



FORMULATION AND DEVELOPMENT OF MINT CONTAINING HERBAL HAND SANITIZER

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ABSTRACT

The main aim for the preparation of a poly herbal hand sanitizer is for "hand hygiene". It is a vital principle in the prevention, control, and reduction of any acquired infection. Mainly hand sanitizer can stop the chain of transmission of micro organisms and other bacteria from hand to different parts of our body. Hand hygiene is important and one of the most critical steps in food production, food service as well as in homes and other day care preparations. Hand sanitizer avoids adverse effects like itching, irritation, dermatitis etc. So, maintaining hand hygiene as the prime criteria-instead of some synthetic formulation, an attempt has been made to formulate an herbal hand wash by using some extracts of commonly available plants like Neem, clove and mint. The formulation was evaluated for its physical parameters. It is sure that these ingredients especially mint on combination behave as an effective hand sanitizer.

KEYWORDS: Hand hygiene, antimicrobial activity, nosocomial infections, herbal sanitizer.

INTRODUCTION

Hygiene is defined as the maintenance of the practice of cleanliness which is of most importance in the maintenance of well-being. Keeping body hygiene and using the cleanser are necessary for a healthy life. These concepts underscore the need to maintain hygiene in disease prevention.^[1]

Although good & simple hygiene technique is single most important, easy and least expensive means of preventing health care-associated (nosocomial) infections and the spread of antimicrobial multidrug resistance; but, unfortunately poor hand-hygiene practices are still observed due to lack of scientific knowledge, unawareness of risks and unavailability of hand-hygiene facilities.^[2] Nosocomial infections are those which are acquired or originated in a hospital or health care setting and are result of high prevalence of pathogens, high prevalence of compromised hosts, efficient mechanisms of transmission from patient to patient. Thus occurrence of nosocomial infections is alarmingly increasing and has emerged as a serious concern in hospital care outcome; resulting in prolonged hospitalization, ample disease and mortality, and excessive costs.^[3]

Escherichia coli, *Pseudomonas* spp., and *Staphylococcus aureus* are commonly involved opportunistic microorganisms that primarily cause nosocomial infections. Generally infectious sites are urinary tract, surgical wounds, respiratory tract, skin, blood,

gastrointestinal tract, and central nervous system. These pathogens also tend to become incorporated into the normal flora of health care workers. *Pseudomonas aeruginosa* is the most commonly detected microorganism in hospitalized patients and immunosuppressed people.^[4]

Usually, microbes residing on the hands are divided into resident and transient flora. Resident flora (e.g. *Corynebacterium diphtheriae*, *Staphylococcus aureus*, *Staphylococcus epidermidis* and *Streptococcus viridans*) colonizing deeper skin layers are more resistant to mechanical removal has lower pathogenic potential. Transient flora (e.g. *Staphylococcus aureus*, Gram-negative bacilli, *Candida species*) colonizes the superficial skin layers for short periods, is usually acquired by contact with a patient or contaminated environment and these microorganisms are easily removed by mechanical means such as hand washing and are responsible for most health care-associated infections and the spread of antimicrobial resistance.

In the current scenario of mechanized life style; a consumer will always prefer ready-made formulation of alcohol hand rub rather than hand washing (application of a non-antimicrobial or antimicrobial soap; and mechanical friction is generated by rubbing the hands together for 1 minute, followed by rinsing with water, and then drying thoroughly with a disposable towel).^[5] Traditional healers have long used plants to prevent or

cure infectious conditions. Plants are rich in a wide variety of secondary metabolites, such as tannins, terpenoids, alkaloids, and flavonoids, which have been found *in vitro* to have antimicrobial properties.^[6] Considering this ultimatum; an attempt has been made to screen classical literature for the herbs with antimicrobial properties and found that *Mentha piperita* (Peppermint leaves), *Eugenia caryophyllus* (Clove flower buds) and *Azadirachta indica* (Neem leaves) has those antimicrobial activities. To formulate and evaluate herbal sanitizer comprise of combination of alcoholic extracts *Azadirachta indica* (Neem) and *Mentha piperita* (Peppermint) along with oil of *Eugenia caryophyllus* (Clove) using suitable excipients; which can be used as a ready-made herbal hand sanitizer.

MATERIALS AND METHODS

Drugs and Chemicals

Leaves of Neem (*Azadirachta indica*), Peppermint (*Mentha piperita*), and flowering buds of clove (*Eugenia caryophyllus*) were collected from the local area of Bareilly region, India. The plant material were identified and authenticated by the Department of Pharmacognosy. The Herbarium specimen (No. 173, 174 and 175) of plant was deposited it has been identified from Shri Ram Murti Smarak College of Engineering and Technology, Bareilly.

Nutrient Agar (For bacterial cultivation, Standard antibiotics (Ampicillin, Amphotericin-B) were purchased from Hi-Media. Culture of microorganisms (overnight incubated), Alcohol were of lab grade. Pathogens selected for evaluation of anti-microbial activity of the test drug were specified below.

Bacterial strains

1. *Escherichia coli* (gram-ve) ATCC 10531
2. *Pseudomonas aeruginosa* (gram-ve) ATCC 25619
3. *Staphylococcus aureus* (gram +ve) ATCC 6538

Table 1: Formulation Composition of alcohol based Herbal Hand Sanitizer.

S.No.	Ingredients	Quantity given (%)	Quantity taken (gm/ml)	Uses
1.	Deionized water	30.0	9.0	Vehicle
2.	Alcohol denatured	62.0	18.6	Antibacterial
3.	Neem leaves extract	1.00	0.3	Antimicrobial
4.	Peppermint extract	1.5	0.45	Antibacterial
5.	Clove oil	0.50	0.15	Antibacterial
6.	Carbopol 940	0.50	0.15	Thickening agent
7.	Triethanolamine	0.70	0.21	Solubilizing agent
8.	Glycerine	2.30	0.69	Emollient
9.	Polysorbate 20	0.50	0.15	Emulsifier
10..	Preservative	0.50	0.15	Preservative
11.	Perfume	0.50	0.15	Fragrance

EVALUATION

Physical Evaluation

The Poly-Herbal Gel Based Hand wash was subjected to Physical evaluation visually. The test parameters were Colour and Odour.

Extraction of plant material^[7]

Ten grams of each dry plant material (Neem and Peppermint leaves) were added separately in 100 ml of methanol solution (9 parts of methanol: 1 part of water).



The mixture was heated on water bath at 60°C for 1 hour, then filtered and plant extract was collected.

Formulation of Herbal Hand Sanitizer

Carbopol was added to deionized water with constant stirring. After uniform mixing, Triethanolamine (TEA) was added with slow stirring to avoid formation of possible air bubbles in the product and kept aside for 24 hrs. Both the Neem and Peppermint extracts along with clove oil were added to denatured alcohol along with glycerine, polysorbate 20 were mixed with aqueous phase. Finally, 0.25% each of Methyl & Propyl Paraben was added as preservative and 0.5% of perfume and mixed with slow stirring to obtain uniform product. Prepared product was stored in air tight HDPE containers. (Table 1)



Fig 1: Herbal Hand Sanitizer.

Colour: It was determined visually.

Odour: It was determined manually.

Appearance and Homogeneity: The Poly-Herbal Gel Based Hand wash was subjected to check manually.

pH: 1 gm of sample of Poly Herbal Gel Based hand wash was taken and dissolved it into 100 ml distilled water. The pH of solution was taken in previously standardized digital pH meter.^[10]

In-vitro Antimicrobial activity by Agar plate diffusion method

In present study the antimicrobial activity of trial drug was carried out by the agar plate diffusion method. Different concentrations were incorporated into an agar medium in a Petri dish. Replicator device was used to inoculate multiple specimens on to a series of plates with varying concentration of antibiotics. Responses of organisms to the trial drugs were measured and compared with the response of the standard reference drug. Ampicillin was the standard reference for antibacterial study.

RESULT AND DISCUSSION

Total three bacterial species were selected in the present study to evaluate the anti- microbial activity of the test drugs. Different concentrations of the products were

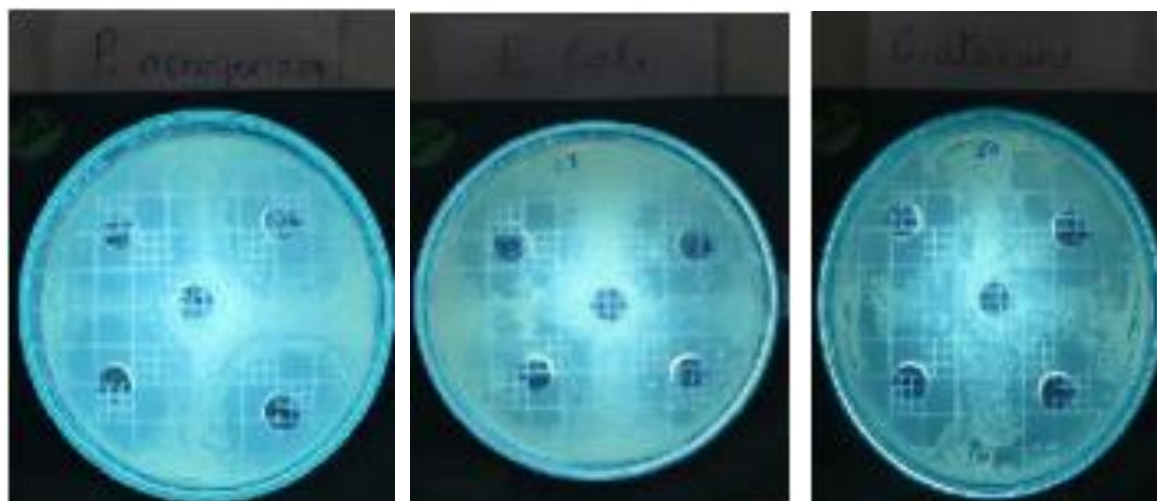
incubated and observed for the zone of inhibition (Table 3). The extracts were mixed in a fixed ratio of 1:1 and antimicrobial activity is carried out by counting the zone of inhibition. Gel at the concentrations 400 µg/ml was observed in two bacterial species (*E. coli*, *S. aureus*). The other one bacterial (*P. aeruginosa*) didn't significant to the maximum concentration (800 mcg/ml) of the gel in current study. This indicates that the hand Sanitizer has anti-bacterial activity particularly against *E. coli*, *S. aureus* and *P. aeruginosa* at minimum concentration of 400 mcg/ml (Fig.2). The resistant organism may respond to the test drugs in further concentrations.

Table 2:Evaluation Parameter.

S.No.	Parameters	Observation
1.	Colour	Green
2.	Odour	Characteristic
3.	pH	6.48
4.	Homogeneity	Homogenous
5.	Appearance	Translucent gel

Table 3: Result of antimicrobial activity of Hand Sanitizer (Mean + SD) (n= 3).

Conc. of Drug (µg/ml)	Zone of inhibition(diameter mm)				
	800	400	200	Control	SD
Organism					
<i>E.coli</i>	36±2.0	32±1.2	29±1.5	0	32±1.1
<i>P. aeruginosa</i>	30±1.7	25±1.5	20±1.7	0	37±1.2
<i>S. aureus</i>	32±1.8	30±2.0	28±1.9	0	31±1.1



1. *E.coli*

2. *P.aeruginosa*

3. *S.aureus*

Fig 2: Result of antimicrobial study of Hand Sanitizer against various pathogens

CONCLUSION

The prepared formulation of peppermint containing herbal hand sanitizer showed significant results at concentrations starting from 400 µg/ml against two bacterial species. The significance was found to be more in comparison to the standard reference. The composition (*Eugenia caryophyllus*, *Mentha piperita* and *Azadirachta indica*) has been attributed with properties like free radical scavenging, anti-helminthic, anti-microbial, anti-inflammatory and analgesic etc. More

concentrations may be needed to get a broad spectrum activity of the test drug. Scientific evidence and ease of use support of alcohol-based hand sanitizers during patient care. It may be concluded that Herbal Hand Sanitizer has a significant anti-microbial effect on the specified microorganisms except *P. aeruginosa*. Thus, there is immense potential in establishing the use of antimicrobial herbal products as a measure to control the multidrug resistant microbes and cheap in cost.

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