SIDDHA PAPERS
A PEER REVIEWED ONLINE JOURNAL
ISSN: 0974-2522

Siddha Papers 2019 (14) (1)
REVIEW ARTICLE
https://www.siddhapapers.com

Anti-microbial potency of *Mul Ilavu* (Bombax malabaricum) Chooranam

P. Pushpa[1], S. Keerthiga[1], R. Dharshini[1], A. Archana[1], Thomas.M. Walter[2].

[1] Second Year BSMS, Government Siddha Medical College, Palayamkottai, Tirunelveli - 627 002, pushpa.sivagami@gmail.com

[2] Lecturer, Government Siddha Medical College, Palayamkottai, Tirunelveli - 627 002.

ABSTRACT

Purpose

Siddha Medicine is one of the Indian's oldest systems of medicine prevalent in South India. *Mul ilavu(Bombax malabaricum*) is a siddha herb that belongs to Bombaceae family. This herb is widely documented in *Gunapadam mooligai, kannu samayam*, etc., It is used to treat urinary disorders , gynaecological disorders, wounds, piles, pimples,etc., So, it is a source of natural drug for microbial devastation. Antimicrobial activity refers to the process of killing or inhibiting the disease causing microbes. Due to the overuse of antibiotics, antibiotic resistance develops. As a result, the standard antibiotics become ineffective. So, the need of traditional formulations is more.

Objective

This study was done to assess the antimicrobial potency of *Mul ilavu chooranam*.

Methodology

Gunapadam mooligai vaguppu describes *mul ilavu chooranam* to assist in the treatment of gonorrhoea, dysuria, haematemesis. The *Mul ilavu chooranam* was tested against some gram negative, gram positive bacteria and fungi by Disc diffusion method.

Result

The results showed that the *Mul ilavu chooranam* is effective against *Streptococcus mutans, Staphylococcus aureus, Klebsiella pneumoniae, Bacillus subtilis, Proteus vulgaris, Candida albicans.*

Conclusion

Thus, the *mul ilavu chooranam* helps in the management of infectious diseases.

KEY WORDS

Mul ilavu(Bommbax malabaricum) chooranam, Antimicrobial activity, Siddha formulation, Disc diffusion method.

INTRODUCTION

The infections caused by microorganism are being treated by the antimicrobial agents. The antimicrobial resistance started appearing in the late 1930s. The human body shows resistance against the antimicrobial agents. As per World Health Organisation (WHO), Antimicrobial resistance (AMR) threatens the effective prevention and treatment of an ever –increasing range of infections caused by bacteria, parasites, viruses and fungi. Without effective antibiotics, the success of major surgery and cancer chemotherapy would be compromised. Besides, the newly developed third and fourth generation antibiotics poses big side effects that are harmful to human health. Hence, the use of herbal formulations as alternative antibiotics is needed. Today, over 80% of the world's population depend on phytomedicine as first line of treatment for various diseases. Until the beginning of 20^{th} century, virtually all medicines were derived from natural sources, most often from plants. These antimicrobial agents derived from higher plants have been reviewed recently. This study shows the antimicrobial potency of Mul *ilavu Chooranam. Mul ilavu chooranam* can be primarily used for gonorrhoea, dysuria, haematemeis. It was tested against the gram positive *(Staphylococcus aureus, Streptococcus mutans, Bacillus subtilis)* and gram negative *(Klebsiella pneumoniae, Proteus vulgaris, Escherchia coli)* and fungi *Candida albicans*. The drug is not effective against *E.coli*.

MATERIALS AND METHODS

Collection

The seeds of *mul ilavu* were collected from *mul ilavu* tree in Krishnagiri, Tamil Nadu, and South India. The fruits of *seeragam* and *vaal milagu* were collected from raw drug store in Tirunelveli, Tamil Nadu, South India.

Ingredients of *mul ilavu chooranam*

Bombax malabaricum - *mul ilavu*
Cuminum cyminum - *seeragam*
Piper cubeba - *vaal milagu*
The drugs were clearly explained in table number 1.

Table 1. The ingredients *of Mul ilavu chooranam*

S.No	Botanical Name	Family	Parts used	Chemicals	Action	Uses
1.	*Bombax malabaricum*	Bombaceae	Seed	β-sitosterol, essential amino acids,phytosterols, palmitic acid,stearic acid.	Aphrodisiac ,Demulcent	Cures Diarrhoea, Dysentry
2.	*Cuminum cyminum*	Umbelliferae	Fruit	Cuminaldehyde, cymene, γ-terpinene, β-pinene, cuminol.	Stimulant carminative, Astrigent, Stomachic	Cures Dyspepsia chronic diarrhea, billous, nausea
3.	*Piper cubeba*	Piperaceae	Fruit	Cubebol, β-caryophyllene, β-elemene, γ-cadinene.	Carminative, diuretic, expectorant	Cures Indigestion constipation bad breath piles, fistula bleeding diseases.

Purification

The seeds of *mul ilavu*, fruit of *seeragam* and *vaal milagu* were washed and dried in shade. and then, they were fried in dry pan separately.

Preparation

10*g* of Bombax malabaricum(mul ilavu) seeds and 5*g* of Cuminum cyminum (seeragam) and 2.5*g* of Piper cubeba(vaal milagu) were powdered separately and mixed well.

***Chooranam* purification**

Based on the siddha literature, this *chooranam* is purified by *vasthirakayam*(Powder is sieved by using tiny pored cloth). It is done to obtain more finer particles of chooranam so as to increase its solubility in solvent and absorbtion into the body. Thus the finely powdered *mul ilavu*

chooranam was prepared and stored in an air tight container to prevent contamination from dust and air. The *mul ilavu chooranam* was tested for antimicrobial analysis.

Microbial analysis

Dilution 0.1*g* in 1*ml*

Test organism

The test microorganisms used for antimicrobial analysis. Microorganism name were purchased from Microbial Type Culture Collection and Gene Bank (MTCC) Chandigarh. The bacterial strains were maintained on Nutrient Agar (NA).

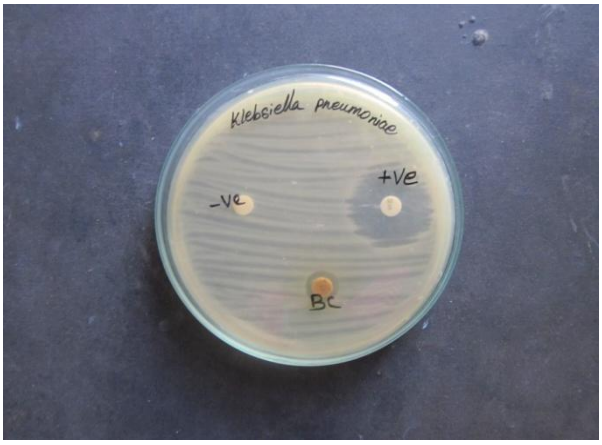
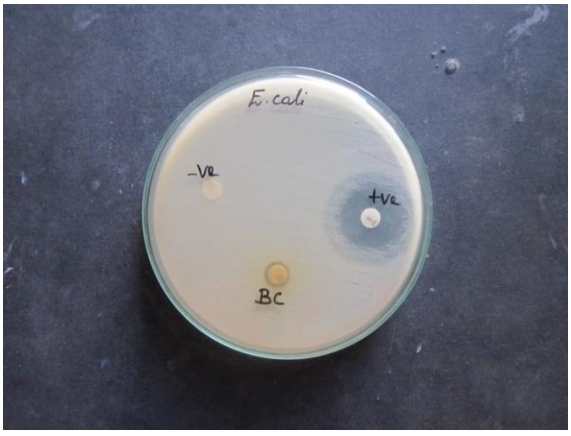
Nutrient Broth Preparation

Pure culture from the plate were inoculated into nutrient Agar plate and sub cultured at 37˚C for 24h. Inoculum was prepared by aseptically adding the fresh culture into 2*ml* of sterile 0.145*mol/L* saline tube and the cell density was adjusted to 0.5 McFarland turbidity standard to yield a bacterial suspension of 1.5×108 *cfu/ml*. Standardized inoculum used for antimicrobial test.

Antimicrobial Test

The medium was prepared by dissolving 3g of Muller Hinton Agar Medium (Hi media) in 1000*ml* of distilled water. The dissolved medium was autoclaved at 15 *Lbs* pressure at 121˚C for 15min (pH - 7.3). The autoclaved medium was cooled, mixed well and poured petriplates (25*ml*/plate) the plates were swabbed with pathogenic bacteria culture viz . Microorganism name finally, The sample or sample loaded Disc was then placed on the surface of Muller-Hinton medium and the plates were kept for incubation at 37˚C for 24hours. At the end of the incubation, inhibition zones were examined around the disc and measured with transparent ruler in millimetres. The size of the zone of inhibition (including disc) was measured in millimetres. The absence of zone of inhibition was interpreted as the absence of activity (Kohner *et.al*., 1994; Mathabe *et al*., 2006). The activites are expressed as resistant, if the zone of inhibition was less than 7*mm*, intermediate (8 - 10 *mm*) and sensitive if more than 11*mm* (Assam *et al.,* 2010) .The zone of inhibition produced in the petriplates against each bacteria is given in the following figure no-1

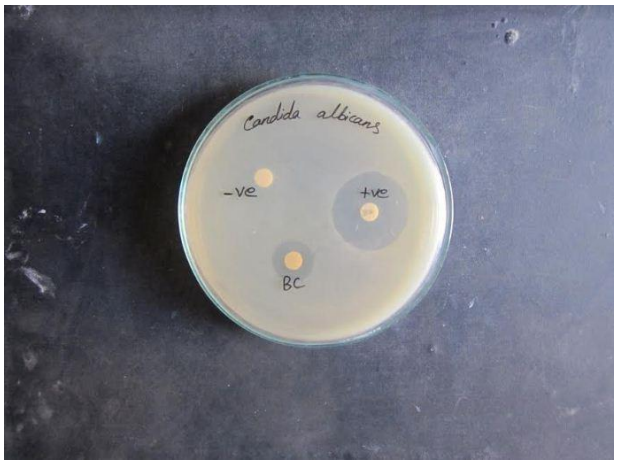
Figure 1. Zone of inhibition for selected test microorganisms

Zone of inhibition for *Staphylococcus aureus*	**Zone of inhibition for *Streptococcus mutans***
	
Zone of inhibition for *Klebsiella pneumoniae*	**Zone of inhibition for *Bacillus subtilis***
	
Zone of inhibition for *Proteus vulgaris*	**Zone of inhibition for *E.coli***
	

Anti-fungi Assay by Disc Diffusion Method

Antibiotic susceptibility tests were determined by agar disc diffusion (Kirby-Bauer) method. Fungi strains (Fungi Name) were swabbed using sterile cotton swabs in SDA agar plate. Up to 40 μl of each concentration of the extract were respectively introduced in the sterile discs using sterile pipettes. The disc was then placed on the surface of SDA medium and the compound was allowed to diffuse for 5 minutes and the plates were kept for incubation at 22˚C for 48 hours. At the end of incubation, inhibition zones were examined around the disc and measured with transparent ruler in *millimetres*. The zone of inhibition produced in the petriplate against fungus is given in the following figure no:2

Figure 2. Zone of inhibition for *Candida albicans*



Some of the common infections caused by selected microorganisms listed in the following Table 2.

Table 2. Infections caused by the selected microorganism

Bacteria	Infection
Staphylococcus aureus	Skin infection, Pneumonia, Heart value infection
Steptococcus mutans	Dental caries, ulcerative colitis, crohn's disease, major inflammatory bone disease.
Klebsiella pneumonia	Pneumonia, Atophic rhinitis, Rhinoscleroma
Proteus vulgaris	UTI, Haematuria, Nosocomial infection.
Bacillus subtilis	Endocarditis, musculoskeletal infection, septicaemia.
Fungi *Candida albicans*	Candidiasis, UTI.

RESULT AND DISCUSSION

The results produced in the test are given in the table.

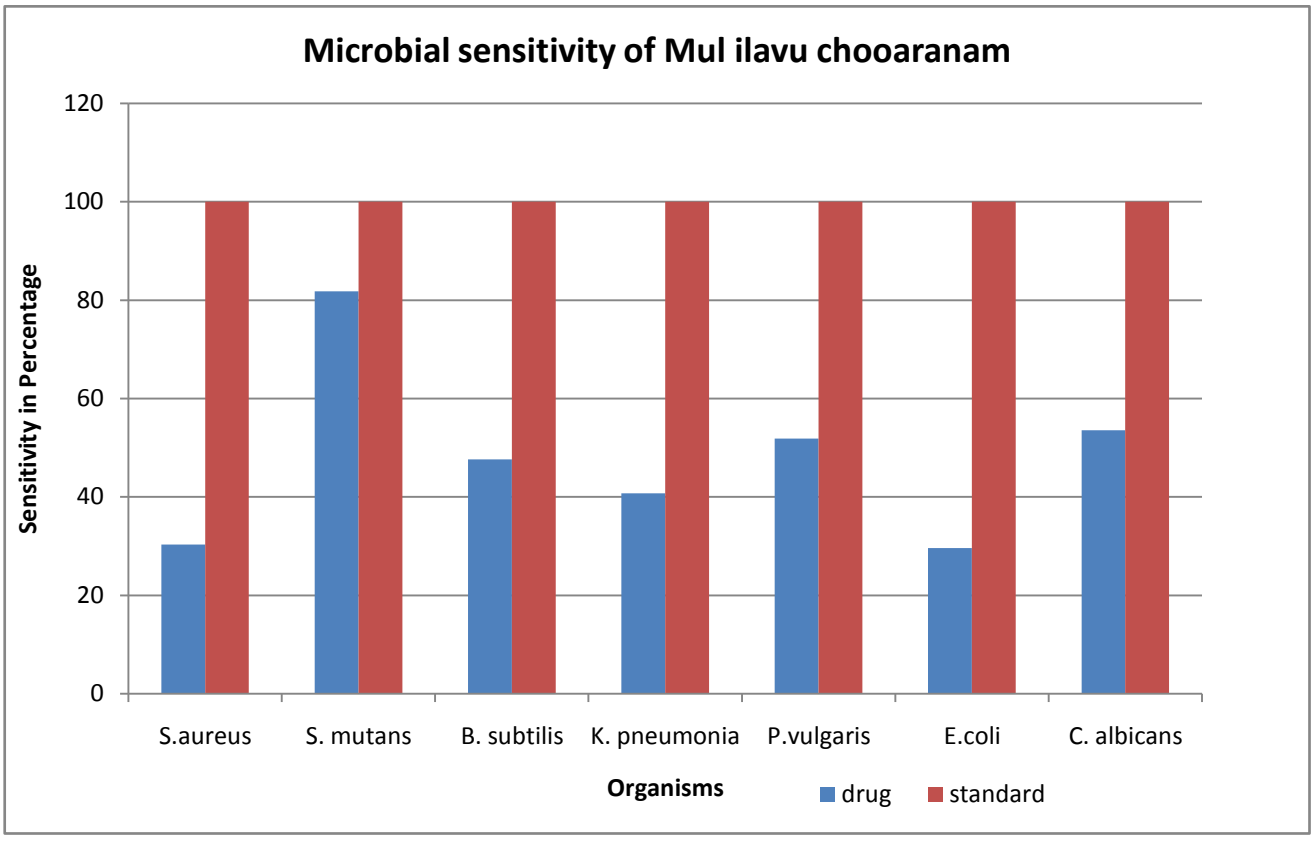
Table 3. Zone of inhibition produced in the test

	Bacteria Strains Name						
Sample	***Staphylococcus aureus* (G+ve)**	***Streptococcus mutans* (G+ve)**	***Bacillus subtilis* (G+ve)**	***Klebsiella pneumonia* (G-ve)**	***Proteus vulgaris* (G-ve)**	***E.coli* (G-ve)**	***Candida albicans* (F)**
Mul ilavu	10	9	10	11	14	8	15
Positive Control	33	11	21	27	27	27	28
Negative control	NZ	NZ	NZ	NZ	NZ	NZ	NZ

Notes: Mm - millimeter, NZ - No zone, G+ - Gram Positive Organism, G- - Gram Negative Organism, F- Fungi

The zone of inhibition against the test microorganism was measured in millimeters. The sensitivity produced by positive control (Streptomycin) is taken as standard. When the data is converted in percentage, the zone of inhibition obtained by positive control is fixed as 100%. The *Mul Ilavu chooranam* shows 30%, 82%, 48%, 41%,52%, 30%, 54% inhibition to *Staphylococcus aureus, Streptococcus mutans, Bacillus subtilis, Klebsiella pneumonia, Proteus vulgaris, E.coli,Candida albicans* respectively. Hence, *Mul ilavu chooranam* is highly sensitive against *Streptococcus mutans, Candida albicans, Proteus vulgaris, Basillus subtilis, Klebsiella pneumonia*. It shows less sensitivity to *Staphylococcus aureus and E.coli.* The graphical representation of antimicrobial results is given in the following figure no-3 shows that the *mul ilavu chooranam,* a Siddha formulation is more sensitive to *Streptococcus mutans, Proteus vulgaris, Candida albicans, Klebsiella pneumoniae.* It is sensitive to *Bacillus subtilis, Staphylococcus aureus*. It is less sensitive to *E.coli.* Hence, The mul ilavu chooranam has antibacterial and antifungal activity. It can be recommended for Bleeding disorders, Gonorrhoea, Dysuria, Haematuria, Pneumonia, Rhinoscleroma, UTI, Endocarditis, Septicemia, Candidiasis diseases.

Figure 3. Microbial sencitivity of Mul ilavu chooranam



CONCLUSION

The study shows that the mul ilavu chooram, a Siddha formulation is more sensitive to *Proteus vulgaris, Candida albicans*, *Klebsiella pneumoniae.* It is sensitive to *Bacillus subtilis, Staphylococcus aureus*. more sensitive to Hence, The mul ilavu chooranam has antibacterial and antifungal activity. It can be recommended for Bleeding disorders, Gonorrhoea, Dysuria, Haematuria, Pneumonia, Rhinoscleroma, UTI, Endocarditis, Septicemia, Candidiasis diseases.

ACKNOWLEDGEMENT

I would take this opportunity to thank our seniors Dr. S. Merish & V.S.Nandhini and the editorial board of Siddha Papers Journal for encouraging our team in completing this work.

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