type Culture Collection (MTCC), Chandigarh. Active cultures were generated by inoculating a loopful of culture in separate 100 mL nutrient/potato dextrose broths and incubating on a shaker at 37oC overnight. The cells were harvested by centrifuging at 4000 rpm for 5 min, washed with normal saline, spun at 4000 rpm for 5 min again and diluted in normal saline to obtain 5 x 105cfu/mL.

2.3 Determination of antimicrobial activity

The crude extracts of the different plant parts of different species were subjected to antimicrobial assay using the agar well diffusion method of [31] modified by [32]. 20 ml of nutrient agar was dispensed into sterile universal bottles these were then inoculated with 0.2 ml of cultures mixed gently and poured into sterile petri dishes. After setting a number 3-cup borer (6 mm) diameter was properly sterilized by flaming and used to make three to five uniform cups/ wells in each petri dish. A drop of molten nutrient agar was used to seal the base of each cup. The cups/wells were filled with 50 μl of the extract concentrations of 100 mg/ ml, 250 mg/ ml, and allow diffusing for 45 minutes. The solvents used for reconstituting the extracts were similarly analyzed. The plates were incubated at 37 ℃ for 24 hours for bacteria. The above procedure is allowed for fungal assays but except the media potato dextrose agar instead of nutrient agar and incubates at 25 ℃ for 48 hours. The zones of inhibition were measured with antibiotic zone scale in mm and the experiment was carried out in duplicates.

3. Results

Table 1: Antimicrobial activity of chloroform and methanol extracts *D. metel*.

In the present study, chloroform and methanol extract exhibited different degree of growth inhibition against tested bacterial and fungal strains. According to Table 1, methanolic extracts of *D. metel* exhibited considerable antimicrobial activity against tested microbial strains. (Table 1) summarizes the antimicrobial activities zone of inhibition of chloroform varies from (9 to 18 mm) where as with methanol (11 to 30 mm) at 100 mg/ml and chloroform (11

Table 1

Antimicrobial activity of chloroform and methanol extracts *D. metel*

Pathogen name	100 mg/ ml DSMO		250 mg/ ml DSMO		
	C	M	C	M	
Aeromonas hydraophylla	17	21	18	21	Zone of inhibition in mm
Alternaria alternate	14	14	16	14	
Asperigellus niger	10	13	12	15	
Acremonium strictum	9	11	11	12	
Erwinia caratovara	17	25	19	30	
Fusarium oxysporum	12	21	14	24	
Lactobacillus acidophilus	12	12	12	14	
Pencillium expansum	10	12	11	14	
Pseudomonas marginalis	12	13	13	15	
Pseudomonas syringae	18	30	19	35	
Pseudomona aeruginosa	12	14	13	16	
Steptococcus mutans	12	12	13	14	
Steptococcus salivarious	13	15	15	18	
Stephylococcus aureus	10	11	12	12	
Ustilago maydis	13	17	15	19	
Xanthomonas compestries	14	18	16	20	

C=Chloroform, M = Methanol

Volume per well: 50 μl, Borer size used: 6mm

to 19 mm) and methanol (12 to 35 mm) with 250 mg/ml DMSO concentrations subsequently. The variation of antimicrobial activity of our extracts might be due to distribution of antimicrobial substances, which varied from fraction to fraction of the crude extract. Among all microorganisms studied *A. hydraophylla, F. oxysporum, E. caratovara* and *P. syringae* showed the considerable growth inhibition with chloroform and methanolic extracts with all considerations.

4. Discussion

Plant based antimicrobial compounds have enormous therapeutical potential as they can serve the purpose without any side effects that are often associated with synthetic antimicrobials. Plants are employed as important source of medication in many traditional medications [33]. Continued further exploration of plant-derived antimicrobials is needed today. It has also been widely observed and accepted that the medicinal value of plants lies in the bioactive phytocomponents present in the plants [34-48]. Much work has been done on ethnomedicinal plants in India [49-51]. The results of the present study clearly showed that plant *D. metel* extracts showed antimicrobial activity against tested pathogenic strains including antibiotic resistant strains. *D. metel* extract is harmless and nonphytotoxic; it has been proved that extracts inhibitory effects on germination and on the viability of fungal spores as well. It showed moderate activity against A. niger as it is a saprophyte in soil causes black mould of onion, garlic and shallot; stem rot of Dracaena; root stalk rot of Sansevieria; and boll rot of cotton; spoilage of cashew kernels, dates, figs, vanilla pods and dried prune. The effectiveness of the active compounds present in plant extracts cause the production of growth inhibition zones that appear as clear are as surrounding the wells. However, plant extract was unable to exhibit antibacterial activity against tested bacterial strains. These bacterial strains may have some kind of resistance mechanisms e.g. enzymatic inactivation, target sites modification and decrease intracellular drug accumulation [52] or the concentration of the compound used may not be sufficient. Lowest activity was observed against *A. strictum* with Chloroform and A. strictum and S. salivarious with methanolic extracts respectively. No inhibition was observed with controls, which proves that solvents could not act as antibacterial agents. In almost all tests, crude methanolic extracts showed better inhibition against all tested bacterial and fungal strains, indicating that active ingredients in plant materials could be extracted into methanol. However, highest antibacterial activity of extract was observed due the presence of secondary metabolites such as alkaloids, flavonoids and steroids against P. syringae (30 mm) with 100 mg/ml and (35 mm) at 250 mg/ml DMSO and the organism is a rod shaped, Gram-negative bacterium with polar flagella. Further research is necessary for successful separation, purification and characterization of biologically active compounds using chromatographic methods and spectroscopic techniques. Further studies are being carried out in order to separate the individual components that are present in plant extracts of *D. metel* using column chromatography to develop Biopesticide which is alternative to synthetic agents.

Conflict of interest statement

We declare that we have no conflict of interest

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