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RESEARCH ARTICLE

EVALUATION OF ANTIMICROBIAL ACTIVITY OF *CANANGA ODORATA* (LAM.)HOOK.F. & THOMSON LEAF EXTRACT: AN *IN VITRO* STUDY

Isaivani Indrakumar*[1], V.Selvi[1], R.Gomathi[1] and S.Karpagam[2]

[1]Research scholar, Dept of Botany, Queen Mary's College, Chennai – 600 004, India, [2]Associate Prof., Dept of Botany, Queen Mary's College, Chennai – 600 004, India.Email - isaivanii@gmail.com

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ABSTRACT

Medicinal plants are important source of potentially useful structures for the development of novel chemotherapeutic agents. The first step towards this goal is the *in vitro* antibacterial activity assay(Tona *et al.,* 1998). The present study was aimed to evaluate the antmicrobial effect of *Cananga odorata* leaf extract on some bacterial cultures such as *Staphylococcus aureus, Salmonella typhi, Escherichia coli, Vibrio cholerae* and fungi such as *Epidermophyton floccosum. Microsporum gypseum and Trichophyton mentagrophytes.* The solvents Methanol, Chloroform and Petroleum ether were used for extraction.The inhibitory effect was assessed by well diffusion method. The zone of inhibition was measured. Among the solvent extracts tested, methanol was more effective than chloroform and petroleum ether.

Key words: *Cananga odorata, Antimicrobial activity, Staphylococcus aureus, Microsporum gypseum.*

INTRODUCTION

Plant derived drugs remains important resource especially in developing countries, to combat serious disease. Approximately 62 – 80% of the world's population still relies on traditional medicines for the treatment of common illness (WHO,2002; Zhang, 2004). Over 50% of all modern clinical drugs are of natural product origin (Stuffness *et al.,* 1982) and natural products play an important role in drug development programs in the pharmaceutical industry (Baker et al., 1995). Medicinal plants represents a rich source of antimicrobial agents Plants are used medicinally in different countries and are a source of many potent and powerful drugs (Srivastava, Lambert and Vietmeyer, 1996). It is estimated that today, plant materials are present in, or have provided the models for 50% of allopathic drugs (Rodders, 1996).

Cananga odorata belonging to the family Annonaceae is a moderate sized fast growing tree, growing up to 20 meters in height. Leaves simple, alternate, entire elliptic-lanceolate, glabrous in the outer side and pubescent in inner side, acuminate and with wavy margins and prominent midrib. Flower arises from the leaf axils, are very fragrant, greenish yellow colored with long petals. Fruits are greenish black in colour, with 6-12 ovoid fruitlets, contain 6-12 small ovoid flattened seeds. *Cananga odorata*, grows as small tree or compact shrub with highly scented flowers. The plant is distributed throughout India, China and Indonesia. The plant is useful against pitta, stomach ailment, fever, inflammation, burning sensation, malarial fever, asthma in aromatherapy, hypertension, anxiety, depression and as a sexual stimulant.

MATERIALS AND METHODS

Collection of Plant material

The leaves of the *Cananga odorata* tree were collected from localities of Chennai. The plant was authenticated at Plant Anatomy Research Centre (PARC) Tambaram, Chennai. The leaves were shade dried for about 15 days and powdered in an electric blender.

Preparation of Extracts

Fifty gm of shade dried, powder of leaf of the medicinal plants was extracted with 200 ml each of Petroleum ether, Chloroform, and Methanol using Soxhlet extractor for 48 hrs. All the extracts were concentrated using rotary flash evaporator. After complete solvent evaporation, each of these solvent extract was weighed and preserved at 5°C in airtight bottles until further use. About 10 mg of each solvent residue was dissolved in 1 ml of Dimethyl sulphoxide (DMSO) which served as the test extracts for antibacterial and antifungal assay.

Test microorganisms

The micro organisms used in the study were bacteria such as *Staphylococcus aureus, Salmonella typhi, Escherichia coli, Vibrio cholerae* and fungi such as *Epidermophyton floccosum. Microsporum gypseum and Trichophyton mentagrophytes.*

Culture medium

Nutrient agar (NA) medium was used to study the antibacterial activity and Potato dextrose agar (PDA) was used to study the antifungal activity.

Anti-bacterial activity assay

Antibacterial activity of solvent extracts was determined by well diffusion method on nutrient agar medium (Anon, 1996). Wells were made in nutrient agar plate using sterile cork borer (5 mm) and inoculum containing bacteria were spread on the solid plates with a sterile swab moistened with the bacterial suspension. The extracts were applied to different wells in volumes of 50-200uL using a micropipette. **Ampicillin** 10 µg/ml was used as the control for bacterial cultures. The plates were labelled, covered and incubated at 37ºC for 24h.

Anti fungal activity assay

The fungal mycelial suspension was mixed with the warm, melted, autoclaved PDA and poured into plates under aseptic conditions. The plates were covered and allowed to cool. When cooled, wells were made at the plate using a 5mm cork borer that was sterilized with alcohol and flame. The extracts were applied to different wells in volumes of 50-200uL using a micro liter syringe. **Nystatin** 10 µg/ml was used as the control for fungal cultures. The plates were labelled, covered, and incubated at 28ºC for 48h.

Results and Discussion

The present investigations revealed that, the different concentrations of the solvent extracts of *Cananga odorata* proved to be effective against both Gram positive and Gram negative bacteria tested. Among them *Staphylococcus aureus* (Gram positive bacteria) showed maximum zone of inhibition (1.62cm in 200µl conc) in methanolic extract compared to the other test organisms like *Salmonella typhi, Escherichia coli, Vibrio cholerae*. The petroleum ether and chloroform extracts of leaves of *Cananga odorata* showed considerably minimum zone of inhibition compared to methanolic extract. Antibiotics provide the main basis for the therapy of bacterial infections. However the high genetic variability of bacteria enables them to rapidly evade the action of antibiotics by developing antibiotic resistance. In recent years development of multidrug resistance in the pathogenic bacteria and parasites has created major clinical problems in the treatment of infectious disease (Davies, 1994). In

general gram negative bacteria were more resistant to antibiotics than gram positive bacteria (Paz *et al.,* 1995). The resistance is due to the differences in their cell wall composition. In this investigation too gram positive bacteria were more susceptible to crude extract than gram negative bacteria.

Table 1: Effect of Methanolic extract of *Cananga odorata* on different organisms

Test organisms	Zone of inhibition in cm (Methanolic extract)				
	50µl	100µl	150µl	200µl	Control(10 µg/ml)
Staphylococcus aureus	0.42±0.08	0.84±0.05	1.12±0.12	1.62±0.21	1.47±0.08*A
Salmonella typh i	0.28±0.08	0.32±0.04	0.68±0.04	0.90±0.04	1.47±0.08*A
Escherichia coli	0.28±0.07	0.40±0.08	0.60±0.10	0.98±0.10	1.78±0.04*A
Vibrio cholerae	0.28±0.07	0.58±0.09	0.70±0.07	1.08±0.10	1.48±0.16*A
Epidermophyton floccosum	0.44±0.05	0.74±0.18	0.98±0.10	1.20±0.12	1.62±0.21*N
Microsporum gypseum	0.54±0.05	0.84±0.05	1.38±0.08	2.14±0.16	2.10±0.04*N
Trichophyton mentagrophytes	0.32±0.04	0.54±0.05	0.74±0.18	1.62±0.08	1.64±0.05*N

All values are mean zone of inhibition ± SD; (-) No zone of inhibition; *A- Ampicillin, *N – Nystatin

Table 2 : Effect of Petroleum ether extract of *Cananga odorata* on different organisms

Test organisms	Zone of inhibition in cm (Petroleum ether extract)				
	50µl	100µl	150µl	200µl	Control(10 µg/ml)
Staphylococcus aureus	0.28±0.08	0.72±0.07	1.24±0.08	1.62±0.08	1.47±0.08*A
Salmonella typii	-	0.28±0.08	0.74±0.18	1.10±0.07	1.47±0.08*A
Escherichia coli	0.32±0.04	0.64±0.05	0.84±0.05	1.12±0.10	1.78±0.04*A
Vibrio cholerae	-	0.60±0.07	0.90±0.04	1.18±0.04	1.48±0.16*A
Epidermophyton floccosum	0.32±0.04	0.70±0.07	0.80±0.05	1.20±0.07	1.62±0.21*N
Microsporum gypseum	0.28±0.07	0.74±0.08	1.18±0.04	2.12±0.18	2.10±0.04*N
Trichophyton mentagrophytes	-	0.42±0.16	0.62±0.15	1.06±0.11	1.64±0.05*N

All values are mean zone of inhibition ± SD; (-) No zone of inhibition; *A- Ampicillin, *N – Nystati

Table 3 : Effect of Chloroform extract of *Cananga odorata* on different organisms

Test organisms	Zone of inhibition in cm (chloroform extract)				
	50µl	100µl	150µl	200µl	Control(10 µg/ml)
Staphylococcus aureus	-	0.18±0.08	0.32±0.04	0.64±0.05	1.47±0.08*A
Salmonella typhi	0.28±0.08	0.44±0.05	0.84±0.05	1.20±0.05	1.47±0.08*A
Escherichia coli	-	0.18±0.10	0.54±0.05	0.84±0.05	1.78±0.04*A
Vibrio cholerae	-	-	0.18±0.05	0.32±0.04	1.48±0.16*A
Epidermophyton floccosum	0.18±0.08	0.54±0.05	0.68±0.04	1.90±0.04	1.62±0.21*N
Microsporum gypseum	-	0.28±0.04	0.60±0.04	1.20±0.05	2.10±0.04*N
Trichophyton mentagrophytes	-	0.32±0.04	0.72±0.07	1.42±0.08	1.64±0.05*N

All values are mean zone of inhibition ± SD; (-) No zone of inhibition; *A- Ampicillin, *N - Nystatin

The effect of plant extracts was different with different fungal strains. The methanolic extract of *Cananga odorata* leaf extract showed best antifungal activity (2.14 cm in 200µl conc) against *Microsporum gypseum* when compared with the other fungus like *Epidermophyton floccosum, Trichophyton mentagrophytes.* The petroleum ether and chloroform extract comparatively showed lesser zone of inhibition when compared to methanolic extract.Thus from the investigation it is clear that Methanolic extract showed best antimicrobial activity with the other solvents used.

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