



Antimicrobial Herbal Soap Formulation

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Authors' contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

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ABSTRACT

Nosocomial infection has been recognized as a crucial issue in the outcome of hospital care, with significant morbidity and mortality results. The primary routes of infection transmission to patients are the hands of health-care workers. This also evokes utilization of antiseptics for hand-washing purposes. Many of the antiseptics commercially available are sanitizers dependent on alcohol that have certain shortcomings or harmful effects. The regular use can cause dermal irritation. The aim of the present study was to formulate an herbal hand-wash using leave extracts of *Terminalia catappa*, fruits of *Curcuma longa*, and rinds of *Garcinia indica*. Using the Disc diffusion process, the antibiotic activity against skin pathogens was evaluated, and results were compared with commercial antimicrobial hand-wash. The results showed that the herbal hand-wash was more effective against *Bacillus subtilis* and *Escherichia coli* than the commercially available antiseptic hand-wash. The effectiveness of herbal hand-wash was analysed using the Glove Juice method, which revealed that herbal hand-wash is effective in reducing pathogenic microbes present on the hands better than commercial hand-wash. These plant extracts can therefore be used for formulating herbal hand-wash to prevent hand borne microbial infections.

Keywords: *Curcuma longa*; *Garcinia indica*; herbal hand-wash; skin pathogen; *Terminalia catappa*.

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1. INTRODUCTION

The most vulnerable part of body, skin epidermis, needs to be protected from being exposed to pathogenic bacteria. Nosocomial infection has become a crucial problem in the outcome of hospital treatment, leading to prolonged hospitalization with the risk of mortality. The Health Care Workers' (HCWs) hands are the main routes of exposure of drug-resistant pathogens and severe infections [1]. This therefore evokes utility of antiseptics for hand cleaning process. Many of the chemicals antiseptic are commercially available as sanitizers consisting alcohol, chlorhexidine and so on. These hand-washes help to control contagious disease transmission associated with health care more effectively but they produce diverse effects on prolonged use. Their repeated application can lead to dermal irritation and also pathogen resistance. Some of the causative agents for skin infections are species such as *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Bacillus subtilis* and *Escherichia coli*. Some researchers have shown that growing resistance in microbes towards chemical antiseptics has led to severe disease outbreaks.

The plants have traditionally been a strong source of anti-infective agents. Plant-based antimicrobials are a huge untapped medicinal source. They are effective in treating infectious diseases while at the same time attenuating many of the side effects often associated with synthetic antimicrobials. The leaves of *Terminalia catappa* (Almond Tree) have antioxidant, sudorific and antibacterial effects, and used as remedy for leprosy. These natural sources have phytochemicals like tannins, quercetin, kaempferol. *Curcuma longa* (Turmeric) has a wide range of medicinal and pharmacological applications and is used as a spice, preservative, coloring matter. It has anti-inflammatory, anti-HIV, antibacterial, antioxidant, nematocidal, anti-parasitary, antispasmodic and anti-carcinogenic activity. Likewise, *Garcinia indica* rinds generally referred to as Kokam are rich source of organic acids primarily hydroxyl-citric acid and other garcinol components [2]. Malic acid, citric acid and tartaric acid are also present in small amounts. The rinds have been used as spice, while the fruit has been reported to be anthelmintic, cardiostonic, and effective in piles, dysentery, tumors, pains and heart attacks.

In the present study, *Terminalia catappa*, *Curcuma longa*, *Garcinia indica* were selected in

formulation of herbal hand-wash. *Terminalia catappa*, *Curcuma longa*, *Garcinia indica* were extracted with methanol to ensure maximum collection of phytochemicals that act on the pathogens of the skin and certify maximum antimicrobial activity. Then, Herbal hand-wash was prepared and screened against the skin pathogen.



Fig. 1. *Terminalia catappa* [3]



Fig. 2. *Curcuma longa* [4]



Fig. 3. *Garcinia indica* [5]

2. REVIEW OF LITERATURE

The extensive literature survey was performed using database platforms like PubMed to study regarding the formulation and evaluation of antiseptic properties displayed by herbal hand-wash Heyam et al. [1], study showed that formulation of herbal hand-wash was done by utilizing *Matricaria chamomilla* flowers along with their evaluation of antimicrobial property effective against *S. aureus* and *P. aeruginosa* skin pathogen and performed by using the disc diffusion method. Efficacy of the synthesized hand-wash was checked and compared to the commercially available hand-washes. Results showed that the herbal hand-wash was more efficient than commercial hand-washes in reducing the number of microorganisms.

Sharma et al. [6], research revealed that formulation and evaluation of nontoxic herbal hand-wash by using barks of the *Madhuca indica*. Evaluation of antimicrobial activity was done by disc diffusion methods and results revealed that synthesized formulation was more efficient than industrial antiseptic soaps.

Joshi et al. [2], study revealed Evaluation of herbal hand-wash formulation. Evaluation of antimicrobial efficacy of the herbal hand-wash was done by glove juice method and it revealed that synthesized herbal hand-wash was more proficient than commercially available hand-wash soap in reducing the number of organisms.

Patel et al. [7], study showed the formulation and evaluation of Herbal hand-wash by using ethanolic extract of *Glycyrrhiza glabra* root. Disc diffusion method was used to evaluate the antimicrobial activity of synthesized herbal hand-wash and its efficacy was evaluated and compared with the standard commercial hand-wash available. Results showed that the synthesized herbal hand-wash using extract of *Glycyrrhiza glabra* root was more efficient than commercially available hand-wash in reducing the organism with no side effects.

Mounika and Jyothi [8] study revealed Formulation and evaluation of poly herbal hand-wash gel which contains essential oils. Pour plate technique was used to evaluate the antimicrobial activity of synthesized poly herbal hand-wash gel. Results showed that the synthesized herbal hand-wash was more

proficient than commercially available hand-wash in reducing the organism with no side effects.

Power et al. [9], research showed the formulation and evaluation of poly herbal antibacterial gel based face wash using the *Garcinia indica* and *Curcuma longa* extract. Antimicrobial activity of formulated poly herbal antibacterial gel based hand-wash was evaluated against skin pathogen and its efficacy was tested using cup plate method. Results concluded that utilization of herbs in the formulation of hand-wash give a better effect with less or no side effects.

In the prior research conducted, hand-wash prepared using different plant extracts were found to be effective but lacked in terms of efficacy and reducing the skin pathogens from hands. Therefore, there is need to develop herbal hand-wash with enhanced efficacy to reduce to number of pathogens after using hand-wash. *Terminalia catappa* shows the antioxidant, antibacterial as well as sudorific properties. *Curcuma longa* possess antimicrobial, antioxidant and anti-inflammatory properties. *Garcinia indica* possess strong antioxidant and antimicrobial properties. So, in the present research *Terminalia catappa*, *Curcuma longa*, *Garcinia indica* were selected in formulation of herbal hand-wash.

3. METHODOLOGY

3.1 Design

To prepare herbal hand-wash, *Terminalia catappa*, *Curcuma longa*, *Garcinia indica* were selected in formulation of herbal hand-wash. *Terminalia catappa*, *Curcuma longa*, *Garcinia indica* were extracted with methanol to insure retrieval of phytochemicals from the natural sources. Then, Herbal hand-wash was prepared, screened against the skin pathogen and effectiveness of synthesized herbal extracts hand-wash was evaluated by in-vitro assays.

3.2 Sample

Plant:

Terminalia catappa, *Curcuma longa*, *Garcinia indica*.

3.3 Preparation of Plant Extracts

The plant materials were collected from the source. 10 g (dry weight) of each plant material

(plant leaves) was added separately in 100 ml solution of methanol. The mixture was heated at 60°C in water and incubated for an hour. The mixture was filtered and used as a methanolic extract for further use (Fig. 4) [10].

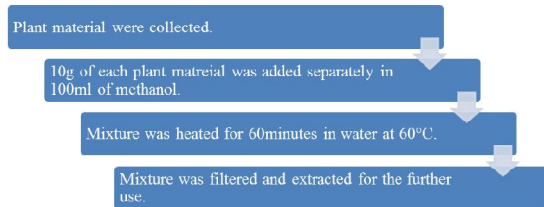


Fig. 4. Preparation of plant extract

3.4 Preparation of Hand-wash

The herbal hand-wash was formulated by adding 4ml methanol extracts of plant material in the 6 ml distilled water. In the final working volume (10 ml), 3 g of the sodium-lauryl-sulphate was used as per the standards. The solution was prepared homogeneously at room temperature and antibacterial activity against microorganisms was evaluated [2] (Fig. 5).

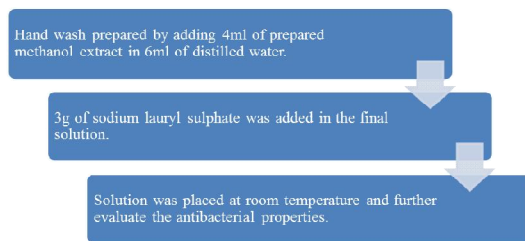


Fig. 5. Procedure of preparation of herbal hand-wash

3.5 Antimicrobial Assay

Pathogens from the hands of HCWs were retrieved. *Bacillus subtilis* and *Escherichia coli* were selected and used for the analysis of this research [11]. The antibacterial study of the combined plant extracts was evaluated by using disc diffusion method. *Bacillus subtilis* and *Escherichia coli* test cultures were used. Sterile 6 mm filter paper discs were filled with herbal hand-wash of 10ml. Care was also taken that the sterile discs efficiently percolated the hand-wash in its matrix. Discs of industrial antiseptic hand-wash and SLS (sodium lauryl sulphate) developed in similar ways. The discs were placed on Mueller Hinton agar, previously inoculated with the test cultures, when completely dried.

3.6 Physical evaluation of Hand-wash

Formulated hand-wash was evaluated for the color, pH, viscosity and spreadability. The observations are mentioned in the Table 3.

3.7 pH

The pH of hand-wash was found to be 6.5 and evaluated by using digital pH meter.

3.8 Viscosity

40 ml of hand-wash was poured in 100 ml of beaker and viscometer tip was dipped into the beaker and measured using digital Brookfield viscometer. Viscosity range of hand-wash was found to be 50-100 mP.

3.9 Spreadability

0.01 gm of the prepared hand-wash was placed into between two glass plates and the diameter was calculated after 1 minute and spreadability of hand was found optimum for topical application.

3.10 Efficacy Study

The efficacy of developed herbal hand-wash was calculated using Glove Juice method [12]. The test would assess product's efficacy in reducing microbial growth. The drug is applied to hands, gloves are provided, sampling solutions are incorporated inside glove and growth of bacterial cells was checked at regular intervals by extracting samples. The greater the reduction in log, the more effective the test product is. For this method *Bacillus subtilis* and *Escherichia coli* were used as test cultures.

3.11 Statistical Analysis

Statistical analysis was carried out by evaluating the standard deviation using Origin Pro software and statistical significance was analyzed by one-way ANOVA test.

4. RESULTS

Disc diffusion results showed that the hand-wash developed using methanol extract from the combination of naturally derived plant material has more efficient action as compared to commercial options of hand sanitizers. Also the sodium lauryl sulphate disc was kept as control.

Table 1. Antibiotic activity of herbal hand-wash, standard hand-wash and SLS. Herbal Hand-wash showed larger inhibition zone than commercial hand-wash against *Escherichia coli* and *Bacillus subtilis*

Organism	Antimicrobial activity as zone of inhibition (mm)		
	Herbal hand-wash	Standard hand-wash	SLS
<i>Escherichia coli</i>	18 ± 2	10 ± 1	14 ± 2
<i>Bacillus subtilis</i>	20 ± 2	8 ± 2	0

Values calculated as means ± SD (n=3): Results are significant (p<0.05)

Table 2. Effect of herbal hand-wash on reducing microbial load. Herbal hand-wash reduced 4 log CFU/ml microorganisms from the hands in comparison to commercially available hand-wash

Organism	Microbial load (CFU/ml)		
	Control	Antiseptic hand-wash	Herbal hand-wash
<i>Escherichia coli</i>	1.10×10 ⁶ ± 1.4	5.12×10 ⁶ ± 0.4	9×10 ³ ± 1.8
<i>Bacillus subtilis</i>	1.04×10 ⁷ ± 0.	1.9×10 ⁷ ± 0.2	7.9×10 ³ ± 0.4

Values calculated as means ± SD (n=3): Results are significant (p<0.05)

Table 3. Physical evaluation of herbal hand-wash

Herbal Hand-wash	Color	pH	Viscosity	Spreadability
	Green	6.64 ± 0.045	94 ± 0.36	13 ± 0.26

Values calculated as means ± SD (n=3): Results are significant (p<0.05)

The zone inhibition results of SLS discs showed that herbal hand-wash antibacterial activity is associated with the synergistic effect of active components not additives such as SLS. Table 1 shows the average result with standard deviation. The synthesized herbal hand-wash was able to reduce 4 log CFU/ml microorganisms from the hands in comparison to commercially available hand-wash which resulted in microbial decline to 2 log CFU/ml (Table 2).

4.1 Physical Evaluation of Hand-wash

The hand-wash was green in colour and translucent in appearance and pH also maintained which was found 6.64 ± 0.045. Spreadability was also measured and found to be less variation. The initial viscosities of developed gels were measured using Brookfield viscometer (Table 3).

5. DISCUSSION

T. catappa fruits are popularly used for therapeutic applications. These natural sources are also rich in complex substances, such as flavonoids, polyphenols, alkaloids, etc. The principal component is tannins. Tannins belong to class of water soluble polyphenols, generally found in two forms – hydrolysable and condensed [12]. Tannins exist in many foods.

The compounds have been reported to be bactericidal against *Bacillus subtilis*. They function upon the organism's membranes. Quercetin is the most important flavonoids. It exists as yellowish crystalline form, which prevents inflammation by interfering with histamine release. Therefore, it has been used as an anti-allergic, antihistamine, and anti-inflammatory drug. Quercetin has poor water solubility which causes the nutrient to be poorly absorbed. It functions as an antioxidant. *G. indica* rinds are rich source of organic acids like hydroxyl-citric acid a powerful inhibitor of ATP citrate lyase with possible effect on bacteria. The plants being studied are rich in these varied compounds in the present context and are therefore more protective against skin pathogens. The extract of methanol efficiently removes the phytochemicals and acids that work on the pathogens. The combination proves beneficial, and is therefore used to prepare herbal hand-wash. The prepared herbal hand-wash was tested for its effectiveness through diffusion of the discs. The findings clearly show that the thus prepared herbal hand-wash is much more successful than commercial antiseptic hand-wash. Consequently, the active components of the developed hand-wash are or eliminating microbial species exposure better than those used in normal hand-washes [13-17]. Thus, to prepare superior antiseptic hand-wash with

minimal side effects, these compounds can be extracted and formulated to develop more effective hand-wash [2]. A new way of overcoming the antibiotic drug resistance of pathogenic species has been developed for providing safer living through aseptic hands. While the exclusion is not 100%, it is possible to reduce a large amount.

6. CONCLUSION

In the prior studies it was noted that Nosocomial infection has been recognized as a crucial issue in the outcome of hospital care, resulting in significant morbidity and mortality. The primary routes of infection transmission to patients are the hands of health-care workers. Many of the antiseptics are sanitizers that dependent on alcohol and can have deleterious effects. Their regular use can cause irritation of the skin.

Therefore, herbal hand-wash was prepared using herbal extract *T. catappa*, *C. longa* and *G. indica*. The present results indicate that the ingredients of *T. catappa*, *C. longa* and *G. indica* extracts and their combinatorial compositions are capable of developing better antiseptic hand-wash against skin pathogens than the commercially available preparations. Therefore, a new way, of combating antibiotic drug resistance of pathogenic organisms and healthier living by germ-free aseptic hands can be found. A significant number of microbial load can be reduced by natural, economic and safe hand-wash. This may serve as the reasonable basis for the preparation of the herbal hand-wash. This has opened new avenues in the production of 'antiseptic hand-wash' replacing the use of chemical substances.

DISCLAIMER

The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

CONSENT

It is not applicable.

ETHICAL APPROVAL

It is not applicable.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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