



IJPPR

INTERNATIONAL JOURNAL OF PHARMACY & PHARMACEUTICAL RESEARCH
An official Publication of Human Journals

ISSN 2349-7203



Human Journals

Research Article

August 2022 Vol.:25, Issue:1

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Formulation and Evaluation of Herbal Hand Sanitizer

 <p>IJPPR INTERNATIONAL JOURNAL OF PHARMACY & PHARMACEUTICAL RESEARCH An official Publication of Human Journals</p> 	
<p>Arun Kumar¹, Vijay Kumar Sharma*¹, Gangeshwar Pratap Singh¹, Rahul Shukla¹, Rohit Kumar Bijauliya²</p>	
<p><i>¹Future Institute of Pharmacy, Bareilly, Uttar Pradesh, India</i></p>	
<p><i>²BIU College of Pharmacy, Bareilly International University, Bareilly, Uttar Pradesh, India</i></p>	
Submitted:	22 July 2022
Accepted:	28 July 2022
Published:	30 August 2022



HUMAN JOURNALS

www.ijppr.humanjournals.com

Keywords: Herbal Medicine, hand sanitizer, infection, skin

ABSTRACT

Microbes and diseases are first spread through the hands. A crucial idea and practice in the prevention, management, and decrease of diseases is hand cleanliness. The COVID epidemic has raised the demand for hand sanitizer, which reduces hand dryness. We created a polyherbal sanitizer utilizing seven plant extracts and other components such as isopropyl alcohol, camphor, hydrogen peroxide, glycerol, and water after considering the demand. The antibacterial properties of the substances were considered before choosing them. The antibacterial properties of the components and sanitizer were assessed, and they demonstrated strong action against gram-positive bacteria. This study focuses on the efficacy of herbal hand sanitizers made using Neem, Eucalyptus, Tulsi, Aloe vera and Mint extract, revealing a positive outcome. The majority of study has been on maintaining cleanliness by preventing germs from entering the body through hands. After discussing the advantages of eliminating microorganisms, the goal of the current investigation is established. Natural herbal hand sanitizers are affordable, effective and environmentally friendly.

INTRODUCTION:

The area of the body that is most exposed to the sun, environmental pollutants, and some pathogen defense is the skin. Eczema (atopic dermatitis), warts, acne, rashes, psoriasis, allergies, etc. are among the most prevalent skin conditions. To shield the skin from damaging microbes and stop the progression of several skin infections (1). Microbes and diseases are first spread through the hands. A crucial idea and practice in the prevention, management, and decrease of diseases is hand cleanliness. Two types of bacteria live on hands: transitory bacteria and resident bacteria (2). The resident flora resides under the stratum corneum and can be found on surface of skin, namely *Staphylococcus epidermis*, *S. hominis*, *Corynebacteria*, *Propionibacteria*, *Dermobacteria*, *Micrococci* and *fungi Malassezia spp.* Hand washing is a crucial safety measure. Instead of using a synthetic preparation, the goal of the current endeavor is to physically examine and manufacture a herbal hand sanitizer from widely available plants (3). In addition to washing your hands with soap and water, hand sanitizer also acts as an antimicrobial. Hand sanitizer comes in a variety of formulations, including gel, foam, liquid solution, and others. (4) A thickening agent, humectants, and other inactive chemicals are typically found in hand sanitizers along with alcohol. When compared to soaps, alcohol-based hand sanitizers are far more efficient in killing bacteria (5). In the US, every hand sanitizer product needs to have a "national drug code" designation.

Plants were the primary treatment for many ailments before modern medicine was developed. As new antibiotics are introduced, bacteria gradually become resistant to them as well (6). These emphasize to researchers the value of plants with antibacterial characteristics. They work to create the unique capacity of various secondary metabolites to exhibit consistent and enduring action against a variety of microorganisms. (7)

The purpose of the present study was to prepare herbal hand sanitizer incorporating the leaves extracts of *Ocimum sanctum* Linn. (Tulsi) and *Eucalyptus globulus* (Nilgiri), the well-known herbal combination with multidimensional activities and to evaluate their respective antimicrobial efficacy and safety of hands. The formulation was evaluated against the specified microorganism (Bacteria- *E. coli*, *P. aeruginosa*, *K. pneumoniae*, *S. aureus*, *M. luteus*, *S. epidermidis* and Fungi- *Candida albicans*) by culture sensitivity test. The significance was found to be more in comparison to the standard reference.

MATERIALS AND METHODS:

In the present study, herbal sanitizer was prepared and its efficacy was checked on different bacterial strains isolated from the hospital premises. The study was carried out in the Department of Pharmacy, Future Institute of Pharmacy, and Bareilly.

Collection of Leaves: The plants leaves were collected for the preparation of sanitizer from in and around the campus the hospital premises. The plant was selected on the basis of its potent antimicrobial activity reported in research articles. The plants used for the study were *Ocimum gratissum* (Van tulsi), *Ocimum sanctum* (Shyama tulsi), *Eucalyptus globules* (Niligiri), *Azadirachta indica* (Neem), *Aloe barbadensis* (Ghritkumari) and *Menthe arvensis* (Mint). The plant leaves collected were weight, washed, cleaned and shade dried in a laboratory. After drying plant extract was prepared in ethanol and used for the preparation of hand sanitizer.

Preparation of Extract for Hand Sanitizer: The plant extracts were prepared by weighing 50 gm of dried leaves of each plant, powdered mechanically and soaked in 100 ml of ethanol overnight. After 24 hours, the extracts were filtered using funnel and filter paper and used for the preparation of hand sanitizer. (8, 9)

Preparation of Herbal Hand Sanitizer:

- Carbopol was added to deionized water with constant stirring.
- After uniform mixing, Triethanolamine was added with slow stirring to avoid the formation of possible air bubble in the product.
- Kept aside for 24 hrs.
- The extract of plants and Carbopol were added to alcohol with glycerine were mixed with aqueous phase.
- Finally, methylparaben was added as a preservative and perfume was added.
- Mixed with slow stirring to obtain uniform product. (10)

The herbal sanitizer was prepared by the following ingredients given below:

Table No. 1: Formulation Chart

Sr. No.	Ingredients	Quantity
1	Neem	2.5 gm
2	Tulsi	2.5 gm
3	Eucalypstus	2.5 gm
4	Aloe vera	2.5 gm
5	Mint	2.5 gm
6	Hydrogen peroxide	2 ml
7	Glycerol	10 ml
8	Ethanol (50%)	18.6 ml
9	Isopropyl alcohol (70%)	21.3 ml
10	Carbopol	0.18 gm
11	Methyl paraben	0.5 gm
12	Triethanolamine	0.07 ml
13	Deionized water	0.9 ml
14	Distilled water	Make upto 100 ml

Physiochemical Characterization and Evaluation of Hand Sanitizer Gels

Organoleptic Test: The prepared samples were inspected visually to check the texture, odor, and color of the gels in semisolid conditions. (11)

pH Evaluation: The pH measurement of the formulated gels was measured using a digital pH meter (Mettler-Toledo pH meter, USA). The pH measurements represent the mean \pm standard deviation (SD) of three replicates. (12)

Viscosity (Rheological Properties): The rheological and flowability properties of the prepared gels were determined at room temperature using a TCV 300 viscometer (Cambridge applied laboratories viscometer, TX,USA). A piston of a range of 1–10 cP was used, as the formulations had a texture equivalent to water, and the temperature was set to room temperature (≈ 24 °C). One mL from each prepared hand sanitizer was filled into the measurement chamber. The chamber was capped for 60 s until it was stable, and then the data were recorded. (13)

Spreadability: According to the methods outlined in, the spreadability of the produced hand sanitizers was assessed by spreading 0.5 gm of each formulation gel over a pre-marked transparent glass with a 2 cm diameter. After that, a second clear glass was added on top, and the contents were distributed over five minutes by adding a 500 g weight. Using this technique, the spreadability was assessed based on the gels' properties of slip and drag. The borders were scraped clean of extra gel (14). The diameter of the spreading area of each formulation was determined and represented by the mean \pm SD of three replicates. The following equation was used to determine the spreading percentage:

$$\text{Spreadability \%} = \frac{A2}{A1} \times 100$$

Where, A1 is initial area before spreading (cm) and A2 is final area after spreading (cm).

Antimicrobial Activity of Hand Sanitizer:

Microbial Suspension Preparation:

The American Type Culture Collection (ATCC) was used to gather gram-positive and gram-negative bacteria, as well as opportunistic pathogenic yeast (*C. albicans*), as reference microorganisms to assess the antimicrobial effectiveness of produced hand sanitizers. *E. coli*, *P. aeruginosa*, *K. pneumoniae*, *S. aureus*, *M. luteus*, and *S. epidermidis* were among the bacterial isolates. The ATCC provided the *C. albicans* yeast. According to [15, 16], Mueller–Hinton broth was used to make the bacterial and yeast suspensions, also referred to as inoculums. Every microbe was cultivated on Mueller–Hinton agar media and left in the incubator overnight at 37 °C.

Antimicrobial Zone of Inhibition Test: The zone of inhibition test against several gram-positive and gram-negative bacterial strains and a yeast was carried out to assess the antibacterial activity of the produced hand sanitizer gels. As experimental controls, three widely accessible hand sanitizers were also evaluated. On the surface of the agar plates, a final concentration of 1 10⁶ CFU/mL inoculum was evenly dispersed. Each hand sanitizer gel was applied on a sterile microbiological disc, which was then placed on the Mueller–Hinton agar plate after briefly drying. At 37 °C, all plates were incubated overnight. The clean region of no growth surrounding each disc's diameter was measured in millimetres (mm). The findings are the mean and standard deviation of three replicates. (17, 18, 19)

RESULTS AND DISCUSSION:

Organoleptic Test: To assess how the created formulations looked physically, an organoleptic test of hand sanitizer gels was performed. The created hand sanitizer gels underwent a visual quality examination, and the findings showed that the tested formulation had satisfactory qualities. The gels were uniform, transparent, and had the expected smell. There was no syneresis, and they were simple to use, light to spread, and had a steady flow. With overnight storage, a bubble-like look developed, but it vanished after a gentle shaking.

- Colour: - Yellowish white
- Odour: - Characteristics
- Appearance:- bubble-like

pH Evaluation: A digital pH metre was used to measure the pH levels of the hand sanitizer gel formulations. The objective of the study was to examine the neutralization of various produced formulations. To prevent skin irritation and inflammation, the optimal requirements for a topical dosage form's pH value should be within the skin's natural pH range, which is 4.0 to 7.0. The produced compositions' pH readings were somewhat acidic, averaging approximately 4.3. This might be as a result of the substantial amount of aloe vera, which naturally has an acidic pH (4.0–4.5).

Viscosity (Rheological Properties): One of the key variables that should be under control is the viscosity of the created gel formulations since it might indicate the consistency and flowability of the gel formulations when applied to the skin. The TCV 300 viscometer was used in this study's viscosity test to measure preparation thickness and investigate the impact of gel components on the rheological qualities of the end products. The viscosities of prepared formulations were greater than those of ethanol and water (0.9 cP). The prepared formulation's viscosity was determined to be 0.4 cp.

Spreadability Study: Spreadability is very important when applying hand sanitizers because it affects customer compliance and the uniformity of the applied gels to fulfill the requirements for topical application quality. To determine if the manufactured hand gels could adequately disseminate when applied to the skin, the gel spreadability test was performed; the ideal gel formulation should have a shorter spreading time (i.e., high spreadability). The formulation's viscosity is one of the key factors that might impact the gel's

spreadability; a lower viscous gel has a higher spreadability. The created hand sanitizer gels spreadability values, which are discovered to be 532 percent.

Antimicrobial Zone of Inhibition Assay: The produced hand sanitizers were tested against gram-negative and gram-positive bacterial strains, as well as *C. albicans* yeast, using the zone of inhibition method in comparison to three commercially available hand sanitizers (T1). Variable-diameter zones of inhibition with clear boundaries were seen.

Neem, eucalyptus, mint, tulsi, and aloe vera hand sanitizer can suppress bacterial and yeast germs by 2.5 percent (v/v), according to the antimicrobial assessment. According to a recent investigation, clove oil, when administered in a concentration range of 1 to 5 percent (v/v), has an antifungal action against certain fungi. Neem, aloe vera, and tulsi have all been linked to antibacterial action when it comes to various pathogens including *E. coli*, *P. aeruginosa*, *K. pneumoniae*, *S. aureus*, *M. luteus*, *S. epidermidis*, and *C. albicans*, according to Nzeako et al. To evaluate the safety of these hand sanitizers after application, herbal hand sanitizers (F1) were chosen for the acceptance research.

Table No. 2: Zone of Inhibition of prepared and commercially available hand sanitizer for gram-negative and gram-positive bacteria

F. Code	E. coli	P. aeruginosa	K. pneumoniae	S. aureus	M. luteus	S. epidermidis
F1	9.7 ±0.65	8.7±7.5	10.87±0.34	9.65±0.45	10.87±0.65	8.09±0.56
T1	11.87±0.65	10.87±0.54	12.98±0.87	11.65±0.27	13.08±0.54	11.76±0.43

Table No. 3: Zone of Inhibition of prepared and commercially available hand sanitizer against *Candida albicans*

F. Code	<i>Candida albicans</i>
F1	11.5 ±1.42
T1	14.76±1.61

CONCLUSION:

We can conclude that the designated microorganisms are significantly bacterially affected by herbal hand sanitizer. As a result, there is enormous potential for developing the usage of antibacterial herbal items as a measure to control the multidrug-resistant germs as well as to

stop their transmission by hands from one geographic location to another. An alternative to chemically manufactured hand sanitizers with active silver nitrates is herbal hand sanitizer. Natural herbal hand sanitizers are affordable, effective, and environmentally and environmentally friendly.

ACKNOWLEDGEMENT: The author is thankful to Mr. Arun Kumar, Assistant Professor, Future Institute of Pharmacy, Bareilly, U.P., and India for their valuable guidance.

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