Biocidal actives and their efficacy against COVID-19 (SARS-CoV-2)

We at Amphochem supply a range of biocidal active compounds for formulation of biocidal products. Three surfactant based biocidal actives in our active biocidal product range are Arquad MCB, Arquad 2.10 and Triameen Y12D. We frequently receive questions concerning the efficacy of these biocidal active products against the currently circulating strain of Corona Virus, COVID-19. As of today, and like most other biocidal actives, none of these actives are tested directly against COVID-19. Their anticipated efficacy is based on an extrapolation of effect shown on other viruses belonging to the same main group, enveloped viruses. Amongst others the virus causing Swine flu (H1N1) some time ago, as well as the (in)famous virus causing the "SARS-outbreak", belong to the same main group. The enveloped viruses have a lipid envelope causing them to be rather sensitive towards various substances and environments (e.g. heat).

All three above mentioned biocidal active products have been tested against the H1N1 influenza virus, as stated above also being an enveloped virus. This does not explicitly prove effect against COVID-19, however is a strong indication effect can be expected against other enveloped viruses as well. Statements concerning testing against the H1N1 virus, as well as a summary of common viruses belonging to the main group of enveloped viruses are attached.

Furthermore, we also frequently receive questions concerning the use of above biocidal products for hand sanitizers. Connected to BPR, both Arquad MCB and Arquad 2.10 have been submitted for authorization within product type 1 (PT1, "human hygiene") with BPR authorities. In Europe by tradition we have used mostly alcohol (Ethanol and Iso-propanol) as biocidal actives for these types of products, however e.g. Benzalkonium chloride-based products (Arquad MCB) have been on the US market for quite some time. There is an ever-ongoing debate on both the efficacy and required contact time for these products as compared to Alcohol-based alternatives. Furthermore, there are publications indicating a long-term effect of these products as compared to alcohol-based ones, in which the alcohol readily evaporates away, whilst Benzalkonium chloride stays and can continue to act. Three articles dealing with the subject are attached for reference.

So are these products effective against COVID-19

Many results indicate they are, however there is no replacement for a real test involving the COVID-19 virus. However, no such test is to be expected within near future so for the time being each acting party needs to decide for themselves, based on existing data, whether they will place a product on the market for the purpose of fighting COVID-19.

For any inquires, how to formulate or others please contact your sales representative!

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Biocides Information sheet

Virucidal Efficacy

Triameen Y12D, Arquad MCB, Arquad 2.10

Viruses are small infectious agents replicating only inside of living cells of their hosts. They can infect all types of life forms like humans, animals, plants and microorganisms. Usually they have a narrow host range.
There are 2 main groups based on their structure: enveloped and non-enveloped viruses. Enveloped viruses have a lipid envelope which is relatively sensitive to desiccation, heat and detergents. They are easier to be inactivated by biocidal active substances than non-enveloped viruses.

Examples for enveloped viruses are listed below:

- Herpesviruses (e.g. Epstein-Barr virus, Herpes simplex, Bovine alphaherpesvirus 1 (causing Bovine Rhinotracheitis), suid herpesvirus 1 (causing pseudorabies=Aujeszky's disease))
- Poxviruses (e.g. smallpox, vaccinia virus)
- Hepadnaviruses (e.g. Hepatitis B virus)
- Asfarviridae (e.g. African Swine fewer)
- Flavivirus (e.g. Hepatitis C, yellow fever virus, Zika virus, dengue virus)
- Alphavirus (e.g. eastern equine encephalitis virus)
- Togavirus (e.g. Alphavirus)
- Coronavirus (e.g. SARS virus, MERS virus, novel coronavirus (2019-nCoV – Wuhan coronavirus outbreak))
- Hepatitis D virus
- Orthomyxovirus (e.g. Influenca virus A (H1N1-swine flu, H3N2-Hong Kong flu, H9N2-avain influenza, H3N8, H5N1, H5N2…), Influenca virus B, Influenca virus C, Influenca virus D)
- Paramyxovirus (e.g. mumps virus, human parainfluenza virus, measles virus, canine distemper virus, rinderpest virus)
- Rhabdovirus (e.g. rabies virus)
- Bunyavirus (e.g. Hantavirus, California encephalitis virus, Congo hemorrharic fever virus)
- Filovirus (e.g. Ebola virus, Marburg virus)
- Retrovirus (e.g. Human Immunodeficiency Virus (HIV), Mouse mammary tumor virus)
- Arteriviridae (e.g. Porcine Respiratory and Reproductive Syndrome Virus (PRRSV))

The biocidal active substances in our products below are effective against enveloped viruses

- Triameen Y12D, Triameen Y12D-30
  (N-(3-aminopropyl)-N-dodecylpropane-1,3-diamine - CAS No.: 2372-82-9)
- Arquad MCB-50, Arquad MCB-80
  (BKC = C12-16-alkyldimethylbenzylammonium chloride - CAS number 68424-85-1)
- Arquad 2.10-50, Arquad 2.10-70 HFP, Arquad 2.10-80
  (DDAC = Didecyldimethylammonium chloride - CAS number 7173-51-5)

This is shown in numerous literature studies.

Own test data are available for Influenza virus H1N1. Details can be received on request.

Customer formulations containing our products will still have to be tested according to national regulations. In case a (biocides) registration scheme is in place customer products might need (national) approval before being placed on the market.

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Statement

Efficacy of ARQUAD® MCB-50 versus influenza virus H1N1 (Swine Flu, Mexican flu)

The efficacy of ARQUAD® MCB-50 was tested in a suspension test following to the European standard DIN EN 14476:2005-08 under clean conditions (0,03% BSA).

ARQUAD® MCB-50 was tested as a 750 ppm, 500 ppm, and 250 ppm solution.
The exposure times were 1 min, 5 min, 10 min and 15 min.

Conclusion: 750 ppm concentration of ARQUAD® MCB-50 (is 375 ppm active substance) is effective against influenza virus A/H1N1/X-179A at room temperature under clean conditions within an application time of 10 minutes.

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Statement

Efficacy of ARQUAD® 2.10-50 versus influenza virus H1N1 (Swine Flu, Mexican flu)

The efficacy of ARQUAD® 2.10-50 was tested in a suspension test following to the European standard DIN EN 14476:2005-08 under clean conditions (0,03% BSA).

ARQUAD® 2.10-50 was tested as a 250 ppm, 125 ppm, and 62.5 ppm solution.
The exposure times were 1 min, 5 min, 10 min and 15 min.

Conclusion: 250 ppm concentration of ARQUAD® 2.10-50 (= 125 ppm active substance) is effective against influenza virus A/H1N1/X-179A at room temperature under clean conditions within an application time of 5 minutes.

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Statement

Efficacy of TRIAMEEN® Y12D versus influenza virus H1N1 (Swine Flu, Mexican flu)

The efficacy of TRIAMEEN® Y12D was tested in a suspension test following to the European standard DIN EN 14476:2005-08 under clean conditions (0,03% BSA).

TRIAMEEN® Y12D was tested as at 300 ppm, 225 ppm, and 150 ppm solution.
The exposure times were 1 min, 5 min, 10 min and 15 min.

Conclusion: 300 ppm concentration of TRIAMEEN® Y12D (is 300 ppm active substance) is effective against influenza virus A/H1N1/X-179A at room temperature under clean conditions within an application time of 10 minutes.

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Effectiveness of a Nonrinse, Alcohol-Free Antiseptic Hand Wash

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This study evaluated the efficacy of a novel surfactant, allantoin, and benzalkonium chloride hand sanitizer using the US Food and Drug Administration's method for testing antiseptic hand washes that podiatric physicians and other health-care personnel use. The alcohol-free product, HandClens, was compared with an alcohol-based product, Purell. Independent researchers from the California College of Podiatric Medicine conducted the study using 40 volunteer students from the class of 2001. The results show that HandClens outperformed Purell and met the regulatory requirements for a hand sanitizer. Purell failed as an antimicrobial hand wash and was less effective than a control soap used in the study. (J Am Podiatr Med Assoc 91(6): 288-293, 2001)

In today's health-care environment, prudent handwashing practices have been adopted to decrease the transmission of bacteria from person to person.[1] However, the conscientious health-care workers, podiatric physicians, and others who follow these guidelines have a greater risk than less conscientious workers of developing a contact dermatitis due to repetitive hand washing and glove changing.[2-7] It has been established that the damaged skin of nurses can carry a greater number of bacterial pathogens associated with nosocomial infections than can healthy, undamaged skin.[2]

The irony of the antisepsis practices' causing dermatologic changes was originally discovered by Halstead.[8, 9] Halstead invented the surgical glove as a means of reducing the hand irritation associated with the antimicrobial agents being used at the time. Although hand-washing formulations have changed since Halstead's time, the contact dermatitis associated with antimicrobial agents has remained.[10]

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The need for a broad-spectrum, long-lasting antimicrobial hand-wash product that protects the natural skin barrier motivated the development of an alcohol-free formulation of surfactants, allantoin, and benzalkonium chloride (SAB) in a hand spritz.[11] In this study, the efficacy of this novel formulation was compared with that of an alcohol-containing hand sanitizer using the US Food and Drug Administration (FDA) protocol for health-care personnel antiseptic hand washes. Independent researchers from the California College of Podiatric Medicine conducted the study using 40 volunteer students from the class of 2001.

Materials and Methods

Test Solutions

For this study, two solutions were evaluated using the FDA protocol (21 CFR 333.470) for health-care personnel antiseptic hand washes.

The first solution was the SAB hand wash, HandClens (Woodward Laboratories, Inc, Los Alamitos, California). The active ingredient in this product is benzalkonium chloride (0.13% vol/vol). Other ingredients are water, hydroxypropylmethyl cellulose, propylene glycol, cocamidopropylbetaine, cocamido-

propylamine oxide, cetyl, trimethyl ammonium chloride, quaternium-12, imidazolidinyl urea, quaternium-15, allantoin, methyl paraben, propyl paraben, eucalyptol, methyl salicylate, and triethanolamine.

The second solution was alcohol-based Purell (GOJO Industries, Akron, Ohio). The active ingredient is ethyl alcohol (62% vol/vol). Other ingredients are isopropyl alcohol, water, emollients, and thickener.

Subjects

Forty volunteer students from the class of 2001 at the California College of Podiatric Medicine participated in this study. All subjects completed a 7-day quarantine period of abstaining from antimicrobial products prior to testing. All subjects had fingernails no longer than 2 mm. To be eligible for inclusion, subjects could have no abrasions or open lesions on their wrists or hands, and the wearing of jewelry was not permitted. Each subject gave informed verbal and written consent before entering into the study.

Protocol

Each subject was asked to perform a practice wash using the control soap, Dove (Lever Pond's, Toronto, Ontario, Canada). Then, 5 g of control soap and 15 mL of sterile phosphate buffer were used to wash the hands for 2 min. Subjects then rinsed their hands under tap water for 30 sec.

Each subject was given a baseline inoculation: 5 mL of *Serratia marcescens* was placed in the subject's cupped hands. The subject rubbed the hands for 45 sec and allowed the hands to air-dry for another 2 min. Next, subjects underwent gloving and sampling. Large powder-free polyethylene gloves, with 50 mL of sterile phosphate buffer in each glove, were placed on the hands, secured with a rubber band at the wrist, and massaged for 1 min (gloving). Finally, both gloves were drained for "glove juice," using a sterile technique, into a sterile sampling tube. Undiluted and diluted samples were placed on agar plates, consisting of a combination of lecithin and polysorbate 20, and stored at room temperature, or 25°C (sampling).

Each subject was given a control wash. Subjects were inoculated once again and given 5 g of control soap to wash with for 2 min. They rinsed for 30 sec and then underwent gloving and sampling.

Each subject was allowed ten hand washes with the appropriate test solution. The steps were similar to those in the inoculation procedure: 5 mL of test solution with 2 min of washing, then gloving, and sampling only on hand washes 1, 3, 7, and 10. For hand washes 2, 4, 5, 6, 8, and 9, glove juice was drained down the sink and all other steps remained the same.

Each sample was placed on an agar plate immediately. An undiluted plate was made. For more accurate counting, a diluted plate of a 1:10,000 dilution for baseline, a 1:1,000 dilution for control, and a 1:100 dilution for the hand-wash trials were made. In order to inactivate the benzalkonium chloride, and to prevent further microbicidal activity, a combination of lecithin and polysorbate 20 was used as the agar media. For inactivation of the alcohol-based sanitizer, simple dilution in the phosphate buffer was sufficient.

A 2-min intermission was employed between each contamination/hand-wash cycle and the next contamination/hand-wash cycle. Subjects refrained from touching any items after the practice wash was initiated. After the final hand wash, subjects were given their choice of antimicrobial scrub or soap.

Data Analysis

Statistical analysis was performed using a Student's *t*-test available with Microsoft Excel (Microsoft, Inc, Redmond, Washington). For the purpose of comparison, mean differences were considered statistically significant if the confidence value returned from the test was 0.05 (ie, $P < .05$).

Results

For this experiment, the efficacy of a novel SAB formulation, HandClens, *versus* an alcohol-based, established formulation, Purell, was evaluated in terms of immediate and residual disinfecting power. Data sheets were kept for all subjects. The number of colony-forming units seen on the lecithin and polysorbate 20 agar plates for both undiluted and diluted samples was recorded for baseline contamination, control soap, wash 1 (test solution), wash 3, wash 7, and wash 10. Data on the individual subjects are provided in Tables 1 and 2. The efficacy of a test solution was calculated as a reduction factor (RF) defined by the FDA as the difference between the $\log_{10}$ of the colony-forming unit for baseline contamination (CFU_B) and the $\log_{10}$ of the colony-forming unit for washes (CFU_W):

$$RF = Log_{10}\, CFU_B - Log_{10}\, CFU_W$$

The FDA requires a minimum reduction factor value of 2 after a single hand wash and a minimum reduction factor value of 3 after the tenth hand wash for a hand sanitizer to be considered an acceptable antiseptic formulation. Statistical analysis was performed using

Table 1. Data Collected for Subjects in the HandClens Group

Subject ID	Baseline	Control	Wash 1	Wash 3	Wash 7	Wash 10
1	450,000	830	275	23	7	160
2	590,000	1,200	120,000	8	3	1
3	770,000	51,000	80	6	1	1
4	[a]	1,600	10,400	312	26	4
5	610,000	800	80,000	1	2	1
6	90,000	400	170	1	2	54
7	1,100,000	2,000	228	800	5	25
8	50,000	11,000	500	102	52	294
9	170,000	800	112	18	1	17
10	10,000	59,000	800	1	27	7
11	3,200,000	800	640	1	1	1
12	120,000	800	280	2	4	1
13	480,000	800	8,000	3	1	2
14	1,840,000	800	260	1	1	1
15	960,000	1,520	1,400	115	27	54
16	1,260,000	1,460	74,800	2	2	1
17	1,030,000	5,000	103	2	3	2
18	440,000	23,000	15,200	1	2,200	1
19	690,000	5,000	49	1	3	10
20	2,960,000	68,000	7,300	68	12	37

Note: Numbers shown indicate the number of colony-forming units with the following dilutions: baseline, 1:10,000; control, 1:1,000; hand wash, 1:100.

[a] Unable to calculate due to baseline sample overgrowth (too numerous to count).

Table 2. Data Collected for Subjects in the Purell Group

Subject ID	Baseline	Control	Wash 1	Wash 3	Wash 7	Wash 10
21	760,000	688	4,000	7,200	23,900	23,500
22	50,000	400	800	12,600	8,200	160,000
23	140,000	800	5,900	23,000	7,000	13,200
24	220,000	1,600	400	800	3,900	400
25	400,000	4,000	800	2,100	2,700	9,000
26	460,000	400	1,600	5,100	10,000	4,900
27	400,000	4,500	9,000	12,000	40,000	28,000
28	130,000	2,400	270	7,600	5,500	26,400
29	400,000	2,200	15,100	3,100	6,800	13,200
30	1,080,000	4,700	464	6,000	9,200	5,800
31	180,000	5,200	3,500	7,200	8,000	12,000
32	1,000,000	6,100	5,200	3,400	40,000	8,700
33	1,980,000	3,700	6,100	6,600	19,100	27,200
34	1,100,000	10,800	17,200	13,100	16,100	9,200
35	1,800,000	11,200	2,400	6,100	9,500	28,000
36	50,000	300	9,400	6,800	14,400	6,600
37	820,000	15,200	28,800	12,600	11,600	35,200
38	670,000	96,000	2,400	4,800	10,600	6,400
39	400,000	9,600	17,200	88,000	80,000	[a]
40	1,120,000	172,000	27,200	23,200	2,800	19,600

Note: Numbers shown indicate the number of colony-forming units with the following dilutions: baseline, 1:10,000; control, 1:1,000; hand wash, 1:100.

[a] Unable to calculate due to baseline sample overgrowth (too numerous to count).

the Student's *t*-test, and the results are shown in Tables 3 and 4.

The results showed that both groups met the minimum requirement for the first hand wash, with an average reduction factor value of 2.6 for HandClens and 2.1 for Purell. Next, an overall trend of sustained disinfecting power was seen for HandClens, as demonstrated by reduction factor values of 2.6, 4.9, 5.0, and 4.9 for hand washes 1, 3, 7, and 10, respectively. These values not only met the first requirement, but surpassed the minimum expected persistent values. This is noticeable at the third hand wash, which exceeds the expected persistence by 1.9 $\log_{10}$ units, and also at the seventh hand wash, as demonstrated by 2.0 $\log_{10}$ units.

In contrast, Purell's performance diminished over time and hand washes, as illustrated by reduction factor values of 2.1, 1.8, 1.6, and 1.5 for hand washes 1, 3, 7, and 10, respectively. Clearly, these values began to plummet as early as the third hand wash (Fig. 1) and failed to meet FDA standards for an antiseptic hand sanitizer. Indeed, by the tenth hand wash, Purell's disinfecting abilities did not meet the minimal requirements with a 1.5 $\log_{10}$ value. In fact, the antimicrobial capacity of Purell by the tenth hand wash was 0.5 $\log_{10}$ less than that of the control soap (Dove). Surprisingly, only a 0.1 $\log_{10}$ difference was found between the disinfecting ability of the nonantimicrobial control soap and that of the alcohol-based antimicrobial Purell. The antimicrobial activity of the alcohol-based hand sanitizer was significantly less (wash 1, $P < .001$; washes 3, 7, and 10, $P < .001$) than that of the alcohol-free HandClens product.

Discussion

The value of using an antimicrobial hand sanitizer with an acceptable disinfecting power defined by the FDA for podiatric physicians and other health-care workers has already been emphasized. A nonirritating sanitizer with residual activity is ideal to preserve the natural skin barrier. This study evaluated the

Table 3. Log Reductions for Subjects Using HandClens

	Control	Wash 1	Wash 3	Wash 7	Wash 10
Subject ID					
1	2.7	3.2	4.3	4.8	3.4
2	2.7	0.7	4.9	5.3	5.8
3	1.2	4.0	5.1	5.9	5.9
4	[a]	[a]	[a]	[a]	[a]
5	2.9	0.9	5.8	5.5	5.8
6	2.4	2.7	5.0	4.7	3.2
7	2.7	3.7	3.1	5.3	4.6
8	0.7	2.0	2.7	3.0	2.2
9	2.3	3.2	4.0	5.2	4.0
10	−0.8	1.1	4.0	2.6	3.2
11	3.6	3.7	6.5	6.5	6.5
12	2.2	2.6	4.8	4.5	5.1
13	2.8	1.8	5.2	5.7	5.4
14	3.4	3.8	6.3	6.3	6.3
15	2.8	2.8	3.9	4.6	4.2
16	2.9	1.2	5.8	5.8	6.1
17	2.3	4.0	5.7	5.5	5.7
18	1.3	1.5	5.6	2.3	5.6
19	2.1	4.1	5.8	5.4	4.8
20	1.6	2.6	4.6	5.4	4.9
Statistics					
Average	2.2	2.6	4.9	5.0	4.9
SD	1.0	1.2	1.0	1.2	1.2
t-value	9.266	9.86	20.62	18.44	17.63
df	18.0	18.0	18.0	18.0	18.0
P value	<.001	<.001	<.001	<.001	<.001

[a] Unable to calculate due to baseline sample overgrowth (too numerous to count).

Table 4. Log Reductions for Subjects Using Purell

	Control	Wash 1	Wash 3	Wash 7	Wash 10
Subject ID					
21	3.0	2.3	2.0	1.5	1.5
22	2.1	1.8	0.6	0.8	−0.5
23	2.2	1.4	0.8	1.3	1.0
24	2.1	2.7	2.4	1.8	2.7
25	2.0	2.7	2.3	2.2	1.6
26	3.1	2.5	2.0	1.7	2.0
27	1.9	1.6	1.5	1.0	1.2
28	1.7	2.7	1.2	1.4	0.7
29	2.3	1.4	2.1	1.8	1.5
30	2.4	3.4	2.3	2.1	2.3
31	1.5	1.7	1.4	1.4	1.2
32	2.2	2.3	2.5	1.4	2.1
33	2.7	2.5	2.5	2.0	1.9
34	2.0	1.8	1.9	1.8	2.1
35	2.2	2.9	2.5	2.3	1.8
36	2.2	0.7	0.9	0.5	0.9
37	1.7	1.5	1.8	1.8	1.4
38	0.8	2.4	2.1	1.8	2.0
39	1.6	1.4	0.7	0.7	[a]
40	0.8	1.6	1.7	2.6	1.8
Statistics					
Average	2.0	2.1	1.8	1.6	1.5
SD	0.6	0.7	0.6	0.5	0.7
t-value	15.75	13.9	12.34	13.08	9.357
df	19.0	19.0	19.0	19.0	18.0
P value	<.001	<.001	<.001	<.001	<.001

[a] Unable to calculate due to baseline sample overgrowth (too numerous to count).

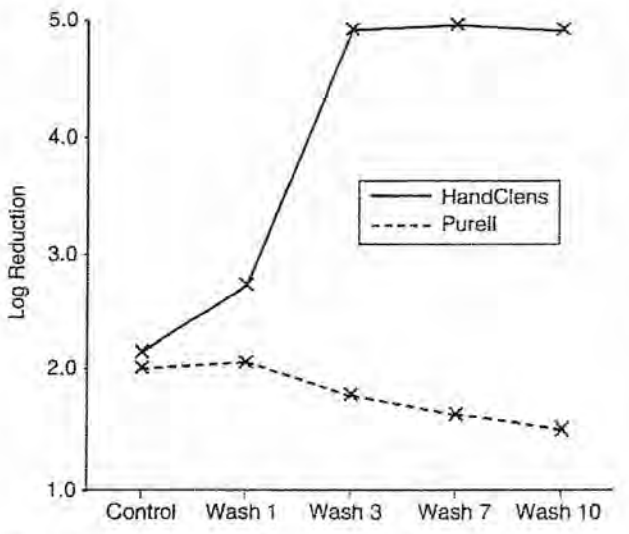

Figure 1. The average log reduction for the HandClens group and the Purell group for control wash using Dove soap and washes 1, 3, 7, and 10.

novel SAB formulation HandClens and compared its efficacy with that of an alcohol-based leading brand, Purell. The novel SAB formulation not only effectively killed microbes after the first wash, but continued to do so at a maximal value. The extent of the reduction factor was limited by the baseline contamination.

The efficacy of HandClens may be attributed to its unique SAB formulation. The combination was hypothesized to complement the natural skin barrier and enhance its performance, whereas alcohol-based formulations cause a deterioration of skin over time and with repetitive use. Clearly, HandClens surpassed the minimum FDA standards for an antiseptic hand sanitizer. Most importantly, the proven efficacy of HandClens will promote its adoption as an adjunctive hand sanitizer for busy podiatric physicians and other health-care professionals. Users of this SAB formulation can be assured of its disinfecting residual power and complementary action on their epidermis. The next concern should be testing the develop-

ment of more SAB products in different vehicles, such as surgical scrubs and hand lotions.

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Evaluating the Effectiveness of Alcohol-based Hand Sanitizers compared to Alcohol-free Hand Sanitizers

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Abstract

Background and Purpose: Hand washing is one of the most important critical control points in public premises in preventing the spread of bacteria and viruses. There is vast research on the effectiveness of alcohol-based hand sanitizers in killing germs. However, the efficacy of alcohol-free hand sanitizers lacks real-world evidence. With little to no guidelines in which one type of hand sanitizers may be more appropriate depending on the types of public premise such as food establishments, hospitals, work place, or schools, Environmental Health Officers(EHOs)/ Public Health Inspectors(PHIs) will need to educate the public and operators on the effectiveness of these hand sanitizers and their advantages and disadvantages. The purpose of the study was to compare the effectiveness of alcohol-based hand sanitizers and alcohol-free hand sanitizers by conducting statistical analyses of the reduction in mean *E.coli* counts.

Methods: 60 pigskins were prepared (30 for alcohol-based hand sanitizers, 30 for alcohol-free hand sanitizers), which were inoculated with *E. coli*, then applied either alcohol-based hand sanitizers or alcohol-free hand sanitizers. After 48 hours of incubation for *E.coli* growth, *E.coli* was counted. The difference in mean *E.coli* counts before applying hand sanitizers and after hand sanitizers was calculated, then compared between the two hand sanitizers.

Results:

The mean *E.coli* reduction count (CFU) from alcohol-based hand sanitizers (30 samples) was 10.200; the median was 11; the standard deviation was 1.7889; the range was 5.0000.

The mean *E.coli* reduction count (CFU) from alcohol-free hand sanitizers (30 samples) was 10.233; the median was 10.5; the standard deviation was 0.8976; the range was 3.0000.

The statistical t-test resulted in p-value of 0.1034.

Conclusion: There was no significant difference between the two types of hand sanitizers. Both the alcohol-based hand sanitizers and alcohol-free hand sanitizers effectively reduced the number of *E.coli* counts (CFU) by averages of 10.2000 (92.7% reduction) and 10.2333 (93.03% reduction) respectively. While the BC Centre for Disease Control recommends 60 percent alcohol hand sanitizers to prevent the spread of germs, this research showed that alcohol-free hand sanitizers with sulfactants, allantoin, and benzalkonium chloride (SAB) formula is just as effective in killing germs. Therefore, EHOs/PHIs can educate the public and operators on the advantages and disadvantages on the two types of hand sanitizers in preventing the spread germs during the flu season and give practical advice or guidance on which type of hand sanitizers would be most appropriate in restaurants for example.

Key words: Alcohol-free hand sanitizers, alcohol-based hand sanitizers, benzalkonium chloride, *E. coli*

Introduction

The simple act of washing your hands correctly could protect you from the spread of disease. According to the British Columbia Centre for Disease Control (BCCDC), eighty percent of common infections are spread by hand, and washing your hands five times a day could drastically decrease the frequency of influenza (flu) and nosocomial infections (BCCDC, 2014). The main pathogens of concern are *Salmonella, Staphlococcus aureus, Streptococi, E.coli, and protozoa* (Fazlar & Ekhtelat, 2012). However, not everyone has frequent access to water and soap, which is why the more convenient hand sanitizers, which some companies claim to kill "99.9% of germs" (Purell, 2015) are increasing in popularity. Even the BCCDC recommends the use of hand sanitizers to supplement hand washing, which will increase its efficacy (BCCDC, 2014).

However, with different types of sanitizers being used in different public premises, the public may not be aware of why one type is preferred over the other, specifically comparing alcohol-based hand rubs (ABHR) and alcohol-free hand sanitizers. There are advantages of using either type of sanitizer but according to the Wall Street Journal, there is insufficient real-world evidence to demonstrate that the alcohol-free hand sanitizer works as well in the real world as it does in laboratory testing (Johannes, 2013).

As a result, it is important to analyze the efficacy of both types of hand sanitizers to determine which is appropriate in different public premises and to educate the public in deciding which type to use during this flu season. This in turn may increase hand washing in general. The purpose of this research will be to determine the effectiveness of alcohol-based hand sanitizers and alcohol-free hand sanitizers by conducting statistical analyses of the reduction in microbes. In particular, the means of *E. coli* counts from using alcohol-based hand sanitizers and from using alcohol-free hand sanitizers will be compared by conducting inferential statistics. There has been extensive research analyzing the effectiveness of alcohol-based hand sanitizers. In particular, the Fraser Health clinical practice guideline concludes that alcohol-based hand rubs (ABHR) of concentration of at least seventy percent inactivates microorganisms and temporarily stops the growth of pathogens (Fraser Health, 2012). However, there is a lack of empirical data of how long the alcohol-free hand sanitizers last on the hands before they become ineffective against bacteria (Johannes, 2013).

Literature Review

The advantages of alcohol-based hand sanitizers and alcohol-free hand sanitizers in different public premises are discussed first below. Depending on the types of public premise such as food establishments, hospitals, work place, or schools, one type of hand sanitizer may be better than the other. Finally, the lack of real-world evidence of the efficacy of alcohol-free hand sanitizers is discussed for comparison.

Hand sanitizers in Food Establishments

Hand washing is one of the most important critical control points in a food premise or establishment in preventing the spread of bacteria and viruses, which ultimately cause foodborne illnesses. However, could any method of hand washing, whether soap and water or alcohol-based or alcohol-free hand sanitizer, be appropriate for food establishment employees? According to the *BC Food Premise Regulation Division 5 Section 21(3)(4)*, each employee must wash his or her hands as often as necessary and the operator of the establishment must supply and maintain adequate number of hand washing stations (FPR, 2008). The *Food Retail and Food Services Code (FRFSC) Section 5 (a)* from the Canadian Food Inspection Systems Implementation Group also gives guidelines on washing hands, vigorously for 20 seconds and then rinsed with clean warm water (FRFSC, 2004). The Food and Drug Administration (FDA) states that hand sanitizers do not replace hand washing with soap and water by food retail workers because hand sanitizers do not reduce fatty and proteinaceous materials that pathogens can survive on (Minnesota Department of Health, 2009). Also, these fatty materials reduce the effectiveness of hand sanitizers and these sanitizers are ineffective against viruses such as norovirus, which can be transmitted from person-to-person (U.S. Food and Drug Administration, 2014). The FDA recommends that hand sanitizers with at least 60 percent alcohol be used after hand washing with soap and water (U.S. Food and Drug Administration, 2014). No further information is available for alcohol-free hand sanitizers and whether these can also be used. All food establishments should therefore have guidelines on hand sanitizers in employee hygiene policy that adhere to the Food Premise Regulations (FPR). It may also be advisable to offer hand sanitizers to customers during the flu season as a further precaution in preventing the spread of germs.

Improved hand hygiene in healthcare settings by using Alcohol-Based Hand Sanitizers

According to the World Health Organization, 1.4 million people around the world acquire infections at hospitals and health-care associated infections (HAI) incur additional 5.7 billion dollars and 90,000 deaths in the United States (World Health Organization, 2007). Therefore, hand hygiene is a fundamental solution to decrease the spread of diseases. Research has shown that multimodal, multidisciplinary strategies that include promoting hand hygiene adherence and alcohol-based hand sanitizer are essential in ensuring patient safety (World Health Organization, 2007). According to the Fraser Health clinical practice guideline, alcohol-based hand rubs (ABHR) of concentration of at least 70 percent inactivates microorganisms and temporarily stops the growth of pathogens and should be available in all areas of the hospital (Fraser Health, 2012). Some of the benefits of using ABHR are quicker application, no need for soap or water, more readily available, and effective in reducing microorganisms on hands (Fraser Health, 2012). Research done by Hilburn et al. studied the efficacy of alcohol-based hand sanitizer in an acute-care facility and determined that the primary infection types were urinary

tract and surgical site infection (Hilburn J, 2003). The use of hand sanitizer resulted in 36.1% decrease in infection rates for the 10-month period of the study and recommends its use ABHR in acute care facilities and hospitals (Hilburn J, 2003).

Effectiveness of Alcohol-free hand Antiseptic Hand Wash among podiatric physicians and healthcare personnel

Healthcare workers are at a greater risk of contacting dermatitis due to constant hand washing and glove changing (Moadab A, 2001). Alcohol-based hand sanitizers can irritate the skin with cuts and chops, dry the skin with overuse, and cause dermatologic changes. As a result, alcohol-free hand sanitizers that contain sulfactants, allantoin, and benzalkonium chloride (SAB) have been studied for their efficacy in immediate and residual disinfecting power (Reichel, 2014). The study followed the Food And Drug Administration (FDA) protocol in evaluating the two solutions and determined that the SAB formula not only killed the microbes after the first wash, but also maintained its residual disinfectant power after ten washes (Moadab A, 2001). This surpassed the minimum FDA standard for antiseptic hand sanitizer and showed that SAB sanitizers are appropriate for podiatric physicians and healthcare personnel (Moadab A, 2001). However, the limitation of this study is that it did not test SAB products in different vehicles as surgical scrubs (Moadab A, 2001) and therefore is not appropriate for hospitals.

Effectiveness of Alcohol-free hand sanitizers in Schools

The use of hand sanitizers should be emphasized in schools where children are in close proximity to each other and there are many factors that could pose increased spread of diseases. These factors include many objects as vehicles of transmission, lack of hand washing facilities, inadequate time requirement for proper hand washing, and education (White CG, 2001). ABHR can be poisonous if children ingest them, can irritate skin with cuts and eyes, and are flammable, which make them hazardous at a school setting (White CG, 2001). As such, alcohol-free hand sanitizers may be a good option. The controlled study that used elementary school students' absenteeism as an indication of effectiveness of alcohol-free hand sanitizer showed that in conjunction with at will hand washing with soap and water, alcohol-free hand sanitizers were just as effective as alcohol-based sanitizers and reduced absenteeism by 31% (White CG, 2001). At school settings, especially among small children who are likely to become ill four or more times a year, hand hygiene is a crucial practice in and outside of school. With the benefits of alcohol-free hand sanitizers, it should be recommended that children use this type of hand sanitizer compared to alcohol-based hand sanitizers.

Use of alcohol-based hand sanitizers in open, nonclinical workplace setting

Acute infectious respiratory and gastrointestinal diseases are among the most common diseases in schools, universities, and workplace settings (Hübner NO, 2010). The crowdedness of the working space, the number of close person-to-person interactions, and constant sharing of public space make transmission of diseases easy. As a result, productivity is greatly diminished due to absenteeism from work (Hübner NO, 2010). According to a prospective, controlled study that followed a cohort for 1230 person months and recorded the use of hand disinfectant, there was a reduced number of illnesses for the majority of gastrointestinal symptoms when using alcohol-based hand sanitizers, thus a reduced absenteeism (Hübner NO, 2010). The use of hand sanitizers, whether alcohol-based or non-alcohol based, should be part of all workers' hand hygiene and company health support programs.

Public Health Significance

No matter which type of public premise, hand washing has been proven to prevent the transmission of diseases between people. Most Health Authorities and Centers for Disease Control recommend using hand sanitizers to fight nosocomial infections and pathogens such as *Salmonella, Staphlococcus aureus, Streptococi,* and *E.coli.*

After reviewing various research studies that examined alcohol-based hand sanitizers and non-alcohol hand sanitizers, there still needs objective empirical evidence of the efficacy of non-alcohol hand sanitizers compared to alcohol-based hand sanitizers. Extensive research shows that alcohol-based hand sanitizers at 70% or higher effectively kills pathogens and reduce infection rates at hospitals (Hilburn J, 2003). However, it may not be suitable for children at elementary schools due to some of the hazards of alcohol-based hand sanitizers such as being

poisonous if ingested, flammable, and irritation to the cut skins (White CG, 2001).

Hand sanitizers are a great supplement to hand hygiene practice and people should be educated in the different types of hand sanitizers, their advantages and disadvantages, and their efficacy in different public premises such as food establishments, schools, hospitals, and workplaces. Although non-alcohol based hand sanitizers seem favorable due their effectiveness, less irritation to the skin, and non-flammable properties, there is still a lack of empirical evidence that shows its effectiveness compared to the vastly researched alcohol-based hand sanitizers. It is still a good idea to be aware of the different types of hand sanitizers.

Methods and Materials

The experiment involved preparation of the *E. coli* dilution (10^{-6}), preparation of 60 pigskins (30 for alcohol-based hand sanitizer, 30 for alcohol-free hand sanitizer), the inoculation of *E. coli* on the pigskin pieces before applying hand sanitizers, and applying the hand sanitizers on the pigskin pieces. The experiment was conducted in the Food Microbiology Laboratory at BCIT Burnaby Campus, under the supervision of Helen Heacock (Environmental Health Program Instructor, BCIT), with guidance and feedback from Melinda Lee (Technical Staff II, BCIT) and Ken Keilbart (Assistant Instructor, BCIT), and with the approval for lab use from Erin Friesen (Food Program Head, BCIT). The procedures were taken from Sophia Yip but altered for the purpose of this research experiment (Yip, 2003)

Description of Materials

***One Step Hand Sanitizer* (236ml, 62% ethyl alcohol)** meets the recommended alcohol concentration by Fraser Health to inactivate the microbes. It is inexpensive at $4.72 CAD, and is widely available at retail stores such as *Walmart*.

***X3 Clean Foaming Hand Sanitizer* (Benzalkonium chloride 0.13%)** contains benzalkonium chloride, a quaternary ammonium compound that is used widely as antiseptic agents due to their cationic amphiphilic property and destabilizing the pathogen's surface (Campanac, Pineau, Payard, Baziard-Mouysset, Baziard-Mouysset, & Roques, 2002). Also, it is inexpensive at $5.79 CAD, and is widely available at retail stores such as *Walmart*.

E. coli is mostly harmless bacteria found in the intestines of humans and animals but some strains such as *E. coli 0157:H7* can cause severe abdominal pain, diarrhea and vomiting (Public Health Agency of Canada, 2015). It makes up 97% of fecal materials in human excretion and is used as an indicator for fecal contamination and unsanitary practices. It can be detected by the enumeration method by using lactose fermentation (U.S. Food and Drug Administration, 2002). *E. coli* culture is available at the BCIT microbiology lab (Food Microbiology Laboratory at BCIT) or can be purchased online.

Pigskin is anatomically similar to human skin in terms of color, hair follicles, sweat glands, and subcutaneous fat (Herron, 2009). Also, since pigs are considered food source, it is widely accepted by the public for its use as laboratory animals. It is also readily available at butcher shops and is relatively inexpensive. Thus, it is frequently used as a model of human skin (Herron, 2009).

***E. coli* 3M Petrifilms**

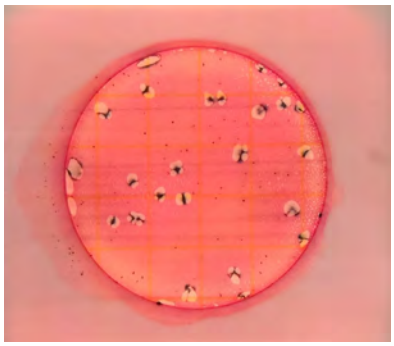
3M Quickswabs

Preparation of *E. coli* culture: *E. coli* culture was obtained from the Food Microbiology Laboratory at BCIT. *E. coli* was transferred into nutrient broths so that it can be diluted to 10^{-6} (Yip, 2003).

Negative Control: This was done to verify the swabs and Petrifilm were sterile by pouring an unused Quickswab onto the Petrifilm.

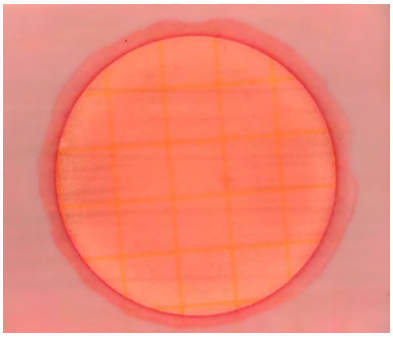
Preparation of Controls: Thoroughly washed the pigskins with tap water to ensure there was no dirt or other debris. Cut into five 10cm x 5cm pieces. Inoculate the pigskin pieces with *E. coli* by swabbing. Incubated all five petrifilm plates for 48 hours at 35C. This was the counted *before data* (Colony Forming Unit). Confirmed *E.coli* coliforms were blue colonies with associated gas bubbles (3M Corporation, 2015) as seen in figures 1.

Figure 1: Blue colonies are the confirmed *E.coli* counts used for *Before Data* (CFU)



Preparation of Sixty pigskin pieces for hand sanitizers application: After inoculating the pigskin pieces *with E.coli,* two types of hand sanitizers were applied for 30 samples each. The pigskins were incubated for 48 hours at 35C. This was the counted *after data* (Colony Forming Unit). Confirmed *E.coli* coliforms were blue colonies with associated gas bubbles (3M Corporation, 2015) as seen in figures 2.

Figure 2: After applying hand sanitizers, most of the blue colonies were eliminated.



Inclusion and Exclusion criteria

The alcohol-based hand sanitizers used in this experiment was *One Step Hand Sanitizer* (236ml, 62% ethyl alcohol) and the alcohol-free hand sanitizer was *X3 Clean Foaming Hand Sanitizer*. The specific breed of pigskin used was Yorkshire pig. The exclusions were any other types or brands of hand sanitizers on the market.

Results

The hypotheses generated are as follows:

H_o: The reduction mean *E. coli* counts on pigskin that is cleaned with alcohol-based hand sanitizers is the **same as or less than** the reduction mean *E. coli* counts on pigskin that is cleaned with alcohol-free hand sanitizers. $\bar{y}_{Alcohol-based} \leq \bar{y}_{Alcohol-free}$

H_a: The reduction mean *E. coli* counts on pigskin that is cleaned with alcohol-based hand sanitizers is **greater** than the reduction mean *E. coli* counts on pigskin that is cleaned with alcohol-free hand sanitizers. $\bar{y}_{Alcohol-based} > \bar{y}_{Alcohol-free}$

Inferential Statistics

The hypothesis test that was used by SAS and Microsoft Excel was one-tailed two-sample t-test. The two-sample t-test was able to compare the two mean reductions in *E. coli* counts and assessed which group was more effective in reducing the *E. coli* counts. After calculating the p-value, if the p-value is less than 0.05, we reject the null hypothesis and conclude that there is statistical significant difference between the two groups. If the p-value was greater than 0.05, we fail to reject the null hypothesis and conclude there is not statistically significant difference between the two groups.

Description of Data

The data for this research was both nominal and numerical data. The type of hand sanitizers (*One Step Hand sanitizer* or *X3 Clean Foaming Hand Sanitizer*) is the nominal data. The *E. coli* count in CFU is the numerical (discrete) data. The mean reduction in *E. coli* count after applying alcohol-based hand sanitizer ($\bar{y}_{DA}$) and that of alcohol-free hand sanitizer ($\bar{y}_{DF}$) was compared to determine if there was significant evidence that the reduction mean *E. coli* counts on pigskin that was cleaned with alcohol-based hand sanitizers was greater than the reduction mean *E. coli* counts on pigskin that was cleaned with alcohol-free hand sanitizers. The mode and the median were not used in this research for statistical analysis, although they may aid in determining how effective hand sanitizers were in reducing the pathogen. The ranges of the mean reduction *E. coli* counts allowed for the determining of the extremes. The standard deviation, which showed how data is spread about the mean, also showed the variation in mean reductions. Table 1 summarizes the descriptive statistics. Table 2 shows the mean reduction *E.coli* counts.

Table 1: Descriptive Statistics

	Variable 0 (Alcohol-based hand sanitizer) (CFU)	Variable 1 (Alcohol-free hand sanitizer) (CFU)
Mean ($\bar{y}$)	10.2000	10.2333
Median	11	10.5
Standard Deviation	1.7889	0.8976
Variance	3.2002	0.8057
Samples	30	30
Minimum	3.0000	8.0000
Maximum	11.0000	11.0000
Range	5.0000	3.0000

Table 2: Mean reduction of *E. coli* count (CFU) (before and after using *One Step Hand*

Sanitizer sanitizer $(\overline{y}_{DA})$ **and** ***X3 Clean Foaming Hand Sanitizer*** $(\overline{y}_{DF})$**)**

Sample	**Difference (CFU)** $\overline{y}_{DA}$	**Difference (CFU)** $\overline{y}_{DF}$
1	**11**	**9**
2	**11**	**9**
3	**11**	**10**
4	**10**	**11**
5	**11**	**10**
6	**11**	**11**
7	**11**	**9**
8	**10**	**10**
9	**11**	**8**
10	**11**	**11**
11	**10**	**10**
12	**11**	**9**
13	**11**	**10**
14	**10**	**11**
15	**10**	**9**
16	**11**	**11**
17	**11**	**10**
18	**11**	**10**
19	**11**	**11**
20	**11**	**11**
21	**11**	**11**
22	**10**	**11**
23	**3**	**11**
24	**6**	**11**
25	**11**	**11**
26	**7**	**11**
27	**10**	**11**
28	**11**	**9**
29	**11**	**11**
30	**11**	**10**

Interpretation

Using SAS software (SAS University Edition, 2015), the normality was tested for the data, which showed p-values all less than 0.05, confirming that the data is not normally distributed, thus a nonparametric test was performed (the Wilcoxon rank sum test). From the Wilcoxon rank sum test, the p-value was 0.1034, which was greater than 0.05, thus we failed to reject the null hypothesis and concluded that there was no significant difference between the two types of hand sanitizers.

Discussion

Both the alcohol-based hand sanitizers and alcohol-free hand sanitizers effectively reduced the number of *E.coli* counts (CFU) by averages of 10.2000 (92.7% reduction) and 10.2333 (93.03% reduction) respectively. There have been many research studies to show the effectiveness of alcohol-based hand sanitizers and alcohol-free hand sanitizers such as the separate studies done by Hilburn et al., Hübner NO et al., Reichel et al, and White CG et al. as mentioned earlier in the literature review. However, none of the studies compared the two types of hand sanitizers and which one has greater effectiveness in reducing *E.coli* counts. Some of these studies used indirect correlation between absenteeism among children at schools (White CG, 2001) and workers at jobs (Hübner NO, 2010) and the effectiveness of hand sanitizers to determine if hand sanitizers significantly contributed to preventing communicable pathogens and thus reducing absenteeism. This research confirmed that each type of hand sanitizers effectively reduced *E.coli* counts through microbiological lab experiment, although the Food and Drug Administration (FDA) protocol for testing sulfactants, allantoin, and benzalkonium chloride (SAB) formula-based hand sanitizers may be more stringent (Moadab A, 2001).

Furthermore, this research compared the two types with statistical analysis, which showed no difference in their effectiveness. While the BC Centre for Disease Control measure (BCCDC, 2014), Fraser Health Authority (Fraser Health, 2012), and the FDA recommend hand sanitizers with at least 60 percent alcohol to supplement hand washing with soap and water at different public premises such as restaurants, hospitals, and schools (U.S. Food and Drug Administration, 2014), this research, using statistical analysis derived from microbiological testing, showed that alcohol-free hand sanitizers can be just as effective as 60 percent alcohol-based hand sanitizers. However, this research did not test for the residual effect of the hand sanitizers, which should be considered to determine which one is more effective in reducing communicable pathogens in the real world and testing on human hands with regular activities such as shaking hands, touching door nobs, and using the computer.

Recommendations

We failed to reject the null hypothesis and concluded that there is no significant difference between the two types of hand sanitizers. There is some practical significance with respect to the field of public health. As discussed in the literature review, with different types of hand sanitizers being used in different public premises, the public may not be aware of why one type is preferred over the other, specifically comparing alcohol-based hand rubs (ABHR) and alcohol-free hand sanitizers. This research study showed, with microbiological evidence, that the effects of hand sanitizers are the same for both alcohol based and alcohol free. For public premises such as food establishments, all

employees are required to wash hands with soap and hot water (FPR, 2008). Whether they use alcohol-based or alcohol-free hand sanitizers is the operators' choice, factoring in the fact that alcohol-free hand sanitizers are slightly more expensive but have other benefits such as no irritation to the skin. The practical significance would be to use the cheaper sanitizers, as it is more cost effective for operators. Neither the *Food Premise Regulation (FPR)* nor the *Food Retail and Food Services Code (FRFSC)* used by health inspectors mentions the effectiveness of hand sanitizers whether alcohol-based or alcohol-free (Canadian Food Inspection System, 2004); there is potential for educating the food establishment operators in the FPR or the FRFSC of the effectiveness of the different types of hand sanitizers and to offer hand sanitizers to customers during the flu season as a further precaution in preventing the spread of germs.

In regards to hand sanitizers at schools, it may be recommended that schools use alcohol-free hand sanitizers since children are more sensitive to skin irritation, have weaker immune systems, and can contract sickness up to four times a year (White CG, 2001). Such benefits of alcohol-free hand sanitizers include less irritation to the skin, non-flammable properties, and not being poisonous to children. The practical significance is educating the children and staff of the proper hand hygiene practice in and outside of school with soap and water, while supplementing the practice with alcohol-free hand sanitizers. Although non-alcohol based hand sanitizers seem favorable due their same effectiveness as alcohol-based sanitizers, there is still a lack of empirical evidence that shows its effectiveness compared to the vastly researched alcohol-based hand sanitizers. It is still a good idea to be aware of the different types of hand sanitizers.

Limitations

Improvements to Study: The validity of the experiment depended heavily on the experimenter's lab techniques and skills. Due to the cost issue, two experimenters with differing microbiological lab experience and techniques performed the experiment and shared the data for alcohol-based hand sanitizers, thus leading to potential human errors. To improve the experiment even further, one experimenter could have performed the entire experiment.

Possible Errors or Bias: Human errors cannot be fully eliminated as this experiment relies heavily on the accuracy of the experimenter (both performing the experiment and counting the *E. coli* colony counts), but can be minimized by having an experimenter who is well trained on laboratory techniques and instruments.

There was no Type I error since the difference was not significant but Type II error could be decreased by increasing the number of samples. However, Type II error occurs when the p-value obtained is only slightly greater than 0.05 (ex. between 0.05 and 0.1). Thus, a p-value as high as 0.1034 suggested that there was simply no difference between the two types of hand sanitizers, regardless of the size.

Due to the vastly researched effectiveness of alcohol-based hand sanitizers such as the studies done by Hübner NO et al. and Hilburn J et al., as well as the recommendation of using 60 percent alcohol-based hand sanitizers by BCCDC, the Fraser Health Authority, and the FDA, there was initial bias towards the alcohol-based hand sanitizers. However, both types of hand sanitizers were applied on the pigskins in the same method and analyzed using the same materials, thus minimizing any bias.

Future Research

This research compared the two types of hand sanitizers by analyzing the mean reduction in *E.coli* counts (CFU). However, it did not measure the residual effect of alcohol-based and alcohol-free hand sanitizers, which is an important property in the real world. The residual effect of hand sanitizers is how long it kills germs after the initial application. For future research, this residual effect, particularly of alcohol-free hand sanitizers, could be studied to show how effective alcohol-free hand sanitizers are in terms of having long or short residual effect. Having long residual effect could be more effective in preventing communicable diseases such as *Salmonella, Staphlococcus aureus, Streptococi, E.coli,* and *protozoa* as compared to short residual effect. There are many studies that show the residual effect on alcohol-based hand sanitizers such as the one by Hilburn et al. but more studies should be conducted using alcohol-free hand sanitizers. Indeed, there is a limited real-world evidence of the efficacy of non-alcohol based hand sanitizers outside the clinical studies before they become ineffective against bacteria (Johannes, 2013).

Conclusion

The p-value was 0.1034, which was greater than 0.05, thus we failed to reject the null hypothesis and concluded that the reduction

mean *E. coli* counts on pigskin that was cleaned with alcohol-based hand sanitizers is the same as or less than the reduction mean *E. coli* counts on pigskin that was cleaned with alcohol-free hand sanitizers. In other words, there is no difference between the types of hand sanitizers.

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Competing Interest

The authors declare that they have no competing interests.

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Testing a New Alcohol-Free Hand Sanitizer to Combat Infection

Following universal precautions is an integral part of OR staff members' responsibilities in perioperative patient care. The precautions mandate routine hand washing with soap and water before and after all patient contact, and especially before invasive procedures. Although intended to reduce the postoperative risk of infection of healing incisions and wounds, universal precautions, including hand washing, are followed only 55% of the time in the nation's hospitals.[1]

The contributing factors behind this insufficient hand washing are manifold; however, identified principal causes include the following.

- Direct patient caregivers are handling an excessive patient load. Conservative estimates indicate that physicians attend to 20 to 30 patients per day, and other health care personnel (eg, nurses, physical therapists, respiratory therapists) may have as many as 200 patient contacts per day.
- The repeated hand washing required for that volume of patients causes dryness and subsequent microabrasions of the skin.[2]
- The skin on the hands has a short period of time to recover between washings.

These factors, among others, have led to a great increase in the use of rinse-free instant hand sanitizers as a supplement to proper hand washing with soap and water.

The most widely used hand sanitizers are gels and foams that rely on alcohol as the main antimicrobial ingredient. Alcohol, however, solubilizes and strips away sebum and lipids that guard against bacterial infections of the skin.[3] Extensive use of alcohol-containing hand sanitizers actually increases the skin's susceptibility to infection by transient disease-causing bacteria. This situation can increase the chances of spreading disease-causing microorganisms among patients.

The threat of spreading disease could be avoided by using alcohol-free hand sanitizers that complement, rather than compromise, the natural barrier functions of the skin. An acceptable alcohol-free formula would require an antimicrobial agent that kills a wide variety of disease-causing microorganisms, including gram-positive and gram-negative bacteria, fungus, and molds. This formula also would need to allow the active ingredient to penetrate the skin while minimizing skin irritation.

A B S T R A C T

Universal precautions require that perioperative health care personnel wash their hands before and after all patient contact. Time constraints, however, can make adhering to universal precautions, including proper hand washing, difficult. Some perioperative health care workers, therefore, routinely use rinse-free hand sanitizers to supplement normal hand washing. This study evaluated immediate and persistent antimicrobial effectiveness of two alcohol-containing hand sanitizers and a novel surfactant, allantoin, benzalkonium chloride (SAB) hand sanitizer using a federally approved effectiveness protocol. Results indicate that all three products were equally effective after a single application. After repeated use, the alcohol-containing sanitizers did not meet federal performance standards, and the alcohol-free sanitizer did. These properties and others illustrated in this article indicate that the nonflammable, alcohol-free SAB hand sanitizer is the most favorable of the rinse-free hand sanitizer formulas for normal hand washing. *AORN J* **68 (August 1998) 239-251.**

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Such a formula is obtained by combining certain surfactants and allantoin with the widely used antimicrobial agent, benzalkonium chloride. This formula is known as a surfactant, allantoin, and benzalkonium chloride (SAB) sanitizer.

ESTABLISHING THE HYPOTHESIS

Based on this information, researchers at Woodward Laboratories, Inc, Los Alamitos, Calif, hypothesized that the prolonged use of alcohol-containing hand sanitizers would be less effective at degerming the skin than an alcohol-free SAB sanitizer. To test this, they used a protocol validated by the US Food and Drug Administration (FDA) for the performance of health care personnel antiseptic hand washes.[4] This protocol was developed specifically to test the degerming effectiveness of handwash preparations with extended use and is accepted as a national clinical standard for such performance testing.

LITERATURE REVIEW

The literature review for this study indicated that rinse-free hand sanitizers are, by definition, intended for degerming skin without the aid of rinsing with water. This type of product has steadily gained popularity in professional circles as a supplement to hand washing with soap and water. The types of rinse-free hand sanitizers generally are grouped into two broad categories:

- alcohol-based products and
- alcohol-free products.

The need for immediate and persistent protection. The FDA clearly seeks both an immediate and persistent degerming activity in antiseptic preparation by its definition of a personnel disinfectant:

> *a non-irritating, antimicrobial-containing preparation designed for frequent use and which will reduce the number of transient microorganisms to a baseline level after adequate washing, rinsing, and drying. Such preparations also are expected to have a broad antimicrobial spectrum, be fast-acting, and persistent.*[5]

A hand sterilizer's immediate antimicrobial effectiveness is based on the physical removal and immediate inactivation (ie, within 60 to 180 seconds of exposure to the antimicrobial agent) of microorganisms residing on the hands. The persistent antimicrobial effectiveness of a hand sanitizer is defined as its microbiocidal activity after up to six hours of the product's application.[6]

Alcohol-based products. These products vary greatly in composition, ranging from 54% isopropanol to 70% ethanol.[7] The choice of this type of rinse-free antimicrobial product often is subjective and mainly based on factors such as cost, presence of emollients in the formula, fragrances, delivery vehicle (eg, gel, foam), size, and marketing. Selection is less often based on the product's effectiveness at eliminating bacteria after a single application.

Although alcohol-based formulas that comply with federal composition standards generally are considered effective, alcohol-based antiseptic handwash preparations are flammable and do not demonstrate persistent antimicrobial activity. Also, repeated use often can cause drying and irritation of the skin.[8] Alcohol strips the skin of essential oils and sebum, which act as a natural protective barrier against bacterial infection and precipitate protein.[9] When applied to wounds or raw surfaces, therefore, it not only increases the risk of injury, but also forms a coagulum under which bacteria may subsequently thrive.[10] It is, therefore, not useful for the disinfection of open lesions or abraded, inflamed skin. Together, these and other adverse properties greatly limit the alcohol-based antimicrobial product's immediate effectiveness and increase the chances for the spread of infection.

Chlorhexidine and hexachlorophane. The persistent antimicrobial activity sought by the FDA has been demonstrated by using the alcohol-free compounds of chlorhexidine and hexachlorophane with a water rinse.[11] These compounds, however, have not been extensively used in rinse-free hand antiseptic application, in part because they are neither absorbed nor dissipated quickly enough to be convenient or user-friendly, and in part because they have aesthetically displeasing side effects such as odor. Additional limitations include a relatively narrow antimicrobial spectrum of certain compounds, such as triclosan.[12]

Benzalkonium chloride. Benzalkonium chloride (BAC) is an alcohol-free, antimicrobial compound that has been widely used in the health care industry for more than 60 years in formulas for preservatives, surface cleansers, sterilizing agents, and topical antiseptic sprays.[13] The chemical properties of BAC

make it a good candidate for persistent antimicrobial activity in mammalian tissue. Extensive exposure to certain nonalcohol antimicrobial agents, including some surfactants, however, can make it have a detrimental effect on the skin unless the active ingredient is formulated with compounds that mitigate this effect.

A unique balance of penetration and nonirritation is attained when BAC is combined with surfactants and allantoin. This type of alcohol-free sanitizer formula is absorbed rapidly into the skin with little impact on the skin's natural barrier function and is predicted to be more useful and effective as a rinse-free hand sanitizer than alcohol-containing formulas.

THEORETICAL AND CONCEPTUAL FRAMEWORK

Purpose of the study. The goal of the study was to provide information about the effectiveness of rinse-free hand sanitizers when used as a supplement to normal hand washing. The study was designed to evaluate the immediate and persistent antimicrobial properties of two types of alcohol-containing, rinse-free hand sanitizers (ie, 62% ethanol, 70% ethanol) and an alcohol-free SAB hand sanitizer (ie, 0.133% BAC, 0.5% allantoin).

Study design. An FDA-mandated protocol was used to measure the effectiveness of sanitizer products on hands that have been heavily contaminated with *Serratia marcescens* bacteria, a pathogen common in hospital-acquired infections. The test is useful for identifying formulas that are effective, first-line defenses against massive personal contamination. The FDA protocol recommends a water rinse; however, the formulas were intended for use without a water rinse. Antimicrobial performance thus was determined both with a water rinse and without a water rinse in separate sets of experiments.

The bacteria *Serratia marcescens* used in this study grows in red-colored colonies, allowing researchers to track only the fate of bacteria introduced on the hands for the purposes of the test. Before testing, all *Serratia* stocks were found to be susceptible to gentamicin, according to National Center of Clinical Laboratory Standards.[14] The experiments were conducted in an environmentally controlled clinical research laboratory, and data was gathered from February to September 1997.

Test solutions. The antimicrobial hand-wash preparations were two commercially available alcohol-based formulas and one alcohol-free SAB formula (Table 1). The nonantimicrobial control hand-wash formula that was used for the initial baseline wash—establishing the mechanical reduction of bacteria—was the commercially available Ivory hand cleanser. Although Ivory soap was used as a representative of nonantimicrobial hand cleansers, other nonantimicrobial cleansers would have served as an adequate control because the principal degerming action of any cleanser that lacks an antimicrobially active ingredient occurs through a mechanical removal of bacteria and not by a direct impact on bacterial viability.

Table 1

HAND SANITIZERS TESTED

Alcohol-based hand washes

Solution 1. Active ingredient: Ethyl alcohol (ie, 62% vol/vol). Other ingredients: Isopropyl alcohol, water, emollients, and thickener.

Solution 2. Active ingredient: Ethyl alcohol (ie, 70% vol/vol). Other ingredients: Emulsifying wax, methyl gluceth 20, polyoxyethylene, stearyl ether, and cylcomethicone.

SAB hand wash

Solution 3. Active ingredient: Benzalkonium chloride (ie, 0.13% vol/vol). Other ingredients: Water, hydroxypropylmethyl cellulose, propylene glycol, cocamidopropyl betaine, cocamidopropylamine oxide, cetyl, trimethyl ammonium chloride, quaternium-12, imidazolidinyl urea, quaternium-15, allantoin, methyl paraben, propyl paraben, eucalyptol, methyl salicylate, and triethanolamine.

Control soap

Ivory hand cleanser. Ingredients: Water, sodium laureth sulfate, sodium lauryl sulfate, cocamidopropyl betaine, and fragrance.

Table 2

DISTRIBUTION OF SUBJECTS AND CONDITIONS

Formula	Condition	Number of subjects
SAB solution	with rinse	21
	without rinse	14
62% ethanol	with rinse	17
	without rinse	16
70% ethanol	with rinse	5
	without rinse	5

Table 3

HAND SANITIZER EFFECTIVENESS PROTOCOL

1. Cultures of *Serratia marcescens* (ie, easy-to-count, red-colored bacteria) are prepared to a concentration of approximately 10^8 bacteria per milliliter of inoculum, and effectiveness is established through a series of contamination and washing cycles.

2. Subjects wash their hands using the control soap.

3. A baseline is determined by inoculation of the subjects' hands followed by immediate sampling using the glove juice method, which is used for each appropriate contamination and wash cycle.

4. A control value for the mechanical degerming activity is sent through a contamination and wash cycle using the control soap.

5. The test subjects proceed through a series of 10 wash cycles with the test solution. Ten minutes pass between each contamination and wash cycle, and the entire series is accomplished in approximately two hours.

6. Glove juice samples to establish antiseptic effectiveness are taken after the first, third, seventh, and 10th contamination and wash cycles, as required by the US Food and Drug Administration.

7. A similar procedure is used for both the rinse and nonrinse protocols.

Subject recruitment and exclusion criteria. In all, 78 healthy adults participated voluntarily in the study and were broken down into test groups for the three formulas (Table 2). The total group comprised 56% men and 44% women, ranging in age from 18 to 47 years. None of the subjects had clinically evident dermatoses or injuries to their hands or had used topical or systemic antimicrobial agents or any other medication known to affect the microbial flora of the skin.

In addition, study participants were required to have a nail length of no greater than 2 mm and were not allowed to wear artificial nails. Initial work had indicated that long or artificial nails sheltered bacteria from the action of the hand sanitizers and significantly skewed results. Similarly, people with nonremovable adornments (eg, rings that could not be removed, bandages) were not allowed to participate in the study because these physical barriers protect bacteria from antimicrobial compounds.

Data collection. The study began with a one-week pretest conditioning period during which subjects were not allowed to use medicated soaps, strong acids or bases, and other antimicrobial products. The antimicrobial effectiveness of the hand sanitizers was judged by a series of hand contaminations that were followed by washes with either a control, nonantimicrobial soap or the test formulas (Table 3).

Glove juice sampling. Researchers used the FDA-approved glove juice sampling technique for bacteria collection.

1) Subjects removed all jewelry and adornments from hands.
2) Five mL of *Serratia marcescens* inoculum were spread over subjects' hands for 45 seconds.
3) Hands were allowed to air dry for two minutes.
4) Polyethylene gloves containing 50 mL of collection fluid each were placed on the subjects' hands and secured above the wrists with rubber bands.
5) Collection fluid was spread over the subjects' hands and massaged for one minute in a standardized manner to ensure uniform recovery of the collection sample.
6) Collection samples from the hands were pooled and immediately plated onto tripticase soy agar (TSA) mediums with both neat samples and serial dilution cultures to guarantee accurate colony counts.

The procedure allowed for a complete sampling of the surface area of each hand below the wrist.

Testing. After the pretest conditioning week, subjects' hands were contaminated as described and then sampled. The number of bacteria recovered from the unwashed hands represented the baseline, which was representative of the maximum bacterial contamination that the unprotected skin could retain. After this, hands were recontaminated and then washed with 5 grams of a nonantimicrobial soap as a control for mechanical degerming action alone. The bacteria remaining on the hands were sampled and plated. After this, subjects' hands were contaminated, washed with 5 grams of the appropriate test sanitizer, and sampled. This last step (ie, contaminate, wash, sample) was repeated 10 times, with five minutes elapsing before the start of the next contamination, wash, and sample cycle. This resulted in a 10-minute recovery period between the subjects' actual washing with the test soap.

In the second series of tests, the 30-second rinse step was omitted, and sampling was performed immediately after washing to test the sanitizers' effectiveness without a water rinse. In each type of test (ie, with or without a water rinse), bacteria remaining on the hands was sampled and plated after the first, third, seventh, and 10th washes.

Collecting cultures. Cultures of FDA-mandated *Serratia marcescens* were prepared according to the method stated in the FDA protocol. Stock bacteria were grown to a concentration of approximately 1 x 10^8 viable bacteria per milliliter of growth medium (ie, Trypsin soy broth). Cultures were agitated before use. No neutralizers were used in the collection fluid; this prevented the buildup in the subjects' skin of neutralizer that would skew the results. The collection fluid had a pH of 7.8 and consisted of

- 0.04% KH_2PO_4,
- 1.0% K_2HPO_4, and
- 0.1% Triton X-100.

Within three minutes of acquisition, samples from both the alcohol-based and SAB antiseptic hand sanitizers were diluted using the collection fluid that contained the appropriate neutralizers and were plated for growth on the TSA medium.[15] Cultures were grown overnight at 37° C (98.6° F) before counting. Washing and rinsing, when applicable, were conducted under running tap water that contained less than one viable bacterium per milliliter.

STATISTICAL ANALYSES AND CALCULATIONS

Statistical analyses were conducted using the Student's *t* test with the aid of Statview statistical analysis software. Data presented in this document represent the mean and the standard error of the mean for the number of subjects in each test group.

All of the raw data (ie, bacterial colony counts) were converted to a $\log_{10}$ scale to be compatible with the calculation model used. Briefly, the $\log_{10}$ scale deals with exponents such that the $\log_{10}$ of 100 (ie, 10^2) is 2. For example, if 1,000 bacteria are counted, that number could be expressed in a power

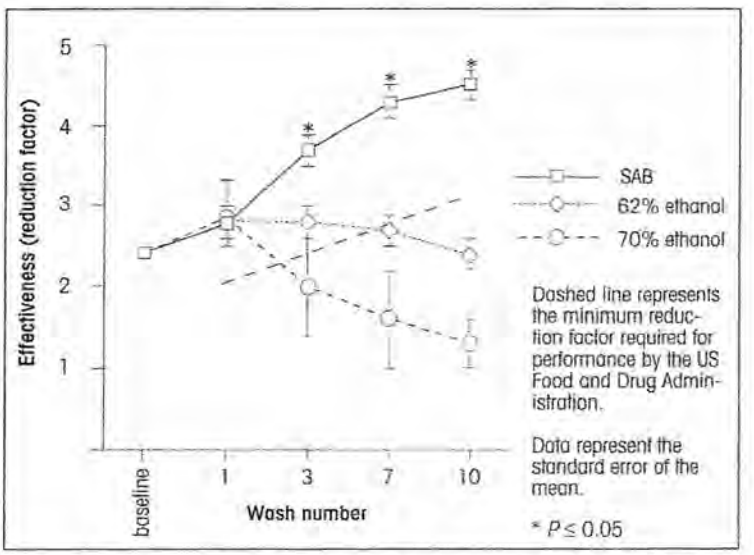

Figure 1 • Hand sanitizer effectiveness performance with a water rinse.

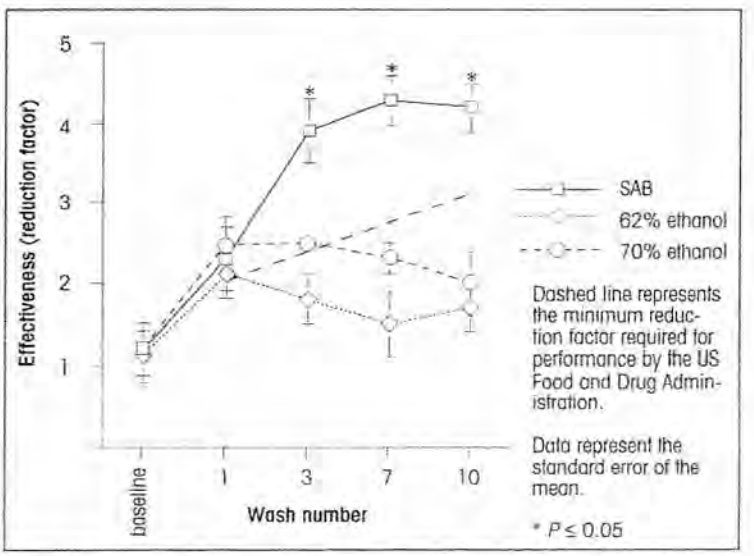

Figure 2 • Hand sanitizer effectiveness performance without a water rinse.

The SAB hand sanitizer's degerming effectiveness increased throughout the hand contamination protocol.

of 10, as 1 x 10^3; thus, the $\log_{10}$ value of 1,000 is 3. This method of converting bacteria counts to log values effectively reduces the statistical variations in bacteria counts from person to person and is useful in comparative studies.

In these experiments, researchers calculated a value called a reduction factor (RF), which is one way to measure how well a test solution decreases the amount of bacteria on subjects' hands. It is calculated as

$$RF = \log_{10}(\text{baseline bacterial count}) - \log_{10}(\text{postwash bacterial count})$$

If 10,000 bacteria, therefore, were recovered from the hands for the baseline, and only 100 were recovered after the wash with the test solution, the RF value would be 2. Another way to look at RF is if the RF is 2, then 99% of all bacteria have been killed; if the RF is 3, 99.9% have been killed, and so on.

A small value for RF means that there was only a small reduction in the number of bacteria on the hands after washing with the test formula. Most nonantimicrobial soaps and sanitizers will give an RF of approximately 2 in this type of test. In contrast, a large value for RF means that there was a large reduction in the number of bacteria on the hands. The FDA-approved protocol used for this study requires a minimum RF of 2 after the first hand wash, and a minimum RF of 3 after the 10th hand wash.

RESULTS

The first series of experiments was performed to compare the effectiveness of the SAB hand sanitizer formula to a commercially available ethanol-based formula (ie, 62% vol/vol) with the inclusion of a 30-second water rinse. The results show that, after a single hand wash, both the SAB hand sanitizer formula and the 62% ethanol-based hand sanitizer formula had a degerming activity that was approximately 20% greater than the degerming activity of the control nonantimicrobial hand wash (Figure 1). Both the alcohol-based and SAB hand-wash formulas demonstrated an RF value of 2.8 ± 0.2.

The degerming efficacy of the alcohol-based hand wash decreased during the remainder of the hand contamination and wash cycles, falling to a level that was below the minimum acceptable FDA standard of RF = 3. In contrast, the degerming effectiveness of the novel SAB hand sanitizer formula increased over the course of the hand contamination and wash cycles required by the protocol.

Rinse-free testing. Both of the hand-wash formulas examined are intended for use without rinsing with water; thus, the above protocol was modified so that the 30-second water rinse was omitted. The results showed that the ethanol-containing hand wash had a moderate degerming action compared to the control nonantimicrobial hand wash after the first hand wash (Figure 2). The degerming effectiveness of the remaining hand contamination and decontamination cycles was markedly decreased for the 62% alcohol-based sanitizer. The degerming activity of the SAB hand sanitizer formula paralleled the results obtained with the rinsing protocol and showed a steady increase in germicidal activity throughout the course of the experiment, exceeding the FDA minimum standard.

Greater ethanol concentration testing. The most antimicrobially active ethanol concentrations lie in the range of 50% to 70% vol/vol in water; thus, researchers were curious to know whether an increased ethanol concentration in an ethanol-based sanitizer would improve antimicrobial performance. The researchers, therefore, examined the effectiveness of a different, commercially available hand sanitizer that also contained emollients, but had an ethanol concentration of 70% (ie, vol/vol).

The results show that, although the 70% ethanol formula initially performed better than the 62% formula, antimicrobial effectiveness decreased as before with successive washes in both the rinse and nonrinse protocols. Although the initial wash with the SAB sanitizer produced approximately the same RF as the 70% ethanol-containing formula in the rinse protocol, subsequent washes with the SAB formula produced bacterial reductions greater than

the 70% ethanol formula. Likewise, in the nonrinse protocol, the SAB formula's effectiveness was approximately the same after the first wash, but was significantly greater than the 70% ethanol formula for subsequent contamination and recontamination cycles.

Subjective testing. In addition to these objective results, subjects were asked to subjectively evaluate the condition of their hands after the completion of the formula tests. A significant number of subjects (ie, 47%) who had completed the test protocol with the alcohol-based hand sanitizer formulas—either in the rinse or nonrinse protocol—reported palmar pain or discomfort. After visual inspection, these subjects were found to have pronounced swelling that was, in some instances, accompanied by erythema of the palmar tissues. Also, the group that used ethanol-containing products tended to indicate some discomfort in palmar surfaces for one to several days after the test. In contrast, none of the subjects that used the SAB hand sanitizer formula reported any pain or discomfort of their hands after completing either the rinse or the nonrinse protocol.

None of the subjects who used the SAB hand sanitizer reported any pain or discomfort after use.

DISCUSSION

The Centers for Disease Control and Prevention (CDC) has stated that hand washing is the single most important factor in the prevention of disease and the spread of infections. Officials at the CDC estimate that one-third of all hospital-acquired infections are avoidable and are caused by a lack of adherence to established infection control practices such as hand washing.[16]

This insufficient hand washing has led to a great increase in the use of waterless hand sanitizers by health care personnel. This study evaluated the effectiveness of two ethanol-containing hand sanitizers and a novel SAB, ethanol-free hand sanitizer using an FDA-approved protocol.

After a single application, the alcohol-free SAB sanitizer and both alcohol-based formulas reduced bacteria more than a control nonantimicrobial handwash formula. When the protocol was repeated omitting the water rinse, similar results were achieved. This illustrated that the first time either of these types of products is used on any given day, degerming activity results that exceeds the federal requirements for antiseptic hand washes.

To be of any value in a health care setting, however, a hand antiseptic should give persistent antimicrobial activity with repeated use. Accordingly, the alcohol-free SAB sanitizer, with or without the water rinse, produced increased antimicrobial effectiveness over time with no adverse effects. In contrast to this, repeated use of the alcohol-based sanitizers produced a decrease in antimicrobial effectiveness over time and was accompanied by swelling, erythema, and discomfort of the palmar surface of subjects' hands. Importantly, by the completion of both the rinsing and nonrinsing protocols, antimicrobial persistence of the SAB formula was so pronounced that its performance exceeded federal requirements for antiseptic hand washes by at least 50%. The tested alcohol-based hand sanitizers, however, failed to meet this federal standard in both the rinse and nonrinse protocols.

In summary, the study showed

- the SAB hand sanitizer formula had a greater sustained degerming activity than the alcohol-containing hand sanitizer formula,
- the alcohol-containing hand sanitizer became less effective with repeated use and irritated the hands of subjects, and
- the SAB hand sanitizer formula became more effective without irritation after repeated use.

LIMITATIONS AND DIRECTIONS FOR FUTURE STUDY

A potential limitation to this study is that it was carried out in the controlled environment of a clinical research laboratory on model pathogens artificially introduced onto the hands of subjects according to a federally approved test protocol. Future research, therefore, would need to include studies of the impact on nosocomially derived infections in clinics, in which either an alcohol-containing or an alcohol-free hand sanitizer was routinely used to supplement normal hand washing.

Also, the interval between washes for each of the sanitizers tested in this study was 10 minutes—an amount of time chosen to model the effects of frequent, acute use, as might occur in a clinical environment that requires 10 to 12 patient contacts per hour. It would be informational, however, to perform the tests described in this document allowing a greater period of time between consecutive washes.

In the same way, the federally mandated time for the actual hand-washing procedure was two minutes, although *in vitro* data indicate that the formulas are effective in a nonskin environment after as little as 10 to 15 seconds. A second investigational parameter for future work, therefore, could include varying the hand wash duration, as well.

RECOMMENDATIONS FOR CLINICAL PRACTICE

Nurses in the OR face a situation that is particularly challenging in terms of maintaining hand sanitization. For example, nurses may at one moment be required to open storage drawers or handle and move equipment, such as lights and foot stools, and at the next moment be required to assist in wound dressing. In cases in which nurses must make the transition between equipment handling and assisting directly with the patient, universal precautions, such as hand washing and the wearing and changing of gloves, should take precedence. In situations in which hands should be sanitized before donning new gloves (eg, inadvertent contamination because of glove tearing) where soap and water are not immediately available, however, this study's results indicate that the alcohol-free SAB formula would be more effective with continued use than the alcohol-based formula at hand sanitization.

It is recommended that perioperative health care personnel who have frequently been using alcohol-based instant hand sanitizers to supplement normal hand washing consider the benefits of using an effective alcohol-free instant hand sanitizer, such as the SAB sanitizer. This formula is quick-acting, does not require a water rinse, and, unlike alcohol-based hand sanitizers, is not flammable—a quality particularly important for perioperative safety in general.

RECOMMENDATIONS FOR EDUCATION

The point for clinical education that may be gained from this study is that, although alcohol-based instant hand sanitizers are widely used in professional and nonprofessional circles, alcohol also is an effective organic solvent. As such, it readily strips away the natural chemical components of the skin (eg, sebum, lipids) that impede water loss and bacterial infection. Frequent and prolonged use of alcohol-containing hand sanitizer products, therefore, can be counterproductive to hand sanitization and can damage the skin.

The results of this study are presented to help perioperative health care professionals choose an appropriate product for rinse-free hand sanitization as a supplement to normal hand washing, not to undermine the fundamental importance of proper hand washing. This study further serves to educate professionals about the limitations of alcohol-containing hand sanitizers and the advantages of alcohol-free hand sanitizers in both a perioperative and general clinical setting. ▲

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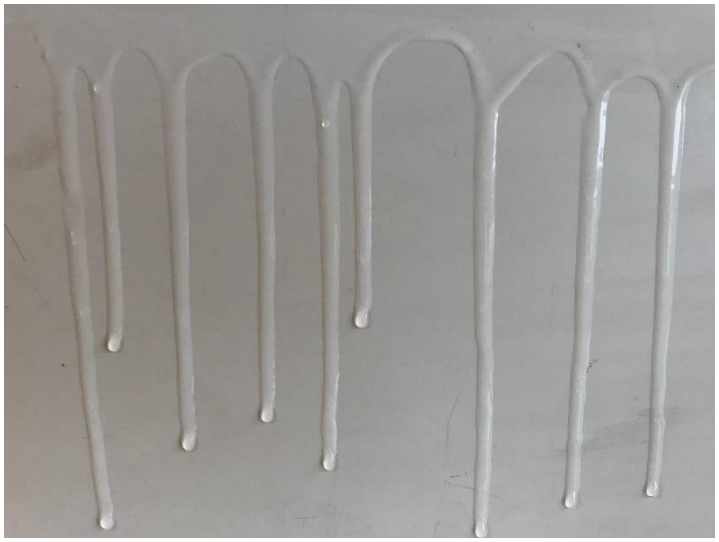
Alcohol free disinfectant

Ethanol and Isopropanol based disinfectants are widely used. In some cases, the use of alcohol might be undesired and one example is when using alcohol based disinfectants long-term and frequently in that it can cause skin irritation.

Arquad MCB (Benzalkonium Chloride) has been found to be effective in the hand sanitation application and indications from tests are also that Arquad MCB can present a lasting anti-microbial effect (Environmental Health Journal, 2016, Song et al).



Disinfectant gel, formulated as per below, allowed to run down a vertical surface

Appearance of ready-made disinfectant gel

Formulation:

Arquad MCB-50	0.25 % (w/w, 0.13% active)
Glycerol (optional)	0.5 % (w/w)
Bermocoll Prime 1000	0.7 % (w/w)
Water	to 100 %

Procedure: To a mixture of Arquad MCB-50 and Glycerol in water, Bermocoll Prime 1000 is added. The mixture is allowed to stir until complete Bermocoll dissolution.

Bermocoll Prime 1000 ratio can be varied to reach the desired rheological properties. Furthermore, as noted above the Bermocoll Prime 1000 is a cellulose based thickener which gives it water/moisture-retaining properties.

Please contact your sales representative for inquires and sample!

Ansvar & Omsorg

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