

Formulation And Evaluation of Herbal Hand Wash

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Abstract: The herbal cosmetic is natural and free from all the harmful synthetic chemicals which generally may turn out to be lethal to the skin. So herbal cosmetic is more referable as compared to synthetic one. The primary purpose of developing an herbal hand wash with a gel basis is to promote personal hygiene. Hand hygiene is a requirement and one of the most critical processes in the preparation of meals, food service, and house and other day care facilities. Current market Alcohol-based cleaning supplies, which are used in antibacterial hand washing, have a variety of drawbacks. To prevent the negative effects of synthetic hand wash preparations, such as itching, dryness, irritation, and dermatitis, and to avoid allergic reactions and any other adverse effects, consumers constantly look for cosmetics made from natural ingredients. The antibacterial activity of the prepared hand wash toward *S. aureus* was determined using the Agar plate diffusion technique. Physical and chemical characteristics such as pH, Viscosity, Foam height, Foam retention, Anti-Microbial Activity, Skin irritation assessment, and other parameters were used to evaluate herbal hand wash, and the results were found to be within normal ranges with minimum to no adverse effects.

Keywords: herbal hand wash, antimicrobial activity, *Azardirachta indica* (Neem), *Spanacia oleracea*.

INTRODUCTION

The cosmetics which are formulated using herbs having cosmetic actions. In cosmetic both natural and Phyto ingredient are used. Natural products include pure constituent obtained by various processes. The area of our bodies that is exposed the most is our skin, which must be shielded from various skin diseases. Hands are the major route of microbe and illness transfer; hand cleanliness or Hand washing is the most efficient way to prevent the transmission of bacteria that cause diarrhoea, influenza, and the common cold. The WHO standard requires people to wash their hands with no antibacterial soap and water. The time duration ranged on average as short as 15 to 30 seconds, including rubbing the backs of hands, wrists, between fingers. Hand washing with herbal hand wash

and water is more effective than washing with water alone in removing bacteria.

Hands are the primary mode of germ and infection transmission. Hand hygiene is thus the most important measure to avoid the spread of harmful germs and the spread of health care associated infections. Hand washing is the act of cleaning one's hands to remove dirt and pathogenic microorganisms and to prevent the transmission of transient microorganisms. According to classical literature for herbs with antimicrobial properties and it was discovered that *Azardirachta indica* is also known as Neem, margosa tree, and Indian lilac. Various neem tree parts have been used in traditional Ayurvedic medicine around the world. Terpenoids, alkaloids, tannin, Saponins, flavonoids, and amino acids are found in neem leaves. It has broad antimicrobial activity against both gram-negative and gram-positive bacteria. It is also used to treat skin diseases, healthy pairs, liver function, blood detoxification, pest and disease control, fever reduction, dental treatments, cough, asthma, ulcers, and intestinal worms. Also, Spinach scientifically known as *Spinacia oleracea* L. extracts have shown antimicrobial activity against various bacteria.

Advantage:

- Safe to use; minimal to no negative effects.
- It is possible to reduce bacterial attack.
- Maintain proper hand hydration.
- It makes the skin feel supple.

Herbal ingredient used in Hand wash:

1] Neem:



Fig: Neem leaves

1] Synonym: Margosa

2] Biological source: It consists of all aerial parts of plant known as *Azadirachta indica* belonging to family *Meliaceae*.

Macroscopic character:

1] Leaves: Alternate, exstipulate, imparipinnate leaflet 5.0 10 cm in length lanceolate closely clustered towards the ends of branches. The leaves have serrate margin, green colour and bitter test.

2] Bark: Moderately thick, rough, brown in colour longitudinally and obliquely furrowed Internally starchy white, laminated with characteristic smell of neem and bitter in taste.

Chemical constituent: good number of chemicals isolated from the plant belong to the classes di terpenes (sugiol), nibiol (bark), triterpenes: B-sitosterol, stigma sterol (leaf), Limonoids: Maliantriol (seed oil) nimbidiol (seed oil), Nimbendiol (seed oil and azadiractin (seed), sulphurous compounds Number of cyclic tri and tetrasulphides (leaves), flavanol glycosides Nimaton, quercetin, myrecetin, kaempferol. Neem leaves contain not less than 1.0% w/w of Rutin.

Uses of Neem:

Antimicrobial anti-inflammatory properties, making it beneficial for combating microbial infections and soothing irritated skin.

2] Spinach:



Figure: Spinach leaves

1] Synonym: *Spinacia domestica* a Borkh

2] Biological source: Dried or fresh leaves, stem and roots of *Spinacia oleracea* Linn. Belonging to family *Chenopodiaceae*.

Macroscopic character:

1] Leaves:

- Shape: The leaves are simple, ovate to triangular, and can be flat or puckered.

- Size: They are variable in size, ranging from 2-30cm long and 1-15cm broad.

2] Plant structure:

- Growth habit: Spinach is an annual plant that grows as a rosette, meaning the leaves emerge from the base of the plant.
- Height: It typically grows to a height of around 30cm long and 1-15cm broad.

Chemical constituent:

The following class of chemical constituents like Flavonoids, Phenolic compounds, Carotenoids, Steroids, Glycosides, Vitamins, Minerals reported.

Uses of spinach leaves:

- Spinach has antimicrobial properties that can help prevent growth of bacteria and other microorganism on the skin.
- Spinach has anti-inflammatory activity that can help soothe and calm irritated skin.

3] Aloes:



Figure: Aloe Vera leaves.

1]Synonyms: Aloe, Musabbar, Kamari.

2]Biological source: Aloes is the dried juice collected by incision, from the base of the leaves of various species of Aloe belonging to family *Liliaceae*.

Chemical constituents:

All the varieties of aloe are the major sources of anthraquinone glycosides. The principal composition of aloe is aloin, which is a mixture of glucosides, among which Barbaloin is the constituent. Along with barbaloin, aloes also contain isobarbaloin, B-barbaloin, aloe-emodin and res The drug also contains aloetic acid, omonataloin, etc.

Uses of aloe Vera leaves:

- It shows anti-inflammatory properties due the chemical contents like salicylates,

carboxypeptidases (inactivating bradykinin) and magnesia acetate (interfering with the conversion of histidine to histamine in the mast cells).

Material and method:

1] Material: Ethanolic neem extract, Ethanolic neem extract, aloe Vera gel, Sodium lauryl sulphate, Glycerine, rose oil, propyl paraben, distilled water.

2] Collection of plant material:

Azardirachta indica and Spinacia oleracea were collected from Amgaon district Gondia.

1] Neem:



Figure: Neem leaves

2] Spinach:



Figure: Spinach leaves

3] Cleaning drying of plant material:

Prior extraction, leaves of neem and spinach were cleaned 2 to 3 times with running water and once with sterilized distilled water then the materials were dried under shade at room temperature ($30\pm 50^{\circ}\text{C}$) for 10 days.

4] Preparation of crude powder:

After about 10 days of shade drying, well dried plants parts of Neem and Spinach were powdered by using electric mixture. Then product was subjected to mass sieving to obtain fine powder.

Method:

Maceration process is used for extraction of neem and spinach

1] Extraction of Neem:

1] Fresh neem leaves are collected and shed dried for 15 days.

2] The dried leaves then powered using mortar and pestle.

3] The powered neem leaves are weighed 25gm and macerated in a beaker using 100ml of ethanol.

4] The prepared mixture is kept covered with aluminium foil and kept for 3 days for maceration while stirring in between, and then the mixture was filtered using filter paper.

5] This mixture was then heated in a water bath at 65°C for 60 minutes.

6] The extract was then filtered again via filter paper to remove any particles.

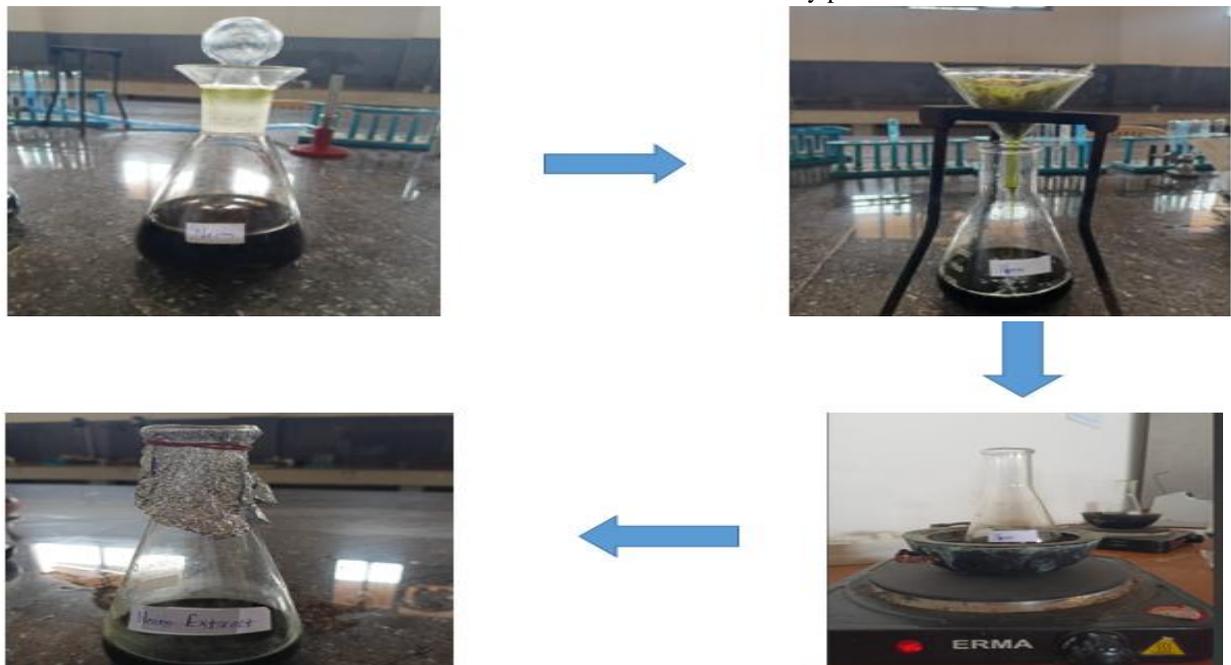


Figure : Process of Extraction of neem

2) Extraction of spinach:

- 1) The collected plant samples were dried and crushed to powder form.
- 2) Then 5gm of powdered plant sample was soaked with 50 ml of ethanol separately.
- 3) The entire mixture was kept for 48 hr. After the period was over, the mixture was filtered by filter paper.
- 4) The mixture was then heated in a water bath to get concentrated Ethanolic extract and were stored at 4°C until further used.

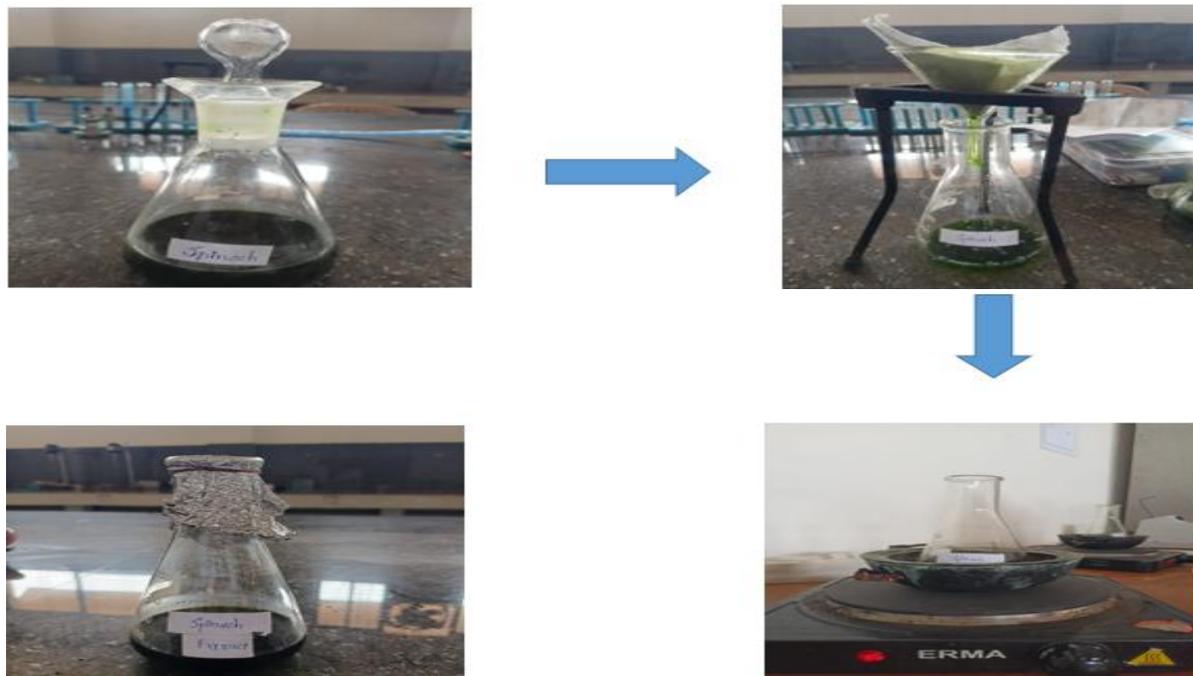


Figure: Process of Extraction of Spinach

Phytochemical test of sample:

Different qualitative test was performed in order to identify the phytochemical nature of each macerate as per standard protocol as follows.

1) Phytochemical test for neem:

A) Saponins – In this process we take 2ml extract of each solvent, 6ml of distilled water were added in a test tube and mixed properly if appearance of bubbles or persistent indicate the presence of Saponins.

B) Tannins- For this we take 2ml extract of each solvent, 10% of alcoholic ferric chloride was added, if formation of brownish blue or black colour indicate the presence of tannins.

C) Phenols- Take 2ml of extract of each solvent, 2ml of 5% aqueous ferric chloride were added. Formation of blue colour indicates the presence of phenols in the sample.

D) Protein - Take 2ml extract of each solvent, 1ml 40% of NaOH and few drops of 1% copper sulphate were added, formation of violet colour indicate the presence of peptide linkage molecule in the sample.

E) Cardiac glycosides- Take 1ml extract of each solvent, 0.5 ml of of glacial acetic acid and 3 drops of 1%aqueous ferric chloride solution were added. Formation of brown ring at the interface indicate the presence of cardiac glycosides in the sample.

F) Terpenoids- Take 1ml of extract of each solvent and add 0.5 ml of chloroform followed by a few drops of concentrated sulphuric acid. Formation of reddish-brown precipitate indicates the presence of Terpenoids in the sample.

G) Carbohydrates- Take 1ml of extract of each solvent, add few drops of molish's reagent and then add 1ml of concentrated sulphuric acid at the side of the tubes. The mixture was then allowed to stand for 2 to 3 minutes. Formation of red or dull violet colour indicates the presence of carbohydrates in the sample.

H) Flavonoids- Take 2 ml of each extract, add few drops of 20% sodium hydroxide. Formation of intense yellow colour is observed. To this few drop of 70% dilute hydrochloric acid were added and yellow colour was disappeared. Formation and disappearance of

yellow colour indicates the presence of flavonoids in the sample.

1] Alkaloids- Take 1ml extract of each solvent add 1 ml of marquis reagent were added and mix properly, appearance of dark orange or purple colour indicates the presence of alkaloid in the sample.

Test	Ethanolic extract result:
1] Saponin	-
2] Tannins	+
3] Phenol	+
4] Protein	-
5] Cardiac glycoside	+
6] Carbohydrate	+
7] Flavonoids	+
8] Terpenoids	+

2] Phytochemical test for spinach:

Phytochemical analysis of the test sample was carried out according to standard methods.

Test for Saponin: About 5 ml of the extract was boiled in 20 ml of distilled water in a water bath and filtered. 10 ml of the filtrate was mixed with 5 ml of distilled water and shaken vigorously for a stable persistent froth. The frothing was mixed with 3 drops of olive oil and shaken vigorously, then observed for the formation of emulsion which confirms a positive presence of Saponin.

Test for tannins: 1 ml of extract was boiled in 20 ml of water in a test and then filtered. A few drops of 0.1% ferric chloride were added and observed green or a blue-black coloration which confirms the presence of tannin. Phenol 5 ml of the extract was pipetted into a 30 ml test tube, then 10 ml of distilled water was added. 2 ml of ammonium hydroxide solution and 5 ml of concentrated amyl alcohol were also added and left to react for 30 min. Development of bluish green colour was taken as a positive presence of phenol.

Test of flavonoids: 3 ml 3 ml of 1% Aluminium chloride solution were added to 5 ml of each extract.

Formulation table of herbal hand wash:

Ingredient	Quantity	Action	Uses and Probable mode of action
Ethanolic neem extract	10ml	antibacterial, anti-inflammatory, and antioxidant	Antimicrobial anti-inflammatory properties, making it beneficial for combating microbial infections and soothing irritated skin.
Ethanolic spinach extract	10ml	Antimicrobial agent, antioxidant	Spinach has antimicrobial properties and Spinach has anti-inflammatory activity.

A yellow coloration was observed indicating the presence of flavonoids. 5 ml of dilute ammonia solution were added to the above mixture followed by addition of concentrated H₂SO₄. A yellow coloration disappeared on standing. The yellow coloration which disappeared on standing indicates a positive test for flavonoids.

Test for cardiac glycosides and cardenolides (Kellerkillani test): 5 ml of each extract was treated with 2 ml of glacial acetic acid containing one drop of ferric chloride solution. This was underplayed with 1 ml of concentrated sulphuric acid. A brown ring at the interface indicates a deoxy sugar characteristic of cardenolides which confirms a positive presence of cardenolides. A violet-green ring appearing below the brown ring, in the acetic acid layer, indicates the positive presence of glycoside.

Test for steroids: 2 ml of acetic anhydride was added to 2 ml extract of each sample followed by careful addition of 2 ml H₂SO₄. The colour changed from violet to blue or green indicate the presence of steroids.

Test for Terpenoids (Salkowski test): 5 ml of each extract was mixed with 2 ml of chloroform, and 3 ml concentrated H₂SO₄ was carefully added to form layer. A reddish-brown coloration of the interface was formed to show positive results for the presence of terpenoids.

Test	Ethanolic extract of spinach result
Saponin	--
Tannins	++
Flavonoids	--
Cardiac glycoside	++
Steroid	+
Terpenoid	--

Legend: +++ (Much abundant); ++ (Less abundant); + (Minute)

Sodium lauryl sulphate	4gm	Foaming Agent	Sodium lauryl sulphate is an anionic surfactant frequently used in household cleaning products as an emulsifying cleaning action.
Aloe Vera gel	2gm	Soothing agent	It shows anti-inflammatory properties.
Glycerine	10ml	Moisturizing agent	Glycerine functions as a humectant, allowing the skin to retain moisture, boost hydration, reduce dryness, and regenerate the skin's surface from the inside out.
Rose oil	1.5ml	fragrance	It enhances complexion brightness and skin tone.
Propyl paraben	0.15gm	Preservative	Used as a preservative.
Distilled water	q.s	Base material	Distilled water is used as a base material where all the other materials are dissolved to prepare a hand wash.

Formulation Development:

Hand wash was prepared by was adding 10 ml filtrate of Ethanolic neem extract, in this 10ml of filtrate of Ethanolic spinach extract, then add 4 gm of sodium lauryl sulphate was added, 2 gm aloe Vera gel, 10 ml glycerine, 0.15 gm propyl paraben, 1.5 ml rose oil was added and volume was made up to 50 ml distilled water.

Ingredient	Quantity
1] Ethanolic neem extract	10ml
2] Ethanolic spinach extract	10ml
3] Sodium lauryl sulphate	4gm
4] Aloe Vera gel	2gm
5] Glycerine	10ml
6] Rose oil	1.5ml
7] Propyl paraben	0.15gm
8] Distilled water	qs

Evaluation of prepared hand wash:



Figure: Prepared hand wash

Physical Evaluation:

The colour, texture, odour, appearance, and Homogeneity of the herbal hand wash were evaluated by physical test.

Spreadability:

On a glass slide, there was one drop of herbal hand wash gel. Another glass slide was kept over it and left aside for 5 minutes. The diameter up to which the formulation was spread, was measured in cm the same procedure was followed for the marketed formulation.



Figure: spreadability test

PH:

1 ml of herbal hand wash was mixed with 100 ml distilled water. This solution was then analysed using a previously calibrated digital pH meter; the pH of the marketed formulation was analysed in the same manner.

Irritancy:

The herbal hand wash 1 was applied to the hands till absorbed. The skin was observed for 1 hour for any signs of irritancy, redness, itching or discomfort, etc.



Figure: Irritation test

Foam Height:

A 100 ml measuring cylinder was filled with 1 ml of herbal hand wash that had been dissolved in 10 ml of purified water. The mouth of the measuring cylinder was tightly covered with the help of the palm, and the cylinder was shaking 25 times. The measuring cylinder was kept still and the height of the foam formed inside the measuring cylinder was noted.



Figure: Foam height.

Foam Retention:

In a 100 ml measuring cylinder, 1 ml of herbal hand wash gel was diluted in 10ml of purified water. Foam height was recorded after every minute for the first five minutes after the cylinder had been shaken for 25 strokes. An ideal formulation should have a foam retention time 6min.



Figure: Foam retention

Cleaning Action:

A piece of wool weighing 10 g was taken and dipped in oil. A solution of 1 ml herbal hand wash gel and 100 ml water was prepared and the prepared piece of wool was placed in this solution. After that, the mixture was shaking for four minutes. The piece of wool was taken out gently, dried, and weighed.



Figure: Cleaning action

Stability:

One sample of the herbal hand wash gel was added to small containers, and it was then maintained at three different temperatures for a week: 25°C, 37°C, and 40°C. The samples were then subjected to evaluation to all the parameters mentioned above.

Grittiness:

The formulation was tested after 1ml of gel was applied to a fingertip and rubbed between two fingertips. Dirt dispersion: The largest test tube, which held 10 ml of distilled water, had two drops of herbal hand wash added to it. 1 drop of Indian ink was added; the test tube was stopper and shaken for 10 times. None, Light, Moderate, or Heavy were the estimated levels of ink in the foam.



Figure: Grittiness test

Evaluation parameter of herbal hand wash:

Sr no.	Evaluation parameter	Formulated herbal hand wash
1	Colour	Dark green
2	Texture	Smooth
3	Odour	Aromatic
4	Appearance	Opaque
5	Homogeneity	Yes
6	Spreadability	1.3cm
7	PH	6..5-6.9
8	Irritancy	No
9	Foam height	4.8cm
10	Foam retention	Stable
11	Cleaning action	40percent

12	Stability	Stable
13	Grittiness	No
14	Dirt dispersion	Moderate

RESULT AND DISCUSSION

We observed that the herbal hand wash showed dark green colour with a bitter smell. The PH of these formulations ranged between 6.5 – 6.9 that's means suitable for the skin and non-irritating. During the stability tests, there was no colour change or phase separation in the prepared hand wash. The antimicrobial efficacy of herbal hand wash formulations was tested on *Staphylococcus aureus* using the agar plate technique. The herbal hand washes have significant antibacterial action, according to the zone of inhibition results. In the culture plates, the activity of herbal hand wash formulation revealed significant inhibition of bacterial growth. It was non-irritating to the skin.

CONCLUSION

The neem extract herbal hand wash was successfully developed with antibacterial properties derived from plant extract like neem, aloe vera, spinach, it not only ensure proper hand hygiene but also nourish the skin. Unlike chemical hand washes, it causes minimal irritation and help in preventing skin dryness. Overall herbal hand wash is a safe, sustainable, and effective choice.

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