

Original Research Article

Formulation of carbomer-based hand sanitizers and comparison with some commercially available brands

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Sent for review: 26 November 2025

Revised accepted: 18 January 2026

Abstract

Purpose: To compare the pharmaceutical properties and antibacterial efficacy of different concentrations of carbomer-based hand sanitizers and some commercial brands available in Iraq.

Methods: Ethanol-containing hand sanitizers were prepared using different concentrations of carbomer (0.5 to 1 %) as a base. Some commercial brands were purchased from pharmacies in Basrah. They were all evaluated for viscosity, spreadability and pH. Furthermore, the following bacteria were tested against each hand sanitizer: *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, and *Staphylococcus aureus*.

Results: There was increase in viscosity as well as a corresponding decrease in pH and spreadability in the formulated hand sanitizers as carbomer concentration was increased from 0.5 to 1 %. All the commercial formulations demonstrated spreadability ranging from 0.4 to 0.62 cm. This was generally lower than the formulated gels (0.88 to 1.2 cm), while pH values of the commercial hand sanitizers were higher than those of the formulated samples. Sample C3 had the lowest pH value of 5.18 and hence was slightly acidic. Brand Z had no activity against all the organisms tested except *P. aeruginosa*, while Brand Y had no activity against *Klebsiella pneumoniae*. C3 was effective against all the organisms tested. However, it was not as effective as Brands H and M. Only one hand sanitizer had no effect (0 %) on all except *Pseudomonas aeruginosa*.

Conclusion: The newly prepared hand sanitizer has the lowest viscosity, pH and highest spreadability when compared to commercial brands. It also demonstrated comparable antimicrobial activity that was greater than that of the controls used. This study has also shown that some commercial hand sanitizers are not effective against common strains of microorganisms, hence would require reevaluation.

Keywords: Hand sanitizers, Viscosity, Microorganisms, Carbomer, Spreadability

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INTRODUCTION

The emergence of new bacterial or viral infections will always result in significant problems for global public health. Severe infections such as severe acute respiratory syndrome coronavirus 2 or SARS-CoV-2, and the most popularly known respiratory infection by COVID-19, are good examples of diseases that

are classified as global pandemics by the World Health Organization (WHO) in the year 2020 [1]. Hands are one of the easiest pathways for microorganisms and disease transmission in humans [2]. Personal hygiene, in addition to hand hygiene, is essential for preventing many infectious diseases. The term "hand hygiene" refers to various practices that involve either washing the hands or using hand sanitizers in

order to remove or eliminate microbes on the hands and fingers [3,4]. Hand sanitizers are available in various forms (gel or liquid) and compositions (alcoholic and nonalcoholic) [5,6]. The antimicrobial ability of antiseptics is a result of their active ingredients. Ethyl alcohol and isopropyl alcohol are active ingredients in alcoholic hand sanitizers [7], while the active ingredients of non-alcoholic sanitizers include triclosan, pyroglutamic acids, benzalkonium chloride, or povidone-iodine chlorinated aromatic compounds [6]. However, studies on their antimicrobial activities are limited.

The mechanism of action of both types is to denature the proteins of microorganisms. Additionally, alcohol denatures lipids and dehydrates microorganisms [8]. Benzalkonium chloride, an active ingredient in non-alcoholic hand sanitizers, has low efficacy against gram-negative bacteria [9]. The pharmaceutical evaluation of hand sanitizers in post-production is necessary to ensure quality [10].

This study aims to prepare carbomer-based, ethanol-containing hand sanitizers and compare their pharmaceutical and microbial activities with some commercially available brands from Pharmacies in Basrah.

EXPERIMENTAL

Materials

Absolute ethyl alcohol (Duksan Korea), carbomer (Alpha Chemika, India), fragrance, glycerin, triethanolamine (Hopkins and Williams Ltd,

England) and distilled water were used to prepare the hand sanitizer (C) that is the subject of this study. Table 1 presents the details of four brands of commercially available hand sanitizers (containing 70 % ethanol V/V, depending on label information), which were purchased from pharmacies in Basrah, Iraq. These products were available during the COVID-19 pandemic and were used before their respective expiration dates.

METHODS

Preparation of carbomer-based hand sanitizer

Carbomer in different ratios (0.5, 0.75 and 1 %w/v) was dispersed in distilled water (10 mL) with glycerin (10 mL). The mixture was agitated using a mechanical agitator till a homogeneous mixture was formed. Afterwards, absolute ethyl alcohol (70 mL) was added with continuous stirring. Finally, two drops of tri-ethanol amine were added to neutralize the dispersion and initiate the gelling properties of the carbomer, and the volume was finally brought to 100 mL using distilled water. The hand sanitizers produced were labelled C1, C2, and C3 for 0.5, 0.75, and 1 %w/v carbomer concentrations, respectively [10].

Measurement of pH

A calibrated pH meter (Inolab, Germany) was used to estimate the pH of the prepared hand sanitizer and the commercial brands. This was done in triplicate (n = 3).

Table 1: Labelled information of commercial hand sanitizers

Brand code	Trade name	Date of manufacture	Expiry date	Manufacturer and country	Labelled materials and their strengths
H	HiGeen®	2019	2022	Sukhtian Households and Toiletries Manufacturing Co. Ltd. Al Hizam Street, Abu Alanda, Amman, Jordan	Ethyl alcohol 70%, acrylates (C10-30 alkyl acrylate cross polymer), carbomer, glycerin, triethanolamine, water, fragrance
M	KONIX®	2019	2022	Turkuaz, Turkey	Ethyl alcohol 70%, acrylates (C10-30 alkyl acrylate cross polymer), glycerine, triethanolamine, water, Isopropyl myristate, tocopheryl acetate, aminoethyl propanol, fragrance
Y	DR. SHAMOOS®	2019	2022	P.R.C	Ethyl alcohol 70%, water, Aloe vera extract, PEG-400, Hydrogenated castor oil, chloroxylenol, carbomer, Tri ethanol amine, fragrance
Z	SHIJING®	2019	2022	China	Ethyl alcohol 70%, water, carbomer triethyl product.

Measurement of viscosity

A viscometer (NDJ-8S viscometer, China) was set up and a suitable spindle was chosen based on the sample to be evaluated. The spindle was immersed in the liquid (the mark of the spindle was on the same level as the liquid, and then adjusted to a horizontal level. After running for two minutes, the readings were recorded [11].

Spreadability test

This evaluation was done using an earlier reported method [11]. Approximately 1 g of the gel was applied between two glass plates and a standardized mass of 100 g was placed on the upper plate. The spreading diameter (d) of 1 g of gel after 1 min was measured. The spreadability (S) was calculated using Eq 1.

$$S = d^2 \times \pi / 4 \dots\dots\dots (1)$$

Microbiological assay

Bacterial strains

Four bacterial strains commonly associated with nosocomial infections (*Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, and *Staphylococcus aureus*) were selected to test the efficacy of the hand sanitizers. The bacterial strains were obtained from the stock culture collection in the Department of Clinical Science, College of Pharmacy, Basrah, Iraq. Then inoculated onto nutrient agar and incubated at 37 °C overnight. For each strain, a bacterial suspension containing 2 mL of sterile normal saline was prepared aseptically and calibrated to a McFarland standard of 0.5 nm absorbance [12].

Disk diffusion method

The bacteria were equally dispersed over the surface of 90 mm Mueller-Hinton Agar plates using a cotton swab and left to dry for approximately 5 minutes within the Biosafety cabinet class II. Two holes in the agar were bored using a sterile 4 mm cork borer. One hole was filled with 100 µL of the selected hand sanitizer using a pipette, and one hole was filled with 100 µL sterile water as a negative control. Separate plates were used for each commercial brand, while 100 µL of laboratory-prepared 70 % ethanol was pipetted into each hole, and a streptomycin antibiotic disc was placed on the agar plate (as a positive control) by boring a hole. This was repeated with all of the tested strains and hand sanitizers. The agar plates were incubated for 24 h at 37 °C. Zones of inhibition

demonstrated the tested organism's resistance or susceptibility (sensitivity) to the anti-bacterial agent and were measured using a digital caliper. For the efficacy measurement, bacterial strains with zones of inhibition greater than or equal to the standard zones (70 % ethanol and 25 µg streptomycin disc) indicated sensitivity, while smaller zones indicated resistance of the bacteria to the hand sanitizer [13,14].

Statistical analysis

Data are reported as means of triplicate determinations ± standard deviation (SD) and were examined using analysis of variance (ANOVA). *P*-value < 0.05 was considered significant.

RESULTS

Table 2 revealed an increase in viscosity as well as a corresponding decrease in pH and spreadability in the formulated hand sanitizers as carbomer concentration was increased from 0.5 to 1 %. The 1 % Carbomer formulation gave the best gel property regarding the combination of pH, viscosity and spreadability. Hence, it was selected for further comparison with other commercially available products.

Spreadability and pH

All the commercial formulations demonstrated spreadability ranging from 0.4 to 0.62 cm. This was generally lower than the formulated gels (0.88 to 1.2 cm), while pH values of the commercial hand sanitizers were higher than those of the formulated samples. Sample C3 had the lowest pH value of 5.18 and hence was slightly acidic.

Viscosity

The results show that all samples follow shear thinning pattern, but with variation in response depending on the intermolecular forces of the sample. Product C3 had the lowest viscosity at a speed rate (60 rpm) while samples H, M, and Z had the highest viscosity, as illustrated in Table 3.

Microbiological properties

Antibacterial efficacy of the four commercially available hand sanitizers (H, M, Y, and Z) and the carbomer-based hand sanitizer (C3) was tested by measuring the average zone of inhibition (ZOI) and standard deviation (SD) values, and comparing them with ZOI produced by 70% alcohol and streptomycin (Table 4).

Table 2: Properties of carbomer-based and commercial hand sanitizers (n = 3)

Parameter	pH	Viscosity (cp)	Spreadability (cm)
C1 (0.5%)	7.18±0.04	300.8±1.8	1.2±0.02
C2 (0.75%)	6.88±0.03	325.7±2.5	0.98±0.02
C3 (1%)	5.18±0.05	352.97±2.2	0.88±0.0 2
H	7.36±0.006	1403.3±15.3	0.44±0.048
M	6.95±0.214	4207.17±16.2	0.41±0.007
Y	7.20±0.265	1733.23±33.8	0.62±0.032
Z	6.51±0.121	3850.07±33.5	0.4±0.036

Values are mean ± SD

Table 3: Effect of shearing rate on viscosity of the various hand sanitizers (Spindle method)

Shearing rate (rpm)	H (cp)	M (cp)	Y(cps)	C3 (cps)	Z (cps)
0.3	No response	No response	86250.7±159	No response	No response
0.6	60321.7±358	No response	46628.7±219	No response	No response
1.5	28820.7±40.8	86486.7±419	22749.3±330	4922.57±188	No response
3	16234.7±45	47670±97.6	13341.7±235	2979.17±96.9	45940.3±31.5
6	9033.77±22.7	25969.7±135	8171.53±20.7	1858.9±49.3	24662.7±145
12	5450.33±88.8	14711.7±296	4966±41.2	1182.23±27.3	13679.3±127
30	2830.17±29.1	7159.87±29.3	2732.17±16.2	680.433±16.7	6535.77±30.5
60	1403.3±15.3	4207.17±16.2	1733.23±33.8	352.967±2.2	3850.07±33.5

Values are mean ± SD; n = 3

Table 4: Zone of inhibition measured in (mm) after 24 h incubation at 37 °C for different hand sanitizers against different test organisms

Formulation	Gram-Negative			Gram-Positive
	<i>Escherichia coli</i>	<i>Klebsiella pneumoniae</i>	<i>Pseudomonas aeruginosa</i>	<i>Staphylococcus aureus</i>
H	16.5±2.12	15.5 ±2.12	28.0±2.83	17.0 ±4.24
M	17.0±1.41	15.0±1.41	17.0±1.41	16.0 ±0
C3	9.0 ±2.83	14.0 ±1.41	18.0±2.83	11.0 ±1.41
Y	17.0±1.41	0	18.0 ± 0	12.0 ±2.83
Z	0	0	18.0±2.83	0
Lab control 70%	7.0±0.81	2.0±0.81	7.0±0.81	2.0±0.81
Streptomycin	4.0±1.15	7.0±0.81	7.0±0.81	6.0±1.63

Values are mean ± SD; n = 3

There were variations in ZOI produced by all tested hand sanitizers within each bacterial strain. Brand Z had no activity against all the organisms tested except *P. aeruginosa*, while Brand Y had no activity against *Klebsiella pneumoniae*. C3 was effective against all the organisms tested. However, it was not as effective as Brands H and M (Table 4).

The highest average zone of inhibition was 28 mm, which was observed in *Pseudomonas aeruginosa* against the H-labelled hand sanitizer. Brands H, M and prepared C3 produced IZD that were generally higher than those produced by 70 % ethanol and streptomycin.

DISCUSSION

Most hand sanitizers have a neutral or slightly basic pH to avoid skin irritation. Although human skin tends to be acidic, excessive use of alkaline hand sanitizer may be problematic; excessive use of acidic hand sanitizers can also irritate the

skin. Some studies have reported that bacterial growth may be affected by pH value, as certain bacteria thrive at high pH [15].

Viscosity is an important property in the design, formulation, and manufacturing process of hand gels [16]. A higher viscosity product may be perceived as higher quality. Low viscosity allows for packaging needs, for example, allowing the gel to be pumped straight from the bottle. A high viscosity product, when stored, gives the product a premium appearance. A drop in viscosity when sheared allows the gel to be dispensed through a pump or nozzle while preserving a premium appearance. Spreadability indicates the extent of the area to which the gel readily spreads on application to the skin. It has a relationship with the therapeutic potency of the formulation. The more the spreadability, the greater the potency of hand sanitizers in killing germs. Moreover, it helps in the uniform application of the product to the skin. Therefore, hand sanitizers must have good spreadability and satisfy the ideal quality in

topical application [17]. The C1 formulation had the highest spreadability because of its low concentration of carbomer. Unfortunately, the very low viscosity makes it undesirable due to lack of elegance and poor retention at the site of action. The chosen formulation C3, although it displayed sufficient viscosity property, still had the lowest viscosity but the highest spreadability when compared to all the commercial brands tested in this study.

In the biological assay, streptomycin and 70 % ethanol were utilized as positive controls or standards in the laboratory. If the zone of inhibition of any of the tested sanitizers is greater than the controls, it is considered effective; whereas zones of inhibition less than these controls are considered less effective. Brand Z Sanitizer was ineffective in reducing the growth of all bacterial strains except *P. aeruginosa*. Hand sanitizers H, M, C3, and Y were effective against all bacterial strains, as their zones of inhibition were more significant than the two positive controls. The concentration of alcohol was indicated on the ingredients of hand sanitizers (H, M, C3), whereas brands Y and Z had unknown concentrations of alcohol. Hand hygiene with sanitizers is one of the most effective strategies to prevent the transmission of pathogens, including bacteria, in the workplace and the community. In recent years, the use of hand sanitizers has become more widespread worldwide because of COVID-19 pandemic, which primarily uses supportive and preventative methods to reduce transmission [18,19].

As a result, several traders and research Institutions in Iraq produced and imported several hand sanitizers. With the increased use of hand sanitizers, it is necessary to evaluate the efficacy of the various products. The antibacterial activity of hand sanitizers routinely used in Iraq was also addressed in this study.

Zones of inhibition for the sanitizers at the manufacturers' concentration ranged between 9 and 28 mm. The sanitizers showed varying inhibitory activity against the bacterial strain. It was observed that the number of tested products with lower inhibitory zones was less effective than previously published works [20]. The decreased effectiveness may be a result of increased consumer demand and compromised manufacturing techniques. The most resistant bacterial strain in the current study was *K. pneumonia*. These findings suggest that not all hand sanitizers or decontaminants are effective against bacteria. An earlier study [21] investigated the effectiveness of three hand sanitizers against *E. coli*, *K. pneumoniae*, *P.*

aeruginosa, and *S. aureus*. The study shows that *K. pneumonia* and *S. aureus* have a higher zone of inhibition when compared to the 70% ethanol positive control [21].

In this study, the formulated hand sanitizer (C3) compared favourably against the commercial hand sanitizers (H and M), although with lower zones of inhibition when tested against *E. coli*, *K. pneumoniae*, *P. aeruginosa*, and *S. aureus*. The C3, however, had a higher ZOI when compared with 70 % ethanol and streptomycin (25 µg). The higher zones of inhibition in this study mean that the bacterial strain cannot grow in the presence of the hand sanitizer compared with streptomycin. Regardless of brand or formulation, hand sanitizer must efficiently minimize microbial contamination on the hands. Hand sanitizer Z had no zone of inhibition against *E. coli*, *K. pneumoniae*, and *S. aureus*. The failure of this hand sanitizer to inhibit their growth may be due to suboptimal alcohol concentration.

CONCLUSION

Carbomer (1 %) based ethanol containing hand sanitizer has the lowest viscosity, pH and highest spreadability when compared to commercial brands. It also demonstrates comparable antimicrobial activity that was greater than that of the controls used. This study has also shown that some commercial hand sanitizers are not effective against common strains of microorganisms, hence would require re-evaluation.

DECLARATIONS

Acknowledgment/Funding

I would like to extend my sincere gratitude to my students Hala Khalid Abdulkareem, Miryam Muhanad Awaid and Zainab Hazim Sabih. I want to thank them for their role in conducting some experiments during the research.

Ethical approval

None provided.

Use of Artificial intelligence/Large language models

We also declare that we did not use Generative artificial intelligence (AI) and AI-assisted technologies in writing the manuscript.

Availability of data and materials

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Conflict of interest

No conflict of interest is associated with this work.

Contribution of authors

We declare that this work was done by the author(s) named in this article, and all liabilities pertaining to claims relating to the content of this article will be borne by the authors.

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