

Formulations of Antimicrobial Polyherbal Hand wash

Miss. Prital Vinayak Phad, DR.Hingane.L.D

Aditya Pharmacy College, Beed

Abstract: The aim of present work was to prepare formulations of polyherbal handwash from the methanolic extracts of leaves of *Mimosa pudica* (touch me not), *Azadirachta indica* (Neem) and lemon juice.

Two formulations of hand wash were prepared and the formulations were evaluated for physical properties like appearance, pH and viscosity.

The antimicrobial activity of prepared formulations of hand wash was checked against skin pathogens *Bacillus subtilis*, *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Escherichia coli* by agar diffusion method.

The results revealed that prepared herbal hand wash formulations showed significant zone of inhibition compared with standard antibiotic drug (Amoxicillin). So these plant materials can be used in the preparation of herbal hand wash on commercial scale.

Keywords: Polyherbal hand wash, Antimicrobial activity, *Mimosa pudica* and *Azadirachta indica*.

INTRODUCTION

Skin being the most exposed part of our body requires protection from skin pathogens. The hands of Health Care workers (HCWs) are the primary routes of transmission of multidrug resistant pathogens and infection to the patients. Hence, it brings up the use of antiseptic for hand wash purpose.

Many of the chemical antiseptics are now available in market as alcohol based sanitizers, chlorhexidine products etc. These soaps or solutions help to reduce health care associated transmission of contagious diseases more effectively but they have some shortcomings or adverse effects. Their frequent use can lead to skin irritation and also resistant among pathogens.

Organisms such as *Staphylococcus aureus*, *Pseudomonas spp.*, *Klebsiella pneumonia* & *Proteus vulgaris* are some of the skin pathogens.

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 food.

Antimicrobial properties of certain Indian medicinal plants were reported based on folklore information and only few reports are available on inhibitory activity against certain pathogenic bacteria and fungi. Use of plants as source of medicine has been inherited and is an important component of the health care system in India.

In these systems of Indian medicine, most practitioners formulate and dispense their own recipes; hence this requires proper documentation and research

Description of Plant Mimosa Pudica



Mimosa pudica (from Latin: *pudica* "shy, bashful or shrinking"; also called **sensitive plant, sleepy plant, action plant, touch-me-not, shameplant**) is a creeping annual or perennial flowering plant of the pea/legume family Fabaceae.

It is often grown for its curiosity value: the compound leaves fold inward and droop when touched or shaken, defending themselves from harm, and re-open a few minutes later. In the UK it has gained the Royal Horticultural Society's Award of Garden Merit.

The species is native to the Caribbean and South and Central America, but is now a pantropical weed, and can now be found in the Southern United States, South Asia, East Asia, Micronesia, Australia, South Africa, and West Africa as well. It is not shade-tolerant and is primarily found on soils with low nutrient concentrations.

USES OF MIMOSA PUDICA

The infusion of the twigs with leaves possess antipyretic, soothing and sudorific properties.

Apart from the toxicity from mimosine, the plant has shown a significant hypotensive, sedative and anti-inflammatory effect.

An alcoholic extract has shown to lower blood glucose concentrations in diabetic rats.

The alkaloidal extracts of the roots is antagonistic to both acetylcholine and histamine and histamine.

A crude ethanol extract of the leaves was found to have insecticidal activity against one *Trilobium* species.

Biology Of Mimosa Pudica

Perennial plant.- It reproduces by seeds.

Fruits- are disseminated by animals. The persistence and spread of sensitive plant is aided by its prolific seed set. There are approximately 110 seeds/g. It can complete its life cycle in 3 months.

At nightfall the leaflets fold together and the rachises bend down.

Flowers- throughout the year.

Seed germinate -**Germinate** within 2 weeks (scarification helps); after 19 years storage, 2-percent still developed.

NEEM PLANT



(Azadirachta indica)

Azadirachta indica, commonly known as **neem**, **nimtree** or **Indian lilac**,^[3] is a tree in the mahogany family Meliaceae.

It is one of two species in the genus *Azadirachta*, and is native to the Indian subcontinent and most of the countries in Africa.

It is typically grown in tropical and semi-tropical regions. Neem trees also grow on islands in southern Iran. Its fruits and seeds are the source of neem oil.

Plant Description Of Neem Plant

Neem trees can reach 15–30 metres (49–98 feet) in height and have attractive rounded crowns and thick furrowed bark.

The compound leaves have toothed leaflets and are typically evergreen but do drop during periods of extreme drought.

The small fragrant white flowers are bisexual or staminate (male) and are borne in clusters in the axils of the leaves. The fruit is a smooth yellow-green drupe and has a sweet-flavoured pulp.

Neem is usually grown from seed but can be propagated from cuttings or root suckers.

The plant is hardy and resilient and grows well in poor, rocky soils.

Neem tolerates a wide variety of environmental conditions but cannot survive freezing temperatures or being waterlogged.

Morphology Of Neem Plant

Neem is a medium-sized tree, reaching 15 to 30 m in height, with a large rounded crown up to 10-20 m in diameter. It is mainly evergreen but sometimes shed its leaves during the dry season.

Neem has a deep taproot and is a mycorrhizal-dependent species. The bark is grey, becomes fissured and flakes in old trees. A sticky foetid sap exudates from old trees in humid climates. The branches are numerous and spreading.

The leaves are alternate, petiolated, clustered at the end of the branches, unequally pinnate, glabrous and dark glossy green at maturity, 20-40 cm in length and bearing 10-20 leaflets.

The leaflets are 5-10 cm long x 1.2-4 cm broad, sickle-shaped and slightly denticulate

The flowers are numerous, fragrant, white and borne in large clusters (up to 30 cm long). Neem fruits are 1-2 cm long drupes, smooth and green with white milky juice when unripe, turning to yellow to brown when mature.

They have a thin epicarp, a mucilaginous fleshy mesocarp and a hard endocarp. They contain a variable number of ovoid (1-2 cm) oil seeds

SIDE EFFECTS OF NEEM PLANT

Neem is **POSSIBLY SAFE** for most adults when taken by mouth for up to 10 weeks, when applied inside the mouth for up to 6 weeks, or when applied to the skin for up to 2 weeks.

When neem is taken in large doses or for long periods of time, it is **POSSIBLY UNSAFE**. It might harm the kidneys and liver.

Uses Of Neem

1. Antibacterial activity

Recent research shows the isolation and identification of the antibacterial active compound from petroleum ether extract of neem oil. The study of Zhong *et al.* showed an antibacterial activity of 9-octadecanoic acid-hexadecanoic acid-tetrahydrofuran-3,4-diyl ester from neem oil. Elavarasu *et al.* studied *in vitro* anti-plaque microbial activity of neem oil.

2. Antiviral

Galhardi *et al.* studied the *in vitro* antiviral property of *Azadirachta indica* polysaccharides for poliovirus. The study of Saha *et al.* showed water extracted polysaccharides from *A. indica* leaves with anti-bovine herpes virus type 1 (BoHV-1) activity. The research of Xu *et al.* showed the *in vitro* antiviral activity of neem seed kernel extracts against duck plague virus.

3. Sexually transmitted disease

Few researchers have focused on neem efficacy in treating sexually transmitted diseases. The reports that have been completed are overwhelmingly positive. Recent research of Shokeen *et al.* showed the evaluation of the activity of 16 medicinal plants against *Neisseria gonorrhoeae*

4. Neem and the immune system

Thoh *et al.* studied that azadirachtin interacts with the tumor necrosis factor (TNF) binding domain of its receptors and inhibits TNF induced biological responses.

5. Anti-inflammatory activity

The study of Alam *et al.* showed the anti-inflammatory activity of epoxyazadiradione against macrophage migration inhibitory factor [45]. Thoh *et al.* found that azadirachtin interacts with retinoic acid receptors and inhibits retinoic acid-mediated biological responses.

6. Antioxidant effect

Manikandan *et al.* researched that antioxidant and protective effects of active neem leaf fractions against hydrogen peroxide induced oxidative damage to pBR322 DNA and red blood cells.

MATERIALS AND METHODS

Collection of plant materials:

The plants *Mimosa pudica* L and *Azadirachta indica* A were collected from the garden area of NGSMIPS Campus, Deralaktte, Mangalore.

Preparation of herbal leaf extracts:

The collected plants *Mimosa pudica* L and *Azadirachta indica* A leaves are taken and coarsely powdered. 10 grams of coarsely powdered leaves of both plants were soaked in 200 ml of methanol and kept for maceration for about 3-4 days. After maceration the extract is filtered and the filtrate was collected and used for making hand wash.

Preparations of herbal hand wash formulations:

Formulation 1 (F-1):

In this formulation the hand wash was prepared using 20 ml of methanolic extract filtrate. To this filtrate 6g of SLS , glycerin 40 ml , 0.3 g of methyl paraben, 5ml of rose merry oil is added and the volume is made up to 100ml with purified water.

Formulation 2 (F-2):

This formulation was prepared by adding 20 ml of lemon juice to 20 ml of methanolic extract filtrate of *Mimosa pudica* L and *Azadirachta indica* A leaves.

The remaining ingredients include all the same as mentioned above in formulation 1.

The formulations of both F1 and F2 is shown in below tables

Table 1: Formulation of Poly herbal hand wash F-1

| INGRIDIENT | QUANTITY |
|--|----------|
| Methanolic extract of <i>Mimosa pudica</i> L and <i>Azadirachta indica</i> | 20 ml |
| Sodium lauryl sulphate | 6 gms |
| Glycerin | 40 ml |
| Methyl Paraban | 0.3 gms |
| Rosemerry oil | 5 ml |
| Purified water | 100 ml |

Table 2: Formulation of Poly herbal hand wash F-2

| INGRIDIENT | QUANTITY |
|---|----------|
| Methanolic extract of <i>Mimosa pudica</i> and <i>Azadirachta india</i> | 20 ml |
| Lemon Water | 20 ml |
| Sodium lauryl sulphate | 6 gms |
| Glycerin | 40 ml |
| Methyl paraben | 0.3 gms |
| Rosmerry oil | 5 ml |
| Purified water | 100 ml |

Table 3: Antimicrobial activities of Polyherbal hand wash formulations

| ORGANISM | ZONE OF INHIBITION IN Cms | | |
|------------------------|---------------------------|-----|-----|
| | Std.Drug | F-1 | F-2 |
| BACILUS SUBTILUS | 1.4 | 3.4 | 3.8 |
| STAPHYLOCOCCUS AUREUS | 2.7 | 3.8 | 4.3 |
| PSEUDOMONAS AERUGINOSA | 2.7 | 3.6 | 4.2 |
| ESCHERICHIA COLI | 1.8 | 3.3 | 3.8 |

Evaluation of antimicrobial activity

The screening of anti-microbial efficacy of the formulated poly herbal hand wash was performed on various microorganisms by using agar plate method as per standard procedure. Four sterile petri plates were taken for testing the anti-microbial activity against four different microorganisms, *Bacillus subtilis*,

Staphylococcus aureus, *Psuedomonas aeruginosa* and *Escherchia coli*. The plates were filled with nutrient agar solution and allowed for solidification. After solidification the microorganisms from the subculture were inoculated into the nutrient agar media and three cavities were made in it.

The first cavity is filled with standard antibiotic amoxicillin, second one with herbal hand wash without lime water (F-1) and third cavity is filled with herbal hand wash with lime water (F-2). It was taken care that sample should be placed at the level of cavity. The plates are placed in incubator at 37°C 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 estimated

RESULTS

The prepared formulations of polyherbal handwash were subjected for physical evaluation and antimicrobial efficacy.

Appearance:

The prepared two formulations of hand wash appear as greenish brown and greenish yellow colour.

pH:

The pH of formulations was measured by digital pH meter. The pH of two formulations was found to be 6.31 and 6.24

Viscosity:

The viscosity of hand wash was determined by using 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 and its viscosity was measured. The viscosity of F-1 and F-2 was found to be 54 and 62 CPS.

Antimicrobial activity:

The Anti-microbial efficacy of the formulations of Polyherbal Hand Wash was tested on *Staphylococcus aureus*, *Psuedomonas aeruginosa*, *Bacillus subtilis* and *Escherichia coli* by agar plate technique.

The results of zone of inhibition showed that the hand wash prepared from methanol extract of the combined plant materials shown significant antimicrobial activity. The hand wash prepared with lemon juice (F-2) showed little higher activity than the formulation prepared without lemon juice (F-1).

CONCLUSION:

The results suggest that methanolic extract of *Azadiracta indica*, *Mimosa pudica* and their combinations with lemon water are capable of giving superior zone of inhibition to protect 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.

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