

## **Antimicrobial Activity, Safety and Acceptability of Formulated Ginger-fortified Hand Sanitizer Gel**

**O. M. David<sup>1\*</sup>, F. J. Olatunji<sup>1</sup>, M. O. Alese<sup>2</sup>, T. O. Babalola<sup>1</sup> and O. O. Alese<sup>3</sup>**

<sup>1</sup>Department of Microbiology, Faculty of Science, Ekiti State University, Ado-Ekiti, Nigeria.

<sup>2</sup>Department of Anatomy, College of Medicine, Ekiti State University, Ado-Ekiti, Nigeria.

<sup>3</sup>Department of Physiology, College of Medicine, Ekiti State University, Ado-Ekiti, Nigeria.

### **Authors' contributions**

*This work was carried out in collaboration between all authors. Authors OMD, MOA and FJO designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors OMD, MOA and OOA managed the analyses of the study. Author FJO managed the literature searches. All authors read and approved the final manuscript.*

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### **ABSTRACT**

Hand hygiene remains the most effective means of breaking transmission of most infectious diseases in and out of hospital settings. Hand hygiene could be achieved by different means. However, in recent time, the use of hand sanitizer which could either be a supplement or an alternative to hand washing has been promoted. The effectiveness of a formulated herbal hand sanitizer was investigated in this study. The herbal (ginger) hand sanitizer was formulated and screened on both bacterial and fungal isolates using different microbiological methods in this study. The skin and eye irritation potential of the sanitizer were conducted on experimental animals. Structured questionnaire was used to test the effects of the product on the skin of consenting human volunteers. The hydro-alcoholic extract of ginger showed a concentration-dependent activity on the test organisms. *Escherichia coli* ATCC 8739 was the most susceptible isolates followed by *Staphylococcus aureus* ATCC 6538. *Serratia marcescens* ATCC 9986 was more resistant to the extract at lower concentrations (0.78 and 1.56 mg/ml). *Aspergillus fumigatus* was the most susceptible out of the three fungi tested followed by *Penicillium chrysogenum*. Herbal hand sanitizer

\*Corresponding author: Email: [davidoluwole5@gmail.com](mailto:davidoluwole5@gmail.com), [david.oluwole@eksu.edu.ng](mailto:david.oluwole@eksu.edu.ng);

(with weighted effectiveness of 3.82) performed better than commercial hand sanitizer (with weighed effectiveness of 3.78). In the glass beads test, both herbal and commercial sanitizers inhibited the growth of *Staphylococcus aureus* ATCC 6538, *Escherichia coli* ATCC 8739 and *Serratia marcescens* ATCC 9986. Though caused eye irritation, the herbal sanitizer produced neither skin irritation nor dryness. The formulated hand sanitizer is economical and found to be safe throughout long period of continued use.

**Keywords:** Sanitizer; ginger; fungi; hand hygiene; pathogens; irritation.

## 1. INTRODUCTION

The recent epidemic of communicable diseases further highlights the need of maintaining good hygiene [1]. Chassin et al. [2] attributed the low level of hygiene to some factors which include lack of awareness, knowledge of risk and unavailability of hand hygiene facilities among others. Hand sanitizers (HS) are antiseptic products usually applied on hands to reduce the number of viable pathogenic microorganisms without causing any damage to the skin [3]. Hand sanitizers could either be supplements or alternatives to hand washing with soap and water [4,5]. Herbal hand sanitizers are natural plant-based alternatives to chemical sanitizers. The active components of medicinal plants are obtained by extraction in suitable solvents, which are evaporated away and the resulting residue further diluted to prescribed concentrations.

Many plants contain bioactive phyto-compounds that inhibit the proliferation of disease causing microorganisms [6,7], tumor [8], inflammation and necrosis [7,9] among others. Herbs and spices are very important and useful as therapeutic agents against many pathological infections [10]. The spices have a unique aroma and flavor which are derived from compounds known as phytochemicals or secondary metabolites [11].

Ginger (*Zingiber officinale*) is a medicinal plant that has been widely used all over the world since antiquity, for a wide array of unrelated ailments including arthritis, cramps, rheumatism, sprains, sore throats, muscular aches, pains, constipation, vomiting, hypertension, indigestion, dementia, fever and infectious diseases [12,13]. Ginger has direct anti-microbial activity and thus can be used in the treatment of both bacterial and fungal infections [14,15]. Ginger belongs to *Zingiberaceae* family, all *Zingiberaceous* plants have strong aromatic and medicinal properties and are characterized by their tuberous or non-tuberous rhizomes [16,17].

Hand sanitizers are effective in reducing gastrointestinal illnesses in households [18], curbing absenteeism in elementary schools and offices, reducing illnesses in university dormitories [19] and reducing infections in healthcare settings. Alcohol-based hand sanitizers are recommended as a component of hand hygiene. The use of herbs in breaking the transmission of pathogenic microorganism has not been explored as that of curative and preventive use of herbs. If encouraged, it will minimize the use of chemicals in hand hygiene which could cause adverse effects.

A good hand sanitizer is that which is economical, simple to use and extremely efficient in operation. It must have a high substantivity effect and safe for long periods of continued use. Also, it should possess antimicrobial activity against a wide range of fungal and bacterial species. This study therefore aimed at formulating an effective hand sanitizer with good quality from a common medicinal plant with no skin and ocular irritation and which also enjoys a high acceptability among prospective users.

## 2. MATERIALS AND METHODS

### 2.1 Collection of Plant Materials

The fresh form of *Zingiber officinale* (ginger rhizome) was purchased at Bode Market in Ibadan, Oyo State, Nigeria. The plant was identified in the Herbarium of the Department of Plant Science, Ekiti State University.

### 2.2 Collection of Test Organisms

The test organisms used in this study include four fungi (*Aspergillus fumigatus*, *Absidia corymbifera* and *Penicillium chrysogenum*), three Gram-positive (*Staphylococcus aureus* ATCC 6538, *Bacillus subtilis* KZN and *Enterococcus faecalis* ATCC 29212) and two Gram-negative (*Escherichia coli* ATCC 8739 and *Serratia marcescens* ATCC 9986) bacteria isolates. All isolates were collected from the Department of

Microbiology, Ekiti State University, Ado-Ekiti, Nigeria.

### **2.3 Preparation of Plant Extract**

The method of David and Afolayan [20] was used to prepare the extract from the plant. Two kilogram of the plant sample (ginger) was macerated with blender and soaked in 2500 ml of 95 % ethanol for pre-extraction. After five days, the mixture was filtered using Whatman No. 1, filter paper. The extract was evaporated to dryness under reduced pressure at 40°C using a rotary evaporator (Laborota 4000 efficient, Heldolph, Germany). The extract was diluted using 5% dimethyl sulphoxide (DMSO) to give 50 mg/ml stock solution. This was then diluted to the required concentrations for the bioassay.

### **2.4 Determination of the Antimicrobial Activity of the Plant Extract**

#### **2.4.1 Minimum inhibitory concentration (MIC)**

Different concentrations of ginger extract was prepared in solution using 5.0% DMSO and the filtration through a 0.23 µm membrane filter. Filter paper discs (5.75 in diameter) were sterilized in hot air oven for 2 hours at 160°C and then impregnated with different concentrations of the extracts. Each of the isolates was grown at 37°C in Mueller-Hinton broth (Oxoid) for 18 h, adjusted to an optical density of 0.5 McFarland Standard and aseptically seeded on the sterile Nutrient Agar (Oxoid). The paper discs containing different concentrations of the extract were carefully placed on it. The plate was incubated at 37°C for 24 h and the zone of inhibition was observed and measured to the nearest millimeter.

#### **2.4.2 Determination of fungicidal activity**

The technique of Nene and Thapilyal [21] was used to screen for antifungal activity of the plant extract. Filter sterilized extract was mixed with sterilized Potato Dextrose Agar (PDA) (Oxoid) to achieve a concentration range of 3.125 mg/ml to 25.000 mg/ml. The inoculation was done at the center of each plate with a 5 mm mycelium block cut from the advancing edge of a five day old culture of the test fungi on PDA. The blocks were placed at the center of each Petri plate in an inverted position to get greater contact of the mycelium with the culture medium. The inoculated plate was incubated at 25°C. Potato Dextrose Agar without extract was also

maintained at the same condition to serve as control. After 72 hours of incubation, the diameter of fungi was measured in mm. The percentage inhibition was calculated using the formula:

$$\% \text{ Inhibition} = [(Control - Test)/Control] \times 100$$

Control= Growth of the test fungus in the control plate while Test = Growth of the test fungus in the test plate

### **2.5 Formulation of the Herbal Hand Sanitizer**

The herbal hand sanitizer was formulated hygienically using: 500 ml of absolute ethanol, 1 g of ginger extract, 200 ml of sterile water, 3 g of thickening agent (Zhejiang Xinyong Biochemical Co., Ltd. China.), 1ml of perfume, 10 ml of humectants and 5 ml of glycerin. The formulation was made to guarantee a minimum of 50% alcohol content to improve its effectiveness and serve as in-solution preservative.

### **2.6 Determination of Antimicrobial Property of the Formulated Hand Sanitizer**

#### **2.6.1 Finger imprint test**

The modified method of David [22] was used to determine the after-use effect of the herbal sanitizer. Convenient sampling method was used to select students in their hostels to participate in the study after obtaining their informed consent. The subjects include 60 students (30 males and 30 females) of Ekiti State University, Ado-Ekiti. Students who have used similar products or been on antibiotic treatment within two weeks to the study period were excluded from the study. The subjects were divided into six groups and treated with either the herbal hand sanitizer or a commercial sanitizer (Germ-X) as shown in Table 1.

The subjects were instructed on how to apply the sanitizers on their fingers to ensure even distribution and press the fingers on the surface of the sterile Nutrient Agar opened slightly in a sterile environment. After inoculation, the plates were incubated at 37°C for 24 h. Then the growth patterns of the finger imprints were observed and recorded as confluent growth, many growths, few growths and no growth and scored as 1, 2, 3 and 4 respectively.

**Table 1. The treatment and distribution of human volunteer subjects**

Treatment	Sex	
	Male	Female
Washed hand (using bland soap) with herbal hand sanitizer	5	5
Unwashed hand with herbal hand sanitizer	5	5
Washed hand (using bland soap) with commercial hand sanitizer	5	5
Unwashed hand with commercial hand sanitizer	5	5
Washed hand (using bland soap)	5	5
Unwashed hand	5	5

The weighted effectiveness was calculated as:

$$WE = (\sum \text{Growth pattern} \times \text{score}) / 100$$

The sanitizer was classified as very poor (if *WE* is between 1 and 1.4), poor (if *WE* is between 1.5 and 2.0), fair (if *WE* is between 2.5 and 3.0), average (if *WE* is between 3.1 and 3.4) and excellent (if *WE* is between 3.5 and 4.0).

### **2.6.2 Glass beads test**

Five beads (3mm) were sterilized in the autoclave (121°C for 15 mins) and carefully introduced into sterile petri dishes containing 100 µl of each of standardized inoculums of the culture of the test bacteria. The plates were covered and gently rocked to ensure all part of the beads was covered with the broth. The excess culture was drained and the beads were allowed to dry. Each of the beads was dipped into the herbal sanitizer, removed and left for 60 s for the excess sanitizer to drain off. Later, the beads were carefully picked by sterile forceps, separately planted into the sterile Nutrient Agar and incubated at 37°C for 24 h. The plates were observed for the sign of growth around the challenged beads.

## **2.7 Determination of Irritation Tests**

### **2.7.1 Animal care and management**

Three nulliparous and non-pregnant female New Zealand White Rabbits weighing 2.0 to 3.5 kg and between twelve to twenty weeks old were purchased from the Animal House of the College of Medicine, Ekiti State University, Ado-Ekiti. The animals were acclimatized for a period of one week and observed daily to ensure there was no abnormality in their general conditions. They were given identification by numbering the inner surface of the ear with a permanent marker. The animals were housed in individual compartment of wooden rabbit cages under standard laboratory conditions of natural light/dark cycle at

room temperature (24.6±4.2°C) and humidity (68±28%); fed on standard rabbit feed (Ladokun Feeds, Ibadan, Nigeria) and given water *ad libitum*.

All animals were handled in accordance with the Guidelines for Animal Research as detailed in the NRC Guidelines for the Care and Use of Laboratory Animals [23]. The methodology was designed according to OECD Guidelines for the Testing of Chemicals No. 404 'Acute Dermal Irritation/Corrosion' (2002) and Method B4 Acute Toxicity (Skin Irritation) of Commission Directive 2004/73/EC.

### **2.7.2 Skin irritation test**

The fur on the dorsal trunk of the each of the test animals was sheared approximately 24 hours prior to the testing. The animals were grossly observed to ensure healthy intact epidermis. Three suitable sites were selected on the back of the rabbits. At each test site, 0.5 ml of the test material was introduced under a 2.0 cm X 2.0 cm 4 ply cotton gauze patch and placed in position on the shorn skin. Each patch was secured with a strip of surgical adhesive tape. The trunk of the rabbits were wrapped in an elastic bandage for the duration of 1 and 4 hour exposure periods. Animals were returned to their cages after treatment. A patch was removed at 3 minutes, 1 hour and 4 hours after each application. After consideration of the skin reactions produced in the first animal, the remaining two animals were treated with 0.5 ml of the test material. A patch was applied to the back of each rabbit and was allowed to remain in contact with the skin for a period of 4 hours. A depilatory produced by Softsheen-Carson® was used as a positive control. The test sites were then observed at 1 hour, 24, 48 and 72 hours following the removal of the patches for evidence of primary irritation and scored according to the Draize scale [24]. Cage-side observations for general condition, appearance and demeanor were made daily.

### **2.7.3 Ocular irritation test**

A little quantity (0.1 ml) of the hand sanitizer was carefully instilled into the cupped conjunctiva sac of the right eye of each rabbit following which the eyelids were gently held together for one second and then massaged for 30 seconds. The left eye served as the negative control. The readings were performed at 1 hour, 24 hours, 48 hours, 72 hours and seven days after the application, and the corneal, iris and conjunctiva alterations were graded according to the Draize scale [24]. The scores were processed differently to decide if the formulated herbal sanitizer was irritant or not and/or to grade the severity of the irritation.

### **2.8 Administration of Questionnaire**

A well-structured questionnaire was given alongside a bottle of the formulated hand sanitizer to a total number of 20 randomly selected subjects recruited in this study. The aim and the objectives of the study were disclosed to them and those that consented to participate were recruited. Subjects who had earlier participated in any part of this study, been on antibiotic treatment or used similar products in the last two weeks were excluded. Also, subjects that have wound on the hands were not included in the study. The subjects include 12 females and 8 males with age range of 20-39 years old (n=18) and 40-59 years old (n=2). The questionnaire sought to test the knowledge of respondents on the use of hand sanitizer (in 6 questions), the effect of the herbal hand sanitizer after application for 15 days (17 questions) and the product satisfaction as it affects hand hygiene practices (4 questions). The questionnaire also assessed the demographic characteristics of the subjects.

### **2.9 Statistical Analyses**

Statistical analysis was done using SPSS (version 17) to determine frequency distribution, analysis of variance (ANOVA) and Dunn's Multiple Comparisons Test. The level of significance was set at 0.05.

### **2.10 Ethical Clearance**

The ethical clearance was also sought from the Research Governing body of the Faculty of Science, Ekiti State University.

## **3. RESULTS**

The activity of the extract on the test organisms was concentration dependent. The extract had the highest activity against *E. coli* ATCC 8739 followed by *Staph. aureus* ATCC 6538. Compared to *Ent. faecalis* ATCC 29212 and *B. subtilis* KZN, *S. marcescens* ATCC 9986 was more resistant to the extract at low concentrations (0.78 and 1.56 mg/ml) (Table 2). The susceptibility of *Ent. faecalis* ATCC 29212 to the extract differs significantly at  $p < 0.05$  from the susceptibility of *E. coli* ATCC 8739.

As shown in Table 3, *A. fumigatus* was the most susceptible of the three fungi, followed by *P. chrysogenum*. At  $P < 0.05$ , the percentage inhibition of all the three fungi tested was significantly different from one another. The effectiveness of the extract on *A. fumigatus* was most pronounced at the 96<sup>th</sup> hour of exposure. At the 48<sup>th</sup> and 72<sup>nd</sup> hours of exposure, *A. corymbifera* and *P. chrysogenum* inhibition were at the peak (Table 3). The commercial hand sanitizer (germ-X) when used after washing with bland soap had a better sanitizing effect than the herbal hand sanitizer. On the other hand, the herbal hand sanitizer performed better than the commercial hand sanitizer on unwashed hand. The herbal hand sanitizer had a weighted effectiveness of 3.82 while that of commercial hand sanitizer was 3.78 (as shown in Table 4). Considering the glass beads method, both herbal and commercial sanitizers inhibited the growth of *Staph. aureus* ATCC 6538, *E. coli* ATCC 8739 and *S. marcescens* ATCC 9986. The herbal sanitizer performed better on *B. subtilis* KZN than germ-X as reported in Table 7.

Table 6 shows the absence of erythema, eschar and edema formation on the rabbit skin on exposure to the herbal hand sanitizer at time interval of 3 minutes, 4 hours, 24 hours, 48 hours and 72 hours respectively. As seen in Table 7, there was no reaction in the cornea and iris of the rabbit's eyes on exposure to the herbal hand sanitizer at time interval of 3 minutes, 1 hour, 24 hours and 72 hours respectively. Plate 1 shows the reactions of the eyes and skin of the experimental animals on exposure to the herbal sanitizer. Conversely, there was severe eye reaction on the conjunctivas of the selected animals at the same time intervals. Six (30%) of the subjects reported that they used the herbal hand sanitizer for 1-2 times daily while fourteen (70%) reported that they used herbal hand sanitizer for 3-5 times daily as shown in Table 8.

**Table 2. The antibacterial activity of the hydroalcoholic extract of ginger (zone of inhibition in mm)**

Test organisms	Concentrations (mg/ml)				
	0.78	1.56	3.125	6.25	12.50
<i>Ent. faecalis</i> ATCC 29212	4.32±0.72 <sup>a</sup>	4.31±0.02 <sup>a</sup>	5.67±0.26 <sup>a</sup>	5.70±1.73 <sup>a</sup>	5.96±0.23 <sup>a</sup>
<i>B. subtilis</i> KZN	4.32±0.31 <sup>b</sup>	5.21±1.00 <sup>b</sup>	6.06±0.09 <sup>b</sup>	6.10±1.94 <sup>b</sup>	6.43±1.94 <sup>b</sup>
<i>Staph. aureus</i> ATCC 6538	6.13±1.49	8.44±0.23	8.27±1.62	8.64±2.56	10.15±2.91
<i>E. coli</i> ATCC 873	6.52±0.63 <sup>ab</sup>	8.86±1.02 <sup>ab</sup>	10.28±2.93 <sup>ab</sup>	10.69±1.94 <sup>ab</sup>	13.23±2.83 <sup>ab</sup>
<i>S. marcescens</i> ATCC 9986	4.32±0.88	4.31±0.37	9.39±1.05	8.02±1.63	9.26±0.25

<sup>a,b</sup>=Data showing significant difference at  $p < 0.01$  using ANOVA**Table 3. Percentage inhibition of test fungi at different concentrations (mg/ml) of ginger**

Time (hours)	Test fungi											
	<i>Aspergillus fumigatus</i>				<i>Absidia corymbifera</i>				<i>Penicillium chrysogenum</i>			
	25.00	12.50	6.26	3.125	25.00	12.50	6.26	3.125	25.00	12.50	6.26	3.125
48	50.00	21.05	12.50	8.33	54.24	22.55	33.33	16.67	41.67	40.75	33.33	15.79
72	63.26	32.63	16.67	8.14	51.00	30.83	20.39	14.27	51.01	47.58	38.76	11.58
96	68.42	34.48	29.41	10.53	44.92	29.41	7.79	12.38	36.84	36.84	21.05	6.90
120	57.89	20.97	27.83	-1.05	20.97	34.78	7.33	2.64	34.21	22.63	20.00	1.21
144	53.33	30.51	18.28	-10.34	37.92	21.41	2.91	-0.03	48.28	18.72	17.24	-1.69

**Table 4. Effectiveness of hand sanitizer using finger imprint test**

Treatment	Growth pattern score					Remarks
	CG	MG	FG	NG	WE	
Washed hand (using bland soap) with herbal hand sanitizer	1	12	13	74	3.60	Excellent
Unwashed hand with herbal hand sanitizer	0	1	16	83	3.82	Excellent
Washed hand (using bland soap) with commercial hand sanitizer	1	8	12	79	3.69	Excellent
Unwashed hand with commercial hand sanitizer	0	7	8	85	3.78	Excellent
Washed hand (using bland soap)	24	29	31	16	2.39	Fair
Unwashed hand	30	52	17	1	1.89	Poor

Key: CG = Confluent growth, MG = Many growth, FG = Few growth, NG = No growth, WE=Weighted effectiveness

**Table 5. Glass beads test on efficacies of the formulated herbal hand sanitizer on test organisms**

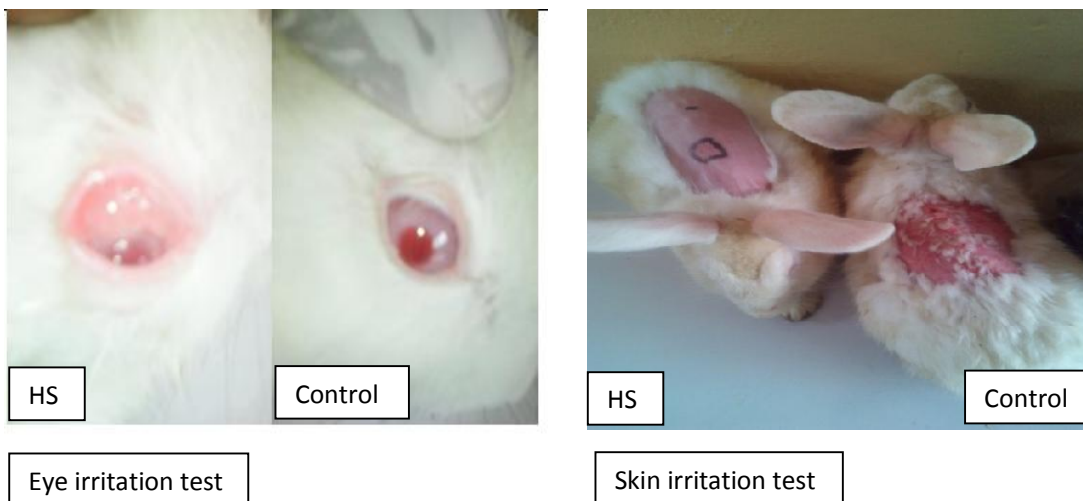
Organisms	Test		Controls			
	Herbal hand sanitizer		Commercial Hand sanitizer (germ-X <sup>®</sup> )		Sterile Distilled water	
	Growth	No growth	Growth	No growth	Growth	No growth
<i>Ent. faecalis</i> ATCC 29212	1	4	1	4	5	0
<i>B. subtilis</i> KZN	1	4	2	3	5	0
<i>Staph. aureus</i> ATCC 6538	0	5	0	5	5	0
<i>E. coli</i> ATCC 8739	0	5	0	5	5	0
<i>S. marcescens</i> ATCC 9986	0	5	0	5	5	0

**Table 6. Assessment of skin irritation test score of formulated herbal hand sanitizer**

Skin reaction	Observation time	Individual score- rabbit number (Test group)				Individual score- rabbit number (Control group)			
		A	B	C	Total	A	B	C	Total
Erythema and eschar formation	3 minutes	0	0	0	0	0	0	0	0
	4 hours	0	0	0	0	0	0	0	0
	24 hours	0	0	0	(0)	0	0	0	(0)
	48 hours	0	0	0	0	0	0	0	0
	72 hours	0	0	0	(0)	0	0	0	(0)
Edema formation	3 minutes	0	0	0	0	0	0	0	0
	4 hours	0	0	0	0	0	0	0	0
	24 hours	0	0	0	(0)	0	0	0	(0)
	48 hours	0	0	0	0	0	0	0	0
	72 hours	0	0	0	(0)	0	0	0	(0)

**Table 7. Assessment of ocular irritation test score of formulated hand sanitizer**

Skin reaction	Observation time	Individual score- rabbit number (Test group)				Individual score- rabbit number (Control group)			
		A	B	C	Total	A	B	C	Total
Cornea	3 minutes	0	0	0	0	0	0	0	0
	4 hours	0	0	0	0	0	0	0	0
	24 hours	0	0	0	0	0	0	0	0
	48 hours	0	0	0	0	0	0	0	0
	72 hours	0	0	0	0	0	0	0	0
Iris	3 minutes	0	0	0	0	0	0	0	0
	4 hours	0	0	0	0	0	0	0	0
	24 hours	0	0	0	0	0	0	0	0
	48 hours	0	0	0	0	0	0	0	0
	72 hours	0	0	0	0	0	0	0	0
Conjunctiva	3 minutes	0	1	1	2	0	0	0	0
	1 hour	1	1	1	3	0	0	0	0
	24 hours	1	1	1	3	0	0	0	0
	48 hours	1	1	1	3	0	0	0	0
	72 hours	1	0	1	2 (26)	0	0	0	0



**Plate 1. The results of eye and skin irritation tests of the herbal hand sanitizer on test rabbits**  
 Key: HS = Herbal sanitizer

**Table 8. Assessment of subjects' response on the quality of the formulated hand sanitizers**

Questions	Reponses	
	Yes	No
Do you receive formal training on how to use hand sanitizer?	20	0
Do you find the hand sanitizer as a quick and easy product to use?	20	0
Do you wear jewellery when applying hand sanitizer?	17	3
Is hand rubbing more rapid for hand cleansing than hand washing?	18	2
Do you use any antibiotics for the last four (4) weeks?	2	18
Do you trust this hand sanitizer to deliver effective hand hygiene?	20	0
Does it cause skin dryness more than repeated hand washing with soap and water?	4	16
Does it cause stinging skin irritation?	0	20
Does it cause more allergy and skin intolerance?	0	20
Does it cause stinging of the hands due to pre-existing skin irritation?	0	20
Does it cause cracking of the skin?	0	20
Does it cause piling of the skin?	0	20
Does it affect the colour of the nail?	0	20
Does it moisturize?	18	2
Does it cause excessive sweating of the palm?	0	20
Does it cause crinkling of the eye?	18	2
Do you perceive the effect after long period of usage?	19	1
Does it cause any involuntary action (e.g. sneezing) when used?	0	20
Does it cause watering of the eye	19	1
Are you pleased with the aroma or odour of the sanitizer?	18	2
How much (in Naira) are you willing to pay for 50 ml bottle which will last for 2-3 weeks upon regular use?		
Less than 500	6	
500-600	8	
700-800	6	
1000 and above	0	
Apart from the germ killing property of the hand sanitizer, what other properties would you like in the product?		
Fragrance	2	
Moisturizing agent	2	
Convenient and attractive packaging	16	
If you were to change one thing about the product to improve hand hygiene, what would it be?		
Fragrance	2	
Moisturizing agent	2	
Convenient and attractive packaging	16	
Please rate your satisfaction with the hand hygiene product		
Not at all satisfied	0	
Averagely satisfied	8	
Extremely satisfied	12	

#### 4. DISCUSSION

Human hands harbour microorganisms ranging from normal microbiota to pathogenic species [25]. Human skin provides optimum growth conditions for most disease causing organisms and also the opportunistic pathogens. These bacteria evidently could develop resistance to the cleaning agents, thus contributing to their persistence in an ecosystem [26]. The use of hand sanitizer is one of the means of reducing the microbial load present on the hands [27].

Ginger has direct anti-microbial activity and thus can be used in the treatment of bacterial infections [14]. Several studies have described the antibacterial and antifungal properties of different herbs and spices [10]. However, there is little information on the exact mechanism of their antimicrobial action [10,28-34]. The antimicrobial activity of medicinal plants is due to specific phytochemicals or essential oils present in them [10,11].

This study revealed the microbial effect of ginger on the different bacterial and fungal tested at



different concentrations ranging from 10 mg/ml to 50 mg/ml. The extract had the highest zone of inhibition on *E. coli* followed by *S. aureus* while *B. subtilis* showed the least susceptibility. These findings compare well with those of Omoya and Akharaiyi [7] who reported maximum antimicrobial activity of ginger extract against *E. coli* and slightly low inhibitory effect on *B. subtilis*.

Ginger extracts have been reported to exhibit antifungal activity against *A. fumigatus*, *Fusarium* sp and *Alternaria* sp. [35,36]. Ficker et al. [37] also isolated and identified the antifungal phytocompounds from ginger. The factors responsible for the high susceptibility of the test organisms to ginger extract are not exactly known but may be attributed to the secondary metabolites and bioactive compounds in higher plants [38].

The formulated herbal hand sanitizer completely inhibited the growth of microorganisms on agar medium. Also, the herbal hand sanitizer exhibited a high antimicrobial efficacy in inhibiting the growth of all the test organisms when used on glass bead. These results agree well with the study of Onyeagba et al. [39], Pankaj et al. [40] which showed the good antimicrobial activity of ginger extract against food borne pathogens. The goal of hand hygiene is a sufficient reduction of microbial load on the skin and the consequent break in the disease transmission route [41]. It is easier to prevent the hands from contamination than to decontaminate already soiled hands. The critical density of microorganisms on the hands needed for the spread of pathogens depends on the type and duration of contact, the type of microorganism, the individual's resident flora and their colonization resistance [2].

The safety of the formulated hand sanitizer is of great importance. A good hand sanitizer must be safe to use. Using the method of classification Draize [24], the herbal hand sanitizer was classified as non-irritant to skin but severely irritant to the eyes. These properties of the formulated herbal hand sanitizer conform well to the standard for healthcare antiseptic products recommended in professional and national guidelines by Center for Diseases Control and Prevention [42].

Compliance to hand hygiene has been reported to break the cycle of transmission of pathogens both in the hospital and at home [43]. Application of sanitizer could therefore be very useful and

acceptable in playing the significant role of bio-burden reduction on the hand as well as preventing irritation due to constant hand washing [44].

The overall assessment of the questionnaire in determination of the efficacy of the formulated herbal hand sanitizer shows a positive result as over 70% of the respondent's confirmed with respect to each of the questions asked. Hand washing removes lipids from the hand which results in adverse reactions such as dryness and irritation of the skin [15,44]. There was no adverse reaction in the formulated herbal hand sanitizer and ginger may have contributed to this.

## 5. CONCLUSION

In this study, the formulated herbal hand sanitizer has proved to be a promising cosmetic hygiene product as it has characteristically reduced microbial load and may serve as an alternative to hand washing. Its use could also aid compliance to hand hygiene in households and among health workers in hospitals since they do not easily cause skin irritation and/or dryness.

## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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