



Decreasing of hand colonization and sterility of refill antiseptic in Dr. Yap Eye Hospital, Yogyakarta

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ABSTRACT

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Dr. Yap Eye Hospital, Yogyakarta uses aseptic gel containing 70% ethyl alcohol to refill antiseptic containers during times of antiseptic scarcity. The study aimed to evaluate the effectiveness and sterility of the refill antiseptics in reducing the number of colonization on the hands of nurses. It was a quasi-experimental using pre- and post-control groups design involving 56 nurses who used antiseptics in a bottle sterilized by plasma device (plasma bottle) compare to those washed using detergent (detergent bottle) before being refilled. Sterility tests were performed every two wk for up to two mo. Colonization pre and post hand hygiene practices were determined as an efficacy test and compared using the Mann-Whitney test in STATA 14. Antiseptic in plasma bottles remains sterile until the week 8th since refilled, longer than detergent bottles (6th weeks). The pre-handrub colorizations were 0.04-16.92 CFU/cm² and decreased significantly post-handrub to 0.00-3.08 CFU/cm² ($p < 0.0001$). Significant decrease pre- vs post-handrub colonization was observed in usage of detergent bottle (0.04-31.04 vs 0.00-10.48 CFU/cm², $p = 0.0007$). There was no significant difference in Δ colonization for two bottles (0.40-15.76 vs 0.04-30.92 CFU/cm², $p = 0.8790$). In conclusion, antiseptic in the plasma bottle remains sterile longer than in the detergent bottle since refilled. Both of them are equally effective in reducing colonization after handrub activity.

ABSTRAK

Rumah Sakit Mata Dr. Yap. Yogyakarta menggunakan antiseptik isi ulang aseptic gel yang mengandung etil alkohol 70% pada masa kelangkaan bahan antiseptik kebersihan tangan. Tujuan penelitian ini adalah untuk mengevaluasi efektivitas dan sterilitas antiseptik isi ulang dalam menurunkan angka kuman di tangan petugas. Penelitian ini merupakan eksperimental semu menggunakan rancangan *pre and post test* dengan melibatkan 56 perawat yang menggunakan antiseptik isi ulang dalam botol yang disterilkan alat plasma (botol plasma) dibandingkan dengan dicuci detergen (botol detergen) sebelum diisi ulang. Uji sterilitas dilakukan setiap 2 minggu selama 2 bulan. Penghitungan angka kuman sebelum dan sesudah cuci tangan dilakukan sebagai uji efikasi. Perbandingan angka kuman dilakukan dengan uji Mann-Whitney menggunakan STATA 14. Antiseptik dalam botol plasma tetap steril hingga minggu ke-8 sejak diisi ulang, lebih lama dibandingkan botol detergen (minggu ke-6). Angka kuman sebelum cuci tangan dengan botol plasma berkisar antara 0,04-16,92 CFU/cm² dan turun signifikan sesudah cuci tangan menjadi 0,00-3,08 CFU/cm² ($p < 0,0001$). Penurunan yang signifikan terhadap angka kuman sebelum dan sesudah cuci tangan juga terjadi pada penggunaan antiseptik botol detergen (0,04-31,04 vs 0,00-10,48 CFU/cm², $p = 0,0007$). Tidak ada perbedaan bermakna pada Δ angka kuman kedua botol (0,40-15,76 vs 0,04-30,92 CFU/cm², $p = 0,8790$). Dapat disimpulkan, antiseptik botol plasma lebih lama steril dibandingkan botol detergen sejak diisi ulang. Kedua antiseptik efektif menurunkan angka kuman setelah aktivitas cuci tangan.

Keywords:

Antiseptic sterility;
antiseptic effectiveness;
hand colonization;
refill antiseptic;
handrub

INTRODUCTION

Infectious diseases are still the leading cause of high morbidity and mortality in the world. Hospital-acquired infections (HAIs) are related directly to patient safety, staff safety, and the hospital environment.^{1,2} The HAIs increase morbidity and mortality, long-term disability, and financial burden on health.¹ The incidence of HAIs is reported to be higher in low- and middle-income countries than in high-income countries. The low incidence of HAIs has become one of the target markers for the quality of hospital services, hospital performance indicators, national hospital accreditation standards, and hospital minimum service standards.^{3,4} According to the regulation of the Ministry of Health of Republic of Indonesia No.129/2008, the standard of HAIs figures should generally not be more than 1.5%.⁵ Meanwhile, the infection control target based on hospital performance indicator dictionary is not more than 5%.⁶ Studies showed HAIs rates ranged between 5-10% worldwide. The World Health Organization (WHO) data showed that the incidence of HAIs in Ukraine, Italy, France were 10%, 6.7%, 6.7-7.4%, respectively. In Indonesia, a study in 11 hospitals in Jakarta in 2008 showed that 9.8% of hospitalized patients has HAIs.⁷

Hand hygiene is the key to infection control. All hospital staff members, patients, and visitors must easily practice and readily get access to hand hygiene facilities while in the Dr. Yap Eye Hospital, Yogyakarta. Various studies have shown that hand hygiene is the most important and most cost-effective way to prevent and reduce the transmission of HAIs. Hands are the most common transmission medium for pathogens in hospitals.⁸ Hand hygiene can be done in two ways: washing hands with water and soap if hands look dirty and using alcohol-based handrub.^{9,10}

Dr. Yap Eye Hospital, Yogyakarta uses an alcohol-based refill antiseptic which was put into refill bottles during times of antiseptic scarcity in COVID-19 pandemic 2020. Preparation of the refill bottles was conducted as needed. High-level disinfection is required to kill spores. One of them uses a low-temperature plasma sterilizer H₂O₂ compatible with a sterilized plastic hand-rub bottle. Bauer-Savage explained the WHO recommendation of alcohol-based hand-rub prepared by putting the hand-rub into a small container (100 - 500 mL) and leaving for 72 h before use so that the spores in the container would die.¹⁰ the World Health Organization (WHO) In this study, the handrub was left for two days after being refilled in a bottle.⁹ However, unfortunately, the WHO guidelines does not explain how to prepare and clean refill bottles. Meanwhile, the cost of sterilization using plasma is relatively high. Therefore, a cheaper method to clean the refill bottles was needed by washing with detergent. The effectiveness of preparing the refill bottle should be compared.

This study aimed to evaluate the effectiveness and sterility of refill antiseptic in reducing the number of germs on the nurses' hands. This study also compared the bottles sterilized with plasma and those washed with detergent. The results of this study are expected to underlie the rationality of using refill antiseptic and its preparation to support the infection prevention control programs.

MATERIALS AND METHODS

Study design

It was a quasi-experimental using pre- and post-control groups design. Refill antiseptic (active ingredient ethyl alcohol 70%) was used. Two refilled antiseptics were treated differently; bottle 1 was sterilized with a plasma

device (plasma bottle), and bottle 2 was washed with detergent (detergent bottle) before refilling. After being refilled with antiseptic gel (the active ingredient ethyl is alcohol 70%), those bottles were left two days before being tested.

Subjects of the study

These subjects were nurses selected by random sampling technique who met the inclusion criteria. Inclusion criteria were Dr. Yap Eye Hospital, Yogyakarta nurses doing WHO standardized hand hygiene and directly contacting patients. Hand hygiene was conducted in 8 steps for about 20-30 sec and supervised by an Infection Prevention Control Nurse (IPCN).^{11,12} Exclusion criteria were nurses performing hand hygiene less than 1 h before and not correctly doing their profession accordingly.

Sterility testing

Sterility tests of those antiseptic ingredients were conducted by smearing those ingredients on 2 blood agar plates every 2 wk up to 2 mo. This research was conducted for 2 mo because the most prolonged use of a hand-rub bottle at Dr. Yap Eye Hospital, Yogyakarta was 2 mo. The agar plate was later incubated at 37^o C for 24 h. Microbe counts were reckoned based on colonization growth on brown agar plate after 24 h incubation. An efficacy test was performed when the antiseptic gel inside the bottle was sterile.⁹

Efficacy testing

Efficacy of the hand-rub was calculated by the number of viable microbes by comparing pre-and post-handrub colonization. Efficacy testing was performed with total sample and subject determination based on consecutive sampling to 2 refilled bottles, plasma and detergent bottles. Each

bottle had seven opportunities for hand hygiene. Therefore, there would have been a total of 56 opportunities for hand hygiene in nurses while comparing pre-and post-handrub colonization using a refilled bottle that had been sterilized by the plasma device (plasma bottle) vs washes by detergent (detergent bottle) before being refilled.

The pre- and post-hand hygiene colonization evaluation was done consecutively and was considered an efficacy test. Palm smears with size dimension 5x5 cm were performed entirely before and after undergoing hand rub. The smear of hands that had been planted on blood agar plate was incubated at 37^o C for 24 h, growing colonizations were counted, and the results were later divided to 25 cm².¹³

Statistical analysis

The statistical analysis used to analyze these research's results was STATA version 14 (Stata Corp., College Station, TX) with a p value < 0.05 regarded as statistically significant. Ultimately, Mann-Whitney test was used to assess differences in colonization pre- vs post-handrub plasma bottle, pre- vs post-handrub detergent bottle, delta colonization between plasma bottle vs. detergent bottle. All colonization data were expressed as median (min – max).

Ethical clearance

This study has been approved by the Law and Ethics Committee of Dr. Yap Eye Hospital, Yogyakarta (Number 02/KEH/EC/II/ 2022).

RESULTS

The study was conducted for two months, considered the most prolonged antiseptic use since it was opened at Dr. Yap Eye Hospital, Yogyakarta. Before the efficacy test, a sterility test was

performed. Sterility test results are presented in TABLE 1 and showed that antiseptic inside plasma bottle (8 wk) were more sterilized than antiseptic

inside detergent bottle (6 wk). If the sterility test had shown that the antiseptic ingredient was sterile, an efficacy test would have been performed.

TABLE 1. Sterility test results between antiseptic in plasma and detergent bottle

Week	Plasma bottle (n=1)	Detergent bottle (n=1)
0	Sterile	Sterile
2	Sterile	Sterile
4	Sterile	Sterile
6	Sterile	Sterile
8	Sterile	Non-sterile

TABLE 2. Efficacy test results of comparison between decreasing of colonization

Week [median (min-max)]	Plasma bottle			Detergent bottle		
	Pre-	Post-	Δ	Pre-	Post-	Δ
0	n = 7 2.40 (0.24-10.20)	0.40 (0.08-1.28)	1.12 (0.04-9.16)	n = 7 2.60 (0.44-16.00)	0.20 (0.04-10.48)	2.04 (0.20-7.00)
2	n = 7 4.4 (0.40-11.20)	0.76 (0.04-2.40)	3.64 (0.20-8.80)	n = 7 0.76 (0.04-7.40)	0.12 (0.00-1.72)	0.24 (0.04-5.68)
4	n = 7 0.96 (0.32-11.44)	0.52 (0.08-3.08)	0.44 (0.00-8.36)	n = 7 1.68 (0.64-12.52)	0.52 (0.04 - 3.24)	1.16 (0.2-11.96)
6	n = 7 2.08 (0.04-7.52)	0.48 (0.00-2.44)	1.52 (0.04-5.08)	n = 7 9.48 (0.08-31.04)	0.40 (0.00-5.00)	5.80 (0.08-30.92)
8	n = 7 3.76 (0.48-16.92)	0.40 (0.12-1.68)	2.08 (0.4-15.76)	Not performed		
Total	n = 35 2.08 (0.04-16.92)	0.48 (0.00-3.08)	1.12 (0.4-15.76)	n = 28 1.78 (0.04-31.04)	0.32 (0.00-10.48)	1.08 (0.04-30.92)

Mann Whitney test: Pre- vs post-handrub plasma bottle (p <0.0001); Pre- vs post-handrub detergent bottle (p = 0.0007); Δ colonization plasma bottle vs. detergent bottle (p = 0.8790). n= handrub practice; Pre-: pre-handrub colonization; Post- : post-handrub colonization; Δ: different colonization between pre and post handrub.

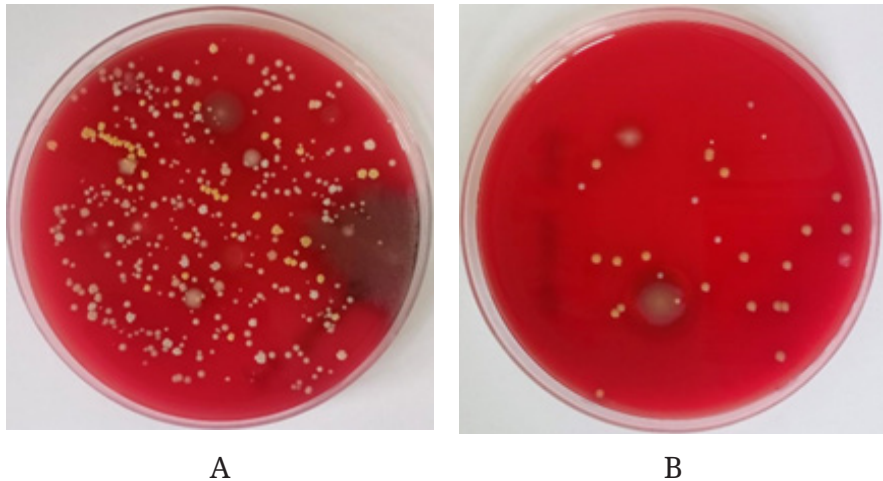


FIGURE 1 A. Pre hand-rubbed colonization; B. Post hand-rubbed colonization. Subject used plasma bottle in week 8th.

There were 56 nurses who fulfilled the inclusion and exclusion criteria. Pre-, post-, and Δ colonization of hand hygiene practice results are displayed in TABLE 2 and FIGURE 1. The result of pre-handrub colonization on 56 nurses was in the range of 0.04-16.92 CFU/cm² and significantly declined on post-handrub and became 0.00-3.08 CFU/cm² ($p < 0.0001$). The significant decline of pre- vs. post-handrub colonization also occurred in using detergent bottle antiseptic (0.04-31.04 vs. 0.00-10.48 CFU/cm², $p = 0.0007$). There was no significant difference in declining delta colonization between the two bottles (0.40-15.76 vs 0.04-30.92 CFU/cm², $p = 0.8790$).

DISCUSSION

Antiseptic ingredients were common antiseptic substances containing alcohol, isopropanol, n-propanol, or a combination of both products. Antimicrobial activities of alcohol derived from its ability to denature protein, and its most efficacious concentration was 60 – 80%. Unfortunately, the higher its concentration, it did not become any more potent. Chlorhexidine decreased microbe counts despite lower concentrations by deteriorating the

cytoplasmic membrane and causing precipitation of what was inside the microbe cells. The antimicrobial activity of chlorhexidine was relatively slower. However, chlorhexidine had a residual activity that was more significant than alcohol.¹⁴

According to WHO, average bacterial counts on medical officers were 3.9-4.6x10⁴ CFU/cm². WHO recommended that alcohol-based handrub as the gold standard in performing hand hygiene to visually or seemingly clean handwashing with soap was recommended to visually dirty hands. The majority of alcohol-based antiseptics contained isopropanol and ethanol. Both agents could have killed most bacteria fast for about 10-20 seconds, including multidrug-resistant organism (MDRO), for instances methicillin-resistant *Staphylococcus aureus* (MRSA) and vancomycin-resistant *Enterococcus* (VRE), *Mycobacterium tuberculosis*, a few fungi, and is also capable of inactivating a few viruses such as herpes groups.¹⁴

Human skin surfaces consisted of varied bacterial colonization. Pre-handrub colonization on the 5x5 cm² size of hands was varied. The usage of antiseptic in plasma and detergent bottles significantly decreased colonization. This finding was similar

to a previous study that confirmed alcohol had the most delicate germicidal activity toward bacterial vegetation and various fungi. Moreover, another study of antiseptic activity had shown that 70% ethanol and 70% isopropanol could decrease enveloped bacteriophage titer more effectively than antimicrobial soap containing chlorhexidine 4%.^{3,14}

In this study, handrub was leftover for two days after being refilled in a bottle, which is one day faster than homemade antiseptic production. According to WHO, it must be left over 72 h after being refilled.⁹ The bottle that was disinfected using plasma was more sterile than the one washed with detergent (8 wk vs 6 wk). Based on this research, it was suggested that a refilled bottle washed by a detergent could be used for up to 1.5 mo until refilled numbers could be readjusted. The WHO hand sanitizer formula for local production has a 2-year expiration date from production. There is no FDA guidance to set expiration dates. Meanwhile, the Center for Disease Control and Prevention (CDC) recommended minimum 60% alcohol content well beyond the affixed expiration date.¹⁵ Commercial hand-rub will expire depending on the bottle type. Alcohol-based hand disinfectant in a dispenser expires in 12 mo after opening. The expiration date is shorter in a dispenser without closing system to cover the neck of the bottle.¹⁶ Hand-rub expires because of the alcohol content which dissolves over time to less than 60% as it evaporates.¹⁷ However, there is no clear explanation why a detergent bottle is not sterile anymore in 8 wk from the refill process.

CONCLUSION

Antiseptic in the plasma bottle remained sterile longer than detergent bottle since refilled. Both of them are equally effective in reducing colonization after hand-rub activity.

However, there is no significant difference in Δ colonization for the two bottles. This study does not identified growing microbes in pre-hand-rub compared to post-hand-rub. Therefore, follow-up study will be essential, particularly by examining microbe identification and using a larger sample at each efficacy test.

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