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ORIGINAL ARTICLE

Efficacy of two mouth rinse sprays in inhibiting *Streptococcus mutans* growth on toothbrush bristles

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KEYWORDS

Mouth rinse sprays;
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Chlohexidine gluconate

Abstract *Objective:* To compare the efficacy of two types of mouth rinse sprays (Periogard and Plax) in inhibiting the growth of *Streptococcus mutans (S. mutans)* on toothbrush bristles used by children.

Methods: An experimental comparative study was performed. The sample included 60 children aged 6–8 years with high caries index. Children were divided randomly into 3 groups (20 each) according to materials applied on toothbrush. Each group was further subdivided into 2 subgroups A and B (10 each) according to the laboratory standards for processing microbiological specimens. Each toothbrush was placed in phosphate buffered saline, vortexed then serially diluted. Mitis salivarius bacitracin (MSB) agar plates were inoculated and incubated for 48 h. *S. mutans* colonies were identified by morphology, gram stain and biochemical tests.

Results: Statistically, significant difference was observed between the three groups either when toothbrushes were processed immediately or when processed after 24 h. Group I showed highest bacterial count followed by group III whereas group II showed least bacterial count. Bacterial counts were significantly decreased by time in group I and group III while in group II no significant decrease as both subgroups showed very low bacterial count.

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Conclusions: Although both mouth rinses were effective against *S. mutans* toothbrush contamination, chlorhexidine gluconate proved to be better.

1. Introduction

The utilization of toothbrushes and/or dental floss is necessary to remove dental biofilm and to prevent tooth decay, gingivitis and periodontitis (Chandki et al., 2011). Although toothbrushes are the most common oral hygiene aid used to encourage oral health and stop dental diseases, the toothbrush head due to its complicated structure is particularly susceptible to a more substantial contamination, which leaves it with an infective charge for the next use (Saini and Saini, 2010).

Millions of microorganisms can get trapped in toothbrushes which are usually kept in bathrooms. The existence and endurance of bacteria on toothbrushes filaments might take part in the etiology of dental infections. Unfortunately, proper care of toothbrushes is often neglected due to lack of awareness among the public regarding toothbrushes maintenance and proper care (Queiroz et al., 2013).

Another risk of cross-infection occurs when microorganisms on one toothbrush are transferred to others kept in close proximity or when more than one person shares the same toothbrush (Nascimento et al., 2008). In the world of organ transplant and alteration of immune system, it is essential to count the toothbrush as an origin of possible microorganisms. Usually people traumatize themselves with their tooth brushes; this trauma may become a possible point of access for organisms (Antunes et al., 2010).

Extended use of the toothbrush enables more infection of it by different oral and environmental microorganisms such as Streptococci, Staphylococci, Lactobacilli, Actinobacilli, Actinomycetemcomitans, Candida albicans (C. albicans), Coliform bacteria that are found in bathrooms and herpes simplex virus type 1 (HSV-1) (Nanjunda Swamy et al., 2011). These microorganisms are involved in causing dental caries, gingivitis, stomatitis, stroke, arthritis, chronic infections and infective endocarditis in a human, affecting both general and oral health (Wetzel et al., 2005).

The presence and survival of *Streptococcus mutans* (*S. mutans*) on the toothbrushes depends on the number of filaments per tuft as well as on the number of tufts themselves. Also, the number of microorganisms varies according to the size of the exposed area and frequency of use (Ferreira et al., 2012).

Many chemical agents were evaluated for disinfection of toothbrushes either by spraying or immersion. Soaking the toothbrush in alcohol was one of the first recommended effective procedures for disinfection in 1920 (Cobb, 1920). Later in 1929, Kauffmann tried to place the toothbrush in a sealed container with a preparation including formaldehyde for its disinfection, but this agent proved to have many drawbacks, such as being an irritant agent, having poor penetration, leaving non-volatile residue and the reduction of its activity in the presence of protein (Kauffmann, 1929).

Many studies and comparisons were done to distinguish the most effective, nontoxic, easily implemented method for elimination of bacterial contamination on toothbrushes. These methods showed varying levels of effectiveness. They include liquid chemical disinfectant, coating of toothbrush bristles with antimicrobial agents during manufacturing, wet or dry heat, and radiation (Nelson-Filho et al., 2011; Belanger-Giguere et al., 2011; Zautner et al., 2013). In addition, some studies were concerned about in vivo microbial contamination of toothbrushes, proposing ways for their disinfection (Sato et al., 2005; Balappanavar et al., 2009).

The use of toothbrushes with medicated tufts did not succeed in avoiding infection by cariogenic and periodontopathogenic bacteria (Quirynen et al., 2003). Also, Silver-coating in the present form did not enhance any antibacterial action against remaining bacteria present on the head of the toothbrush (Al-Ahmad et al., 2010).

The objective of this study was to compare the efficacy of two types of mouth rinse sprays; Periogard (0.12% chlorhexidine gluconate) and Plax (0.03% triclosan) in inhibiting the growth of *S. mutans* on toothbrush bristles used by children. The null hypothesis tested was that there was no difference among the effect of the two types of mouth rinse sprays in inhibiting the growth of *S. mutans* on toothbrush bristles used by children.

2. Methods

2.1. Ethical considerations

The Ethical Committee at the Faculty of Dentistry, Alexandria University, Egypt approved the research protocol. A signed informed consent was obtained from all participants' parents or guardians before the investigation. The informed consent procedure was approved by the Institutional Ethical Committee.

2.2. Study design

An experimental comparative study was performed.

2.3. Sample size estimation

A power of 80% was used to detect a clinically meaningful difference of *S. mutans* finding of toothbrushes after use of Periogard and Plax = 13.2%, precision of 7%, effect size = 0.95 and alpha error = 0.05. The minimal required sample size was found to be 60 persons over groups with allocation proportion = 1:1.

2.4. Study sample

Sixty participants were enrolled in this study (32 males and 28 females). They were selected from the outpatient clinic of

Pediatric Dentistry and Dental Public Health Department, Faculty of Dentistry, Alexandria University, Egypt.

Eligibility criteria for participants: (1) Children's age ranging between 6–8 years, (2) They did not receive antibiotic treatment for a period of 30 days before sample collection, (3) Children with high caries index (dfs and DFS scores more than 5), (4) All primary molars and first permanent molars were present.

Sample grouping: The sample of 60 children was divided randomly into 3 groups (20 each) according to materials applied on toothbrush (Colgate Palmolive, China) as follows: Group I (control), used fluoridated toothpaste (Colgate Palmolive, Egypt). Group II (Periogard), used fluoridated toothpaste, followed by spraying the toothbrush with Periogard mouth rinse (Colgate, 2017) (Colgate Palmolive, USA). Group III (Plax), used fluoridated toothpaste, followed by spraying the toothbrush with Plax mouth rinse (Tanomaru et al., 2008) (Colgate Palmolive, USA).

Each group was further subdivided into 2 subgroups A and B (10 each) according to the laboratory standards for processing microbiological specimens: Subgroup A, where microbiological processing was done within 1–2 h of tooth brushing. Subgroup B, where microbiological processing was done after 24 h of tooth brushing.

2.5. Method

All children included in the study were instructed not to brush their teeth for one day before sample collection. An operator performed tooth brushing using toothbrushes with standardized dimensions, bristles, and trademarks for all the children and a microbiologist who was blinded to materials applied on toothbrush performed the identification of *S. mutans* colonies by morphology, gram stain and biochemical tests. The microbiologist was a faculty staff member from the Microbiology Department, High Institute of Public Health, Alexandria University. Regarding *S. mutans* identification and counting, intra-examiner agreement was determined using the Kappa statistic and was considered excellent (K = 0.92).

For each child, teeth were brushed for one minute using Roll technique (Hughes and Dean, 2016). A standardized size (0.1 ml) of fluoridated toothpaste was used. After brushing, the toothbrushes were treated as follows: Group I (control), each toothbrush was rinsed with 20 ml of sterile distilled water. Group II (Periogard), each toothbrush was rinsed with 20 ml of distilled water followed by spraying the toothbrush 6 times by Periogard mouth rinse from a 5 cm distance (Nascimento et al., 2008). Group III (Plax), each toothbrush was rinsed with 20 ml of distilled water followed by spraying the toothbrush 6 times by Plax mouth rinse from a 5 cm distance.

Microbiological processing: After tooth brushing, the used toothbrushes were divided into 2 subgroups: Subgroup A, each toothbrush was placed immediately (within 3 min) in test tube containing 20 ml phosphate buffered saline (as a transport media) and all toothbrushes were transported to the laboratory and processed within 1 or 2 h. Subgroup B, the toothbrushes were kept in rank in room temperature in the laboratory for 24 h. They were placed in an upright position keeping head up. After 24 h, each toothbrush was placed in test tube containing 20 ml phosphate buffered saline for processing.

Preparation of mitis salivarius bacitracin agar plates (selective media for S. mutans) (Momeni et al., 2014)*:* Ninety grams of mitis salivarius dehydrated agar (Difco Laboratories, Becton, USA) was mixed with one liter of distilled water and 20% w/v sucrose (El-Nasr Chemicals, Egypt). The mix was boiled for one minute then autoclaved for 15 min at 121 °C. When temperature of the mix reached 50–55 °C, 1% potassium tellurite (Difco Laboratories, Michigan, USA) and 0.2 U bacitracin (Sigma Chemicals, St Louis, MO, USA) were added. Agar was poured in plates, sealed and stored at 4 °C to prevent contamination till use.

Sample culturing (Momeni et al., 2014)*:* The test tube of each toothbrush was vortexed by vortex machine for 30 s to disperse bacteria, then serially diluted (ten-fold) from 1:10 to $1:10^3$. Each toothbrush had four plates. Fifty μL volume of each dilution was pipetted by automatic pipet onto each agar plate and evenly distributed on the agar surface using sterile spreaders. Plates were incubated at 37 °C under anaerobic conditions (Anaerobic jar) for 24–48 h.

S. mutans identification: After 48 h, the plates were removed from the incubator and the isolated colonies were examined and identified (Figs. 1–3) based on: (Hardie and Whiley, 2013) (1) Morphological criteria on plates: *S. mutans* colonies showed small, raised, irregularly margined and adherent figure, (2) Gram staining of colonies under light microscope (power of the lens × 40): *S. mutans* colonies are gram positive bacteria that appeared dark blue or violet on gram staining. Colony units organized as pairs or short to medium length chains. Each unit took the form of coccobacillus which was more oval than spherical, (3) Biochemical confirmatory tests: (a) Thioglycollate broth gave positive results which turned from clear to turbid solution, (b) Positive mannitol and sucrose fermentation tests as color turned from red to yellow.

Counting of bacteria colonies: A Petri dish was placed on the electronic pressure pad of colony counter (SC6 Plus, Stuart, UK). A transmission light array with magnifier was used to help in counting colonies. Counted colony-forming units (CFU) were marked with a felt tip pen on the plate cover to discriminate counted from uncounted colonies or to avoid double counting. Touch pressure caused a count to be registered on the digital display and an audible tone confirms each count made. The sensitivity of the electronic pressure pad is adjustable to suit the user. The examining data were recorded.

2.6. Statistical analysis

Data were fed to the computer using IBM Statistical Package for the Social Sciences (SPSS) software, version 20.0 (SPSS Inc., Chicago, IL). The distributions of quantitative variables were tested for normality using Shapiro-Wilk test. For abnormally distributed data, non-parametric test (Mann-Whitney test) was used to analyze two independent populations. Data was expressed using median, minimum and maximum. While on comparing more than two groups, Kruskal-Wallis test was used which was non-parametric ANOVA. Significance test results were quoted as two-tailed probabilities. Significance of the obtained results was judged at the 5% level.

3. Results

Table 1 shows comparison between subgroups regarding median bacterial colonies. There was statistically significant

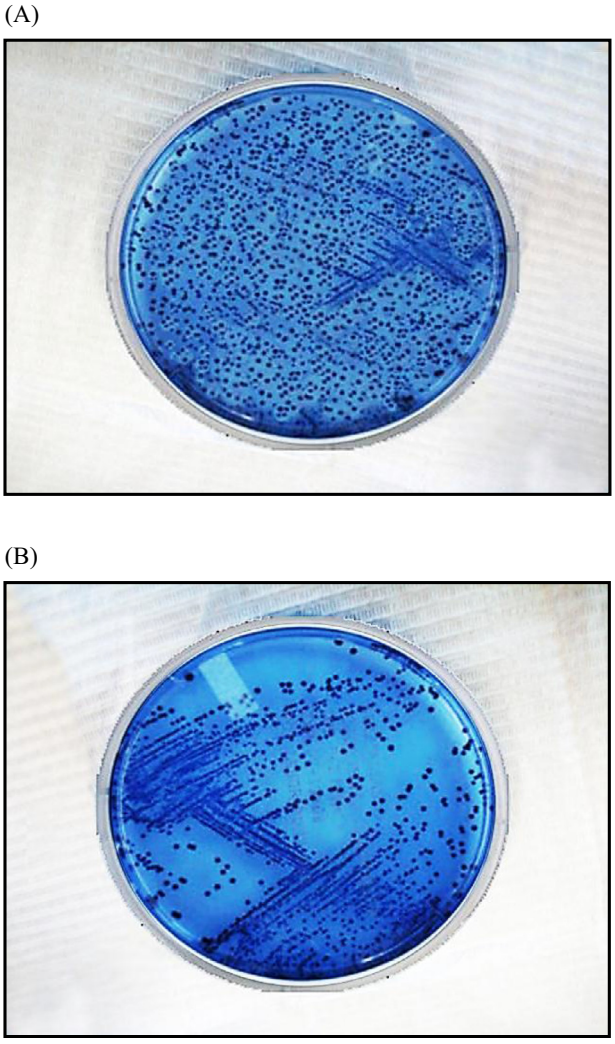


Fig. 1 Identified *S. mutans* colonies in group I (control) plates (Direct plating of the sample): (A) Subgroup A and (B) Subgroup B.

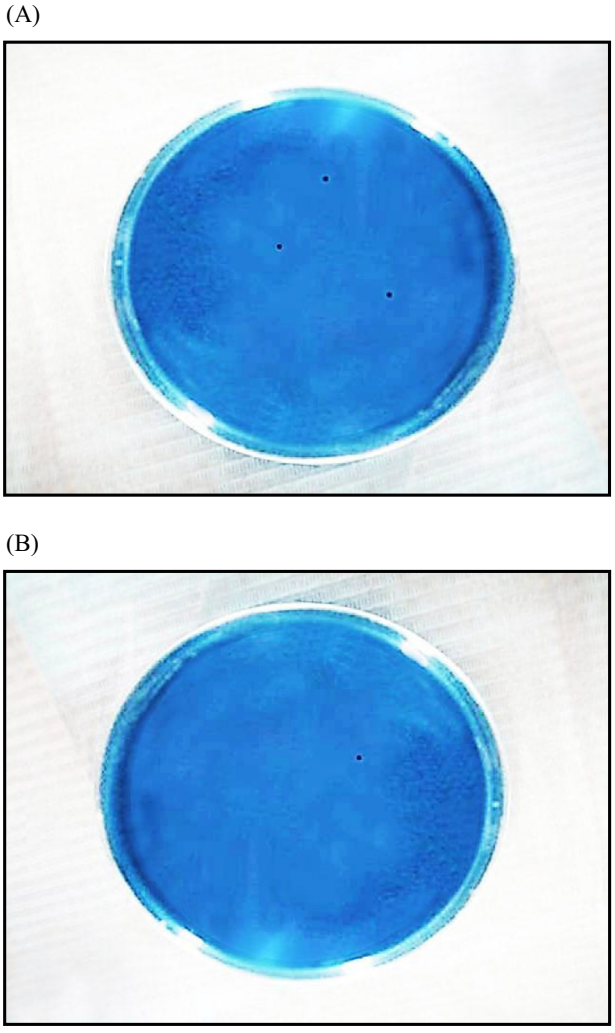


Fig. 2 Identified *S. mutans* colonies in group II (Periogard) plates (Direct plating of the sample): (A) Subgroup A and (B) Subgroup B.

difference between subgroups of group I (control) and group III (Plax) ($P = 0.001$, $P = 0.005$ respectively), while there was no statistically significant difference between subgroups of group II (Periogard) ($P = 0.423$) because the bacterial count in both subgroups was very low due to the bactericidal effect of Periogard mouth rinse (Fig. 4).

In general, the highest bacterial count was observed in group I toothbrushes that were only rinsed with distilled water that processed instantly. Groups II and III toothbrushes that were treated by Plax and Periogard respectively showed lesser bacterial count. While the lowest bacterial count was reported in group II with statistically significant difference.

Table 2 shows comparison between subgroups A (immediate processing) in groups I, II and III regarding median bacterial colonies. Group I (control) showed the highest number of bacterial colonies, while group II (Periogard) showed least bacterial colonies count with statistically significant difference between the three groups ($P = 0.0001$). There was statistically significant difference between subgroups IA and IIA, subgroups IA and IIIA and subgroups IIA and IIIA ($P = 0.0001$).

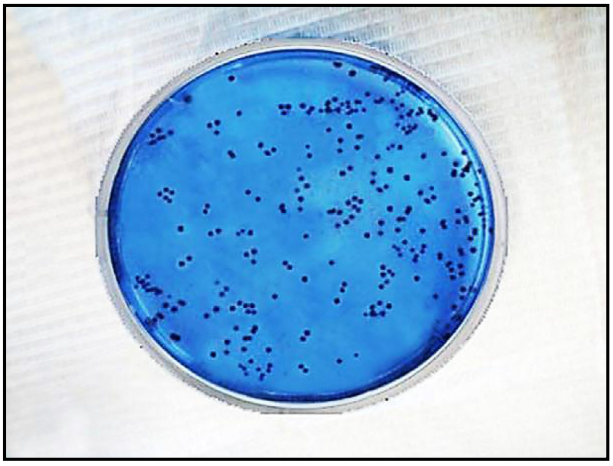
The results of our study revealed that after one-minute tooth brushing, toothbrushes in immediately processed control group IA loaded high count of *S. mutans*, which decreased by time when toothbrushes were kept in clean ventilated area. This might have been due to the need of humidity for *S. mutans* growth.

Table 3 shows comparison between subgroups B (after 24 h processing) in groups I, II and III regarding median bacterial colonies. Group I (control) showed the highest number of bacterial colonies, while group II (Periogard) showed least bacterial colonies count with statistically significant difference between the three groups ($P = 0.001$). There was statistically significant difference between subgroups IB and IIB, subgroups IB and IIIB and subgroups IIB and IIIB ($P = 0.001$).

Our results showed that the bacterial count in control group that processed after 24 h was significantly high when compared to Periogard and Plax groups indicating the need to use toothbrush disinfectant agent to obtain a clean non-infective toothbrush.

By comparing between all three groups, we found that Periogard is the best toothbrush disinfectant because it can prevent

(A)



(B)

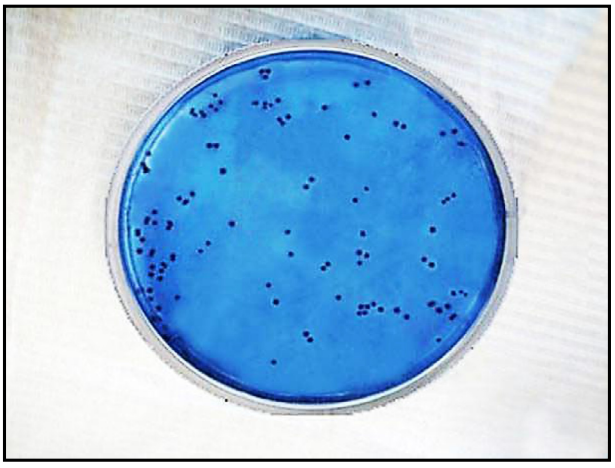


Fig. 3 Identified *S. mutans* colonies in group III (Plax) plates (Direct plating of the sample): (A) Subgroup A and (B) Subgroup B.

toothbrush bacterial contamination within a short period different from Plax that required more time and was less potent than 0.12% chlorhexidine gluconate.

4. Discussion

Periogard (0.12% chlorhexidine gluconate) and Plax (0.03% triclosan) were chosen because they have the characteristics of a disinfectant material which is highly effective, non-toxic, non-irritant, quick and easy to perform. The spray form was also chosen as it is more economic than immersion in chemicals (Al-Talib et al., 2008).

Periogard works against a broad spectrum of gram positive, gram negative organisms and fungi at pH from 5 to 8 and bacterial spores are prevented from germination (Nanjunda Swamy et al., 2011). Sari and Birinci (2007) concluded that chlorhexidine gluconate mouth wash significantly decreased the salivary *S. mutans* count.

Triclosan is a broad-spectrum antibacterial agent. Being a non-ionic molecule makes it suitable to be formulated in traditional toothpastes and mouth washes, but it does not adhere to the oral surfaces for a long period of time and therefore does not provide a prolonged level of anti-plaque activity. In Plax, 0.20% polyvinyl methyl ether/maleic acid (PVM/MA) copolymer is used jointly with triclosan (2, 4, 4′-trichloro-2′-hydroxy diphenyl ether) to increase its retention on oral surfaces (plaque, teeth and mucosa) up to 12 h (Panagakos et al., 2005). Elshibly et al. (2014) found that the use of Plax mouth wash showed significant reduction in salivary *S. mutans* count.

Children age has been limited from six to eight years to insure good cooperation from them during brushing and also to obtain the highest levels of *S. mutans* which were attached to teeth surfaces as in this age all primary and first permanent molars are present.

To standardize high level of *S. mutans* only children with high caries level (dfs and DFS more than 5) were taken. They were informed not to brush their teeth 24 h before sample collection to ensure that the brushes would contain highest level of *S. mutans*.

One of the exclusion criteria was the previous antibiotic treatment in the last month as it gives false low count of oral *S. mutans* than normal (Efstratiou et al., 2007). This exclusion criterion also was taken in consideration in many studies about toothbrush decontamination (Efstratiou et al., 2007; Turner et al., 2009; Nascimento et al., 2012).

In our study, for each child, teeth were brushed by the same operator to standardize the duration and motion force of

Table 1 Comparison between subgroups regarding median bacterial colonies.

Number of bacterial colonies	Group I (control)		Group II (Periogard)		Group III (Plax)	
	A	B	A	B	A	B
Min	200	75	0	0	6	3
Max	1200	500	7	6	263	100
Mean	785.0	271.5	3.00	2.80	111.6	24.9
Median	336.4	164.3	2.53	2.0	125.0	9.5
SD	±336.4	±164.3	±2.53	±1.94	±79.7	±32.8
U	5.36		0.82		10.121	
P	0.001*		0.423		0.005*	

SD: Standard deviation, U: Mann-Whitney test.
* Statistically significant at $P \leq 0.05$.

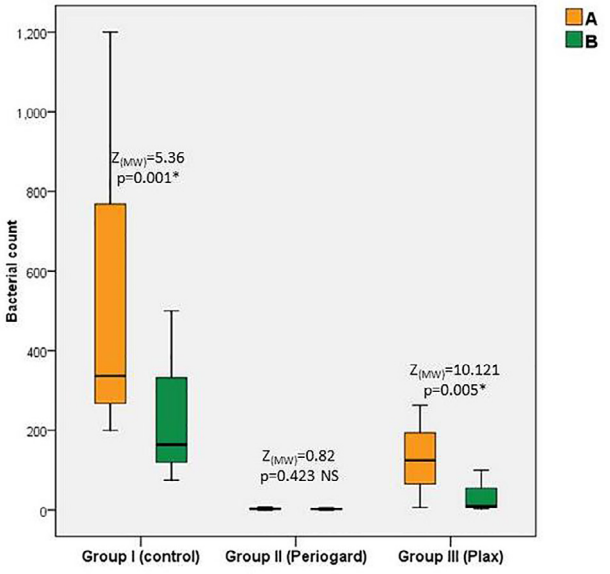


Fig. 4 Box and whisker graph of bacterial count in the studied groups, the thick line in the middle of the box represents the median, the box represents the interquartile range (from 25th to 75th percentiles), and the whiskers represents the minimum and maximum.

Table 2 Comparison between subgroups A (immediate processing) in groups I, II and III regarding median bacterial colonies.

Number of bacterial colonies	IA (control)	IIA (Periogard)	IIIA (Plax)
Min	200	0	6
Max	1200	7	263
Mean	785.0	3.0	111.6
Median	336.4	2.53	125.0
SD	±336.4	±2.53	±79.7
K	45.090		
P	0.0001*		
U1	8.961		
P	0.0001*		
U2			11.26
P			0.0001*
U3		9.25	
P		0.0001*	

SD: Standard deviation, K: Kruskal-Wallis test, U: Mann-Whitney test, U1: Comparison between subgroups IA and IIA, U2: Comparison between subgroups IA and IIIA, U3: Comparison between subgroups IIA and IIIA.
* Statistically significant at $P \leq 0.05$.

Table 3 Comparison between subgroups B (after 24 h processing) in groups I, II and III regarding median bacterial colonies.

Number of bacterial colonies	IB (control)	IIB (Periogard)	IIIB (Plax)
Min	75	0	3
Max	500	6	100
Mean	271.5	2.80	24.9
Median	164.3	2.0	9.5
SD	±164.3	±1.94	±32.8
K	6.852		
P	0.001*		
U1	22.8		
P	0.001*		
U2			6.58
P			0.001*
U3		0	
P		0.001*	

SD: Standard deviation, K: Kruskal-Wallis test, U: Mann-Whitney test, U1: Comparison between subgroups IB and IIB, U2: Comparison between subgroups IB and IIIB, U3: Comparison between subgroups IIB and IIIB.
* Statistically significant at $P \leq 0.05$.

brushing. The fluoridated toothpaste was used in this study to simulate normal brushing condition; this agreed with the opinion of Himaratul-Aznita and Fathilah (2006) and Macari et al. (2011). In contrast, Nelson-Filho et al. (2006) and Nascimento et al. (2008) did not use toothpaste in their studies as they considered it as a disinfectant material that may affect toothbrush bacterial count results.

According to the American Dental Association (ADA) recommendation, pea size toothpaste is the appropriate amount that should be used for 6–8 years children (American Dental Association Council on Scientific Affairs, 2014). In the present study, a syringe was used to standardize the pea size amount and it was equivalent to 0.1 ml.

Periogard mouth rinse, Plax mouth rinse and the toothpaste used in the present study were chosen to be manufactured by the same company (Colgate) to ensure the same level of quality standards of these products.

In accordance to the study by Sato et al. (2005), following brushing, each toothbrush was washed with distilled water instead of tap water to guarantee that no microorganisms would be present in the water rinse that could affect the results, and thus mask the real contamination level.

In the present study, time was considered as an important variable and its relation with bacterial count was evaluated. In each group the treated toothbrushes were subdivided into two subgroups A and B according to time of microbiological processing. In group I (control), immediate microbiological processing (subgroup A) was giving us an idea about initial bacterial count without any disinfectant. This initial bacterial count was considered as a reference count to which short duration (3 min) Periogard and Plax efficacy against *S. mutans* was evaluated. For subgroup B, uncapped treated toothbrushes were placed in upright position for 24 h after brushing and before microbiological processing to simulate normal storage conditions in-between brushing time. Toothbrushes were kept in clean ventilated area to prevent their contamination from any other sources. By comparing between immediate processing (subgroup A) and 24 h before processing (subgroup B) in the three groups, time variable can be evaluated.

For each toothbrush in transport media, ten-fold serial dilution was done. It was used to decrease the difficulty in counting aggregated bacteria in plates. By working back from

an easily counted plate and utilizing the proper dilution factor, the number of bacteria in the first plate (original concentration) was calculated (Bogdanov et al., 2014). The technique of serial dilution was more beneficial with group I (control) as bacterial count in the first plate was difficult to be counted. While in group II (Periogard) and group III (Plax) bacterial count was easily identified from first plate.

Mitis salivarius bacitracin (MSB) was used as a selective media for *S. mutans*. Bacitracin antibiotic in MSB media suppress all types of bacteria but allow *S. mutans* to grow. Sucrose in MSB media increases the adherence of *S. mutans* to agar plate (Wan et al., 2002). The anaerobic jar system was used to create an oxygen-free environment that stimulates growth of facultative anaerobic *S. mutans* (Wang et al., 2011).

The results of our study showed that after one-minute tooth brushing, toothbrushes in immediately processed control group IA loaded high count of *S. mutans*. This count was significantly decreased by time when toothbrushes were kept in clean ventilated area for 24 h (group IB). This might have been due to the need of humidity for *S. mutans* growth. Our finding agreed with Sogi et al. (2002) who found that bacterial count decreased by time when toothbrushes were kept in ventilated area. Also, Borso et al. (2004) concluded that toothbrushes when covered increased humidity and so increase bacterial retention on toothbrushes rather than ventilated uncapped brushes.

However, bacterial count in control group that processed after 24 h was still significantly high when compared to group II (Periogard) and group III (Plax) indicating the need to use toothbrush disinfectant agent to obtain a clean non-infective toothbrush. Many studies were in consistence with this finding, in which normal dryness of toothbrushes was not enough to decontaminate them (Nascimento et al., 2008; Nelson-Filho et al., 2011).

In group II (Periogard) both subgroups showed no statistically significant difference between the bacterial count as both were considered very low. This was in accordance with Nelson-Filho et al. (2006) study as 0.12% chlorhexidine gluconate effectiveness surpassed Brushtox which was the most commercially famous toothbrush disinfectant. In chlorhexidine gluconate (Periogard), chlorhexidine salts were dissociated and released the positively charged chlorhexidine cation. Its antibacterial effect was a result of adherence of this cationic molecule to negatively charged bacterial cell walls. At low concentrations of chlorhexidine, this result in a bacteriostatic effect, while at high concentrations as in our study, it has a bactericidal effect which resulted in membrane disruption and cell death (Leikin and Paloucek, 2008).

In group III (Plax) there was a statistically significant decrease in the bacterial count after 2 h from application of Plax disinfectant spray on toothbrushes than immediate processing. Nevertheless, Plax (triclosan) bactericidal effect was less than Periogard (Kumar et al., 2013). Similar result was recognized by Nascimento et al. (2008). This study was the only one to compare between Periogard and Plax in spray form as toothbrushes disinfectants. Periogard also proved its worth in Rodrigues et al. (2012) study as a toothbrush disinfectant agent.

By comparing between all three groups, we concluded that Periogard is the toothbrush disinfectant of choice as it can counteract toothbrush bacterial contamination within a short period (3 min) as it reached up to 99.7% and long lasting for 24 h in contrast to Plax that needed more time (from 85.8% efficacy in immediate processing to 96.8% after 24 h) and was less effective than 0.12% chlorhexidine gluconate.

Generally, the highest bacterial count was noted in group I toothbrushes that were only rinsed with distilled water that processed immediately. Groups II and III toothbrushes that were treated by Plax and Periogard respectively showed lower bacterial count. While the lowest bacterial count was reported in group II with statistically significant difference, thus rejecting the null hypothesis.

Furthermore, Nascimento et al. (2014) in their study found that 8 h immersion in Periogard solution was highly effective against bacterial toothbrush contamination. According to their study we can determine that Periogard in spray form was considered more economical than 8 h immersion in Periogard solution with the same efficacy.

This study had a limitation which is the difficulty to accurately standardize the base line bacterial count between all groups due to individual variability. To overcome this difficulty in further studies, in vitro artificial contamination of the toothbrushes by standardized count of *S. mutans* need to be performed.

5. Conclusions

Based on this study's results, the following conclusions can be made:

1. Periogard (0.12% chlorhexidine gluconate) and Plax (0.03% triclosan) significantly reduced *S. mutans* count on toothbrushes.
2. The effect of Periogard was more significant than Plax in reducing bacterial count.

Conflict of interest

The authors have no conflict of interest to declare.

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