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Research Article

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Evaluation of Anti-fungal and Anti – bacterial effect of Siddha Formulation *Vanga vennai* against Selected Human Pathogens

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Abstract

Siddha system of medicine more popular in South India particularly Tamilnadu and also wherever the Tamil civilization are living. In this traditional system mostly herbals, metals and minerals are used for the preparation of various medicines. In Siddha, medicines have been classified into *Aga marunthukal* (Internal use) and *Pura marunthukal* (External use) having 32 form of medicines in each. The external medicines are very useful for topical ulcers, skin diseases like eczema, psoriasis and fungal infections. *Vanga vennai*, one of the external preparations of Siddha medicine, is being used for ulcers and fungal infections. This study is aimed to evaluate the antimicrobial and antifungal against selected human pathogens by disc –diffusion method in different concentrations. The result of this study shows that *Vanga vennai* possess significant anti fungal effect against *Candida albicans* (zone of inhibition 10 mm) and moderate anti – bacterial effect against *Klebsiella pneumoniae, Enterococcus fecalis* , *Staphylococcus aureus* (zone of inhibition 12mm, 12mm and 8mm respectively) when compared with standard drug. This study substantiated the textual reference of the test drug Vanga vennai. Further clinical studies are needed to evaluate its therapeutic effect.

Keywords: Siddha medicine; Vanga vennai; Anti-fungal; Anti-bacterial.

Introduction

An antimicrobial is an agent that kills microorganisms or inhibits their growth [1]. Antimicrobial medicines can be grouped according to the microorganisms they act primarily against. For example, antibacterials (commonly known as antibiotics) are used against bacteria and antifungal are used against fungi. Use of substances with antimicrobial properties is known to have been common practice for at least 2000 years.

Long before mankind discovered the existence of microbes, the idea that certain plants had healing potential, indeed, that they contained what we would currently characterize as antimicrobial principles, was well accepted. Since antiquity, man has used plants to treat common infectious diseases and some of these traditional medicines are still included as part of the habitual treatment of various diseases [(2)].

In the past few decades, a worldwide increase in the incidence of fungal infections has been observed. The majority of clinically used anti-fungal have various drawbacks in terms of toxicity, efficacy and cost, and their frequent use has led to the emergence of resistant strains The challenge has been to develop effective strategies for the treatment of candidiasis and other fungal diseases, considering the increase in opportunistic fungal infections in human immunodeficiency virus-positive patients and in others who are immune compromised due to cancer chemotherapy and the indiscriminate use of antibiotics[3].

Candidiasis, the main opportunistic fungal infection has steadily increased over the past 30 years [4, 5]. Among the many species, *Candida albicans* is the most important pathogen and oral candidiasis is an oral lesion caused by this organism. It has been estimated that more than 90% of human immunodeficiency virus (HIV) infected patients develop oral candidiasis often debilitating infection at some time during the progression of their disease [6, 7]. Infections caused by *C. albicans* in immune competent individuals may include oral thrush, vulvar rash, vaginitis, conjunctivitis, endophthalmitis, diaper rash, and infections of the nail, rectum, and other skin folds [8,9].

Candida spp. are the fourth leading cause of nosocomial infections in the USA and elsewhere in the world, and in those patients with candidemia, the attributable mortality rate is up to 35% [10]. Treatment of this fungal infection presents several problems. Besides the toxicity presented by amphotericin B, the widespread use of antifungal agents induced resistance to amphotericin B [11, 12].

Nature has been a source of medicinal agents for thousands of years and an impressive number of modern drugs have been isolated from natural sources; many of these isolations were based on the uses of the agents in traditional medicine. This plant-based, traditional medicine system continues to play an essential role in health care, with about 80% of the world's inhabitants relying mainly on traditional medicines for their primary health care. [13, 14]

Due to the development of resistance in known fungal pathogens and the emergence of fungal pathogens intrinsically resistant to the currently available antibiotics, it is important that novel antifungal agents be identified and developed [15].

Such a novel drug has been used in Siddha system of medicine to this problem for a long time is *Vanga vennai* [16], which is one of the external preparations and used for all types of skin infections. Hence, this study is aimed to assess the anti- fungal and anti-bacterial effect of *Vanga vennai in-vitro*.

Materials and Methods

Vanga vennai:

The test drug *Vanga vennai* has been prepared according the reference text.

Ingredients:

Vanga chenduram (Red lead)	-175 gm
Mirudarsringi (Litharge)	-175 gm
Thurusu (Blue vitreol)	-87.5 gm
Vennai (Butter)	-1400 gm

Process:

Finely powder the ingredients of *Vanga vennai* and mixed together then grounded with butter.

Anti- Microbial activity:

Disc-diffusion method:

The antibacterial activities of the sample VV were carried out by disc diffusion method. The concentrations of the test compounds were used at the concentration of 500, 1000, 2000 and 4000 µg. The target microorganisms were cultured in Mueller–Hinton broth (MHB). After 24 h the suspensions were adjusted to standard sub culture dilution. The Petri dishes containing Muller Hinton Agar (MHA) medium were cultured with diluted bacterial strain. Sabouraud dextrose was utilized for the growth of fungal strains. Disc made of Whatman No.1, diameter 6 mm was pre-sterilized and was maintained in aseptic chamber. Each concentration was injected to the sterile disc papers. Then the prepared discs were placed on the culture medium. Standard drug Streptomycin (10µg) was used as a positive reference standard to determine the sensitivity of each microbial species tested and 20 µl of DMSO was used as vehicle control. Then the inoculated plates were incubated at 37^{o} C for 24 h for bacteria and 72 h for fungus. The diameter of the clear zone around the disc was measured and expressed in millimeters as its anti-microbial property. The results were depicted in Table.No.1 & 2.

Results and Discussion

The antimicrobial efficacy of *Vanga vennai* samples was initially evaluated by the agar well diffusion method, using five strains *Escherichia coli*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Enterococcus fecalis*, *Candida albicans*. Table.1 shows the zones of inhibition (mm) of samples at different concentrations (500µg, 1000µg, 2000µg and 4000µg).

Table. No: 1. Anti- bacterial activity of *Vanga vennai*

Concentrations	Zone of Inhibition (ZOI) in mm			
	Organism			
	Escherichia coli	*Staphylococcus aureus*	*Klebsiella pneumoniae*	*Enterococcus fecalis*
VV (500 µg)	-	-	-	6
VV (1000 µg)	-	-	-	7
VV (2000 µg)	-	6	10	10
VV (4000 µg)	-	8	12	12
Streptomycin	23	23	17	27

Table. No : 2. Anti-Fungal activity of *Vanga vennai*

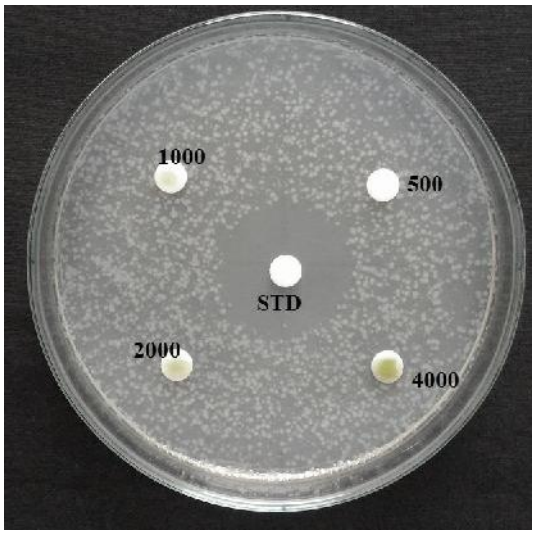
Organism	Zone of Inhibition (ZOI) in mm			
	Candida albicans			
Concentration	500 µg	1000 µg	2000 µg	4000 µg
VV	7	7	7	10
Amphotericin B 20 µg	10			

It was found that the test drug *Vanga vennai*, gave a zone of inhibition of around 6-12 mm, showing moderate activity against *Enterococcus fecalis* and *Klebsiella pneumoniae*. VV gave a zone of inhibition of around 6-8 mm, showing less effect against *Staphylococcus aureus* and no activity to *Escherichia coli*.

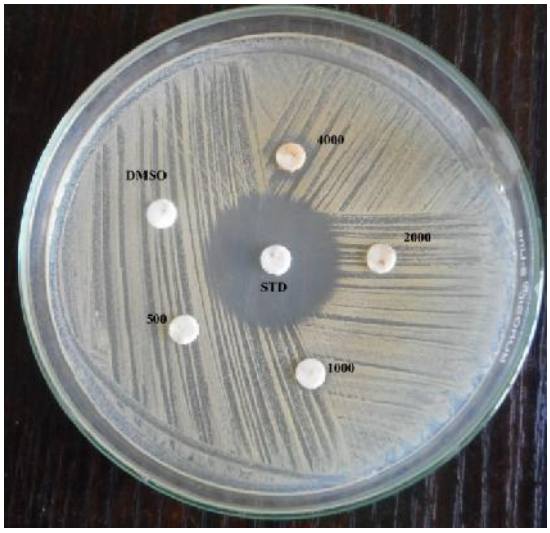
In anti fungal study, the test drug *Vanga vennai* gave a zone of inhibition around 7-10 mm, showing good activity against *Candida albicans* when compared with Amphotericin B (zone of inhibition 10 mm).

In Siddha literature the test drug *Vanga vennai* is indicated for all kind of skin infections which have been used for the same clinical condition by traditional healers and Siddha physicians from ancient time but, there is no scientific data on this test drug. This study results confirming whatever knowledge/ information were mentioned in ancient Siddha literature is true. This study is an initial step for validation of Siddha formulations. This result may encourage our researcher to concentrate more on traditional medicine for exploring the traditional knowledge.

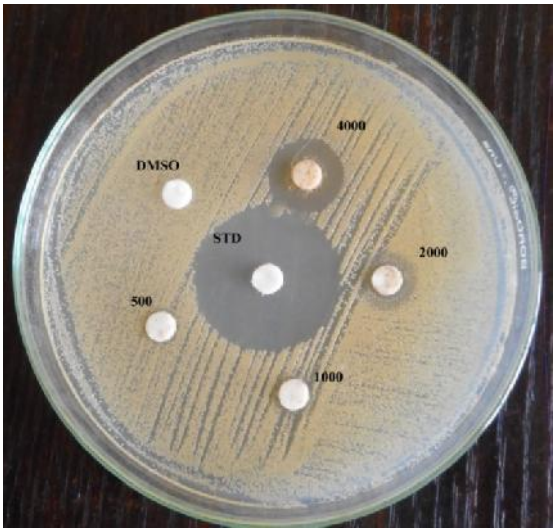
Effect of VV against ***Escherichia coli***



Effect of VV against ***Staphylococcus aureus***

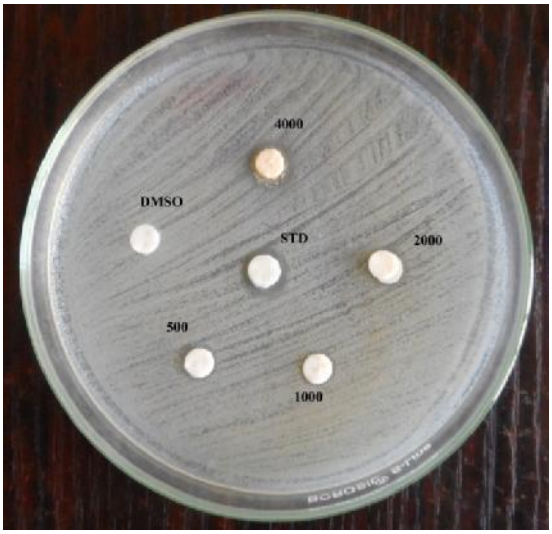


Effect of VV against ***Enterococcus fecalis***



Effect of VV against ***Klebsiella pneumoniae***

Effect of VV against ***Candida albicans***



Conclusion

The present study showed that *Vanga vennai* is effective against *C. albicans*. This investigation has opened up the possibility of the use of this Siddha preparation *Vanga vennai* in drug development for human use for various infectious diseases related to skin.

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