



Formulation and Evaluation of Herbal Hand Sanitizer

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ABSTRACT: This research paper focuses on the formulation and evaluation of herbal hand sanitizer is designed for prevention, control and reduction of microbes and infections. Hands are the first mode of transmission of microbes and infections. Hand hygiene is a key principle and exercise in the prevention, control and reduction of infections. This review highlights the research results aimed at assessing the antimicrobial potential of medicinal plants such as the Averrhoa Bilimbi and Aloe vera. This study also subjected to a series of organoleptic and physicochemical evaluations to determine its quality and stability and these evaluations confirm that the herbal hand sanitizer formulation is stable, safe, and potentially effective for preventing the transmission of microbes and infections.

KEYWORDS: Hand Sanitizer, Averrhoa Bilimbi, Aloe vera, Antimicrobial, Antibacterial, Disinfectant, Isopropyl alcohol.

I. INTRODUCTION

Hand sanitizer, also called hand antiseptic, hand rub, or hand rub, agent applied to the hands for the purpose of removing common pathogens (disease-causing organisms). Their use is recommended when soap and water are not available for hand washing or when repeated hand washing compromises the natural skin barrier. Hands are the first mode of transmission of microbes and infections. Hand hygiene is a key principle and exercise in the prevention, control and reduction of infections. The bacteria resides on hands are classified in two categories namely resident or transient. The resident flora are 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.

Several strategies have been used to prevent the pathogenic microorganism transmission and infection, especially during corona virus outbreaks. Hands are needed to be clean to reduce

the chance of getting the disease through multiple routes, such as nose, mouth, and others. Hand sanitizer is one of the widely used tools during the COVID-19 outbreak as it effectively kills many microorganisms, safe, and available in markets. However, with high demand, their price were markedly increased. It was noticed that the demand of hand sanitizers has increased manifold since the inception of COVID 19. The alcohol (ethanol or isopropanol) based hand sanitizers were recommended by scientist, doctors and health agencies across the world.

Natural ingredient Alcohol-based sanitizers are very effective at quickly destroying a variety of pathogens and that too without the need for water, plumbing, and drying facilities. According to the World Health Organization (WHO), alcohols have an excellent activity against gram-positive bacteria, gram-negative bacteria, enveloped viruses, non-enveloped viruses, mycobacteria, and even fungi. Numerous studies have also documented the in-vivo antimicrobial activity of alcohols and the effectiveness in removing clinical strains of Acinetobacter baumannii, methicillin-resistant Staphylococcus aureus, Escherichia coli, Pseudomonas aeruginosa, and Candida albicans from profoundly contaminated hands of human. When the COVID-19 pandemic spread worldwide, the use of alcohol-based hand rubs (ABHRs) increased exponentially, causing a lack of sanitizers available in the market. To respond to the severe shortage in hand sanitizers, pharmaceutical companies and cosmetic industries, breweries, and perfumeries have started in an unprecedented move to produce hand sanitizers.

Therefore substituting these toxic chemicals additives and synthetic fragrances in hand sanitizer formulations with natural and sustainable ingredients with evident safety and efficacy data is highly commendable. Hence, we have developed an alcohol based herbal hand sanitizer formulation with Aloe vera and Bilimbi extract as natural ingredients.

This hand sanitizer exhibits enhanced antimicrobial effectivity and offers additional benefit of free from

toxic ingredients, which might be quite beneficial for health and environment as well.

II. METERIALS AND METHODS



Front view of Aloe vera and Averrhoa bilimbi

Aloe vera is a plant with height of almost 60-100 cm containing very short stem or stemless long leaves and belongs to family Liliaceae. Aloe vera is used as a disinfectant since it is a natural product that is easily available, inexpensive, has few to no adverse effects, and most importantly, is completely biodegradable and does not harm the environment. When combined with aloe vera, alcohol, which can be abrasive on its own, is less likely to irritate your skin. Aloe vera can be a valuable addition to hand sanitizers, offering moisturizing, anti-inflammatory, and antimicrobial benefits. Additionally, it makes your alcohol last longer, allowing you to utilize the same chemicals to get rid of more germs. The anti-inflammatory, anti-bacterial, anti-viral, antiseptic, and wound-healing effects of aloe vera are well documented. However, it's crucial to consider the concentration, formulation, and potential interactions with other ingredients to ensure the product's effectiveness and safety.

Averrhoa bilimbi is a small tropical tree reaching up to 15m height. It is often multitrunked, quickly dividing into ramifications. The bilimbi fruit is ellipsoidal, elongated, measuring about 4 – 10 cm and sometimes faintly 5-angled. The skin is smooth to slightly bumpy, thin, and waxy turning from light green to yellowish-green when ripe. The flesh is crisp and the juice is sour and extremely acidic and therefore not typically consumed as fresh fruit by itself. Bilimbi mainly used as a folk medicine in the treatment of diabetes mellitus, hepatoprotective, thrombolytic, antioxidant and as an antimicrobial agent.

Camphor is a waxy, white solid with a strong odour. It is found in the wood of the camphor laurel tree, which is native to East Asia. Camphor act as an antimicrobial agent in sanitizers, meaning it has the ability to kill bacteria due to its inherent antibacterial properties. It is often added to sanitizers for its potential to further enhance the germ killing power of the primary disinfectant. Providing a more comprehensive cleansing effect.

III. FORMULATION

Ingredients	Quantity
Aloe vera extract	6 ml
Bilimbi extract	6 ml
Glycerol	0.5 ml
Camphor	0.01 gm
Hydrogen peroxide	0.2 ml
Ethanol	35 ml
Isopropyl alcohol	50 ml
Perfume	0.2 ml
Distilled water	Make up to 100 ml

IV. METHOD OF PREPARATION

Extraction of bilimbi: Fruits with good condition (color, size, shape, no defects and decay) at each maturity stages were selected. Then the ripening stage of the fruits was distinguished through physical observation. To obtain fruits juice, water extraction method was conducted. Selected fruits according to their maturity were cut into halves, and 50 g fruit were weighed and blended with 100 ml water. To obtain clear juice, the blended fruits were filtered through a muslin cloth or a stainless steel filter with small porosity. The filtered fruit extracts were immediately stored in freezer at 0°C.

Extraction of Aloe vera: The good quality of Aloe vera was collected by physical observation and wash with good water. The peeling of Aloe vera is done with a sharp blade or knife. Then slice down the Aloe gel and strain the Aloe slices.

Preparation of Hand sanitizer: To make a hand sanitizer formulation that includes plant extracts, alcohol, glycerol, camphor and hydrogen peroxide, the ingredients are typically combined in a specific order and sequence to ensure a consistent result. The alcohol, glycerine, and aqueous phase are usually mixed together first, and then the plant extracts and camphor are added to the mixture. The hydrogen peroxide helps to sterilize the solution and to give disinfectant action. After the plant extracts and camphor are added, the mixture is typically stirred slowly and carefully to ensure that the ingredients are evenly dispersed and any air bubbles are eliminated. This is important to achieve a uniform consistency throughout the hand sanitizer, which is critical for its effectiveness in killing bacteria and viruses. 3 formulations were made according to increasing order of concentration of plant extracts as Formulation 1, Formulation 2 and Formulation 3.

V. EVALUATION TESTS FOR HERBAL HAND SANITIZER

1. Organoleptic Test

The organoleptic test of hand sanitizer gels was conducted to evaluate the physical appearance of the prepared formulations. Following the visual quality inspection of the prepared hand sanitizer gels, the results indicated good characteristics observed for the tested formulations as follows. The sanitizer was reported to be homogeneous, transparent, and easy to use with a light and continuous flow. They did not exhibit any syneresis which is a positive attribute. Visually inspect sanitizer's color, smell and

texture. Then evaluated the sanitizer's homogeneity to check whether it appears uniformly colored and free of clumps.

2. PH Evaluation

The pH values of the hand sanitizer formulations were measured using a digital pH meter. The aim of the study was to evaluate how various manufactured formulations were neutralized, which likely refers to the process of adjusting the pH of the formulation to a desired level. The pH measurements in Table 06 showed that all prepared formulations were slightly acidic, with pH values around 3.9 to 4.1. This might be due to the large proportion of Bilimbi and Aloe vera (90% v/v) with a natural acidic pH (4.0–4.5). Therefore, the slightly acidic formulations could be advantageous in the antimicrobial applications, as they can be more effective against pathogenic microbial growth.

3. Spreadability Study

Spreadability is an important factor to consider when developing hand sanitizer formulations, as it can affect both customer compliance and the effectiveness of the product. A hand sanitizer with poor spreadability may not be applied evenly, which can result in areas of the skin being missed and potentially leaving areas of the skin unprotected. Skin sensitivity of the sanitizer was checked on different individuals and feedback was collected. The individuals gave positive response with mesmerizing odor and soothing effect after using sanitizer. The individuals were asked to observe redness, irritation, burning sensation and dryness. But no side effects were seen in any individuals after using sanitizer. Like other commercial sanitizer, our sanitizer gave soothing effect and no dryness was observed.

4. Stability Testing

The stability trials involved 4 weeks of storage at various temperatures, including 40°C, 25°C, and 37°C. The sanitizer was found stable at all the three different temperatures. The physical stability including pH, color and odor was stable. As well as consistency and viscosity was also found stable. The turbidity was checked in sanitizer and no turbidity was also seen. High temperature promotes the growth of bacteria and fungus, but sanitizer made by us did not showed any turbidity or growth in sanitizer.

5. Microbial Testing

The agar disc diffusion using Mueller-Hinton was used for evaluating the antimicrobial efficacy of hand sanitizers. The test was conducted to evaluate the antimicrobial efficacy of the prepared hand sanitizers (F1, F2, and F3) against gram-negative and gram-positive bacterial strains, as well. Well-defined zones of inhibition were observed with variable diameters. It was shown that the antimicrobial effectiveness of F3 was significantly higher than F1 & F2 hand sanitizers. Additionally, the antibacterial activity of F3 was significantly higher than the other anti bacterial hand sanitizers (F1 and F2).

Agar Diffusion Test: To determine the susceptibility test of selected test organisms for each hand sanitizer well variant agar diffusion

method was used. This agar diffusion method was done in duplicates for each hand sanitizer by inoculated sterile Mueller Hinton agar plates using sterile cotton swab which was immersed into a tube contain standardized test organisms. After Mueller Hinton agar was inoculated, it was allowed to remain at room temperature to dry for only some minutes and 3 equally spaced holes were bored in the agar plate with the aid of a sterile 6mm cork borer. The 3 holes were filled with hand sanitizer and the Mueller Hinton agar was incubated for 24 hours at 37° C. The zones of inhibition (susceptibility or resistance) of the hand sanitizers to each test organism were examined with the help of a ruler in millimeter by evaluating the readings that were found from duplicates of agar diffusion test for each hand sanitizers.

VI. OBSERVATION AND RESULTS

Sl.No	Parameters	Observation
1	Colour	Colourless
2	Sensitivity	No irritation
3	Grittiness	Fine and Smooth
4	Sedimentation	No sedimentation
5	Stability	No phase separation
6	pH	5.1

Agar Disc Diffusion Test

Test Organism	Concentration 80%	Concentration 50%	Concentration 30%
Staphylococcus aureus	18 mm	15 mm	11 mm



Zones of inhibition

The compound exhibit strong antimicrobial activity against *Staphylococcus aureus* in our study. In the study, at 80% concentration (18 mm zone) high activity was found and moderate activity was found at 50% concentration (15mm zone). Lack of antimicrobial activity at 30% concentration suggest a minimum inhibitory concentration between 30-50%. The test was conducted to evaluate the antimicrobial efficacy of the prepared hand sanitizers (F1, F2, and F3) against gram-negative and gram-positive bacterial strains, as well. Well-defined zones of inhibition were observed with variable diameters. It was shown that the antimicrobial effectiveness of F3 was significantly higher than F1 & F2 hand sanitizers. Additionally, the antibacterial activity of F3 was significantly higher than the other anti bacterial hand sanitizers (F1 and F2). Hence, by increasing the concentration, greater antimicrobial activity was found in the Herbal hand sanitizer.

VII. CONCLUSION

Due to the emergence of COVID 19, hand sanitizer has become an essential commodity in home, workplace and public places. The SARS-CoV-2 is an enveloped virus that can be certainly inactivated by ethanol at concentration ranging from 60–80 %. Hence, alcohol based hand sanitizers are considered as quite effective against its transmission as suggested by health practitioners and medical agencies around the globe. However, taking into consideration the impact of frequent usage of hand sanitizer's formulations with chemical ingredients on human health and environment is quite worrisome. Since synthetic chemical ingredients, fragrances may be responsible for various dermal syndromes like allergic contact dermatitis and irritant contact dermatitis with mild to severe reactions. Therefore, the effectivity of natural and sustainable ingredients in the formulations has been scrutinized. The extracts of Bilimbi and Aloe vera clearly improved the antimicrobial potency of hand sanitizer formulation. Bilimbi fruit extracts at all stages of ripening has some inhibitory activities against selected bacterial strains. However, extracts from younger fruits are more effective against the bacteria. The high content of bioactive compounds in the first stages of fruits might contribute to the antibacterial properties of the fruits. Bilimbi contains oxalic acid which is strong acid that might have an action on bacterial growth. Hence, it has established that these natural ingredients are quite

beneficial for health as well as environment over long term usage.

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