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Preparation and Evaluation of Gel Based Polyherbal Handwash

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ABSTRACT

Infectious diseases (ID) circulating in the home and community remain a significant concern. Two factors are largely responsible first is the constantly changing nature and range of pathogens to which we are exposed and, second is the demographic changes occurring in the community, which affect our resistance to infections. The impact of hand hygiene is important to reduce transmission of ID in the home and community. The antibacterial soap as they contain triclosan which have huge amount of disadvantage and also alcohol base sanitizer which causes irritation due to skin drying, so we can replace it with herbal antimicrobial ingredients. The gel base Polyherbal handwash which prepared by using herbs to avoid the side effects are Azadirachta indica, Citrus Limonis, Aloe vera. The antimicrobial activity of prepared gel base polyherbal handwash was checked by using cup plate method. The microorganism used in antimicrobial test is staphylococcus auries, Candida albicans & bacillus subtillis. The result showed that gel based polyherbal handwash give more inhibition than available commercial synthetic handwash.

Keywords:-Polyherbal handwash, Azadirachta indica, Citrus Limonis, Aloe vera, Antimicrobial activity.

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INTRODUCTION

The Hands are primary mode of transmission of microbial and infections, hygiene of hands are therefore the most important thing to avoid the transmission of harmful germs and prevent the infections. Hand hygiene is the single most important, simplest, and least expensive means of preventing nosocomial infections¹. Not only hand washing is critical in foodservice and food production operations, it is also important in homes and day care operations. Reported a study that demonstrated a decline in diarrheal illnesses in day care centers .when employees were taught to use good hand washing procedures². Hand washing is an important way to help fight the spread of disease. Hand washing removes visible dirt from hands and reduce the number of harmful microorganisms. Harmful bacteria and viruses such as, E. coli and Salmonella can be carried by people, animals or equipment and transmitted to food³.

The hands of health care workers (HCWs) are the primary routes of transmission of multidrug resistance pathogens and infections to patients. Hence it brings up the use of antiseptics for hand washing purposes⁴ Many of the chemical antiseptics are now available in market as alcohol base sanitizers. This soaps or solutions help to reduce healthcare associated transmission of contagious this is more effectively⁵. But they have some short coming or adverse effects. Their frequent use can lead to skin irritation and also resistance among pathogens.⁶ Organisms such as *Staphylococcus aureus*, *Pseudomonas spp.*, *Bacillus subtilis*, and *Candida albicans* are some of the positive agents of skin infections⁷

Azadirachta indica (Neem tree):

Neem tree also found in Indonesia in several areas such as Bali, Lombok, West Java, Central Java, and Nusa Tenggara Barat⁸. From the Meliaceae family, also known as Margosa or Indian lilac. Various parts of the Neem tree have been used as traditional Ayurvedic medicine in India. Neem leaves mainly contain quercetin (flavonoid) and nimbosterol (β - sitosterol) as well as number of liminoids (nimbin and its derivatives). Quercetin (a polyphenolic flavonoid) is known to have antibacterial and antifungal properties. Neem oil, the bark and leaf extracts have been therapeutically used as folk medicine to control leprosy, intestinal helminthiasis, respiratory disorders, and constipation and also as a general health promoter. Neem oil finds use to control various skin infections. Bark, leaf, root, flower and fruit together cure blood morbidity, biliary afflictions, itching, skin ulcers, burning sensations and it also reported to possess immune stimulant activity, hypoglycemic activity, antifertility activity, antiulcer activity, antimalarial

activity, antifungal activity, antiviral activity, anticancer activity, antioxidant activity and antibacterial activity⁹⁻¹³.

Aloe Vera:

Aloe vera gel obtains from plant aloe barbadensis which belongs to the family Lilliaceae. It is traditionally used for cosmetic and wound healing properties. It contain chemical constituents are antheracene glycosides, acemannan, anthraquinones including aloemodin and mixed anthrone-c and O glycoside including alongside A and B and flavonoids which show anti-microbial activity against pathogenic bacteria. The other activities reported are antifungal, anti-diabetic, anti-inflammatory, anticancer, immunomodulatory and gastro protective properties, which recommend its use in medicinal, cosmetic and food industries¹⁴⁻¹⁵. The Aloe vera gel has been attributed to Mannose-6-phosphate which show wound healing activity¹⁶.

Lemon juice:

Which is obtained from fruits of Citrus Limonis, belonging to the family Rutaceae The fruit juice mainly contain fruit acid, citric acid (18%), sugar. The inner layer of lemon fruit contain verity of coumarin derivatives and bitter flavones glycoside¹⁷. It mainly used for the cleaning purpose due to its disinfectant properties? Lemon juice is also used as a short term preservative in some food preparations. Lemon juice is used in Indian medicinal systems because of the anti-microbial properties of lemon. It is also used as a flavouring agent in food¹⁸. The hand wash was prepared from the methanolic extracts of Azadirachta indica, lemon juice, aloe Vera juice.

Antimicrobial Activity of plant extract

Cosmetic and pharmaceutical industries have an increasing interest in replacing synthetic antimicrobials in topical products. Besides the growing consumer interest for natural agents, microbial resistance to conventional antimicrobials is increasing¹⁹. Phenolic compounds are present in plants and it synthesized by plants for defense mechanisms²⁰. They can act by interacting with the microorganism's cell membrane or cell wall, leading to changes in membrane permeability, and resulting in cell destruction^{21,22}. Phenolic can also penetrate into bacterial cells and promote the coagulation of their content. In another way, phenolic compounds as natural antimicrobials could improve the shelf life of different products, inhibiting the growth of pathogenic microorganisms²³.

PROCEDURE

1) Preparation of extract:

The plant materials were collected from local market. The taxonomical identification and authentication was done by Department of botany, Y. C. Institute of Science, Satara. The hand

wash was prepared from the methanolic extracts of *Azadirachta indica*, Lemon juice, aloe Vera juice. 20g of the powdered leaves of neem plant were extracted with 100 ml of methanol solution (9 parts of methanol and 1 part of distilled water) by means of extraction. This mixture subjected to hot continuous extraction in a Soxhlet apparatus for 5-6hours. After complete extraction the solvent was evaporated and concentrated dry residue can obtained.

Table 1:Preparation of Gel Base for polyherbal handwash

Ingredients	Quantity Taken	Role
1. Cabopol-940	0.3 gm	Jelling agent
2. Purified Water	20 ml	Vehicle
3. Triethanolamine	q.s	Neutralizer

Table 2: Formulation of Gel Based Hand Wash using extracts of *Azadirachta indica*, *Aloe barbadensis* juice, *CitrousLimonis*juice.

Ingredients	Quantity Taken	Role
Gel base	20ml	To increase viscosity
Extract	2gm	Antibacterial and Anti-inflammatory agent.
Lemon juice/water	4ml/20ml	Antiseptic agent, cleanser
Aloe Vera juice	5ml	Healing agent
Honey	5 ml	Humectant, Moisturizing agent
Sodium lauryl sulphate	0.7gm	Surfactant
Peppermint oil	0.2ml	Cooling agent
Orange peel oil	0.5ml	Flavouring agent

Herbal gel was prepared using carbopol-934 as a gelling agent in 1% w/w concentration with deionized water overnight. Then the swelled polymer was stirred using a mechanical stirrer to ensure the uniform dispersion of the polymer. The pH was adjusted to 7.0 by the addition of minute quantities of Triethanolamine with continuous stirring. In mortar paste triturate methanolic extracts of *Azadirachta indica* and dissolve in 6-8 ml of methanol, then add 4ml lemon juice in 20 ml of distilled water and aloe vera juice, sodium lauryl sulphate according to above table. In mortar paste then add above gel preparation, honey and peppermint oil, perfumes or flavoring agent and add preservative in sufficient quantity. To prepare a formulation of hand wash gel as per Table 1 and 2. Then formulation was undergone organoleptic evaluation.

EVALUATION

Physical parameter

1. Colour: It was determined visually.
2. Odour: It was determined manually.
3. Appearance : It was determined visually
4. Homogenicity: It was determined visually

Chemical Parameter

A) pH determination

pH determine by using pH meter.

B) Viscosity

The viscosity of Poly Herbal Gel Based hand wash was determined by using digital Brookfield viscometer.

C) Foam Height

0.5gm of sample of Poly Herbal Gel Based hand wash was taken and dispersed in 25 ml distilled water. Then, transferred it into 500 ml stoppered measuring cylinder; volume was make up to 50 ml with water.

25 strokes was given & stand till aqueous volume measured up to 50ml& measured the foam height; above the aqueous volume.

D) Foam Retention

50 ml of the Poly Herbal Gel Based hand wash was taken into a 200ml graduated cylinder & shaken 10 times. The volume of foam at 1-minute intervals for 4 minutes was recorded.

Foam retention should remain stable for at least 5.

ANTI MICROBIAL STUDIES

The screening of anti-microbial efficacy of the formulated poly herbal hand wash and extracts was performed on various microorganisms by using cup plate method as per standard procedure.

Three sterile petri plates were taken for testing the anti-microbial activity against three different Micro-organisms, *Candida albicans*, *Staphylococcus aurous*, and *Bacillus subtiles* organism. The plates were filled with sterile nutrient agar solution and allowed for solidification. After solidification the microorganisms from the subculture were inoculated into the nutrient agar media under aseptic condition and two cavities were made in it. The first cavity is filled with gel base polyherbal handwash as a test and second one marketed commercial available synthetic handwash solution as a standard one. It was taken care that sample should be placed at the level of cavity means filled with 0.1ml solution of handwash. The plates are placed in incubator at 37⁰C to test the activity. After 24 hours the plates were observed for the formation of zone of inhibition. From the zone of inhibition the anti-microbial activity of formulation is estimate.

RESULTS AND DISCUSSION

All the observation data for evaluation of Polyherbal handwash presented as following.

Table 3: Evaluation of Polyherbal handwash

Sr.No.	Test	Observation
1)	Physical parameter	
	Colour	Brown
	Odour	Characteristics
	Appearance	Translucent
	Homogenicity	Good
2)	Chemical Parameter	
	p ^H	6.7
	Viscosity	45 c Pascal's
	Foam Height	290 ml
	Foam Retention	22.5 ml

Antimicrobial activity screening tests

The Anti-microbial efficacy of the formulated Herbal Hand Wash was tested on *Candida albicans*, *Staphylococcus aureus*, and *Bacillus subtilis* by cup plate technique. The results of cup plate method showed that the handwash prepared from methanol extract of the combined plant materials show greater activity than the activity of the commercially available hand wash shown in table 4.

Table 4: Zone of Inhibition

Sr.no	Micro organism	Zone of inhibition of Std. handwash(mm)	Zone of inhibition of Test handwash(mm)
1.	S.A	23	39
2.	B.S	25	45
3.	C.A	30	42

S.A - *Staphylococcus aureus*, B.S. - *Bacillus subtilis*, C.A. – *Candida albicans*

Plates showing zone of inhibition.

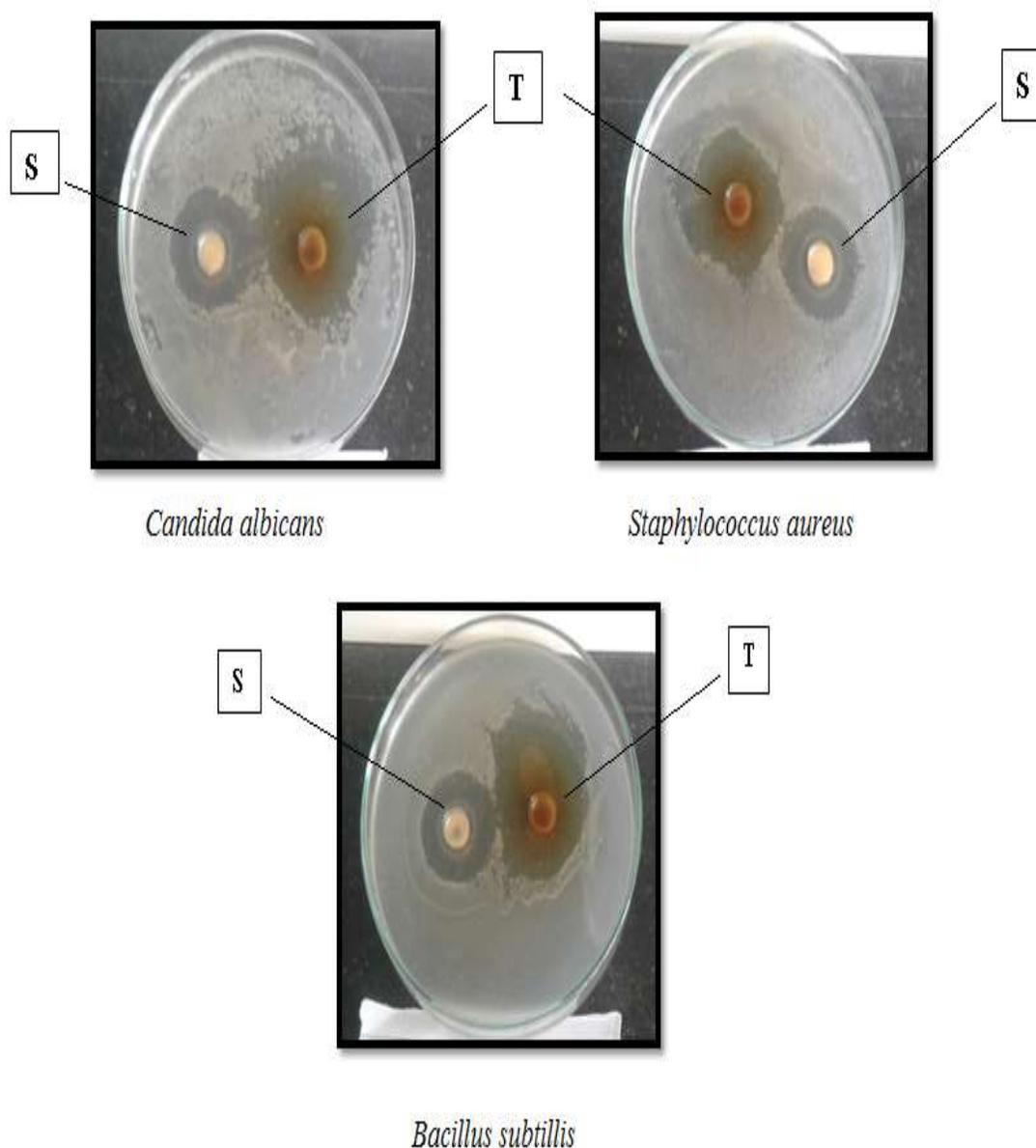


Figure 1: Photographs of Antimicrobial Activity

S:-Standard sample.

T:-Test sample.

DISCUSSION

Polyherbal gel base handwash formulation proved to be beneficial with excellent activity against all the tested microorganisms. Hence it was encouraging to be used as a potent antiseptic preparation.

CONCLUSION:

The results suggest that the constituents of the various extracts of *Azadirachta indica*, *Citrus Limonis* and *Aloe vera*, their combinations are capable of giving superior inhibition than the

commercially available antiseptic handwash against the skin pathogens. This might be rational basis for use of herbs in preparation of handwash. The herbal handwash prepared was checked for its efficacy using cup plate method. The results clearly proved that the prepared herbal handwash is more effective than the commercially available synthetic handwash. Thus, these compounds can be extracted and incorporated in bases, in order to prepare superior anti-microbial handwash with less or no side effects due to low conc. of surfactant and alcohol than, that of commercially available formulations. Hence a new way can be found to come back antibiotic resistant of pathogenic organism and provide safe and healthy living through germ free hand all though the removal is not 100% but a major number can be reduced.

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