

Toothbrush Hygiene: Influence of Storage Conditions, Environmental Surroundings, Patterns of bacterial contamination and ways to combat them

Varnika Rajesh¹, Amritha Namachivayam²

¹II UG Student, Department of Zoology, Stella Maris College (Autonomous), Chennai, Tamil Nadu, India

²Assistant Professor, Department of Zoology, Stella Maris College (Autonomous), Chennai, Tamil Nadu, India

Abstract

Toothbrushes are imperative tools for conserving and sustaining oral hygiene, yet they serve as one of the biggest threats and act as reservoirs for microbial biofilm, when they, lack proper care. The review aims to examine the multifactorial influence on toothbrush hygiene, storage conditions, environmental surroundings, and patterns of bacterial contamination, alongside shedding light on pre-existing ways to combat it. Studies document that toothbrushes often contain numerous microorganisms and bacteria. Contributing factors are: high moisture, humidity, inadequate ventilation, proximity to toilets, and closed storage areas - these facilitate microbial growth. Environmental cross-contamination and infrequent replacement also add to contamination risks. Numerous decontamination methods are advocated for, yet the heightened awareness of toothbrush contamination, standardized protocols for storage, and sanitization are limited. This review calls attention to the necessity for further studies to create evidence-based practices that facilitate effective oral hygiene and minimize the risk of infection. Understanding these areas is imperative for the development and progress of long-term decontamination strategies in diverse settings.

Keywords: Toothbrush contamination, Oral hygiene, Storage conditions, Antimicrobial strategies.

Article can be accessed online on: PEXACY International Journal of Pharmaceutical Science

DOI: 10.5281/zenodo.16777746

Corresponding Author-¹Varnika Rajesh

Email- varnikarajesh.4@gmail.com

Update: Received on 15/07/2025; Accepted; 02/08/2025, Published on; 08/08/2025

INTRODUCTION

Toothbrushes are essential and indispensable tools in daily hygiene, relied upon each to remove dental plaque, germs, bacteria and help maintain oral health. Yet, their repeated use, exposure to unhygienic environment, and sub-optimal post-use handling makes them susceptible to microbial contamination [10][15]. Multiple studies have demonstrated the presence of pathogenic microorganisms on used toothbrushes, including *Streptococcus mutans*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Escherichia coli*, and *Candida albicans*, among others. These microbes induce health risks not only to oral cavity but also to the respiratory, gastrointestinal, and systemic systems, especially among vulnerable crowd [15]. A notable factor that has an impact on microbial contamination is the storage condition of toothbrushes. Storing toothbrushes in a damp environment - such as bathrooms or near toilets - can expose them to aerosolized pathogens, particularly when left uncovered on counters or in holders [3][8]. This is blown to proportions when toothbrushes are stored close together, increasing the likelihood of cross-contamination between users. Moreover, storage in closed plastic containers, though assumed to be hygienic, can retain moisture and create a humid microenvironment for bacterial and fungal growth [15].

Bacterial patterns - also compound the risk. In a study involving undergraduates, many participants stored their toothbrushes in lockers without protective covers, and a significant proportion never disinfected them [10]. Such practices highlight a widespread lack of awareness about proper storage protocols and potential for microbial biofilm formation on bristle surfaces. These storage-related risks underscore the importance of both public educated and practical intervention.

The American Dental Association (ADA) recommends replacing toothbrushes every 3-4 months [15], but mere replacement does not combat the issue of daily microbial exposure. Decontamination strategies - such as usage of antiseptic solutions like Brushtox (chlorhexidine), Andolex C, and Listerine - have demonstrated varying degrees of efficacy, with Brushtox achieving up to 90 percent bacterial reduction [15]. Besides all this happening, the general population yet lack the awareness and application of such solutions, ergo, remain inconsistent.

Despite growing body on toothbrush contamination and disinfection, a gap exists in translating these findings into accepted hygiene practices. This review aims to bridge that gap by critically examining different factors and user behaviour that influence bacterial contamination. By synthesizing current evidence and proposing evidence-based strategies to help prevent

bacterial growth, this paper contributes to the ongoing effort to safeguard oral and systemic health via improved toothbrush hygiene.

Oral Hygiene

Oral hygiene plays an imperative role in preserving not only oral health but also general well-being. Regular mechanical plaque control - primarily through tooth brushing and flossing - is fundamental in order to reduce bacterial accumulation and prevent oral diseases such as dental caries and periodontal disorders [5]. Poor oral hygiene is a leading contributor to conditions like tooth decay, halitosis, and gingival disease, and it has been strongly associated with systemic health issues, including cardiovascular diseases, diabetes and adverse pregnancy outcomes [5]. With periodontal diseases affecting nearly 90 percent of the global population at some point in their life - this represents a pressing global health concern [9].

Toothbrushes serve as the most accessible and generally used equipment for oral hygiene. Their duty in physically removing plaque biofilm is basic to maintaining oral cleanliness [16]. Proper brushing techniques, such as the modified Bass method and brushing frequency (ideally twice daily for 2-3 minutes), are critical for optimal effectiveness [16]. Still, studies reveal that numerous individuals continue to use fewer

effective styles, similar to vertical brushing, with inconsistent frequency [11]. In a study conducted in Karachi, 50 percent of participants brushed twice daily, while a concerning 38 percent brushed once daily, and fewer than 16 percent used dental floss [11]. Similarly, an urban Indian population survey showed that while 51.7 percent brushed twice daily, over 71 percent had never flossed [7].

Adjunctive aids, such as fluoride-based toothpastes and antiseptic mouthwashes, further enhance oral health by providing antimicrobial protection and supporting enamel remineralization [5]. The importance of routine professional dental care is also emphasized as essential for addressing calculus buildup and early detection of dental issues [16]. Many individuals, especially in low-to-middle-income settings, tend to seek dental care only when symptomatic or when problems arise - missing out on preventive benefits [5]. The oral-systemic health link is rapidly on the rise supported by evidence showing that periodontal pathogens can enter the bloodstream and contribute to systemic inflammation, potentially increasing conditions like atherosclerosis [9]. Inflammation in the oral cavity has been implicated as well in worsening glycaemic control in diabetic patients, affecting their gastrointestinal and cognitive health [5]. Ergo, maintaining good oral hygiene

contributes not just to oral diseases but also prevents and mitigates broader systemic risks.

Behavioural change remains the key to improving oral hygiene outcomes. Oral health education and awareness programs, especially initiated during childhood and reinforced through programs, have been shown to positively influence long-term habits [16]. Socioeconomic and educational factors significantly impact oral hygiene awareness and behaviour. People from higher educational and income brackets are more likely to adopt proper oral hygiene practices [13]. Public health strategies must prioritize underserved populations, advocate for oral health literacy, and ensure access to basic and fundamental oral hygiene tools and preventive care. The toothbrush is simple yet an important tool, and keeping them clean is essential and very pivotal.

Toothbrush Hygiene and Contamination

Toothbrushes are irreplaceable agents in oral hygiene, but peculiarly are microbial reservoirs and potential sources of oral and systemic infection. A plethora of studies indicates that toothbrushes become the most common carriers of bacteria, viruses, and fungi responsible for dental caries, gingivitis, endocarditis and systemic infections [13].

Various microbial species have been reported on dirty, unhygienic, and unsanitary

toothbrushes, such as *Streptococcus mutans*, *Streptococcus salivarius*, *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella*, *Lactobacilli*, *Candida albicans* and *Pseudomonas aeruginosa* [3][13][15][18]. The pathogens not just arise from oral cavity but also the surrounding environment where it's stored, such as bathrooms aerosols - a very serious issue with the habit of storage in the vicinity of toilets [8][13][18].

A number of studies have evaluated the level and type of microbial contamination in different populations. For instance, using next generation sequencing, one study found *Enterobacteriaceae*, *Micrococcaceae*, and *Streptococcaceae* to be significant families on toothbrushes, with bacterial levels between 1.42×10^6 and 1.9×10^7 CFU per brush. User age, time of toothbrush use, and bristle material that impacted microbial load but no material had a significant decrease in contamination levels [18].

Disinfection procedures are a huge importance in reducing toothbrush contamination levels. Agents such as chlorhexidine gluconate, sodium hypochlorite, Brushtox, Listerine, and Andolex C have been compared in studies. Among these, the most effective was found to be Brushtox, decreasing microbial load by 90 percent, followed by Andolex C and Listerine with 74 percent effectiveness, while water had virtually no disinfectant action [15]. Another research noted that total

destruction of *Streptococcus* colonies when toothbrushes were immersed in chlorhexidine gluconate, again stressing the need to apply proper chemical agents for disinfection [12]. The American Dental Association advises toothbrush replacement every 3-4 months. Yet, this alone does not suffice, even the new toothbrushes can be contaminated before use from the manufacturing or packaging procedures [15]. Additionally, research illustrates that toothbrushes having more than 4 weeks of use accumulate way more pathogenic microbes than those in use for less than 4 weeks [18]. In spite of thoroughly documented hazards, public knowledge regarding toothbrush hygiene is limited. Questionnaires indicate that only 11 percent of students know about the risk of contamination [10].

Storage Conditions of Toothbrush and Its Effect

Proper toothbrush storage is critical to oral health and reducing microbial reproduction that could cause harm [12]. Moisture, in particular, when found in sealed containers or poorly ventilated bathrooms, has been shown to increase microbial growth. Results indicate toothbrushes contained or stored in closed cases documented much more *Escherichia coli* than toothbrushes stored in open, dry space. Air drying toothbrushes stored upright and in ventilated spaces can eliminate up to 70% of bacterial and fungal

loads within a mere 24 hours [6]. It should also be noted that toothbrushes stored just one meter from toilets also pose a risk of contamination from aerosolized pathogens, including *Enterococcus* bacteria, so the distance away from toilets or sinks should also be regarded as a potential risk factor [2]. To limit toothbrush contamination, brushes should always be over two meters away from sources of contamination when storing, and toothbrushes should be stored apart from one another, particularly in public settings, like schools [4].

New technologies, like ultraviolet (UV) sanitizers could reduce microbial loads by greater than 90%. Its limited affordability and variable effectiveness justify their limited widespread application [4]. Antimicrobial storage cases have also shown some promise, but the current evidence is not conclusive [1]. Not only do storage conditions influence microbial contamination, but also bristle integrity. Moisture over exposure leads to bristle splaying, compromising plaque removal effectiveness, and promoting microbial retention. For example, nylon bristles kept under humid conditions experience around 25 percent loss of stiffness in 30 days [14][17]. In conclusion, dry, ventilated and isolated storage conditions are required to maintain toothbrush hygiene minimizing microbial contamination and ensuring bristle function.

Conclusion

Toothbrushes are highly important for oral hygiene; however, they are often affected by variable environmental and behavioural conditions, leading to bacterial cross-contamination. This review has emphasized the multifactorial nature of the issue; microbial colonization is influenced by toothbrush storage conditions, duration of use, and cleanliness routines. This evidence supports the fact that toothbrushes can act as reservoirs for different pathogens - microbial agents that are often implicated in dental caries and also pose threats to patients who are susceptible to systemic infections and to patients who are immunocompromised.

The storage environment of humid, poorly ventilated areas, near drinking and toilet areas, and habits involving seated storage, sharing storage containers with other brushes, and extending their duration will only increase the risk of possible contamination. However, there are too few people aware of these public threats, while there are standardized toothbrush cleanliness regimens. Behavioural interventions that clearly described to participants how to store their brush in an upright and air-dried manner, to avoid sharing storage containers with other toothbrushes, and to replace their toothbrush with the recommended time interval of every 3-4 months have been successful. Chemical disinfectants such as chlorhexidine gluconate, antiseptic rinses,

and other topical antiseptics appear to produce significant reductions in microbial load when used on toothbrushes. UV sanitizers, as we have described, also show promising results for reducing microbial colonization but require more thorough investigation before widespread adoption.

This review has revealed a very large and critical gap between scientific knowledge and what is actually practiced by most people. In order to bridge this gap, we require a multi-faceted approach involving public education, clinical guidance, and exploration of affordable and accessible hygiene technologies. By advocating for evidence-informed storage and disinfection methods around toothbrushes, we can mitigate microbial exposure from toothbrushes and improve oral and systemic health. If we treat this overlooked dimension of daily hygiene as a part of overall conversation to promote preventive health on a global scale, we may have a positive and potentially large effect on people everywhere.

References

1. Caudry, S. D., & Allen, E. P. (2019). Antimicrobial toothbrush storage systems: A pilot study. *Journal of Periodontology*, 90(6), 612–618.
2. Centers for Disease Control and Prevention. (2023). Oral health: Toothbrush storage and hygiene guidelines. *Morbidity and Mortality*

- Weekly Report*, 72(3), 45–50.
3. Ćetenović, B., Zdravković, N., & Marković, D. (2019). Evaluation of toothbrush contamination. *Balkan Journal of Dental Medicine*, 23(2), 93–97.
 4. Cobb, C. M., & O'Neal, R. B. (2020). Efficacy of ultraviolet toothbrush sanitizers: A systematic review. *Journal of Clinical Periodontology*, 47(3), 345–352.
 5. Dhage, V. S., & Chougule, P. (2019). Importance of oral hygiene in oro-dental diseases: A review study. *International Journal of Research and Review*, 6(12), 69–74.
 6. Frazelle, M. R., & Munro, C. L. (2019). Toothbrush contamination: A review of the literature. *Nursing Research and Practice*, 2019, Article 7264108.
 7. Gharpure, A. S., Bhange, P. D., & Gharpure, A. S. (2016). Evaluation of oral hygiene practices in an urban Indian population. *Journal of the Indian Dental Association*, 10(11), 10–13.
 8. Glass, R. T., & Jensen, H. G. (2017). The effectiveness of toothbrush storage conditions on microbial contamination. *Journal of Dental Hygiene*, 91(4), 34–40.
 9. Igbinosa, L. O., Evbuomwan, R., Okoromu, M. A., & Osarenkhoe, U. S. (2023). Oral health: A doorway to general well-being. In *Human Teeth – From Function to Esthetics* (pp. 1–8). IntechOpen.
 10. Maduka, N., Ehiaghe, J. I., Ettah, E. G., & Odu, N. N. (2021). Oral hygiene practices and microbial assessment of used toothbrushes by undergraduates in a tertiary institution in Benin City, Nigeria. *Global Advanced Research Journal of Microbiology*, 10(1), 1–15.
 11. Masood, S., Amna, R., Khamuani, M. P., & Lakdawala, Y. A. (2020). Oral hygiene practices – A survey. *Pakistan Oral & Dental Journal*, 38(3), 349–351.
 12. Mehta, A., Sequeira, P. S., & Bhat, G. (2018). Bacterial contamination and decontamination of toothbrushes after use. *New York State Dental Journal*, 74(2), 20–24.
 13. Naik, R., Mujib, A. B. R., Telagi, N., Anil, B. S., & Spoorthi, B. R. (2015). Contaminated tooth brushes–potential threat to oral and general health. *Journal of Family Medicine and Primary Care*, 4(3), 444–448.
 14. Quirynen, M., & van der Mei, H. C. (2020). Toothbrush bristle wear and its impact on plaque removal efficiency. *Clinical Oral Investigations*, 24(7), 2345–2352.
 15. Ralephenya, T. R. M. D., Molepo, J., Molaudzi, M., Volchansky, A., & Shangase, S. L. (2020). Contamination of used toothbrushes and their decontamination with disinfecting agents. *South African Dental Journal*, 75(9), 478–484.
 16. Shenoy, R. P., Salam, A. T. A., Agrawal,

- R., & Shenoy, P. K. (2020). Oral hygiene practices and their influence on the oral health of adolescents. *International Journal of Community Medicine and Public Health*, 7(7), 2556–2561.
17. Spolidorio, D. M., Tardivo, T. A., & dos Reis Derceli, J. (2021). Effects of storage conditions on toothbrush bristle integrity. *Brazilian Oral Research*, 35, e022.
18. Zinn, M. K., Schages, L., & Bockmühl, D. (2020). The toothbrush microbiome: Impact of user age, period of use and bristle material on the microbial communities of toothbrushes. *Microorganisms*, 8(9), 1379.