

Antibacterial Activity Of *Sida Acuta* (Burm.F) Against Methicillin Resistant *Staphylococcus Aureus* And Other Multi Drug Resistant Wound Pathogens

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Abstract

Now a days, drug resistance in bacterial pathogens is a serious threat all around the world. There is a need of alternative sources to avoid this problem. In India, mostly traditional medicine was taken from plants. The present study aimed at evaluating the antibacterial activity of solvent extracts of leaves from *Sida acuta* (Burm.f) against methicillin resistant *Staphylococcus aureus* and other multidrug resistant wound pathogens such as *Pseudomonas aeruginosa*, *Salmonella sp*, *Klebsiella sp* and *E. coli*. The GCMS analysis of ethanol extract showed various bioactive compounds. Ethanol extract showed highest antibacterial activity against multidrug resistant pathogens that isolated from wounds than chloroform extract.

Key words: multidrug resistant pathogens, wound infection, antimicrobial agents etc.

INTRODUCTION

In India, ayurveda is thought to have been practiced for 5000 years and it is the most well-known medical system in the world. Utilization of plants as medicines is seen from the start of human civilization. The medicinally useful plant substances are derived from the secondary plant products, which are typically a mixture of metabolites. Plants have been crucial to preserving human health^{1,2,3}. Medicinal plants have been used as traditional medicine and has been used in treatment of various diseases and worldwide ethnomedicine^{4,5}. These traditional remedies' purported therapeutic properties may be linked to the presence of lignans, alkaloids, flavonoids, triterpenoids, and other phenols⁶. The term "wound" refers to the loss of cellular tissue and the interruption of functional continuity as a result of chemical, physical, microbiological, or immunological harm to the living tissue⁷⁻⁹. Blood cells, soluble mediators, extracellular matrix, and parenchymal cells all play important roles in the multi-step, intricate process of wound healing^{10,11}. Plant metabolites have been shown to have biological properties like antibacterial, anticancer, and antioxidant¹². Due to the rise in drug-resistant infections and their usage in traditional medicine across the world, the pharmaceutical and scientific communities have been driven to investigations based on the potential antimicrobial activity of plant-derived compounds, an unexplored source of antimicrobial chemotypes¹³. *Sida acuta* of Malvaceae family and being used for renal problems, reproduction ailments and rheumatism^{14,15}. The present study was done to evaluate the antibacterial activity of *S. acuta* against multidrug resistant wound infection causing bacteria.

Materials and Method

Collection of wound Sample

Swab sample from human wound infection was collected in a sterile container at local clinical laboratory, Tuticorin, Tamil

Nadu, India. Collected sample was preserved in refrigerator till it was used for the isolation of pathogens in a microbiology laboratory.

Isolation and identification of microorganisms

Collected swab sample was streaked on some specific media like Xylose Lysine Deoxycholate Agar, Pseudomonas Isolation Agar, Eosin Methylene Blue Agar, Mannitol Salt Agar, Macconkey Agar. Plates were incubated at 37°C for 48 hours. Visible colonies were taken for further analysis. Identification of microorganisms were done using microscopical / biochemical characteristics.

Collection of plant materials and extraction

Fresh leaves of *Sida acuta* (Burm.f) free from disease were collected from Tuticorin, Tamil Nadu, India, washed in tap water and then with sterile distilled water. Leaves were dried in shade for five days and then powdered with the help of blender. Each 20 gm of dried leaf powder added to 80 ml of ethanol and another 80 ml of chloroform. Both were kept in a conical flask for 24 hours at room temperature, the extract was filtered and dried at temperature at 45 °C for removal of ethanol and chloroform to obtain dense extract. Both dried extracts were stored in glass bottles.

Antimicrobial activity

For the antimicrobial tests, Ethanol and chloroform extracts were diluted in 70% di-methylsulfoxide (DMSO) to a concentration of 20 mg/ml. Antimicrobial activity of different extracts were evaluated by the agar well diffusion method¹⁷. The samples showed antibacterial activity were subjected to minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC)¹⁷.

6. Gas Chromatography and Mass Spectrometry Analysis

The 2 µL *Sida acuta* (Burm.f) extract was injected into the GC-MS system and the condition was as per the earlier method¹⁸. Compounds were identified by MS data library of NIST library and Wiley library.

RESULTS AND DISCUSSION

The antimicrobial potential of *Sida acuta* (Burm.f) was evaluated was recorded as zone of inhibition against various pathogens. Ethanol extract showed high degree of inhibition where zone of inhibition against *S. aureus* was around 13 mm. where *Salmonella spp*, *Pseudomonas aeruginosa*, *Klebsiella spp*, *Escherichia coli* were showing zone of inhibition of 10 mm, 11 mm, 9 mm, 6mm respectively (Fig 1). Various flower extracts of medicinal plants have been shown to have activity against wound pathogens¹⁶. Chloroform extract showed maximum inhibition zone against *S. aureus* with 9 mm in diameter. It shows minimal activity in *Salmonella spp* for about 2 mm in diameter (Fig 2).

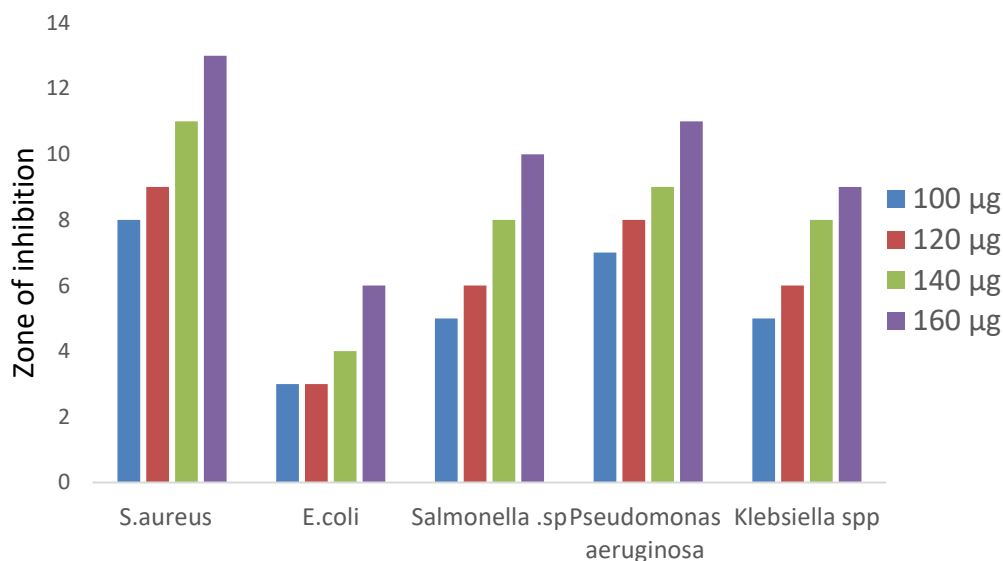


Figure 1. Antibacterial activity of ethanol leaf extract against various pathogens

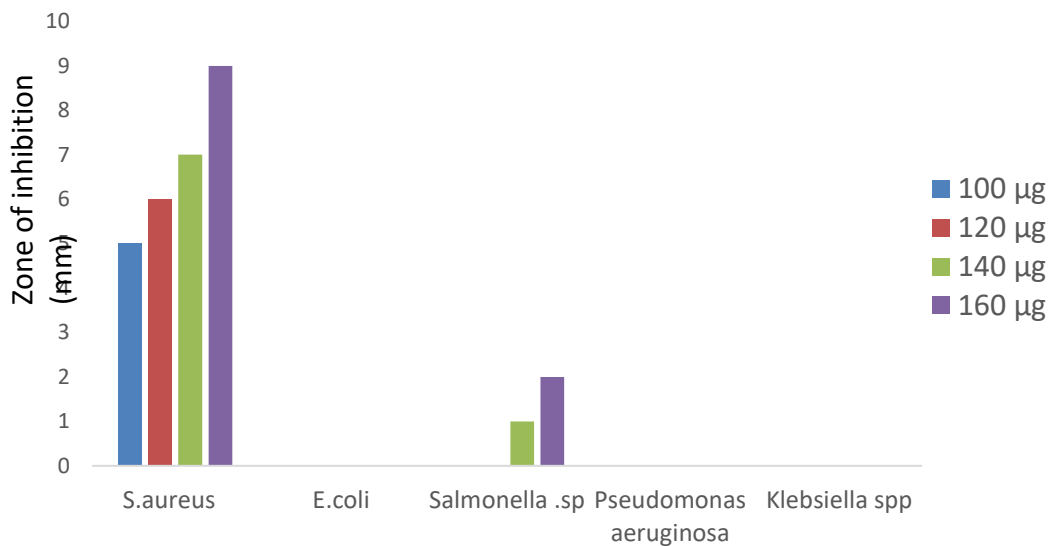


Figure 2. Antibacterial activity of chloroform leaf extract against various pathogens

Ethanol extract of *Sida acuta* (Burm.f) showed MIC value about 125 µg/ml against *S. aureus* and 200 µg/ml for all the wound pathogens (results not shown here). The least MBC value 1mg/ml was observed in ethanol extract against *S. aureus* (Fig. 3). The plant extract of jackfruit has also been shown to have high antibacterial action against *S. aureus*, *Bacillus* sp., and *Pseudomonas aeruginosa*¹⁹. Abirami et al¹⁶ has also reported that ethanol extract of *Pedaliium murex* was also reported to have antibacterial property against *Pseudomonas aeruginosa* and *S.aureus*. Medicinal plants have been well reported to have antibacterial effects²⁰⁻²⁶.

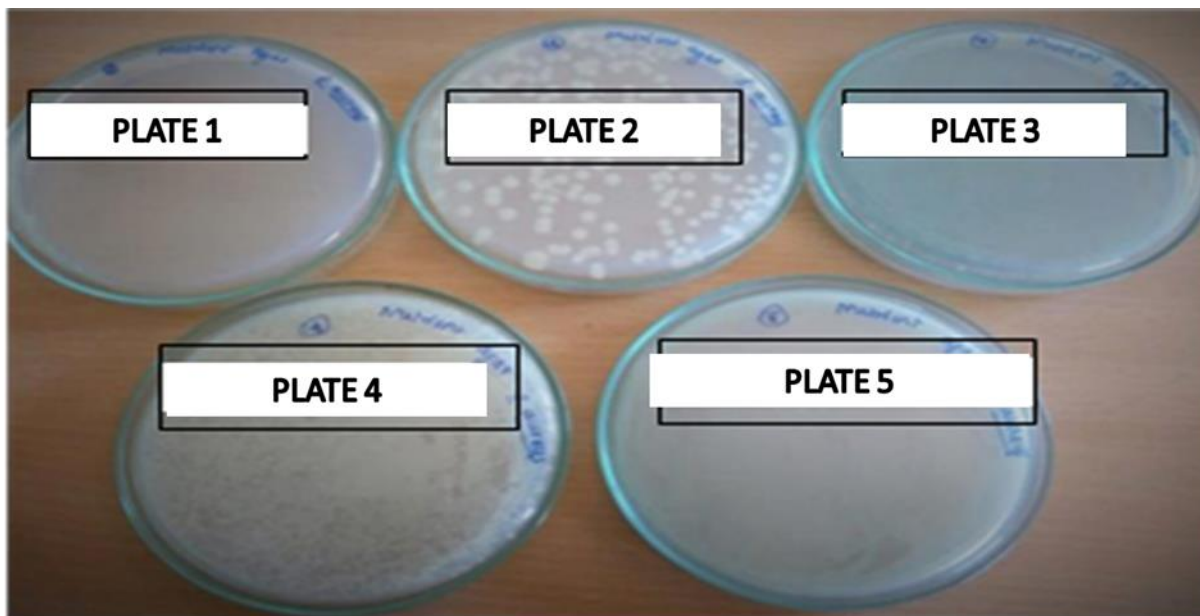


Fig 3. MBC Results of ethanol extract of *Sida Acuta* (Burm.f) against *S. aureus* with concentration of: Plate 1=1 mg/ml; Plate 2=0.5 mg/ml; Plate 3=0.25 mg/ml; plate 4 = 0.125 mg/ml; Plate 5 = 0.625 mg/ml

GC-MS Analysis

The GC-MS analysis of *Sida acuta* (Burm.f)leaves exposed the existence of 12 major compounds (Fig 4). The major compounds like Cyclohexanone, 3-methyl-, oxime; Pyridine; Benzyl Benzoate; Pentadecanal-; Hexadecanoic acid, ethyl ester; Phthalic acid, 2,7-dimethyloct-7-en-5-yn-4-yl isobutyl ester; 9,12,15-Octadecatrienoic acid, ethylester, (Z, Z, Z)-;3-[(4-Methoxy-benzoyl)-hydrazono]-N-pyridin-3-yl butyramide; Silane,trimethyl[5-methyl-2-(1methylethyl)phenoxy] are expected to be responsible for antimicrobial property of *Sida acuta* (Burm.f).

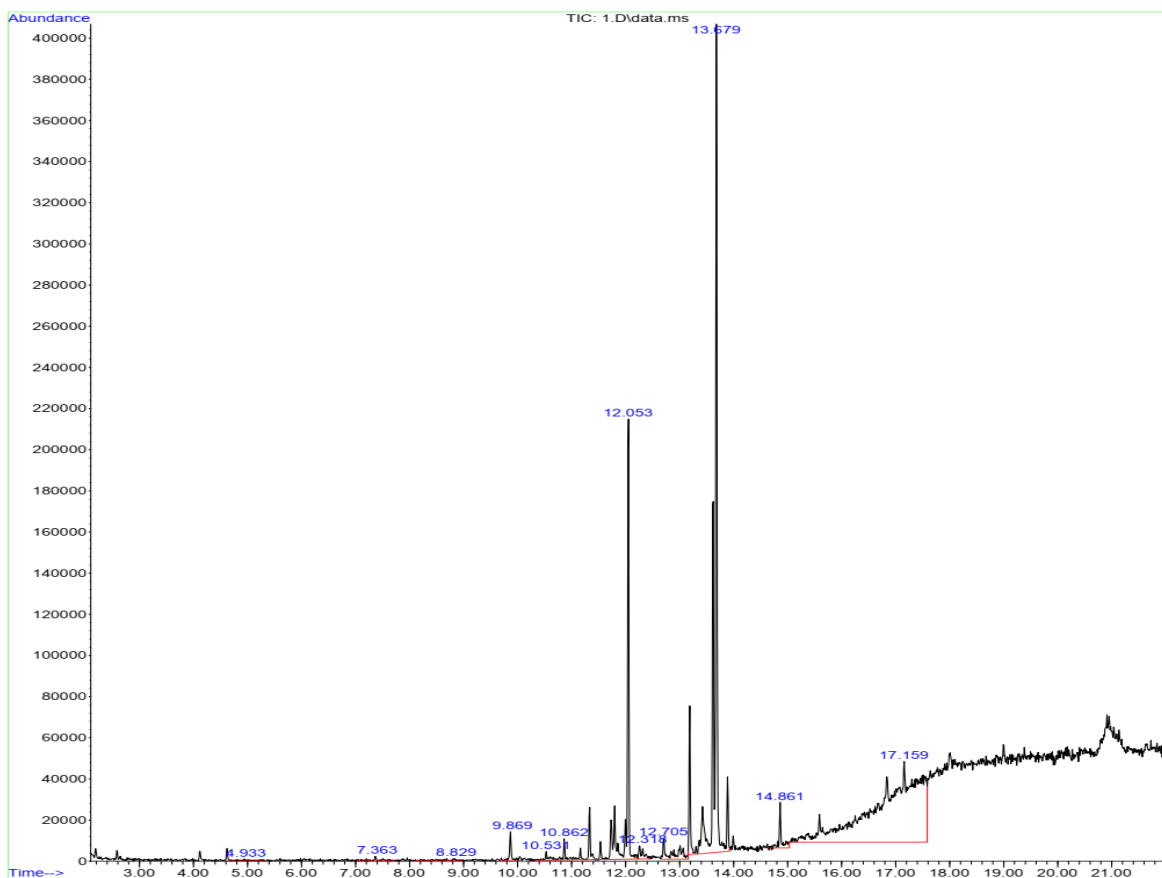


Fig.4 GC analysis *Sida acuta* (Burm.f) leaves

CONCLUSION

To overcome multidrug resistance of bacteria, extracts of *Sida acuta* (Burm.f) were tested against MRSA and wound infections causing multi drug resistant wound pathogens and MRSA strains. This preliminary investigation showed that all ethanol extracts of *Sida acuta* (Burm.f) active against the isolated wound pathogens like MRSA strains, *Pseudomonas aeruginosa*, *Salmonella spp*, *Klebsiella spp*, *E. coli*. MIC was found to be 125µg/ml against *S. aureus* and 200 µg/ml for all the wound pathogens. *Sida acuta* (Burm.f) leaves were subjected to GC-MS analysis and revealed twelve major phytochemicals.

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