REVIEW ARTICLE

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Isolation and Identification of Bacteria in the Mouth Before and After Ablution

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ABSTRACT

Background: The oral is the gateway for the entry of various kinds of microorganisms into the body, with the prevalence of people having dental and oral problems in Indonesia increasing every year. The normal flora of the oral acts as a body defense, but it can cause disease due to predisposing factors, namely oral hygiene. Therefore, it is necessary to find an alternative in maintaining oral health. Islam is a religion that emphasizes personal hygiene, such as performing ablution.

Content: The types of bacteria found in the oral before ablution was 33.33% *Pseudomonas* sp., 6.67% *Lactobacillus* sp., 3.33% *Streptococcus* sp. and 0.14% *Staphylococcus* sp. while the types of bacteria found in the oral after ablution were 26,8% *Pseudomonas* sp., 20% *Lactobacillus* sp., 5% *Streptococcus* sp. and 2% *Staphylococcus* sp.

Conclusion: There was a change in the number of bacteria, namely an increase in gram-positive bacteria in the oral after ablution.

Keywords: Isolation; identification; bacteria; ablution



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Introduction

Health is the most important part of human life, which is physically and spiritually healthy. Health that needs to be considered in addition to general body health, is also dental and oral. Dental and oral health can affect overall body health because dental and oral health is an integral part of overall body health that cannot be separated from general body health.⁽¹⁾

Based on the Basic Health Research (RIKESDAS) in 2018, the percentage of the population who had dental and oral problems in Indonesia had quite increased from the previous year, which was around 57.6%. By province which Having these problems, South Sulawesi ranks second out of the total.⁽²⁾

The mouth and nose are nests of harmful bacteria, if you don't clean them regularly, they will cause various diseases. The bacteria will be more fertile by the remaining food debris that is between the teeth and is not cleaned. However, not all microorganisms are pathogenic in the oral cavity, microorganisms that enter will be neutralized by anti-bacterial substances and normal flora bacteria. However, the presence of normal flora is not always beneficial, under certain conditions normal flora can cause disease, for example, if there is a change in substrate or displacement from the proper habitat. Research has proven that 90% of those who suffer from tooth decay are caused by mistakes in cleaning the mouth. Therefore, it is necessary to find alternatives in maintaining oral health.⁽⁵⁾⁽⁶⁾

Islam is a religion that always emphasizes personal hygiene before worship. This can be seen from the emphasis on bathing and ablution before performing obligatory worship, such as prayer. So Muslims must always maintain cleanliness because cleanliness will manifest physical and spiritual health. As a form of self-cleaning, when we perform ablution, we are also advised to rinse our mouth and siwak (toothbrush), clean the nose and, clean between the beards, as well as the fingers and toes.⁽⁷⁾⁽⁸⁾

Wudhu is a cleaning process carried out by a person to wash the body parts five times a day. According to research conducted by Herika Laksmi Safitri K. with the title "Isolation, Identification and Sensitivity Test of Antibiotic Germs in the Mouth Before and After Wudhu on Students of the 2016 Faculty of Medicine, Muslim University of Indonesia" concluded that there was a decrease in the number of gram-negative bacteria in the mouths of students after ablution. So that there will be a difference between students who have performed ablution and those who have not. Where the development of bacteria is more inhibited after ablution.⁽⁹⁾⁽¹⁰⁾

By looking at various facts regarding the benefits of ablution to public health, this research was carried out to prove and examine how the development of bacteria in the mouth after ablution and knowing the types of bacteria found in the oral cavity.

Subheading

A study conducted by Armiati, IGK in 2018 discussed the decrease in the number of *Streptococcus mutans* colonies in the oral cavity by ethanol extract of aloe vera leaf skin. The sample in this study was

obtained by isolating oral bacteria where people tried to gargle with distilled water for 60 seconds without swallowing and the results of the mouthwash were discarded.⁽²³⁾

Samples were taken using the swab method, where a sterile cotton swab was rubbed on the teeth on the labial surface of the upper right first incisor, the labial surface of the lower left first incisor, the buccal surface of the upper right first molar, the buccal surface of the upper left first molar, and the lingual surface. The lower left first molar and the lingual surface of the lower right first molar. This swab is done before and after gargling. The swab results were inserted into TSB media and cultured on Mueller Hinton Blood media. The culture results were seen after 24 hours and counted the number of bacterial colonies that grew and identified the bacteria by testing the bacteria with the Gram stain test, catalase test and mannitol test. So that the results obtained the number of Streptococcus mutans bacteria before gargling was 6114.80±2733.93 CFU/ml and after gargling was 3683.34±921.63 CFU/ml. This proves that there is a decrease in Streptococcus mutans bacteria after gargling with distilled water. At the same time, Ernawati, K. L (2018) conducted a study discussing kombucha tea, which can reduce the number of *Streptococcus* mutans bacteria in caries patients in 2019. Djohari et al conducted a study that discussed the identification and test of the inhibitory activity of ethanol extract areca nut (Areca catechu L.) against gum bacteria isolates. Where, in these two studies, sampling was carried out in the same way, namely by using a swab technique and placing it on the culture medium, then gram staining and biochemical tests were carried out to identify bacteria.⁽²³⁾⁽²⁴⁾⁽²⁵⁾

In addition, a study by Da-Young et al in 2017, compared the acidity of the oral pH of each study subject, the patient's hygiene performance index score, and the number of bacteria in the oral cavity of each study subject before and after gargling and brushing teeth. In this study, 10 ml of saliva was taken from each subject, after that, it was diluted with agar, a mixture of Sigma Aldrich and agar powder (Samjeon Chemistry), with distilled water 7 times, and mixed with superior agar medium. The mixture was then grown in a culture medium at 37° C for 72 hours (3 days), after which the number of colonies was counted. The comparison of the number of bacteria before and after gargling showed that the number of bacteria decreased after rinsing with water. The average difference in the PHP index (p<0.05) before and after rinsing with water was 2. 25 and 1.94. From this study, it was found that there was no significant difference in the number of bacteria before and after rinsing with water.⁽²⁶⁾

Research conducted by Titi Lasmini in 2020 used the Spread plate method of bacterial isolation. This research was conducted by isolating bacteria on MSA and MacConkey medium, purification, and testing for biochemical reactions. The bacteria found in the oral cavity after gargling were bacteria of the *Staphylococcaceae* group (*Staphylococcus aureus* 3.85%, *Coagulase Negative Staphylococcus* 30.77%), *Streptococcaceae* (*Enterococcus* sp. 3.85%), HACEK Group (*Aggregatibacter* sp. 3.85%), *Enterobacteriaceae* (*Klebsiella* sp. 19.23%, *Citrobacter* sp. 3.85%), and *Nonfermenting Gram-Negative* Publisher: Faculty of Medicine Universitas Muslim Indonesia 134

Bacilli (Acinetobacter sp. 7.69%, *Pseudomonas* sp. 26.92%). The results showed that *Coagulase Negative Staphylococcus* (30.77%), *Pseudomonas* sp. (26.92%), and *Klebsiella* sp. (19.23%) were the most common species in the oral cavity of non-smokers.⁽²⁷⁾

Another study conducted by Utami and Suryani in 2016 found the decrease in the number of germs was shown by counting the number of germs. The type of bacteria found in the mouth was *Staphylococcus* sp. with the number of colonies were 277 ± 178 CFU/ml before ablution and 270 ± 240 CFU/ml after ablution. The *Streptococcus* sp. colonies were also found in the mouth around 116 ± 95 CFU/ml before ablution and 83 ± 82 CFU/ml after ablution. The results of the Wilcoxon statistical analysis showed that the *p*-value for *Streptococcus* sp. in the mouth was *p*=0.30 (*p*>0.05) and *Staphylococcus* sp. was 0.002 (*p*<0.05). This result shows that there is a decrease in the number of germs before and after ablution against *Staphylococcus* sp. and *Streptococcus* sp. in the nurse's mouth.⁽²⁸⁾

In addition, research conducted by Safitri K, H. L. in 2019 shows that a variety of bacterias, such as *Escherichia* sp. 40%, *Pseudomonas* sp. 33.33%, *Lactobacillus* sp. 6.67%, *Streptococcus* sp. 6.67%, *Fusobacterium* sp. 6.67%, *Veillonella* sp. 6.67% and the bacteria found after ablution was *Escherichia* sp. 13.33%, *Pseudomonas* sp. 26.67%, *Lactobacillus* sp. 20%, *Streptococcus* sp. 13.33%, and *Veillonella* sp. 26.67%. And also shows that there is an increase in gram-positive bacteria which is a normal flora in the mouth after ablution.⁽¹⁰⁾

A study conducted by Manurung in 2020, about the effectiveness of gargling steeped green tea (*Camellia sinensis*) on the number of bacterial colonies as an asepsis measure, found that the average number of colonies in the group after gargling with green tea was 90.83 ± 38.73 CFU/ml and the number of colonies in the mineral water gargling group was 218.55 ± 120.86 CFU/ml.⁽²⁹⁾

In addition, a study conducted by Febriyanti in 2018 aimed to compare the number of anaerobic bacterial colonies in the saliva of children who were rinsed with peatland water and PDAM water. In the saliva of children who were rinsed with PDAM water, colonies of *Streptococcus* sp. and *Lactobacillus* sp. were found. In this study, the number of anaerobic bacterial colonies in peatland water was 217 CFU/ml, while the number of anaerobic bacteria colonies in PDAM water was 133 CFU/ml. Based on the results of the independent t-test (0.000) (p<0.05) showed that there was a significant difference between the number of anaerobic bacteria colonies in the saliva of children who rinsed with peatland water was higher than the number of anaerobic bacteria colonies in the saliva of children who rinsed with PDAM water.⁽³⁰⁾

And research conducted by Mahgaidren in 2018 discussed the effectiveness of gargling a 15% forest honey solution and 1% povidone-iodine solution to reduce the number of oral bacteria in students of the Faculty of Dentistry, University of North Sumatra. In this study, aqua was used as a negative control where

the average number of bacteria before treatment (pretest) was $188.43 \times 103 \pm 57.665 \times 103$ CFU/ml and after treatment (posttest) was $128.14 \times 103 \pm 37.791 \times 103$ CFU/ml. from these results indicate that there is a decrease in the number of bacteria in the oral cavity before and after gargling aqua.⁽³¹⁾

From some of these studies, the results showed that by carrying out ablution activities specifically meant in this case was gargling, it was found that there was a decrease in the number of bacteria in the mouth after gargling. Based on the description above, the bacteria found in the mouth before and during ablution can be classified into two types, namely gram-positive bacteria and gram-negative bacteria. The number of gram-negative bacteria tends to decrease after ablution, while the number of gram-positive bacteria increases. This happens because gram-positive bacteria are normal flora in the oral cavity that has an important role in the body's defense. By gargling, you can clean the oral cavity from bacteria and microbes before they spread and cause disease, and prevent the potential for dental and oral diseases. This proves that ablution can minimize the occurrence of problems in dental and oral health.

Conclusion

From the results of the analysis of a systematic study, it can be concluded that the types of bacteria present in the oral cavity before ablution are 33.33% *Pseudomonas* sp., 6.67% *Lactobacillus* sp., 3.33% *Streptococcus* sp. and 0.14% *Staphylococcus* sp. in the oral cavity after ablution were 26.8% *Pseudomonas* sp., 20% *Lactobacillus* sp., 5% *Streptococcus* sp. and 2% *Staphylococcus* sp. this indicates that there is an increase in gram-positive bacteria in the oral cavity after ablution.

Conflict of Interest

No potential conflict of interest relevant to this article was reported

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References

- 1. Kusumanto H. The Relationship between Periodontal Health Status and Learning Achievement in Students aged 10-12 Years at Mim Klaseman Gatak Sukoharjo. 2016;53(9):1689–99.
- 2. Indonesian Ministry of Health. Basic Health Research (RIKESDAS). Jakarta: Indonesian Ministry of Health Research and Development Agency; 2018. Available from: http://www.yankes.kemkes.go.id/assets/downloads/PMK No. 57 of 2013 concerning PTRM.pdf
- 3. Ren W, Zhang Q, Liu X, Zheng S, Ma L, Chen F, et al. Exploring the oral microflora of preschool children. J Microbiol. 2017;55(7):531–7.
- 4. Elkhaira R, Kasuma N, Son AE. Number of Lactic Acid Bacterial Colonies in a Healthy Oral Cavity. J Health Andalas. 2020;8(4):157–61.
- 5. Jawetz E, Melnick JL AE. Medical Microbiology. 27th ed. Jakarta: EGC; 2017. 199 p.
- 6. Chaturvedi M, Punj A. Human Oral Microflora. Int J Curr Adv Res [Internet]. 2017;7(7): 14065–70. Available from: http://dx.doi.org/10.24327/ijcar.2018.14070.2539
- 7. Maawiyah A. Thaharah as the Key of Worship. Sarwah J Islam Civiliz Thought. 2016;15(2).
- Ajib M. Fiqh of Wudhu Version of the Syafi'i School [Internet]. South Jakarta: Fiqh Publishing House; 2019. 1–38 p. Available from: http://repo.iainbatusangkar.ac.id/xmlui/bitstream/handle/123456789/12275/1569222648762_Fiqih Wudhu Version of Madzhab Syafiiy.pdf?sequence=1
- 9. Afif M, Khasanah U. The Urgency of Wudhu and Its Relevance for Health (Study of Ma'anil Hadith) in the Perspective of Imam Musbikin. Riwayah J Study Hadith. 2019;3(2):215.
- Safitri K HL. Isolation, Identification and Sensitivity Test of Antibiotic Germs in the Mouth Before and After Wudhu on Students of the 2016 Faculty of Medicine, Indonesian Muslim University. Indonesian Muslim Univ Makassar. 2019;
- Ansory I. Wudu' Rasulullah SAW According to 4 Schools. South Jakarta: Fiqh Publishing House; 2018. 1–77 p.
- 12. Zulfa FF. The Benefits of Wudu on Health from the Perspective of the Hadith of the Prophet SAW. (A Study of Tahlili Hadith). State Islamic University of Alauddin Makassar. 2019;
- 13. Lumowa SVT. Bacteriology. East Java: RADe.Rozarie; 2016.
- 14. Padoli. Nursing Microbiology and Parasitology. Ministry of Health of the Republic of Indonesia: Agency for the Development and Empowerment of Health Human Resources; 2016.
- 15. MH's daughter, Sukini, Yodong. Dental Nursing Teaching Materials. Microbiology. Ministry of HealthRepublic of Indonesia: Agency for Health Human Resources Development and Empowerment; 2017.
- 16. Hegde MN. Dysbiosis of Oral Microflora: A Review. J Heal Allied Science NU. 2018;08(04):034–9.
- 17. Samaranayake L, Matsubara VH. Normal Oral Flora and the Oral Ecosystem. Dent Clin North Am. 2017;61(2):199–215.
- 18. Adnyani NP, Made I, Artawa B. Effect of Dental and Oral Diseases on Halitosis. J Dental Health. 2016;4(1):24-8.
- 19. Cruz Quintana SM, Diaz Sjostrom P, Arias Socarrás D, Mazón Baldeón GM. Microbiota of oral cavity ecosystems. Rev Cubana Estomatol. 2017;54(1):84–99.
- 20. Puspitasari AM, Ratnawati DE, Widodo AW. Classification of Dental and Oral Diseases Using the Support Vector Machine Method. J Developer of Information Technology and Computing Science. 2018;2(2):802–10.
- 21. Andriyani, Permatasari TAE, Purnamawati D, Putri A, Maududi AA Al. APPLYING CLEAN AND HEALTHY BEHAVIOR BY WUDHU FOR HEALTH BENEFITS. Indonesia J Islam Public Heal. 2021;1(1):1–7.
- 22. Shankhdhar K. Diabetic foot amputation prevention: wudu could be a boon in disguise. Diabetes Foot J. 2021;24(1):26–9.
- 23. Armiati IGK. Reduction in the number of Streptococcus mutans colonies in the oral cavity by ethanol extract of aloe vera leaf bark. Interdental J Dentistry. 2018;14(1):1–4.

- 24. Ernawati KL. KOMBUCHA TEA REDUCE THE NUMBER OF STREptococcus BACTERIA. Faculty of Dentistry, Mahasaraswati Univ, Denpasar. 2018;6–10.
- 25. Djohari M, Lestari R, Hasti S. Identification and test of the inhibitory activity of ethanol extract of areca nut (Areca catechu L.) against gum bacteria isolates. J Researcher Farm Indonesia. 2019;7(2):61–9.
- 26. Da-Young Kwak, Na-Yeon Kim, Hye-Jin Kim, Seung-Yeon Yang, Ji-Eun Yoon I-AH and S-HN. Changes in the oral environment after tooth brushing and oral gargling. Biomed Res. 2019;28(6):1–5.
- 27. Lasmini T. Identification of Mouth Cavity Bacteria of Smokers and Non-Smokers in Pekanbaru. J Anal Health Clinic Science [Internet]. 2018;8(1):17–27. Available from: http://jurnal.univrab.ac.id/index.php/klinikal/article/view/525/361
- 28. Utami VHB, Suryani L. The Effectiveness of the Application of Wudhu in Reducing Germs Numbers on the Hands, Mouth and Nose of Nurses. Med Pearl. 2016;13(1):43–8.
- 29. Manurung DM. The Effectiveness of Gargling with Green Tea (Camellia Sinensis) on the Number of Bacterial Colonies as Asepsis in the Department of Oral and Maxillofacial Surgery, Faculty of Dentistry, University of North Sumatra. University of North Sumatra, Medan. 2020;
- 30. Febriyanti E, Kania TP D, Aspriyanto D. Comparison of Anaerobic Bacterial Colony Numbers in Children's Saliva Gargling with Peatland Water and PDAM Water. Dentist. 2018;II(1):113–7.
- 31. Mahgaidren R. The Effectiveness of Gargling with 15% Forest Honey Solution and 1% Povidone Iodine Solution on Reducing the Number of Bacteria in the Oral Cavity in Faculty Students. 2018; Available from: http://repositori.usu.ac.id/handle/123456789/5178