ORIGINAL ARTICLE

Journal of Clinical Microbiology and Infectious Diseases (*JCMID*) 2022, Volume 2, Number 1: 11-15
E-ISSN: 2808-9405

Published By : the Indonesian Society for Clinical Microbiology

The effectiveness of patchouli oil as hand sanitizer: a comparative study between two antiseptic brands

Zinatul Hayati[1*], Dedy Syahrizal[1,2], Nisrina Nurhikmah[3], Fauzul Husna[4], Wilda Mahdani[1], Ade Oktiviyari[5], Tjut Mariam Zanaria[6]

[1]Department of Microbiology, Faculty of Medicine, Universitas Syiah Kuala, Darussalam, Banda Aceh 23111, Indonesia
[2]Department of Biochemistry Faculty of Medicine, Universitas Syiah Kuala, Darussalam, Banda Aceh 23111, Indonesia
[3]Faculty of Medicine, Universitas Syiah Kuala, Darussalam, Banda Aceh 23111, Indonesia
[4]Department of Pharmacology, Faculty of Medicine, Universitas Syiah Kuala, Darussalam, Banda Aceh 23111, Indonesia
[5]Department of Oral Biology, Faculty of Dentistry, Universitas Syiah Kuala, Darussalam, Banda Aceh 23111, Indonesia
[6]Department of Parasitology, Faculty of Medicine, Universitas Syiah Kuala, Darussalam, Banda Aceh 23111, Indonesia

*Corresponding to:
Zinatul Hayati. Department of Microbiology, Faculty of Medicine, Universitas Syiah Kuala, Darussalam, Banda Aceh 23111, Indonesia;
contact number: 081360335302
hayatikarmil@unsyiah.ac.id

Received: 2022-03-28
Accepted: 2022-05-30
Published: 2022-06-10

ABSTRACT

Introduction: Hand hygiene is a non-pharmacological intervention recommended worldwide to prevent and control infectious diseases. Hand sanitizer is an antiseptic that contains active agents and could eradicate pathogenic microorganisms caused by infectious diseases. The addition of patchouli oil as an active agent in hand sanitizers has been proven to have a bactericidal and bacteriostatic effect against bacteria and viruses. This study aims to compare Hand sanitizer A's effectiveness (which does not contain patchouli oil) with Hand sanitizer B (containing patchouli oil) by comparing the number of normal hand flora colonies before and after using both hand sanitizers.

Methods: This study is a pre-experimental design with a static group pretest and posttest design. There were 16 Medical Laboratory Technologists (MLT) enrolled in the study. Each MLT received two interventions, using Hand sanitizer A and B. The hand swabs were collected before and after using both hand sanitizers. The swabs inoculated on the media, incubated, and colony-forming units were counted.

Result: This study showed a significant difference between hand sanitizers containing and not containing patchouli oil in reducing the number of normal hand flora colonies with a p-value = 0.033 ($\alpha < 0.05$). The median value of total colonies decreased in Hand sanitizer B is 15, lower than the median value of Hand sanitizer A, which is 36.

Conclusion: Hand sanitizer B containing patchouli oil possessed preferable effects to Hand sanitizer A, which does not contain patchouli oil in reducing the number of normal hand flora colonies.

Keywords: hand sanitizer, patchouli oil, colony count, normal flora.
Cite This Article: Hayati, Z., Syahrizal, D., Nurhikmah, N., Husna, F., Mahdani, W., Oktiviyari, A., Zanaria, T.M. 2022. The effectiveness of patchouli oil as hand sanitizer: a comparative study between two antiseptic brands . *Journal of Clinical Microbiology and Infectious Diseases* 2(1): 11-15.

INTRODUCTION

The living microbe on the skin can be either resident flora (microbes that colonize and settle permanently) or transient flora (microbes that live temporarily. Hands frequently get microbes contamination, so they have a crucial role in transmitting infectious disease agents. The most effective method of preventing such transmission is hand hygiene. Hand hygiene aims to eliminate transient flora transmitted through skin-to-skin contact.[1–4]

The World Health Organization (WHO) has recommended hand sanitizer containing 60-95% alcohol and 3-6% hydrogen peroxide. Alcohol has optimal activity against several bacteria and viruses but has feeble sporicidal activity. Antibacterial and sporicidal activity can be generated by adding chemical compounds such as hydrogen peroxide.[5] Various recent innovations for hand sanitizers have been developed by utilizing local natural plants such as patchouli.

Patchouli (*Pogostemon cablin Benth*) is a tropical plant that produces essential oils called patchouli oil. In Indonesia, there are three types of patchouli plants that have patchouli oil with different levels: *Pogostemon cablin Benth* (Aceh Patchouli), *Pogostemon heyneanus Benth* (forest patchouli), and *Pogostemon hortensis Benth* (Javanese patchouli). Aceh patchouli produces an essential oil with a higher concentration than other patchouli plants. Initially, patchouli oil was merely used as a fixative agent by binding and preventing fragrant substances' evaporation in cosmetics, perfume, and others. Several recent studies related to patchouli oil have found therapeutic effects resulting from the constituents present in patchouli oil, such as antibacterial, antifungal, antiseptic, anti-inflammatory, and other effects.[6,7]

Its active and inactive ingredients influence the antimicrobial effectiveness of hand sanitizers. For this study, the researchers found two hand sanitizers in the local market: Hand sanitizer A (containing 75% alcohol and hydrogen peroxide) and Hand sanitizer B (containing 75% alcohol, hydrogen peroxide and patchouli oil). Hand sanitizer A is generally circulating in the market. Meanwhile, hand sanitizer B is a product issued by the Atsiri Research Center (ARC), Universitas Syiah Kuala (USK) research institute. Both hand sanitizer formulations follow

WHO recommendations using alcohol and hydrogen peroxide. Thus, researchers are interested in conducting a study to compare the effectiveness of the two hand sanitizers.

METHODS

The research design was a pre-experimental design using two treatment groups. Group A was taken palm swab before and after performing hand hygiene using hand sanitizer A, which contained 75% alcohol and hydrogen peroxide. In comparison, group B used hand sanitizer B, which had 75% alcohol, hydrogen peroxide and patchouli oil. Both hand sanitizers were treated on the same person for seven days.

The subjects in this study were the Medical Laboratory Technologist (MLT) who worked in the Clinical Microbiology Laboratory of dr. Zainoel Abidin Aceh with a total of 16 people. The sterile cotton swab was swept over the entire surface of the subject's palms, then inoculated on PCA media using a back-and-forth motion technique and incubated at 35 ± 2°C for 24 hours. Furthermore, the number of colonies that grew was counted. Moreover, the number of colonies that grew was measured manually using a colony counter.

The effectiveness of hand sanitizers A and B is obtained from the median total plate count before and after using the hand sanitizer. The colony count results were then analyzed statistically using the Wilcoxon Matched-Pairs Test (dependent sample) and the Mann-Whitney U-test (independent sample). The research data were not normally distributed. The p-value <0.05 indicates a significant difference.

RESULT

Number of bacterial colonies after using hand sanitizer A

The results of inoculation of germs from palm swabs before and after performing hand hygiene using hand sanitizer A showed a decrease in the number of colonies that grew before (HSAx) and after (HSAy) using hand sanitizer A (Figure 1). The calculation of the number of colonies before and after using hand sanitizer A can be seen in Figure 2.

Based on the Wilcoxon Matched-Pairs Test, the significance value (Sig.2-tailed) was 0.008 (p <0.05). This value indicates a significant difference between before and after using hand sanitizer A, which contains 75% alcohol and hydrogen peroxide. The results of the statistical analysis can be seen in Table 1 below.

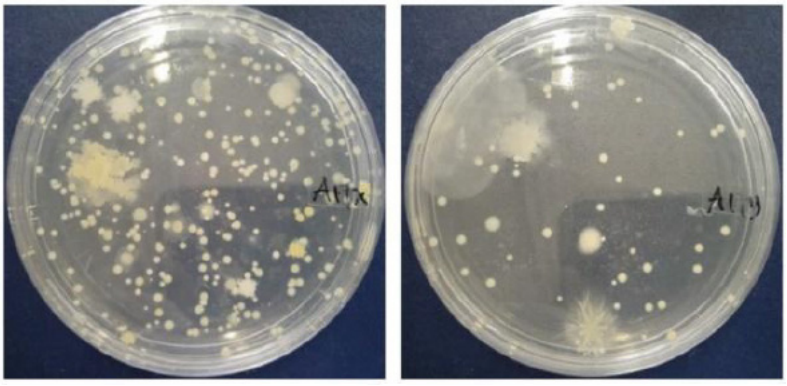

Figure 1. Before (left) and after (right) using hand sanitizer A

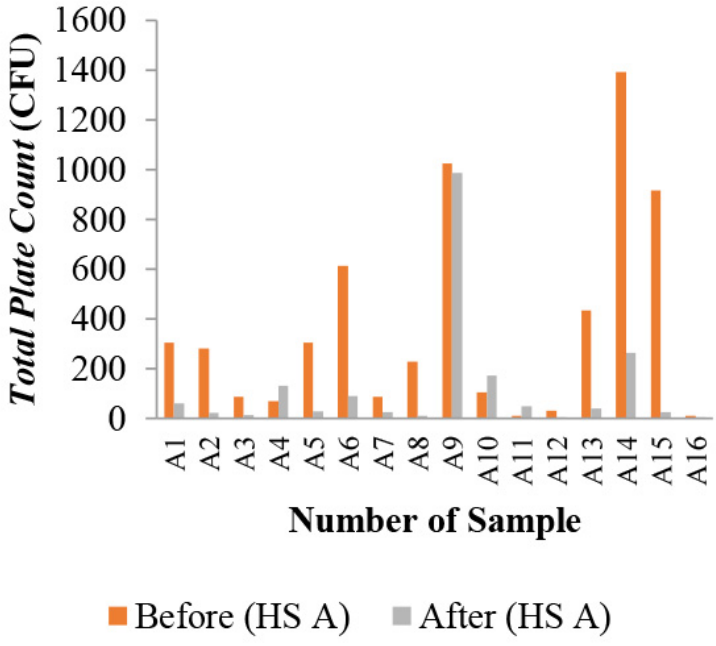

Figure 2. The difference before and after using hand sanitizer A

Table 1. Descriptive and statistical analysis result of hand sanitizer A

Group	N	Median (Min-Max)	Mean±SD	P
HSAx	16	255 (12-1393)	369.25± 413.461	0.008
HSAy	16	36 (4-986)	121.31± 241.354	

Number of bacterial colonies after using hand sanitizer B

After media incubation, the number of bacterial colonies growth was calculated manually using a colony counter (Figure 3). The results can be seen in Figure 4. Figure 4 shows a decrease in the number of colonies before (HSBx) and after (HSBy) using hand sanitizer B.

Based on the Wilcoxon Matched-Pairs Test, the significance value (Sig.2-tailed) was 0.000 ($p < 0.05$). This value indicates that there is a significant difference between before and after using hand sanitizer B. The results of the statistical analysis can be seen in Table 2 below.

Comparison of the effectiveness of hand sanitizer A and B

The comparison of the effectiveness of hand sanitizers A and B is illustrated in the box plot diagram (Figure 5). The significance of decreasing the number of hand sanitizer colonies A and B can be seen from the difference in the median value after using the two-hand sanitizers. The median HSAy value was 36, while the HSBy median value was 15. Based on this median value, the decrease in the number of colonies was more obtained after using Hand sanitizer B with a lower median value than the median value after using hand sanitizer A.

Based on the Mann-Whitney U-test, the significance value (Sig. 2-tailed) was 0.033 ($p < 0.05$). Based on this value, it can be concluded that there is a significant difference between the number of colonies after hand sanitizer B compared to hand sanitizer A. The results of the statistical analysis can be seen in Table 3 below.

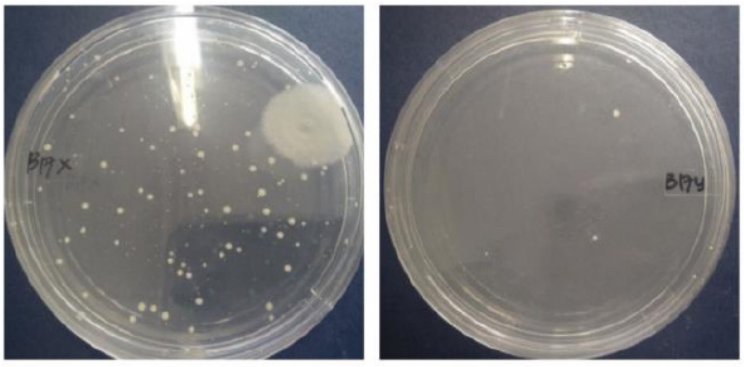

Figure 3. Before (Left) and After (Right) Using Hand Sanitizer B

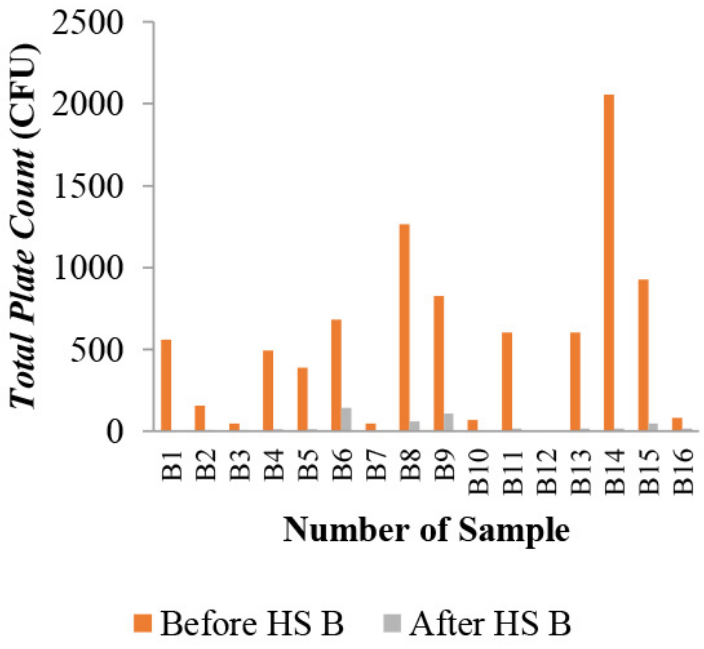

Figure 4. Difference before and after using hand sanitizer B

Table 2. Descriptive and statistical analysis result of hand sanitizer B

Group	N	Median (Min-Max)	Mean±SD	P
HSBx	16	528 (4-2056)	550.75± 544.724	0.000
HSBy	16	15 (1-146)	30.06± 41.719	

DISCUSSION

Hands contain microorganisms ranging from skin microbiota, such as normal flora, to pathogens that cause infection. The skin provides optimal growing conditions for both infectious and opportunistic pathogens. The use of hand sanitizers can reduce the number of microorganisms found on the hands to break the chain of infection transmission.[8]

This study compared the effectiveness of using two hand sanitizers with different active ingredients. Hand sanitizer A contains 75% alcohol and hydrogen peroxide. Meanwhile, hand sanitizer B contains 75% alcohol, hydrogen peroxide and patchouli oil. Hand sanitizers containing alcohol and hydrogen peroxide have reduced the number of typical hand flora colonies. On the other hand, patchouli oil also has good bactericidal and bacteriostatic activity against several bacteria and viruses.[7,9,10]

Patchouli Oil is an essential oil derived from the *Pogostemon cablin* (Blanco) Benth plant, *Lamiaceae* family. This plant is a wild plant found in many tropical countries such as Southeast Asia. The main content of patchouli oil is sesquiterpene compounds (40-45%) such as patchouli alcohol (> 30%), pogostone, α-bulnesene, α-patch-outlene, β-carophyllene, β-patchoulene, globulol, carophyllene oxide, spathulenol, and others. Among all these compounds, only patchouli alcohol and pogostone produce antibacterial effects.[6,11] This study indicates that Hand sanitizer B containing 75% alcohol, hydrogen peroxide, and

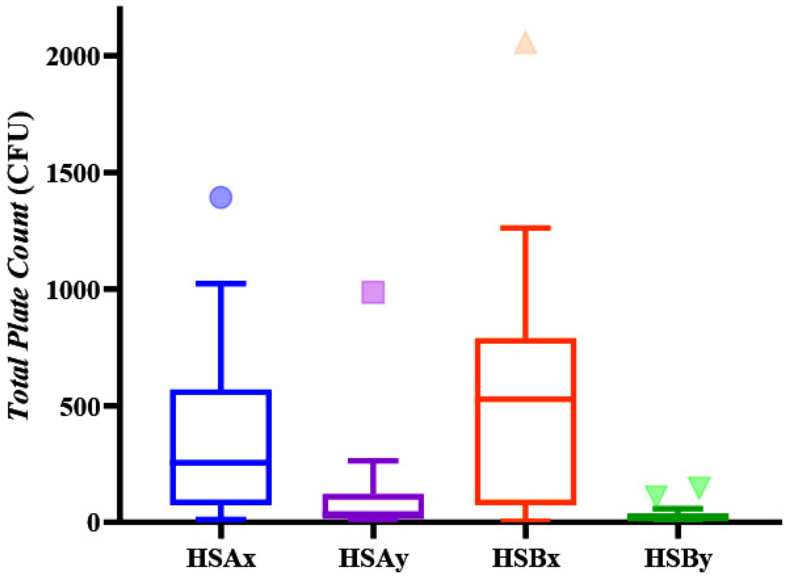

Figure 5. The comparation of colonies number after and before using hand sanitizer A and B

Table 3. Descriptive and statistical analysis results of hand sanitizers A and B

Group	N	Median (Min-Max)	Mean±SD	P
HSAy	16	36 (4-986)	121.31± 241.354	0.033
HSBy	16	15 (1-146)	30.06± 41.719	

patchouli oil is more effective and superior in reducing the number of normal hand flora colonies. Adding patchouli oil as an active ingredient to hand sanitizers containing alcohol and hydrogen peroxide can increase the effectiveness of bactericidal and bacteriostatic, making the effect much more significant.

The antibacterial mechanism of patchouli oil is influenced by its main constituents, such as patchouli alcohol and pogostone, using molecular docking technology and in vitro testing. Patchouli oil's antibacterial activity targets include penicillin-binding proteins (PBP), dihydrofolate synthetase (DHPS), dihydrofolate reductase (DHFR), DNA gyrase and RNA polymerase. These enzymes function for the biosynthesis of the primary cell structures such as cell walls, nucleoplasm cell membranes, and folic acid. Patchouli oil and its constituents can also inhibit the growth of bacterial biofilms due to the increased absorption of patchouli oil's active components by the extracellular biofilm matrix.[6]

Other studies have found that the effects of patchouli oil constituents such as patchouli alcohol and pogostone are more effective against gram-positive bacteria than gram-negative bacteria. Research conducted on 107 strains of gram-positive bacteria and 20 strains of gram-negative bacteria, found patchouli alcohol to have a better bacteriostatic effect against gram-positive bacteria (MIC = 1.5-200 µg / ml) than gram-negative bacteria (MIC = 25- 768 µg / ml). This study also shows that patchouli alcohol effectively against antibiotic-resistant bacteria such as MRSA and MRSE.[6,11]

Other research also states that patchouli oil has antioxidant activity and can inhibit the growth of bacteria that cause Community-Acquired infections and Healthcare-Associated Infections. The ethanol extract from the leaves of *Pogostemon cablin* Benth inhibited the growth of sensitive and resistant Staphylococcus aureus with inhibition zones of 10.33 ± 2.52 and 11.67 ± 1.53 mm. *Pogostemon cablin* Benth extract also inhibited the growth of *Streptococcus pyogenes* with an inhibition zone of 10.33 ± 1.15 mm.[12]

Based on this study's results, Hand sanitizer B's effectiveness was more effective in reducing the number of hand bacterial colonies. However, hand hygiene is the most critical aspect of infection prevention and control efforts. Hand hygiene can break the chain of infection transmission and significantly reduce the incidence of infectious diseases in health care centers and the community. WHO recommends using alcohol-based hand sanitizer (ABHS) as a method of hand hygiene and replacing the practice of washing hands using soap in certain situations. ABHS also has broad-spectrum microbicidal activity against bacteria and viruses, making the use of ABHS can be used as a hand hygiene method in breaking the transmission chain.[13,14]

CONCLUSION

Based on the calculation of the number of colonies from the hand swab of 16 MLT in RSUD dr. Zainoel Abidin uses Hand sanitizer A, which contains 75% alcohol and hydrogen peroxide, and Hand sanitizer B, which contains 75% alcohol, hydrogen peroxide and patchouli oil. It can be concluded that Hand sanitizer B containing patchouli oil has better effect in reducing the number of normal flora Hand sanitizer A does not contain patchouli oil.

LIMITATIONS

Some limitations of this study: First, colony count is done manually, thus allowing errors in calculating the number of colonies to occur. Second, this study did not identify microorganisms, so it is not confirmed what types of microorganisms grow on the media.

ACKNOWLEDGEMENTS

This work was facilitated by the Clinical

Microbiology Laboratory of dr. Zainoel Abidin General Hospital, Banda Aceh, Indonesia.

CONFLICT OF INTEREST

We have no potential conflicts of interest to report. All authors read and approved the final manuscripts.

FUNDING

None.

AUTHOR CONTRIBUTION

All of the authors contributed equally to this study.

ETHICAL CONSIDERATION

This research has received *ETHICAL APPROVAL* from the Health Research Ethics Commission of the Faculty of Medicine, Universitas Syiah Kuala – dr. Zainoel Abidin Hospital (*KEPPKN Registration Number: 1171012P)* by number*: 162/EA/FK-RSUDZA/2020*

REFERENCES

1. US Chemical. The Science of Handwashing [Internet]. 2016. Available from: http://www.uschemical.com/wp-content/uploads/2016/04/L000253_SCIENCE_OF_HANDWASHING.pdf
2. Warren-Gash C, Fragaszy E, Hayward AC. Hand hygiene to reduce community transmission of influenza and acute respiratory tract infection: a systematic review. Influenza Other Respi Viruses. 2012/10/08. 2013;7(5):738–49. Available from: https://pubmed.ncbi.nlm.nih.gov/23043518
3. Jumaa PA. Hand hygiene: simple and complex. Int J Infect Dis. 2005;9(1):3–14. Available from: http://dx.doi.org/10.1016/j.ijid.2004.05.005
4. Gould D, Gammon J, Donnelly M, Batiste L, Ball E, Carneiro De Melo AMS, et al. Improving hand hygiene in community healthcare settings: the impact of research and clinical collaboration. J Clin Nurs. 2000;9(1):95–102. Available from: http://dx.doi.org/10.1046/j.1365-2702.2000.00334.x
5. World Health Organization (WHO). WHO Guidelines on Hand Hygiene in Health Care [Internet]. 2009. Available from: http://whqlibdoc.who.int/publications/2009/9789241597906_eng.pdf
6. Reiza Y. Potensi Antibakterial Nilam Aceh (Pogostemon cablin Benth.). Cermin Dunia Kedokt. 2018;45(11):2–6.
7. Aisyah Y, Hastuti P, Sastrohamidjojo H, Hidayat C. Komposisi Kimia dan Sifat Antibakteri Minyak Nilam (Pogostemon cablin). Maj Farm Indones [Internet]. Vol. 19. 2008. p. 151–6. Available from: https://repository.ugm.ac.id
8. Gupta B, Kotia P, Gupta R, Sharma A, Raina K, Nissar I. Comparing the antimicrobial efficacy of different hand sanitizers: An In vitro Study. J Indian Assoc Public Heal Dent. 2019;17(3):253. Available from: http://dx.doi.org/10.4103/jiaphd.jiaphd_97_18
9. Swamy MK, Sinniah UR. A Comprehensive Review on the Phytochemical Constituents and Pharmacological Activities of Pogostemon cablin Benth.: An Aromatic Medicinal Plant of Industrial Importance. Molecules. 2015;20(5):8521–47. Available from: https://pubmed.ncbi.nlm.nih.gov/25985355
10. Das P, Dutta S, Begum J, Anwar MN. Antibacterial and Antifungal Activity Analysis of Essential Oil of Pogostemon cablin (Blanco) Benth. Bangladesh J Microbiol. 2016;30(1–2):7–10. Available from: http://dx.doi.org/10.3329/bjm.v30i1-2.28446
11. Wan F, Peng F, Xiong L, Chen J, Peng C, Dai M. In Vitro and In Vivo Antibacterial Activity of Patchouli Alcohol from Pogostemon cablin. Chin J Integr Med. 2016;27(2):125–30. Available from: http://dx.doi.org/10.1007/s11655-016-2452-y
12. Dechayont B, Ruamdee P, Poonnaimuang S, Mokmued K, Chunthorng-Orn J. Antioxidant and Antimicrobial Activities of *Pogostemon cablin* (Blanco) Benth. J Bot. 2017;2017:1–6. Available from: http://dx.doi.org/10.1155/2017/8310275
13. Jing JLJ, Pei Yi T, Bose RJC, McCarthy JR, Tharmalingam N, Madheswaran T. Hand Sanitizers: A Review on Formulation Aspects, Adverse Effects, and Regulations. Int J Environ Res Public Health. 2020;17(9):3326. Available from: https://pubmed.ncbi.nlm.nih.gov/32403261
14. Nasution TA, Yunita R, Pasaribu AP, Ardinata FM. Effectiveness Hand Washing and Hand Rub Method in Reducing Total Bacteria Colony from Nurses in Medan. Open access Maced J Med Sci. 2019;7(20):3380–3. Available from: https://pubmed.ncbi.nlm.nih.gov/32002055