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RESEARCH ARTICLE

SCREENING OF ANTIMICROBIAL ACTIVITY OF VARNYA LEPA LOTION – A POLYHERBAL FORMULATION

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Abstract

Objective: To screen the antimicrobial activity of Varnya lepa lotion against skin infecting organisms like Staphylococcus aureus and Staphylococcus epidermis. In Ayurvedic literature, Varnya lepa is one of formulation which includes seven drugs like Raktachandan, Manjista, Kustha, Lodhra, Priyangu, Vatankura and Masura. It is mainly indicated for melasma, enhances complexion and hyperpigmentation.

Methods: The antimicrobial activity of Varnya lepa lotion was testing by using Agar well diffusion method/ Cup-plate method and Minimum inhibitory concentration(MIC).

Results: Through the Disc diffusion method, Varnya lepa lotion has shown no antibacterial activity but by MIC method has minimum antibacterial activity against Staphylococcus aureus and Staphylococcus epidermis organisms.

Conclusion: As herbal products are growing in the market tremendously and concerns surrounding their safety are also increasingly recognised. In this article, it is concluded that Varnya lepa lotion was having a minimum antibacterial effect against the organisms like Staphylococcus aureus and Staphylococcus epidermis and would protect skin from different infections.

Introduction:-

Healthy skin is the desire of every individual which enhances the pleasure and satisfaction of skin. [1] In current era, different type of skin infections like pimples, premature ageing..etc is very commonly occurring due to microbes such as bacteria, yeast..etc. For treatment failures to skin infections related to multidrug-resistant bacteria and it has become a global concern to public health. [2] [3] The various synthetic chemical creams, lotions like aminobenzoic acid derivatives, anthranilates, benzophenones, cinematics, salicylates, inorganics like titanium dioxide and zinc oxide may cause mild to a moderate allergic reaction in some individuals. Some of these may absorb into the bloodstream. [4] As a Herbal formulations have more acceptance because of their no side effects and they are less irritant. [5] Varnya lepa is one of the formulations which includes seven drugs like Raktachandan, Manjista, Kustha, Lodhra, Priyangu, Vatankura and Masura. It is mainly indicated for melasma, enhances complexion and hyperpigmentation. [6] The application of several lepas, creams, lotions will be based on skin

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temperature, duration and thickness. The common problems with lepas are smell, stains after applications, time management, fluidity and Greasiness. To overcome all these problems, the necessity of developing Varnya lepa lotion. [7] Lotions are liquids for external application that are intended to be applied to the unbroken skin without friction. They are aqueous, polyols or alcohol-based solutions or suspensions containing topically active therapeutic agents. [8] A Microorganism is available naturally in the surrounding environment; therefore they can easily reach while processing and packaging. [9] Microbes are responsible for activating certain skin infections and also leads to the decreased shelf life of products. Consequently, attention is devoted to safety, efficacy and antimicrobial activity on the growth of microorganisms like Staphylococcus aureus and Staphylococcus epidermis. The microorganisms susceptibility can change with time even during therapy with a specific drug. [10] Thus, it is essential for the physician to know the sensitivity of the pathogen before treatment.

Aim and Objective:-

To screen the antimicrobial activity of Varnya lepa lotion by Cup-plate/Disc Diffusion Test and MIC method.

Material and Methods:-

The raw drugs for Varnya lepa lotion like Raktachandan, Manjista, Kustha, Lodhra, Priyangu, Vatankura and Masura was procured from GMP certified pharmacy and made into hydroalcoholic extract (6:4) for further preparation of lotion. [11]

Methods:-

Disc Diffusion Test/ Cup-plate Method: [12]

Chemicals:

The chemical used for testing sample is DMSO(dimethylsulfoxide).

Test Organism:

The Organisms like Staphylococcus aureus and Staphylococcus epidermis were used for antibacterial activity testing.

Media Used:

The media was used as Brain Heart Infusion agar.

Temperature :

The Agar plates were bring to room temperature before use.

Inoculum preparation :

i. First step is using a loop or swab, transfer the colonies to the plates.
ii. Then, visually adjust turbidity with broth to equal that of a 0.5 McFarland turbidity standard that has been vortexed. On other hand, the suspension was standardize with a photometric device.

Inoculation of Agar plate:-

i. The inoculum was adjusting within 15min to a McFarland 0.5 turbidity standard, dip a sterile cotton swab into the inoculum and rotate it against the wall of the tube remove excess inoculum from above the liquid.
ii.Swab entire surface of agar plate three times, rotating plates approximately 60º between streaking to ensure even distribution. Keep away from hitting sides of petriplate and creating aerosols.
iii.Allow inoculated plate to stand for atleast 3 minutes but no longer than 15 min before making wells.

Stock solution preparation:-

Prepare the stock solution weighing 10mg of compound and dissolve it in 1ml of DMSO

Addition of compound into plate :-

1. Take hollow tube of 5mm diameter, heat it and press it on above inoculated Agar plate and remove it immediately by making a well in the plate. Similarly, make five well on each plate.
2. The help of micropipette add 75µl, 50µl, 25µl, 10µl and 5µl in each well.

Incubation :-

1. Incubate plates within 15 min of compound application.
2. The plates should inverted and stack them no more than five high.
3. Incubate for 18-24 hrs at 37 ºC in incubator.

MIC Test (Aerobic) :- [12]

1. The 9 dilutions of each drug have to be done with BHI for MIC.
2. The 20microliter of drug was added into the 380microliter of BHI broth in the initial tube.
3. For the dilutions 200microliter of BHI broth was added separately into the next 9 tubes.
4. The 200microliter was transferred to the first tube containing 200microliter of BHI broth from the initial tube. This was considered as 10-1 dilution.
5. 5) To make 10-2 dilution 200microliter was transferred from 10-1 diluted tube to second tube.
6. 6) For the each drug serial dilution was repeated up to 10-9 dilution.
7. 7) From the maintained stock cultures of required organisms, 5microliter was taken and added into 2ml of BHI (brain heart infusion) broth.
8. 8) The 200microliter of above culture suspension was added in each serially diluted tube.
9. 9) The tubes were incubated only for 24 hours and observed for turbidity.

Results And Observation:-

In the present study, varnya lepa lotion were tested for their antibacterial properties against the organism like Staphylococcus aureus and Staphylococcus epidermis. Result showed in Fig 1 and Fig. 2.i.e Through Disc diffusion method Varnya lepa lotion has showed no antibacterial activity but by MIC method has minimum antibacterial activity against Staphylococcus aureus and Staphylococcus epidermis organism.

MIC Results: Fig 1.

Sl. No.	Samples	100 µg/ml	50 µg/ml	25 µg/ml	12.5 µg/ml	6.25 µg/ml	3.12 µg/ml	1.6 µg/ml	0.8 µg/ml	0.4 µg/ml	0.2 µg/ml
	Lotion										
01	Staph aureus	S	S	R	R	R	R	R	R	R	R
02	Staph epidermidis	S	S	R	R	R	R	R	R	R	R

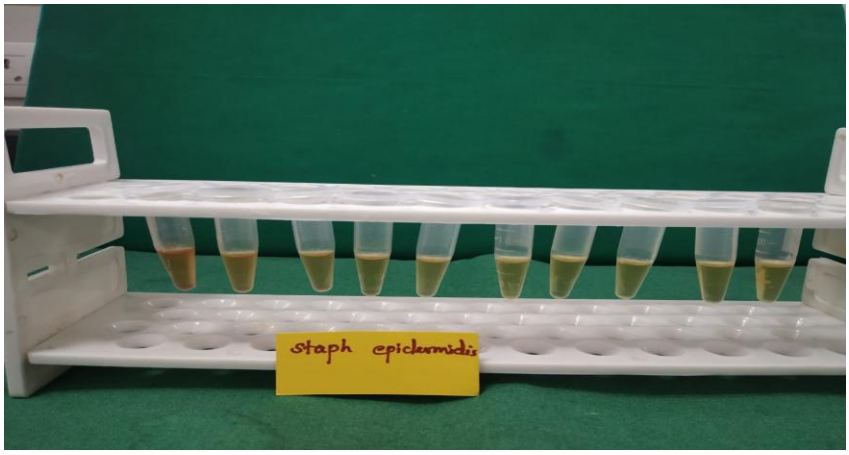

Fig:1:-

Disc Diffusion Results:Fig 2.

Sl. No.	Samples	75µl/ml	50µl/ml	25µl/ml	10µl/ml	5µl/ml
	Lotion					
1	Staph aureus	R	R	R	R	R
2	Staph epidermidis	R	R	R	R	R

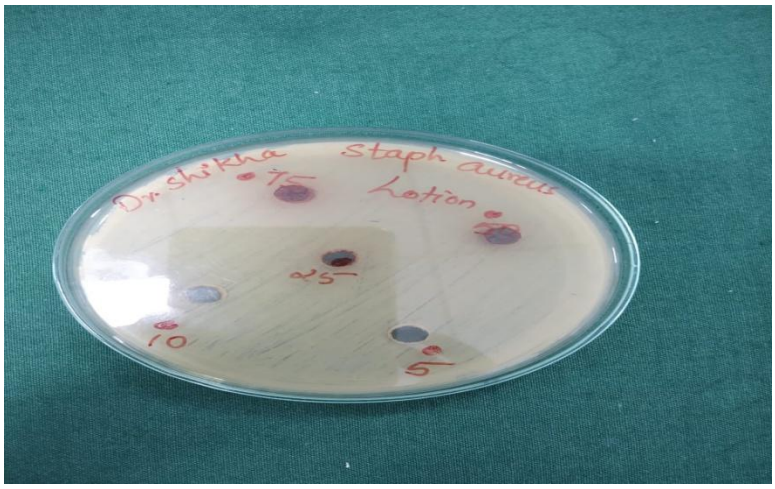

Fig:2:-

Note:
S- Sensitive
R- Resistant

Standard Values
For MIC

Ciprofloxacin:	
Staphylococcus aureus	2µg/ml
Staphylococcus epidermis	2µg/ml

For Disc diffusion

Ciprofloxacin:	
Staphylococcus aureus	26mm
Staphylococcus epidermis	25mm

Discussion and Conclusion:-

The role of herbal products in everyday life met greater acceptability. As herbal products are growing in market tremendously and concerns surroudings their safety are also increasingly recognised. In Ayurvedic literature describes various herb and other natural ingredients for external application in the form of herbal waters, packs, oils, powders etc. In this study, it is concluded that the varnya lepa lotion was developed and being evaluated for its safety concern. The results justified that Varnya lepa lotion was having minimum antibacterial effect against the organisms like Staphylococcus aureus and Staphylococcus epidermis and would protect skin from different infections.

References:-

1. Hazra J, Panda AK (2013) Concept of Beauty and Ayurveda Medicine. J Clin Exp Dermatol Res 4: 178. doi:10.4172/2155-9554.1000178.
2. A. Guschin, P. Ryzhikh, T. Rumyantseva, et al., Treatment efficacy, treatment failures and selection of macrolide resistance in patients with high load of Mycoplasma genitalium during treatment of male urethritis with Josamycin, BMC Infect. Dis. 15 (2015) 1–7.
3. I. Martin, P. Sawatzky, G. Liu, et al., Antimicrobial resistance to Neisseria gonorrhoeae in Canada: 2009–2013, Can. Commun. Dis. Rep. 41 (2015) 40–41.
4. RajendraJangade and S.J Daharwal. Herbal sunscreen : An overview. Research J Topical and Cosmetic Science.(2) July-Dec.2011.
5. Harsha Solanki, Vinay Sagar Verma, Mukesh Sharma, Ajay Singh, Garima Sharma, ManishaMajumdar, Sujata Gupta, Sandhya, Vandana Sahu, Kalyani Dewangan, D.K. Tripathi, Amit Alexander, Ajazuddin. Natural Humectants in Formulation of Calamine Lotion: Its Evaluation and Comparison. Research J. Topical and Cosmetic Sci. 2016; 7(2):41-45. doi: 10.5958/2321-5844.2016.00007.8

6. Rao G. Prabhakar A text book of SharangadharaSamhita, edition First, 2013, Chaukhambha Sanskrit Sansthan pg no- 396.
7. Hazra J, Panda AK (2013) Concept of Beauty and Ayurveda Medicine. J Clin Exp Dermatol Res 4: 178. doi:10.4172/2155-9554.1000178
8. Harsha Solanki, Vinay Sagar Verma, Mukesh Sharma, Ajay Singh, Garima Sharma, ManishaMajumdar, Sujata Gupta, Sandhya, Vandana Sahu, Kalyani Dewangan, D.K. Tripathi, Amit Alexander, Ajazuddin. Natural Humectants in Formulation of Calamine Lotion: Its Evaluation and Comparison. Research J. Topical and Cosmetic Sci. 2016; 7(2):41-45. doi: 10.5958/2321-5844.2016.00007.8
9. Faraja D. Gonelimali, Jiheng Lin, [...], and Shaimaa R. Hatab Antimicrobial Properties and Mechanism of Action of Some Plant Extracts Against Food Pathogens and Spoilage Microorganisms.
10. Mounyr Balouiri, Moulay Sadiki, and Saad Koraichi Ibnsouda Methods for **in vitro** evaluating antimicrobial activity: A review☆
11. Parekh J, Chanda S. In vitro antibacterial activity of the crude methanol extract of Woodfordia fructicosa Kurz. flower (Lythraceae) Braz J Microbiol. 2007;38:204–207. [Google Scholar].
12. Antimicrobial susceptibility testing protocols, Schwalve, Moore and Goodwin, cec Press 2007.