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A Swift Description on Antimicrobial Action of *Sarcostemma intermedium*, an Extraordinary Scarce Medicinal Plant against Few Pathogenic Microorganisms.

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ABSTRACT

Medicinal plants are regarded better since last many years due to development of multi drug resistance in many of the species, large side effects and high cost of antibiotics. In the present study antimicrobial activity of methanolic extract of *Sarcostemma intermedium* was evaluated against few pathogenic microorganisms viz. *Escherichia coli*, *Klebsiella pneumoniae* and *Staphylococcus aureus*. Further the extract was also assessed for its potency against two dermatophyte fungal strains viz. *Microsporum spp.* and *Trichophyton spp.* During the present study this extract was found to be more efficacious against *Escherichia coli* and *Klebsiella pneumoniae* with zone of inhibition of 15 mm and 12 mm respectively as compared to such zone in *Staphylococcus aureus* (8 mm). Interestingly, the same extract was not able to show any zone of inhibition against dermatophytes evaluated during the present study.

**Keywords**: Antimicrobial, Soxhlet apparatus, Zone of inhibition, *Sarcostemma intermedium*

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INTRODUCTION

Medicinal plants are prosperous source of antimicrobial compounds. The antimicrobial compounds are secreted as part of their normal physiology and named as secondary metabolites. Most of these compounds are secreted during the stationary phase [1]. The antimicrobial compounds secreted by the plant execute imperative biological functions and are used as defenders against many pathogenic microorganisms. Although, a major variety of the plant species have been assessed for their antimicrobial activities but still their evaluation is at its infancy [2].

In today’s era the major threat is the development of multi drug resistance by most of the pathogens therefore it has become necessary to evaluate some new and potent antimicrobial compounds for newly emerging and re-emerging diseases [3,4].

The newly developed compounds may follow different pathways that may hit the target in different possible way and win the battle against the respective disorders. The type and level of biological activity the plants bear depends upon various factors like geographical conditions under which the plant being thriving, part of the plant chosen, harvest timings, method chosen for drying, moisture content [5]. Different parts of the plant like stem, root, exudates, twigs of the same plant may have different medicinal value. Thus a single plant might be useful in providing multiple solution for a variety of ailments.

*Sarcostemma intermedium* is an important medicinal plant belonging to the family Asclepiadiceae. It is used as customary medicine for various disorders mainly for body swelling. Other species of *Sarcostemma* are reported for diverse therapeutic action. In a report described recently *Sarcostemma secamone* extract was used as an agent for gargle in throat and mouth infection. Few other reports indicated that its fresh roots are used in treatment of jaundice [6]. Furthermore, it is also revealed that leaf powder of *Sarcostemma secamone* stimulates regulatory system, increases secretion of urine and activates uterus [7]. A noteworthy report states that *Sarcostemma acidum* is used in treatment of cardiac diseases, viral infections, fever, mental disorders and show anti inflammatory activity [8, 9]. As explained above there are very scarce reports on the medicinal value of *Sarcostemma intermedium* and there are scanty data available about it so the endeavor of the present study was to assess the antimicrobial activity of *Sarcostemma intermedium* which was conducted to fetch some significant information for deciphering novel ingredients based on the basic results gained during this work. Nevertheless, it may be noted that such studies are to be repeated to effusively evaluate phytochemical properties of this plant.

MATERIAL AND METHODS

Collection of plant and extract preparation

The healthy whole plants of *Sarcostemma intermedium* were obtained from Department of Forest, Rewa, MP (Forest Nursery, Rewa, MP, India). Plant sample was cleaned and dried for 72 hours in laboratory. Thereafter sample was ground to powder using mortar pestle. The coarsely powdered form was packed in the soxhlet apparatus and
extracted with methanol as solvent. The extract was then filtered. The 1 gm of extract was mixed with 5 ml of methanol to make stock solution for further usage.

**Microorganisms and its maintenance**

Few pathogenic bacteria viz. *Staphylococcus aureus*, *Klebsiella pneumoniae* and *Escherichia coli* were obtained from culture collection and were maintained in nutrient broth (Peptone: 0.5%, Beef extract: 0.5%, NaCl: 0.5%) and were incubated at 37°C for 24 hrs under shaking conditions. The cultures were maintained in nutrient agar slants and stock cultures were preserved at 4°C. Furthermore, two fungal strains which are dermatophytes viz. *Trichophyton spp.* and *Microsporum spp.* were grown at 27°C for 48 hours on potato dextrose agar medium (Hi-Media Laboratories, India).

**Antimicrobial assay**

The antimicrobial activity was evaluated against above mentioned species. The assay was performed on Muller Hinton agar medium by Kirby baeur method [10]. The 25 ml of the Muller Hinton agar was inoculated with the respective culture for each plate and allowed to solidify. Two wells were made with sterile borer onto the plates each for test and control. The test well was filled with the extract of *Sarcostemma intermedium* and control well contained the only methanol. Further the plates were incubated at 37°C for 24 hours for further assessment.

**Agar well diffusion method**

This method was introduced to evaluate the efficacy of extract of *Sarcostemma intermedium* For this purpose, Potato dextrose agar (PDA) plates as described above were used for inoculation. The plant extract (200 mg/ml) was poured in two different concentrations of 5mg/ml and 2.5mg/ml in two different agar petridishes and were incubated at 28°C for 24 hours.

**RESULTS AND DISCUSSION**

During the present prelude study it was noticed that extract of *Sarcostemma intermedium* was found to be more efficacious against *Escherichia coli* and *Klebsiella pneumoniae* with zone of inhibition of 15 mm and 12 mm respectively as compared to such zone in *Staphylococcus aureus* (9 mm). It revealed a considerably fair antibacterial activity against gram negative species with maximum zone (15 mm) appearing for the *E. coli*, which is most common enteric pathogen, at 12.5 mg/ml (Fig 1, Table 1) followed by *K. pneumonia* which displayed a zone of 12 mm (Fig 2). Moreover, it was further noticed that, *S. aureus* presented intermediate sensitivity to this extract with a smaller zone of inhibition of 8 mm (Fig 3). On the other hand, the same extract was not able to demonstrate any zone of inhibition against *Trichophyton spp.* and *Microsporum spp.* the dermatophytic fungi evaluated in the present study. In most cases, the anti bacterial activity increases with increasing concentration of the extract as authenticated by zone of inhibition. As per available resources, diverse studies are conducted to validate that some medicinal plants are considered potential sources of new antimicrobial agents [11]. To continue with it a
study by agar cup plate method on *Sarcostemma acidium* provided the fact that it can serve as broad spectrum antibiotic as it presented sensitivity against many of the pathogenic species viz *Streptococcus pneumoniae*, *Bacillus cereus*, *Salmonella paratyphi* and *Klebsiella pneumoniae* [12]. In a recent report, ethyl acetate extracts of *Sarcostemma acidium* at 6 mg/ml exhibited 68% anti-inflammatory response *in vitro* as confirmed by cell membrane stabilization method [9]. In a study conducted by Mothana et al. it was noticed that the methanolic extracts are found as superior in contributing the antimicrobial activity [13]. In this way our study conducted on antimicrobial action of *Sarcostemma intermedium*, against few pathogenic microorganisms gives further prominence of revealing the phytochemical ingredients of this rare plant.

![Figure 1: Zone of inhibition in *E. coli*](image1)

![Figure 2: Zone of inhibition in *K. pneumoniae*](image2)
CONCLUSION

It is concluded that from the present study that the methanolic extract of *Sarcostemma intermedium* posses significant antibacterial activity. This plant is more effective against gram negative bacteria and has intermediate action against gram positive bacteria. This medicinal plant has broad spectrum antibacterial activity and therefore supports the use in medicine. Further the secondary metabolites present the extract may be identified and purified to a certain level so that it achieves the characteristics like a potent antibiotic.

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