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**Original Research**

# MICROBIAL CONTAMINATION OF TOOTHBRUSH IN RELATION TO THEIR STORAGE PLACE

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ABSTRACT

To emphasize the relation between types and level of microbial bio burden of tooth brush and site of storage. **Methods:** Thirty participants were asked to keep their toothbrush inside the bathroom with combined toilet after brushing and another 30 participants were asked to keep the tooth brush outside the bathroom after brushing. The toothbrushes were then examined for certain groups of microorganisms. Data was analyzed using SPSS. **Results:** There was a statistically significant difference in the mean $\log_{10}$ values of plate count of "Mac" between test and control groups. On the other hand, there was no statistically significant difference in the mean $\log_{10}$ values of plate count of "Sabroud" and "Blood" between test and control groups. **Conclusion:** Tooth brushes kept inside the bathroom can be a potent source of contamination with fecal-oral E.colias this could be an unrecognized cause of many health problems whether oral or systemic.

## Introduction

Although, tooth brushes play an essential role in oral hygiene by prevention of biofilm accumulation and accordingly fighting against tooth decay, it can itself lead to dental diseases as well as many other systemic diseases, including septicemia and gastrointestinal, cardiovascular, respiratory, and renal problems, if not properly stored and maintained. [1]In spite of millions of tooth brushes sold throughout the world each year, there is very little public awareness that tooth brush can be contaminated with use. Millions of microorganisms thrive on contaminated tooth brush. Tooth brushes should not be kept in bathrooms especially those which have combined toilet which harbor potential pathogens. Lot of tooth brushes should not be kept in one container; they will rub against each other and spread germs. Moreover, tooth brushes should not be exchanged between individuals, which could happen by mistake if kept in one container. [2]

A review identified multiple concepts related to toothbrush contamination to include contamination, methods for decontamination, storage, design, and environmental factors. It was found that toothbrushes of healthy and oral diseased adults become contaminated with pathogenic bacteria from the dental plaque, design, environment, or a combination of factors.[3]In healthy adults, contamination of toothbrushes occur early after initial use and increases with repeated use. American Dental Association (ADA) in 1996 has recommended the change of tooth brushes after every 3 months. Patients undergoing chemotherapy should change their tooth brushes after every 3 days.[4]Those subjected to major surgery have to change tooth brushes every day and those who are sick should change brushes at the beginning of illness,

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Figure 1.Bacterial isolates from the tooth brush

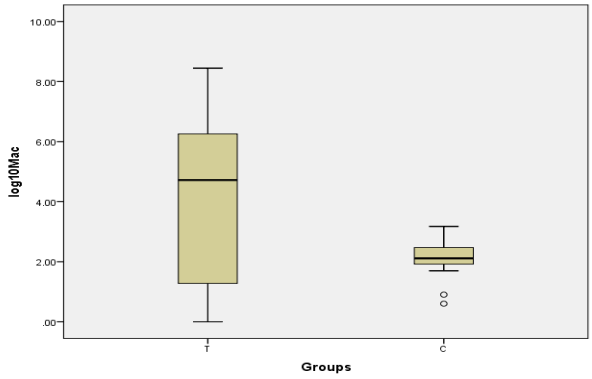



Figure 2. Box plot showing $\log_{10}$ mean values of Mac between test and control groups

when they first feel better, and when they are completely well.[5]The aim of this study is to emphasize the relation between types and level of microbial bio burden of tooth brush and site of storage.

## Methods

A total of 60 participants of Riyadh colleges of dentistry and pharmacy (RCsDP) students were recruited. RCsDP ethics committee approved the protocol for this study. Purpose of the study was explained to the students and informed consent obtained. Test group of thirty participants were asked to keep their tooth brush inside the bathroom with combined toilet after brushing during the study period. Control group of thirty participants were asked to keep the tooth brush outside the bathroom after brushing for the same period. The toothbrushes were then examined for certain groups of microorganisms.

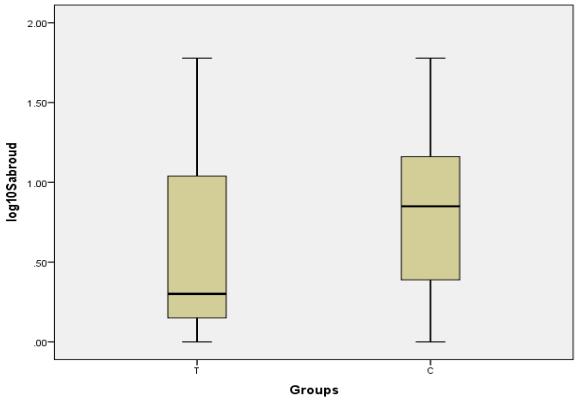
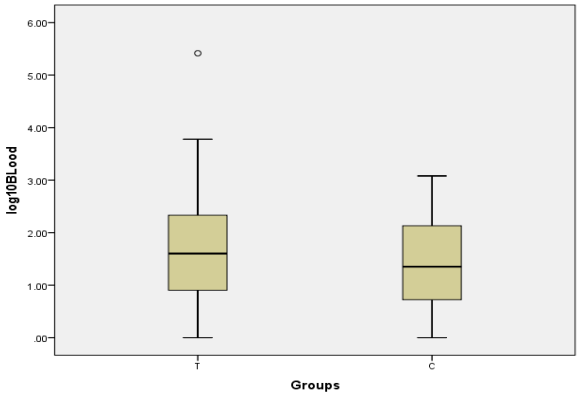



Figure 3. Box plot showing $\log_{10}$ mean values of Sabroud between test and control groups




Tooth brushes were immersed in 10 ml sterile nutrient broth and vortexes for two minutes. Then serially diluted and applied 1ml to plates of selective and specific media for the identification of certain groups of microorganisms, McConkey Agar (McCA) for enterobacteria, Sabouraud Agar (SBA) for yeasts, and Blood Agar for streptococci.[6]After incubation at 35-37 °C for 24-48 hours, microbial growth was examined and quantified comparing the test group with the control group. Data were analyzed using SPSS, Version 21. A p value of $\leq 0.05$ was considered as statistically significant.

## Results

Figure 1 shows bacterial isolates (Gram +, Gram -, and Candida) from the tooth brush. There was a

| Groups | $Log_{10}$ Mac Mean (SD) | t-test for Equality of Means | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | t-value | df | p value | Mean Difference | St Error | 95% Confidence Interval of the Difference | |
| | | | | | | | *Lower bound* | *Upper bound* |
| Test<br>Contol | 4.22(2.83)<br>2.10(0.66) | 2.909 | 30 | 0.007 | 2.11499 | .72699 | 0.63027 | 3.59970 |

Table.1 : Comparison of $log_{10}$ mean values of Mac between test and control groups

| Groups | $Log_{10}$Sabroud Mean (SD) | t-test for Equality of Means | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | t value | df | p value | Mean Difference | Std. Error | 95% Confidence Interval of the Difference | |
| | | | | | | | *Lower bound* | *Upper bound* |
| Test<br>Control | 0.69(0.95)<br>0.82(0.57) | -.284 | 9 | .783 | -.12913 | .45521 | -1.15888 | .90063 |

Table.2: Comparison of $log_{10}$ mean values of Sabroud between test and control groups

| Groups | $Log_{10}$ Blood Mean (SD) | t-test for Equality of Means | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | t value | df | p value | Mean Difference | Std. Error | 95% Confidence Interval of the Difference | |
| | | | | | | | *Lower bound* | *Upper bound* |
| Test<br>Control | 1.69(1.24)<br>1.40(0.89) | .925 | 46 | .360 | .28899 | .31249 | -.34002 | .91799 |

Table.3: Comparison of $log_{10}$ mean values of Blood between treatment and control groups

statistically significant difference in the mean $log_{10}$ values of plate count of "Mac" between test (T) and control (C) groups (p=0.007). The mean $log_{10}$ values were significantly higher in test group when compared with control group (Figure 2). The 95% confidence interval for the mean difference also indicates statistically significant difference between two groups as the interval does not include the value of "0", which is of no difference between groups (Table1).

On the other hand, there was no statistically significant difference in the mean $log_{10}$ values of plate count of "Sabroud" and "Blood" between test (T) and control (C) groups (Figure 3 and 4). The mean $log_{10}$ values of test group when compared with control group were not significantly different with each other (p=0.783) (p=0.360) respectively. The 95% confidence interval for the mean difference also indicated no statistically significant difference between two groups as the

interval include the value of "0", which is of no difference between groups. (Table 2 and 3).

**Discussion**

This study showed a significant difference between the numbers of microbial contaminants of lactose fermenting and non-lactose fermenting enterobacteriaceae between the tooth brushes stored during the period of the experiment inside the bathroom-combined toilet and those used and kept outside as reported by a previous study. However that study was comparing the type of coli form bacteria on the toothbrush and the normal flora of the user of the brush.[4]While there was no significant difference between the number of both candida sp. and streptococci isolated from toothbrushes kept during the period of the experiment inside and outside the bathroom-combined toilet revealing that the effector is the flora of the user and not the external source of splashed bacteria from the environment.

The results of the present study are in comparison with a study by Naik et al.in 2015.[2]The significance of storing and maintaining tooth brushes such as disinfecting or discarding it at regular intervals is important for wellbeing of an individual.[7]A study at American society for microbiology reported that 60% of shared bathroom toothbrushes contain fecal matter. [8] Another study found that diarrhea-causing bacteria from a lidless flush flew as high as 10 inches above the toilet. [9] Furthermore, Contreras et al. in 2010 concluded that closeness to the toilet, aerosols created during toilet flushing and humid environment of the bathroom may facilitate the toothbrush contamination.[10] Studies with larger sample sizes would be beneficial in future studies.

**Conclusions**

Tooth brushes kept inside the bathroom can be a potent source of contamination with fecal-oral E.coli. Dentists play an important role of advising the patients with the protocol of tooth brush usage and the site of storage as this could be an unrecognized cause of many health problems whether oral or systemic caused by fecal-oral E.coli microbes in the environment of toothbrush if it is bathroom-combined toilet..

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