



# Formulation and Evaluation of Hydroalcoholic Polyherbal Hand Sanitizer

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## 1. Abstract [1] [2] [3]:

Maintaining effective hand hygiene will essential for preventing the spread of infectious microorganisms. Increasing concerns regarding the frequent use of synthetic hand sanitizers, such as skin irritation and dryness, have encouraged the development of safer, plant-based alternatives. The presented study focused on the preparation and evaluation of a polyherbal hydroalcoholic hand sanitizer formulated using natural ingredients with proven antimicrobial activity. The formulation was designed by incorporating neem (*Azadirachta indica*) extract obtained by aqueous extraction, clove (*Syzygium aromaticum*) oil extracted through hydro-steam distillation, and fresh aloe vera (*Aloe barbadensis*) gel, along with 70% ethanol as the primary antimicrobial agent. Tween 80 was used as a surfactant, glycerine as a humectant, and rose oil for fragrance. The combination of neem and clove was select to achieve a synergistic bactericidal effect, enhance the overall antimicrobial efficiency of the formulation. The prepared sanitizer was evaluate for physicochemical parameters including pH, spreadability, appearance, and stability.

**Key Words-** Polyherbal Hand Sanitizer, Hydroalcoholic Formulation, Herbal Antimicrobial Activity, Hand Hygiene, Clove, Neem and Aloe vera Hand sanitizer, Natural Antiseptic Preparation, Skin Friendly Evaluation

## 2. Introduction [6] [5]:

Hands are the main reason for transmitting diseases also poor hygiene can lead to increase the risk for Fetal diseases Along with the Contaminated water food can contains large numbers of microorganism so it will increase the chances of causing disease/infection. Among all preventive measures, proper hand hygiene is considered the simplest, most cost-effective, and highly efficient method to reduce the spread of infections, particularly in healthcare settings where the risk of hospital-acquired (nosocomial) infections is significantly higher so best way to control it maintaining hygiene

Microorganisms present on human skin are broadly classified into resident and transient flora. Resident flora, such as *Staphylococcus epidermidis*, *Corynebacterium* species, and *Micrococci*, normally inhabit the skin and may provide the protective role by limiting the growth of harmful pathogens. In contrast, transient flora consists of microorganisms that temporarily colonize the skin surface through environmental exposure and are often responsible for infections. This microbe can be easily transmitted through direct contact, especially when proper hygiene practices are not follow by individuals.

Although handwashing with soap and water remains the standard method for maintain hygiene, alcohol-based hand sanitizers are widely used due to their convenience and rapid action. However, frequent use of synthetic sanitizers may lead to adverse effects such as skin dryness, irritation, and disruption of the natural skin barrier. These limitations have led to increasing interest in herbal-based formulations that are safer for repeated use and provide additional skin benefits

Polyherbal formulations utilize multiple plant-derived ingredients to achieve enhanced therapeutic effects. Medicinal plants such as neem (*Azadirachta indica*), clove (*Syzygium aromaticum*), and aloe vera (*Aloe barbadensis*) are well known for their antimicrobial, antioxidant, and soothing properties. Neem and clove, in particular, are reported to exhibit strong antibacterial

activity, and their combined use may result in a synergistic effect and improve the overall efficacy of the formulation. Aloe vera contributes to skin hydration and helps minimize irritation caused by alcohol content, making the product more suitable for frequent application so it overcomes the problem for those whom frequently use hand sanitizer which can cause dryness but here clove oil and glycerin can prevent dryness and improve effectiveness

In additionally skin is the outer most layer of the body that specifically expose such as pollutants, sunlight, and allergens, which can lead to various dermatological conditions including dryness, rashes, and infections. Maintaining proper hygiene and using skin-friendly products are therefore essential for preserving skin health. Despite the availability of various synthetic products, lack of awareness, inadequate hygiene practices, and limited access to proper healthcare can facilities continue to contribute to the spread of infections and antimicrobial resistance which can further cause serious and fetal conclusions to body

Therefore, the present study focuses on the development and evaluation of a polyherbal hydroalcoholic hand sanitizer formulated using natural ingredients. The aim is to provide an effective antimicrobial formulation that ensures safety, skin compatibility, and suitability for frequent use, offering a promising alternative to conventional synthetic hand sanitizers Also this study focus on the synergistic effect which provided by the Herbal constituents

## 2.1 ANATOMY OF SKIN [7] [8]

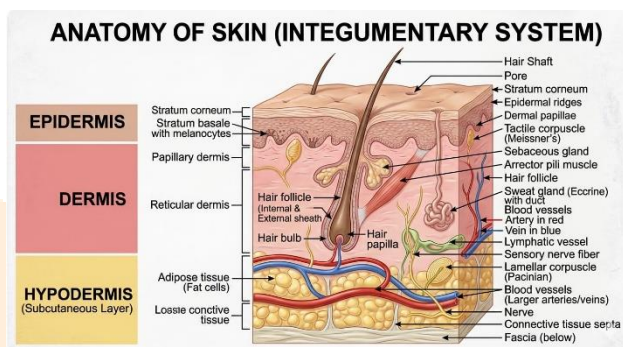


Fig no 1 (Source- this image generated by using Ai Tool)

### Layers Of Skin

#### 1 Epidermis [8]:

The epidermis outermost layer of the skin and functions as the first line of defences against environmental factors such as microorganisms, chemicals, and physical injury. It has an average thickness of about 0.1–0.2 mm, depending on the location of the body, and plays an important role in maintaining skin hydration by preventing excessive water loss

Structurally, the epidermis is composed of multiple layers arranged from the deepest to the most superficial: stratum Basale, stratum spinosum, stratum granulosum, stratum lucidum (present in thick skin), and stratum corneum. The stratum corneum is the outermost layer and consists of dead, flattened keratinized cells that form a tough and water-resistant barrier. This layer is essential for protecting the body against microbial invasion and external damage.

#### 2 Dermis [9]

dermis is the layer of skin located beneath the epidermis and provides structural support and flexibility to the skin. It is significantly thicker than the epidermis and is mainly composed of connective tissue rich in collagen and elastin fibers, which are responsible for maintaining skin elasticity and mechanical strength.

Structurally, the dermis is divided into two regions: the superficial papillary dermis and the deeper reticular dermis. The papillary dermis consists of loose connective tissue and contains fine collagen fibers along with capillaries that supply nutrients to the epidermis. In contrast, the reticular dermis is denser and contains thicker collagen bundles, providing durability and resistance to external stress

#### 3 Hypodermis [9]

The hypodermis, also known as the subcutaneous layer, is the deepest layer of the skin located beneath the dermis. It primarily consists of loose connective tissue and adipose (fat) tissue, which play an important role in supporting and protecting the body. This layer helps anchor the skin to underlying structures such as muscles and bones, allowing flexibility and movement.

One of the major functions of the hypodermis is energy storage, as the adipose tissue present in this layer stores lipids that can be used as a reserve source of energy when required. In addition, it acts as a thermal insulator, helping to regulate body temperature by reducing heat loss. The cushioning effect provided by this layer also protects internal organs and tissues from mechanical injury

## 2.2 Advantages of POLYHERBAL HYDROALCOHOLIC HAND SANITIZER [5]

1. Polyherbal hand sanitizers offer several advantages compared to conventional commercial formulations, especially in terms of efficacy
2. major advantages is reduced skin irritation. Commercial hand sanitizers, particularly those with high alcohol content and synthetic additives, may cause dryness, irritation
3. the synergistic antimicrobial effect. Herbal components like neem and clove possess well-documented antibacterial properties.
4. Polyherbal sanitizers also offer additional therapeutic properties, including antioxidant and anti-inflammatory effects.

### 3. Objectives [2] [4] [5]

1. To formulate and evaluate a polyherbal hydroalcoholic hand sanitizer using natural ingredients
2. To achieve a synergistic antibacterial effect by combining neem and clove
3. To develop a formulation that is safe and suitable for frequent use, minimizing common side effects such as skin dryness and irritation
4. To provide skin-friendly properties, including moisturization and soothing effects
5. To evaluate the prepared sanitizer for its physicochemical characteristics, stability, and overall acceptability
6. To develop an effective, safe, and herbal-based alternative to commercially available hand sanitizers

### 4. Pharmacognostic account of Herbal Drug:

Pharmacognosy is the branch of science that deals with the study of natural drugs obtained from plant, animal, or mineral sources, with a major focus on plant-derived substances having therapeutic value. It includes the evaluation of physical, chemical, and biological characteristics of crude drugs, which helps in their identification, authentication, and standardization. These properties play an important role in ensuring the safety, quality, and efficacy of herbal formulations used for medicinal purposes

#### Herbal Ingredients used in Hand sanitizer:

1. **Neem** [2] [11]: -



Fig no.2: Neem

**Synonym:** *Azadirachta indica*

**Biological source:** Obtained from the seeds and other parts of *Azadirachta indica* Linn.

**Family:** Meliaceae

**Uses:**

1. Neem is widely recognized for its broad-spectrum antimicrobial properties. It is effective against bacteria, fungi, and viruses, making it highly suitable for hand hygiene formulations.
2. The presence of active compounds such as nimbidin and azadirachtin contributes to its ability to inhibit microbial growth
3. In addition, neem helps maintain skin moisture, preventing dryness commonly associated with alcohol-based sanitizers.
4. It also exhibits anti-inflammatory activity, which helps in reducing skin irritation and minor rashes in frequent use

### 2) CLOVE [3] [12]

**Synonyms:** *Syzygium aromaticum*

**Family:** Myrtaceae

**Biological Source:** Dried flower buds of *Syzygium aromaticum* (L.) Merr. & L.M. Perry



**Fig no.3: clove**

1. Clove is widely used in herbal formulations due to its strong antimicrobial and antiseptic properties. It is effective against a variety of microorganisms, making it suitable for maintaining hand hygiene
2. The presence of eugenol as a major active component contributes to its ability to control bacterial growth.
3. clove possesses antioxidant and mild analgesic properties, which help in protecting the skin and reducing minor irritation during frequent use. Its natural aroma also improves the overall acceptability of the formulation

### 3) Aloe vera [4] [13]:

**Synonyms:** *Aloe barbadensis*

**Family:** Asphodelaceae

**Biological Source:** Fresh leaves of *Aloe barbadensis* Miller



(Source the image downloaded from [www.istock.in](http://www.istock.in))

**Fig no. 4: Aloe vera**

#### Uses:

1. Aloe vera is widely used in topical formulations due to its moisturizing and skin-protective properties.
2. prevents dryness, especially in formulations containing alcohol
3. The plant is also known for its soothing and anti-inflammatory effects
4. Help to reduce skin irritation and promote comfort during frequent use of hand sanitizers. In addition, aloe vera possesses mild antimicrobial and antioxidant properties, contributing to overall skin health and protection

### 4) Glycerin [14]:

**Synonyms:** Glycerol

**Biological source:** Obtained from natural fats and oils

#### Use:

1. Glycerin is widely used in topical formulations as a humectant, which helps retain moisture in the skin
2. hand sanitizer formulations, it plays an important role in preventing dryness caused by alcohol
3. It improves skin hydration, enhances smoothness, and provides a soft feel after application. Glycerin also contributes to better spread ability of the formulation, making it more user-friendly for frequent use

## 5) Ethanol <sup>[15]</sup> <sup>[16]</sup>

**Synonyms:** Ethyl alcohol

**Uses:**

1. Ethanol is the primary active ingredient in most hand sanitizers due to its rapid and broad-spectrum antimicrobial activity.
2. hydroalcoholic formulations, ethanol (typically at 60–70% concentration) is highly effective in reducing microbial load on the skin.

Chemical Name: Ethyl alcohol

Chemical Formula: C<sub>2</sub>H<sub>5</sub>OH

Nature of Ethanol: Ethanol is a clear, colorless, and highly volatile liquid with a characteristic odor. It is flammable in nature and evaporates quickly at room temperature

## 5. Formulation Table

NO	INGREDIENTS	QUANTITY
1	ETHANOL	70ml
2	Neem Extract	10ml
3	Clove Oil	2ml
4	Aloe vera gel	5ml
5	Tween 80	1ml
6	Rose oil	Q.S
7	glycerine	3ml
8	Distilled water	Q.S

## 6. MATERIAL AND METHOD

### 6.1 Extraction of Clove Oil (Hydro-distillation Method) <sup>[17]</sup> <sup>[18]</sup>

1. Approximately 150 g of dried clove buds (*Syzygium aromaticum*) were accurately weighed and coarsely powdered in mortar and pestle to increase the surface area for efficient extraction. The powdered material then transferred into a 350 mL round bottom flask.
2. About 250 mL of distilled water was added to the flask, and the mixture was subjected to hydro-distillation using a condenser setup. The contents were heated using a heating mantle or burner, allowing the volatile constituents of clove to vaporize along with water.
3. The vapors were condensed and collected, resulting in the separation of clove oil. The process was continued until sufficient extract was obtained. The collected extract was then separated and stored in a clean, airtight container for further use.



Fig no 5 (Clove extract Obtained By Hydro distillation process)

### 6.2 Preparation of Neem Extract by Aqueous extraction method <sup>[17]</sup> <sup>[23]</sup>:

1. Fresh neem leaves (*Azadirachta indica*) weighing approximately 25 g were collected and thoroughly washed with distilled water to remove dust and impurities. The cleaned leaves were then crushed using a mortar and pestle to obtain a paste-like mass
2. The paste was transferred into a beaker, and approximately 150 mL of distilled water was added. The mixture was heated at a temperature of around 90–100°C with continuous stirring to facilitate the extraction of bioactive compounds.

3. The heating process was continued until the volume of the solution was reduced to about half of its original volume. The resulting extract was then cooled and filtered using muslin cloth or filter paper to remove solid residues. The filtrate obtained was used as neem extract for formulation.



**Fig no 6** (Neem Extract Evaporation to remove water)

## 7. Methodology <sup>[19]</sup> <sup>[20]</sup>:

**A.** Fresh Neem (*Azadirachta indica*) leaves and Aloe vera (*Aloe barbadensis*) leaves were collected from the surrounding areas. Clove (*Syzygium aromaticum*) dried flower buds were obtained from a local market. The plant materials were selected based on their reported antimicrobial properties.

**B.** The collected plant materials were washed thoroughly with clean water to remove dust and impurities. The neem leaves and clove buds were then dried at room temperature in the laboratory for several hours to remove moisture while preserving their active constituents

**C.** The dried neem leaves and clove buds were crushed using a mortar and pestle to obtain a coarse powder. This process increases the surface area of the plant material and improves the efficiency of extraction.

**D.** Approximately 100 g of neem leaves were crushed and transferred into beaker containing 500 ml of distilled water. The mixture was heated on a burner and maintained at a temperature of 80–95°C for about 45–60 minutes to facilitate extraction of the active constituents. The mixture was then allowed to cool. (extraction procedure done by using aqueous extraction method)

**E.** About 150 g of clove buds were crushed into coarse powder and transferred into a round bottom flask containing 500 ml of distilled water. The mixture was heated at 80–90°C for approximately 60 minutes to extract the bioactive components present in clove. (extraction procedure done by using Hydro steam distillation)

**F.** Fresh aloe vera leaves were washed thoroughly. The outer green peel was removed using a sterile knife, and the transparent inner gel was carefully collected. The gel was then crushed to obtain a smooth and uniform consistency. After extraction, the mixtures were allowed to cool to room temperature and then filtered using muslin cloth to remove solid plant residues. The clear filtrate obtained was collected and used as the herbal extract for further formulation

**G.** 70 ml of ethanol was taken in a clean beaker and Then Neem extract and clove extract were added gradually with continuous stirring and Aloe vera gel was incorporated to provide moisturizing and soothing properties

**H.** Tween 80 (1–1.5 ml) was added as a surfactant to ensure uniform mixing of the ingredients. And along with that Glycerine was added as a humectant to prevent dryness of the skin. Rose oil was added in a small quantity to impart fragrance

**I.** Distilled water was added to adjust the final volume of the formulation. The mixture was stirred continuously until a homogeneous polyherbal hand sanitizer was obtained. The prepared sanitizer was transferred into clean and sterile containers and stored at room temperature for further evaluation

## 8. Evaluation Tests <sup>[21]</sup> <sup>[22]</sup>:

### 8.1 Organoleptic Evaluation

Sr.no	Test	Inference
1.	Colour	Light Olive Green
2.	Odour	Aromatic & Pleasant
3.	Appearance	Clear
4.	Texture	Smooth Liquid

## 8.2 P.H Determination

The P.H of Poly Herbal Hand Sanitizer was Found to Be 5.5 which is totally Comfortable for skin

## 8.3 Viscosity

Used Brookfield viscometer to measure the viscosity of the Sanitizer Viscosity 1.17cp

## 8.4 Spreadability test

The given sample is Easily Spreadable on Skin

## 8.5 irritation Test

On first application and on subsequent Application the polyherbal hand sanitizer show No Irritation On skin

## 8.6 Washability Test:

The Sanitizer is Easy to Wash After Immediate Application and Intermediate Time interval and it Does Not Produce Smell After Washing

## 8.7 Anti-Bacterial Activity

The Antibacterial activity of Polyherbal hand sanitizer against E.coli.



Fig no 7 (Zone of inhibition)

### Observation of Antibacterial Activity

SR.NO	SAMPLES	ZONE IN DIAMETER (mm)
1	Control	00
2	Standard (Streptomycin)	25
3	PHS01	20

**Conclusion of Antibacterial Activity-** antibacterial profile of **PHS 01** was evaluated by measuring the zone of inhibition against *E. coli*(ATCC 25922) bacterial strains via well diffusion method The compound PHS 01 exhibited moderate antibacterial activity as compared to the standard streptomycin.

## 9.8 Stability Study

### After 15 Days

Colour	NO Change
Odour	NO Change
P.H	NO Change
Phase Separation	NOT OCCURRED

**After 30 Days**

Colour	NO Change
Odour	NO Change
P.H	NO Change
Phase Separation	NOT OCCURRED

**10. Conclusion:**

The present study concludes that the formulated polyherbal hand sanitizer containing Neem (*Azadirachta indica*), Clove (*Syzygium aromaticum*), and Aloe vera (*Aloe barbadensis*) demonstrated significant antimicrobial activity along with satisfactory physicochemical properties. The formulation exhibited good spreadability, maintain uniform consistency without phase separation, and showed no signs of skin irritation during the study

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