

# Formulation and Evaluation of Herbal Hand Wash Gel

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## I. INTRODUCTION

Herbal medicine also called botanical medicine or phytomedicine refers to using a plant's seeds, berries, roots, leaves, bark, or flowers for medicinal purposes. Herbalism has a long tradition of use outside of conventional medicine. It is becoming more main stream as improvements in analysis and quality control along with advances in clinical research show the value of herbal medicine in the treating and preventing disease.

Hand hygiene is a fundamental principle and practice in the prevention, control, and reduction of healthcare-acquired infection. Good and simple hygienic practice i.e. correct hand washing and drying techniques will stop the chain of transmission of deadly pathogens (from the contaminated surface/site) from hands to other parts of the body.

Hygiene is defined as maintenance of healthy practices. Modern society refers hygiene as cleanliness. However with the development of the germ theory of disease, hygiene refers to any practice, which reduces the harmful bacteria to sub-pathogenic levels so that it does not cause any disease. Good hygiene is an aid to health, beauty, comfort, and social interactions. It aids in disease prevention and isolation. The human skin covers the external surface of the body and varies according to its function like thermoregulation, sensation secretion of substances and serves as matrix for harboring a variety of microbes. These in turn metabolize these secretions and produce specific odorous compounds responsible for characteristic skin odor.

Hands perform the majority of functions of the human's body and are exposed to a variety of substances which include soil during farming, food during cooking, touching raw and contaminated

food material, during personal hygiene. Clean hands stop the spread of germs; therefore hand washing is often emphasized as the single most important measure in any infection control program for preventing cross transmission of microorganisms between patients. Hand washing is the act of cleaning the hands with or other liquid with or without the use of soap or other detergent to remove dirt or loose transient flora thus preventing cross-infection<sup>1</sup>.

Today microbes are getting refractory to antimicrobial drugs and these drug resistant microbes are responsible for occurrence of chronic diseases. Methicillin resistant *S. aureus* (MRSA) is one of the predominant microbes reported to be responsible for hospital acquired (nosocomial) or community-acquired infections. A parallel increase has also been registered in the numbers of Vancomycin resistant *Enterococci* (VRE), Vancomycin intermediate *S. aureus* (VISA) and Vancomycin resistant *S. aureus* (VRSA). MRSA has been isolated from nosocomial environment as well as from the community. Transmission of MRSA from patient to patient after direct contact with asymptomatic healthcare providers or diseased individuals in hospitals as well as community has been traced and documented. Hands of health care workers (HCW's) are the primary mode of transmission of MRSA and other pathogens to new admitted patients as well as in community. Viable microorganisms are easily shed from the patient, or health worker or reservoir's skin, bed linen, bed furniture and other objects in the patient's immediate environment can easily become contaminated with patient flora<sup>2</sup>.

Prevention is better than cure and one of the best ways to avoid any sort of biological and chemical contamination is by adopting proper

methods of hand washing. Hand washing is absolutely essential to prevent contagious diseases and illness. In hospitals, proper hand washing techniques become more crucial as the surgeons, doctors and nurses come in frequent contact with various infected patients. With the frequent outbreaks of infections like swine flu and bird flu, hand washing has assumed a greater significance and hand sanitizers have become a part of every household. Although, hand sanitizers have become very popular, there has been an ongoing debate about the hand sanitizer ingredients. The effectiveness of hand sanitizers has been marketed

as '99.99% germ killing' and as a 'way to wash your hands'.<sup>2</sup>

Hand Washing removes visible dirt from hands and reduces the number of harmful microorganisms such as E.coli and salmonella may be carried by people, animals, or equipment & transmitted to food. To defend the skin from harmful microorganisms and to avoid spreading of various contagious diseases, hand washing is extremely important precaution<sup>3</sup>.

## II. MATERIALS AND METHODS

### HERBAL INGREDIENTS

#### 1. NEEM

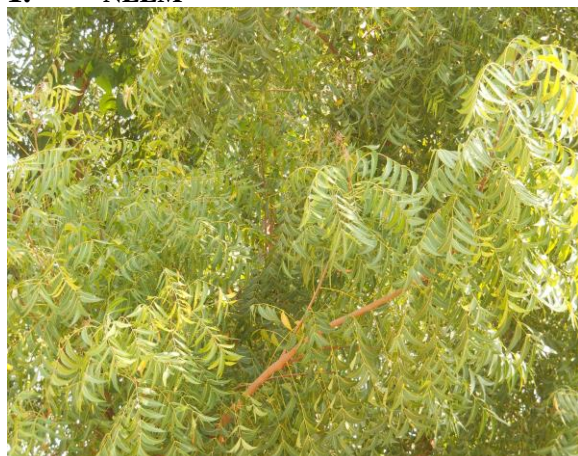


Fig 1: Habitat of Neem tree

#### BIOLOGICAL SOURCE:

It consists of the dried as well as fresh leaves of the plant *Azadirachta indica*.

#### FAMILY:

Maliaceae

#### CHEMICAL CONSTITUENTS:

The major chemical constituents of Neem are Azadirachtin, Nimbin, Nimbodin, Nimbolides, Nimbodin, Nimbosterol<sup>4</sup>

#### MEDICINAL USES:

Various parts of the neem tree have been used as traditional Ayurvedic medicine in India. Neem oil and 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. It is also used for the treatment of rheumatism, chronic syphilitic sores and indolent

ulcer. 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 immunostimulant activity, hypoglycaemic activity, antifertility activity, antiulcer activity, antimalarial activity, antifungal activity, antiviral activity, anticancer activity, antioxidant activity and antibacterial activity<sup>7-9</sup>.

Oil from the leaves, seed and bark possesses a wide spectrum of antibacterial action against gram-negative and gram-positive microorganisms, including *M. tuberculosis* and streptomycin resistant strains. *In vitro*, it inhibits *Vibrio cholera*, *Klebsiella pneumoniae*, *M. tuberculosis* and *M. pyogenes*. Antimicrobial effects of neem extract have been demonstrated against *Streptococcus mutans* and *S. faecalis*<sup>5</sup>.

#### SOME SPECIAL FEATURES:

1. As per Ayurveda, Neem is one of the most powerful blood-purifiers and detoxifiers.
2. Neem is used for manufacturing of many health and beauty care products. These products include bath powders, soaps, shampoos, cream and lotions.
3. Neem is the most effective and environmentally friendly source of pest.
4. Dry neem leaves are used in libraries to keep off book-eating worms. They are placed in between the books in the shelves.
5. Traditionally the twig of neem is being used as the tooth brush as it is believed to take care for teeth<sup>6</sup>.
6. Methanolic extraction from the flowers have shown prenylated flavonoids (5,7,4'- trihydroxy- 8- prenylflavanone, 5,4'- dihydroxy- 7- methoxy- 8- prenylflavanone, 5,7,4'- trihydroxy- 3',8- diprenylflavanone, 5,7,4'- trihydroxy- 3',5'- diprenylflavanone).

Other known constituents present in flowers are triterpenoid (trichilenone acetate), flavanones, nimbaflavone, 3'-prenylaringenin and 4- (2- hydroxyethyl) phenol<sup>10</sup>.

7. Biochemical analysis done on leaf extracts has revealed high presence of proline, which is a current treatment for neurodegenerative diseases

## 2. ALOE VERA



Fig 3: Habitat of Aloe vera plant

### BIOLOGICAL SOURCE:

It consists of the fresh juice of the plant called *Aloe barbadensis*.

### FAMILY:

Liliaceae

### CHEMICAL CONSTITUENTS:

Antheracene derivatives: anthrone-10-c-glycosyls including aloin A, aloin B, 7-hydroxyaloin A and 1, 8-dihydroxy ions include aloe emodin, 6 cinnamic acid esters of these compounds. 2-alkylchromones: including aloe resins B, C and D. Flavonoid Compounds: Aloe Capensis 5-hydroxyaloin and dihydroxy anthraquinones including aloe-emodin and mixed anthrone-c and O glycoside including aloin A and B 2-Alkylchromones: Including aloe resins A, B, C and D Flavonoids.

### USES:

Aloe Vera is used for enhancing skin growth.

Acid inside Aloe Vera is used as effective pain killers.

Antiseptic made from Aloe Vera is used to kill mold, bacteria, fungi and viruses.

Aloe Vera uses include help in skin blemishes.

Acne is treated by Aloe Vera gel and it's really effective.

Aloe Vera also helps in stopping baldness.

like Alzheimer's and Parkinson's disease, Type 2 Diabetes Mellitus and Polycythemia<sup>10</sup>.

8. The leaf of the Neem contains glycoproteins showed immune-modulatory activity, providing the potential to restrict tumor growth by modulating local and systemic immunity<sup>10</sup>.

Important Aloe Vera uses include relief from cuts, bruises and burns.

Aloe Vera uses include aphrodisiac.

Uses of Aloe Vera include expelling our worms.

Act as purgative for lazy and weak people.

Aloe Vera used to help healing blisters.

Jaundice is also healed by Aloe Vera plants<sup>11</sup>.

### SPECIAL FEATURES

1. Aloe vera gel coating could be an effective process of managing berries fruit quality and increasing shelf life. The fruit covered with Aloe vera gel exhibited maximum anti-oxidant ability, total phenol content and total anthocyanin as compared to non-coated fruits.

2. The compounds in Aloe are effective against other viruses through processes such as virus enzyme interaction, viral envelope destruction etc.

3. Minerals like Zinc, which have been demonstrated to have an effect on SARS-CoV-1, Acemannan in Aloe vera is responsible for anti-viral action. It has been demonstrated to decrease herpes virus infection. Aloe-emodin is used as anti-viral agent against different viral infection including herpes virus, influenza virus, pseudorabies virus and varicella zoster virus<sup>12</sup>.

## 3. SOAP NUT

It is used for the foaming activity. Biological source is *Sapindus mukorossi*. The pericarp or the shell of the fruit contains saponins (10%) having anti-microbial, anti-tumor, and surfactant properties. The major phytoconstituents include triterpenoidal saponins like tirucullane, oleanane (Sapindoside A and B), and dammarane. Due to its satisfactory detergency and antimicrobial properties, the extract of soap nuts was utilized to formulate the hand wash, being an excellent replacement for synthetic surfactants<sup>13</sup>.

## 4. LEMON JUICE

Citrus Limon belongs to the family Rutaceae. It is traditionally used as disinfectant. Lemon juice is used as a preservative for short time in some food preparations. Lemon juice is

used in traditional system of medicine because of the antimicrobial properties of lemon<sup>14</sup>.

### GLYCERIN

In its purest form, glycerin is nothing but alcohol called as glycerol. When glycerol is used as an ingredient in various cosmetic and pharmaceutical products, it is modified in concentration and is called as glycerin. Glycerin is a colorless, sweet tasting and thick liquid and it forms a thick paste on freezing. Glycerin easily dissolves in water or alcohol but not in oils. Glycerin is hygroscopic in nature that is it absorbs water from the air. One of the hand sanitizer ingredients, glycerin softens the skin when it is added in water. Glycerin maintains or balances the moisture levels of the skin. Know more on glycerin uses for skin. Alcohol and water are the two main hand wash ingredients and there has been some controversy on the high alcohol content in some of the hand wash.

### CHEMICAL INGREDIENTS

#### 5. CARBOPOL

Gel base

#### 6. METHYL PARABEN

It is used as a preservative.

#### 7. SANDAL WOOD OIL AND AMARANTH

Sandal wood oil is used for the flavoring and Amaranth is used for coloring agent.

### FORMULATION

**Table 1:** Formulation

| S. NO | INGREDIENTS     | QUANTITY (100 gm) |
|-------|-----------------|-------------------|
| 1     | Neem Extract    | 30ml              |
| 2     | Aloe Vera Gel   | 8g                |
| 3     | Lemon juice     | 4ml               |
| 4     | Carbopol        | 500 mg            |
| 5     | Glycerin        | 6ml               |
| 6     | Triethanolamine | 0.5 ml            |
| 7     | Soap nut powder | 5gm               |
| 8     | Propyl paraben  | 0.5mg             |
| 9     | Sandal wood oil | q.s               |
| 10    | Distilled water | q.s               |

### EXTRACTION OF NEEM LEAVES

10 g of the powdered Neem leaves were extracted with 100 ml of methanol solution (9parts of methanol and 1 part of distilled water) by means of maceration. This mixture was heated in water bath at 60°C for 1 hour. The content was filtered through

Whatman filter paper in order to get particle free extract. Filtrate was used as methanol extract<sup>15</sup>.

### FORMULATION OF GEL BASE

Gel was formulated using Carbopol 940 (Harisaranraj et al., 2010). The gelling agent was dispersed in a small quantity of distilled water with stirring and glycerin was added slowly<sup>16</sup>.

### METHOD OF PREPARATION

30 ml of Neem leaves extract and aloe vera gel (8gm) were are taken and it was dispersed in gel base. To the above mixture Soap nut powder (5gm), Propylparaben (0.5mg) and lemon juice (4ml) and glycerin (6ml) were added. The solution was made homogenous under room temperature. Finally sufficient quantity of sandal wood oil as flavoring agent and amaranth as colouring agent were added.

### EVALUATION METHODS FOR FORMULATED HERBAL HAND WASH GEL

#### i) Skin Irritation Test:

There was no irritation or Erythema produced in any of the Human volunteer during the stability period of 21 days, where the formulation was applied every 7 days once.

#### ii) Stability :

The stability studies were carried out with two different types of hand wash formulations i.e one with lemon juice and the other without lemon juice and it was stored at different temperature conditions like 4°C, 25°C & 37°C for 1 week, then its stability was observed.

During the stability studies no change in color and no phase separation were observed in the formulated hand wash. Also the formulations withstand its activity.

#### iii) P<sup>H</sup>

The P<sup>H</sup> was determined by using digital P<sup>H</sup> meter. 60ml of herbal hand wash is taken in beaker and dipped the bulb of the P<sup>H</sup> meter into the formulation and the P<sup>H</sup> was measured .

The P<sup>H</sup> of herbal hand wash was found to be **6.66**

#### iv) Viscosity

The viscosity of hand wash was determined by using digital Brookfield viscometer. 50ml of herbal hand wash is taken into 100ml of beaker and the tip of viscometer was dipped into the beaker containing hand wash formulation its viscosity was measured.

The viscosity range of hand wash was found to be 40 – 120 m.pascals



| SNo. | Micro organisms              | Zone of inhibition in Cm |                |
|------|------------------------------|--------------------------|----------------|
|      |                              | T <sub>1</sub>           | T <sub>2</sub> |
| 1    | <i>Staphylococcus aureus</i> | 0.9                      | 0.7            |
| 2    | <i>Bacillus subtilis</i>     | 0.6                      | 0.5            |

**Table 3: Showing zone of inhibition**

**BIOLOGICAL EVALUATION TEST**

**I. ANTI MICROBIAL TEST:**

- The screening of Anti-microbial efficacy of the formulated Poly Herbal Hand Wash and extracts was performed on various micro organisms by using Dip well method as per standard procedure.
- Two sterile Petri plates are taken for testing the anti microbial activity against two different microorganisms i.e *Staphylococcus aureus*, and *Bacillus subtilis*.
- First nutrient broth solution was prepared and inoculated the micro organism and kept in incubator at 37<sup>0</sup> for 24 hours to grow the micro organism.
- The next day nutrient agar solution was prepared and allowed for solidification. After solidification the microorganisms were inoculated into the nutrient agar media and five cavities were made in it.
- The first cavity is filled with marketed herbal soap solution as a standard one and second one with herbal hand wash.
- It was taken care that sample should be placed at the level of cavity. The petri plates are placed in incubator at 37<sup>0</sup> C to test the activity.
- Next day the petri plates were observed for the formation of zone of inhibition. From the zone of inhibition the anti microbial activity of formulation is estimated.

T1= Herbal soap solution (standard)

T2 = Herbal hand wash gel

**Figures showing zone of inhibition**



Fig:4: Plate showing inhibition

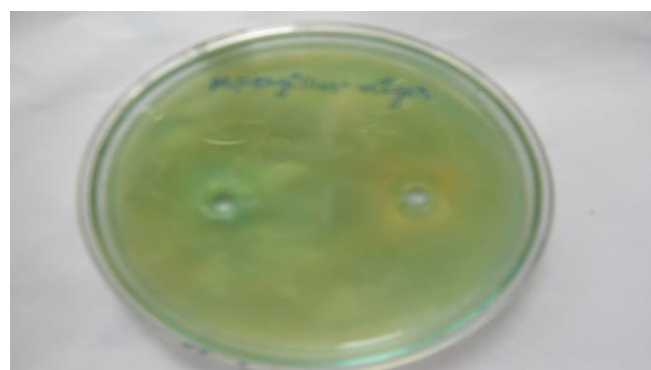


Fig:4: Plate showing inhibition

**III. RESULTS**

**ANTI MICROBIAL STUDIES:**

The Anti-microbial efficacy of the formulated Herbal Hand Wash was tested on *Staphylococcus aureus* and *Bacillus subtilis* by Dip well technique. The results of dip well method showed that the prepared hand wash showed good antimicrobial activity when compared with the commercially available hand wash. The results of experiment are as follows:

**IV. DISCUSSION**

Neem is reported to contain diterpenoids like nimbinone and nimbine which possess antibacterial activity against various gram positive and gram negative organisms. The results from dip well method showed that the hand wash prepared from methanol extract of the neem showed very good antimicrobial activity and the results were comparable with commercially available hand wash. The activity of the prepared formulation may be due to the combined activity of the phytoconstituents present in the neem and the

citric acid which is present in the lemon juice. Citric acid is reported to have potential effect on bacteria. In the present context the plants under study are rich in these varied compounds and hence are more effective against skin pathogens.

## V. SUMMARY AND CONCLUSION

The results suggest that the constituents of the extracts of *Azadirachta indica*, and lemon juice are capable of giving good inhibition against the skin pathogens. This might be rational basis for use of herbs in preparation of hand wash and use of these compounds in making antiseptic lotions or soaps in place of chemicals. The leaves of *Azadirachta indica* are widely used for medicinal purposes. The herbal wash prepared was checked for its efficacy using dip well method. The results clearly proved that the efficacies of prepared herbal hand wash. Thus, these compounds can be extracted and incorporated in bases in order to prepare superior anti microbial hand wash with less or no side effects. 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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