

Effectiveness of Various Antiseptic Hand Soaps in Reducing Skin Bacterial Counts

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ABSTRACT

Hands are the body part most frequently exposed to environmental bacteria and play an important role in disease transmission. Handwashing with antiseptic soap is an effective method for reducing bacterial contamination on the skin. Antiseptic soaps containing chloroxylenol, povidone-iodine, and chlorhexidine gluconate are widely used due to their broad-spectrum antimicrobial activity. However, the comparative effectiveness of these three antiseptic agents remains unclear. This experimental study employed a group pre-test and post-test design involving 27 students from the Faculty of Dentistry, Trisakti University. The respondents were divided into three groups based on the type of antiseptic soap used: 0.5% chloroxylenol, 7.5% povidone-iodine, and 0.5% chlorhexidine gluconate, with nine respondents in each group. The number of bacterial colonies on the hands was assessed before and after handwashing using the swab method. Data were analyzed using the Wilcoxon signed-rank test and the Kruskal–Wallis test. All three antiseptic soaps demonstrated a significant reduction in bacterial colony counts. In the chloroxylenol group, the median colony count decreased from 100 CFU to 22 CFU ($p = 0.018$); in the povidone-iodine group, from 11 CFU to 0 CFU ($p = 0.008$); and in the chlorhexidine gluconate group, from 200 CFU to 61 CFU ($p = 0.008$). Based on the percentage reduction, povidone-iodine showed the greatest decrease (99.74%), followed by chloroxylenol (79.30%) and chlorhexidine gluconate (51.97%). However, comparisons of bacterial colony reduction among the three groups showed no statistically significant difference in effectiveness ($p = 0.481$).

INTRODUCTION

The skin is the largest organ of the human body and serves as the primary barrier against various environmental factors. In addition to functioning as a physical protective barrier, the skin also provides a habitat for diverse microorganisms that constitute the skin microbiome. The microbiome consists of resident flora that colonize the skin surface and transient flora acquired from the surrounding environment, both of which contribute to maintaining the balance of the body's defense system (Ma et al., 2024). Hands are among the body parts most frequently exposed to environmental contaminants and therefore have a high risk of microbial contamination. Daily activities, such as touching objects, shaking hands, using public facilities, and contacting contaminated surfaces, can facilitate bacterial transfer to the hands and contribute to the chain of disease transmission. Therefore, maintaining hand hygiene is one of the most effective measures for preventing the spread of microorganisms (Fitriyani & Murlistyarini, 2022; Sari et al., 2023; Kartika et al., 2017; Meena et al., 2019; Costa, 2024).

The World Health Organization (WHO) recognizes hand hygiene as a key component of infection prevention and control. In addition to removing visible dirt, handwashing helps

reduce the number of microorganisms attached to the skin, thereby lowering the risk of disease transmission. The effectiveness of handwashing is influenced by several factors, including handwashing technique, duration, type of soap used, and individual skin conditions (Ratmaja et al., 2023; Zulfa & Patricia, 2023; Kim et al., 2015).

Soap is a cleansing product containing surfactants that facilitate the emulsification of oils and removal of contaminants from the skin surface. In addition to conventional soaps, various antiseptic soaps containing antimicrobial active ingredients have been developed to enhance hand-cleaning effectiveness. These products not only remove contaminants mechanically but also inhibit or eliminate microorganisms present on the skin surface. Common antiseptic agents used in hand soaps include chloroxylenol, povidone-iodine, and chlorhexidine gluconate (Alum, 2024; Jalaluddin et al., 2023; Abbas et al., 2016; Ayuningtyas et al., 2021; Nuswantoro et al., 2020).

Chloroxylenol is a phenolic compound with antimicrobial activity against various Gram-positive and Gram-negative bacteria. Its mechanism of action involves protein denaturation and disruption of bacterial cell membrane integrity, leading to bacterial cell death (Kristy et al., 2021). Povidone-iodine is an iodine complex widely used as an antiseptic due to its broad-spectrum antimicrobial activity. It functions by releasing free iodine, which oxidizes proteins, enzymes, and essential structural components of microbial cells. Damage to these components disrupts cellular metabolism and ultimately results in microbial death (Eggers et al., 2018; Riza et al., 2019). Chlorhexidine gluconate is an antiseptic with broad antimicrobial activity and a relatively prolonged residual effect compared with several other antiseptic agents. Its mechanism involves electrostatic interactions between positively charged chlorhexidine molecules and negatively charged bacterial cell membranes, resulting in membrane disruption and leakage of cellular contents (Eggers et al., 2018; Rundle et al., 2019; Pratiwi et al., 2022).

Previous studies have demonstrated that antiseptic soaps are effective in reducing bacterial counts on the hands. Riza et al. (2019) reported that chloroxylenol 4.8% and povidone-iodine 7.5% had comparable effectiveness in reducing bacterial counts after handwashing procedures. Meanwhile, Aftab et al. (2023) found that chlorhexidine gluconate effectively reduced microbial counts on the skin due to its strong antimicrobial activity.

The urgency of this research lies in the need for evidence-based recommendations regarding hand hygiene products, particularly amid increased attention to handwashing practices during infectious disease outbreaks. Understanding the comparative effectiveness of commonly available antiseptic soaps can assist consumers and healthcare professionals in selecting appropriate hand hygiene products. This study contributes to the existing literature by providing direct comparative evidence regarding the effectiveness of three commonly used antiseptic agents in a nonhospital setting.

The novelty of this research lies in its direct comparison of three widely used antiseptic agents within a standardized handwashing procedure using concentrations commonly found in consumer products. Furthermore, this study employed a pre-test and post-test design by measuring bacterial counts before and after handwashing to directly evaluate the reduction achieved by each antiseptic soap.

Although previous studies have demonstrated the effectiveness of individual antiseptic ingredients, research directly comparing the effectiveness of chloroxylenol, povidone-iodine,

and chlorhexidine gluconate in reducing skin bacterial counts remains limited. Therefore, further comparative investigations are needed to evaluate the relative effectiveness of these antiseptic soaps. This study aimed to determine the effectiveness of various antiseptic hand soaps in reducing skin bacterial counts and to analyze differences in effectiveness among soaps containing chloroxylenol, povidone-iodine, and chlorhexidine gluconate. The findings are expected to provide scientific evidence regarding the selection of effective antiseptic soaps for maintaining hand hygiene and serve as a basis for future research on the antimicrobial activity of various antiseptic agents.

METHOD

This study employed an experimental research design using a pre-test and post-test group design. The research was conducted in May 2026 at the Microbiology Laboratory, Faculty of Medicine, Trisakti University, West Jakarta. The study involved 27 students from the Faculty of Dentistry, Trisakti University. Respondents were selected based on predetermined inclusion and exclusion criteria, including active students who were willing to participate, had no skin diseases or hand wounds, were not sensitive to antiseptic agents, and had not previously washed their hands or used antiseptic products before sample collection.

The sampling technique used was purposive sampling, in which respondents were selected based on the established criteria. The respondents were divided into three groups according to the type of antiseptic soap used, with nine respondents in each group. Data collection involved obtaining informed consent, collecting hand swab samples before and after handwashing, and counting bacterial colonies after incubation. The collected data were analyzed using SPSS software to evaluate the effectiveness of different antiseptic hand soaps in reducing skin bacterial counts and to compare the effectiveness among the tested antiseptic agents. Statistical analysis was performed using the Wilcoxon signed-rank test and the Kruskal–Wallis test because the data did not meet the assumption of normal distribution based on the normality test.

RESULTS AND DISCUSSION

Table 1. Distribution of study subject characteristics (n=27)

Variabel	Frequency (n)	Percentage (%)
Age (years)		
18 years old	12	44,4%
19 years old	14	51,9%
20 years	1	3,7%
Gender		
Male	12	44,4%
Women	15	55,6%

Based on table 1 above, the majority of respondents were 19 years old, namely 14 respondents (51.9%) and the average respondent who participated in the study was female (55.6%).

Table 2. The effectiveness of various types of antiseptic hand soap in reducing the number of skin bacteria

Variabel	Types of antiseptic soaps		
	<i>Chloroxylenol</i> 0,5%	<i>Povidone Iodine</i> 7,5%	<i>Chlorhexidine</i> <i>Gluconate</i> 0,5%
Decrease in the number of bacteria			
Median Pre	100	11	200
Median Post	22	0	61
Value p	0,018	0,008	0,008

Uji Wilcoxon Signed-Rank Test

Based on the results of table 2 using the *Wilcoxon Signed-Test*, it was found that 0.5% *chloroxylenol* soap had a median handwashing result of 100 CFU to 22 CFU, then in *povidone iodine* soap 7.5% it was 11 CFU to 0 CFU while in *chlorhexidine gluconate* soap 0.5% of 200 CFUs became 61 CFUs. All soap groups experienced a statistically significant decrease in the number of colonies after washing hands using antiseptic soap, namely *chloroxylenol* type soap 0.5% obtained a p value of 0.018, while *povidone iodine* soap 7.5% and *chlorhexidine gluconate* 0.5% had a p value of 0.008.

Table 3. Percentage decrease in the number of bacterial colonies in different types of antiseptic hand soap

Variabel	Types of antiseptic soaps		
	<i>Chloroxylenol</i> 0,5%	<i>Povidone Iodine</i> 7,5%	<i>Chlorhexidine</i> <i>Gluconate</i> 0,5%
Total colony bacteria			
Pre	1.681	779	1.653
Post	348	2	794
Decrease (%)	79,30%	99,74%	51,97%

Based on the results of the table above, it was found that the percentage decrease in the number of different bacterial colonies between soaps with the largest percentage decrease was in *Povidone Iodine* soap 7.5%, which was 99.74%, followed by *Chloroxylenol* 0.5% of 79.30%, and *Chlorhexidine Gluconate* 0.5% of 51.97%.

Table 4. Differences in the effectiveness of different types of antiseptic hand soap on reducing the number of skin bacteria

Variable	Types of antiseptic soaps			Value p
	<i>Chloroxylenol</i> 0,5%	<i>Povidone</i> <i>Iodine</i> 7,5%	<i>Chlorhexidine</i> <i>Gluconate</i> 0,5%	
Decrease in the number of bacteria				
N	9	9	9	
Δ CFU median	86	11	133	0,481

Uji Kruskal Wallis-Test

Based on the results of the *Kruskal Wallis-Test* test in table 3, it was found that the median decrease from the 0.5% *chloroxylenol* soap was 86 CFU, then for *povidone iodine* soap 7.5% was 11 CFU, while in *chlorhexidine soap* 0.5% was 133 CFU. The result of the p value obtained was 0.481 which means that no statistically significant difference in

effectiveness was found between the three types of antiseptic soap in reducing the number of bacterial colonies.

This study showed that all types of antiseptic soaps, namely chloroxylenol 0.5%, povidone iodine 7.5%, and chlorhexidine gluconate 0.5%, were able to significantly reduce the number of skin bacterial colonies after hand washing. These results were proven by the Wilcoxon Signed-Rank Test which showed a $p < 0.05$ value in all treatment groups. These findings indicate that the use of antiseptic soap is effective in reducing bacterial contamination of hands and supports the importance of handwashing practices as one of the infection prevention strategies (Ladyani et al., 2024).

In the 0.5% chloroxylenol soap group, there was a decrease in the number of colonies with a median value before hand washing of 100 CFU to 22 CFU. These findings are in line with research by Riza et al. (2019) who reported that chloroxylenol 4.8% was effective in reducing the number of bacteria after hand washing. The antimicrobial activity of chloroxylenol is obtained through the mechanism of protein denaturation and damage to the bacterial cell membrane thereby inhibiting the survival of microorganisms (Kristy et al., 2021).

Then the povidone iodine soap group of 7.5% showed the number of colonies from 11 CFUs to 0 CFUs. These results are in line with the research of Eggers et al. (2018) which showed that povidone iodine 7.5% has strong bactericidal activity against various microorganisms. Povidone iodine works by releasing free iodine that oxidizes proteins, nucleic acids, and other important components in microbial cells thus causing cell death (Lepelletier et al., 2020).

The largest decrease was found in the chlorhexidine gluconate soap group of 0.5%, from 200 CFU to 61 CFU. These results are similar to the research conducted by Aftab et al. (2023) which stated that chlorhexidine gluconate is effective in reducing the number of microorganisms on the skin. The mechanism of action of chlorhexidine gluconate involves the interaction between the positive charge of chlorhexidine molecules with the negatively charged bacterial membrane resulting in damage to the cell membrane (Thangavelu et al., 2020).

Although all soap groups showed a significant decrease in the number of bacterial colonies, the results of the *Kruskal-Wallis* test showed no statistically significant difference in effectiveness between the three types of antiseptic soap ($p = 0.481$). Meanwhile, based on the percentage reduction results of each soap, the type of povidone iodine soap 7.5% showed the result of reducing the number of bacterial colonies with the largest number of bacterial colonies, which was 99.74%, followed by chloroxylenol 79.30% and chlorhexidine gluconate 51.97%.

The results of this study support the research of Vallejo et al. (2018) which reported that chlorhexidine 4% and chloroxylenol 3% had comparable bactericidal effectiveness as well as the study conducted by Riza et al. (2019) which showed no significant difference between chloroxylenol 4.8% and povidone iodine 7.5% in reducing the number of bacteria in the hands.

The absence of a significant difference in effectiveness can be influenced by several factors. Variations in skin microbiota between individuals are one of the factors that play a role. The amount and type of bacteria present in the hands are influenced by daily activities, environmental exposure, personal hygiene habits, age, gender, and skin conditions. In addition, the skin's natural antimicrobial activity affected by pH, skin temperature, and lactic acid levels

can affect the number of bacteria that survive after handwashing (Zapka et al., 2017; Boxberger et al., 2021; Hayashi et al., 2021).

This research also has limitations, among which is the relatively small number of samples so that the statistical power of the research is limited. In addition, respondent activity prior to sampling could not be fully controlled, allowing for variations in the number of initial bacteria between individuals. And the relatively similar characteristics of respondents in terms of age also limited the results of the study to a wider population. Clinically, the results of this study show that *chloroxylenol* 0.5%, *povidone iodine* 7.5%, and *chlorhexidine gluconate* 0.5% can be used as an antiseptic soap option to maintain hand hygiene because all three have been shown to be effective in reducing the number of skin bacterial colonies.

CONCLUSION

Antiseptic soaps containing 0.5% chloroxylenol, 7.5% povidone-iodine, and 0.5% chlorhexidine gluconate were proven to be effective in reducing bacterial colony counts after handwashing. The percentage reductions in bacterial colony counts were 79.30% for chloroxylenol, 99.74% for povidone-iodine, and 51.97% for chlorhexidine gluconate, respectively. Among the three antiseptic soaps, povidone-iodine demonstrated the highest percentage reduction in bacterial colony counts (99.74%). Therefore, all three antiseptic soaps may be considered potential options for supporting hand hygiene practices.

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