

Effectiveness of a single-tuft toothbrush for control of newly formed dental biofilm

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Abstract

Aim: To compare the effectiveness of a single-tuft toothbrush (STB) with conventional toothbrushes (CT) to control dental biofilm neof ormation in the dentogingival area. **Methods:** For this cross-sectional prospective blind study, 20 periodontally healthy subjects were selected and randomly divided into 4 groups: STB; CT; CHX - chlorhexidine mouthwash (positive control) and PS - placebo mouthwash (negative control). The subjects were instructed to use only the assigned care method for 72 h with a 7-day washout period between experiments. The evaluated parameters were visible and disclosed plaque indices (PI and DPI), gingival bleeding index (GBI) at baseline (T-0) and at the end of each experimental period (T-72). **Results:** Data analysis demonstrated that at T-0 no difference was observed for any of the parameters ($p>0.05$); after 72 h, CT, STB and CHX showed equivalent effectiveness at controlling biofilm. When the PI data were analyzed, between T-0 and T-72, STB was similar to CT and CHX ($p<0.05$), whereas for DPI, STB was significantly superior to the other methods. Except for PS, all methods yielded similar results for GBI ($p<0.05$). **Conclusions:** The tested STB was effective at controlling short-term dental biofilm neof ormation on the dentogingival area.

Keywords: Dental Plaque. Biofilms. Oral Hygiene.

Introduction

Gingivitis is primarily caused by dental biofilm, which must be controlled in order to achieve and maintain periodontal health¹. The bacteria in biofilm are mostly in balance with the host, which denotes a state of persistent periodontal health. When such homeostasis is broken due to inadequate oral hygiene, gingivitis settles in, which may progress into periodontitis². From the pathophysiological viewpoint, no individual is immune to gingivitis, provided biofilm is allowed to accumulate over time, breaking the gingival homeostasis.

When intrasulcular homeostasis is broken, visible clinical changes begin to emerge, such as spontaneous gingival bleeding, bleeding on brushing, erythema, swelling and changes in gingival texture^{2,3}. It is therefore paramount to concentrate efforts at tackling the root of the problem using methods of oral hygiene and consequently halting dental biofilm formation⁴. The gold standard for prevention of gingivitis is mechanical removal of biofilm by regular toothbrushing⁴⁻⁶. For hygiene to be performed according to the instructions by dental professionals, biofilm control strategies must be tailored to the needs of each individual.

Toothbrush and toothpaste are undoubtedly the most widespread tools for mechanical removal of plaque and debris from the tooth surface. Thus, to meet specific individual needs several devices were developed, for instance, interdental brushes and dental floss for interdental areas, and single-tuft brushes, for intra-sulcular and even buccal/lingual/palatal areas⁷⁻¹⁰.

In normal circumstances, teeth cleaning solely with a conventional toothbrush will not remove biofilm equally from all surfaces¹¹. Complementation is therefore

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required using auxiliary devices, such as dental floss or tape, interproximal brushes and/or single-tuft brushes, according to the shape, size and access to the cleaned site. Single-tuft brushes are delicate and may be advised for specific areas. Usually, they are recommended for difficult to access sites, such as furcations, distal surfaces of molars, areas of amputated roots, buccal or lingual surfaces with irregular gingival margin, crowded areas and proximal surfaces of isolated teeth¹². Additionally, because it is relatively uncomplicated to direct STB towards the gingival sulcus, they may be the most effective method to remove biofilm from deep pockets.

The flowchart of evidence on the role of supragingival bacterial biofilm is complete when biofilm control leads to gingival health¹. Evidence derived from large cohort studies have demonstrated that high standards of oral hygiene will ensure stability of the periodontium¹³. Both short-term and long-term cross-sectional as well as longitudinal studies have shown that the incidence of gingivitis and biofilm accumulation still seems to be high even among the adult population that brush their teeth frequently¹⁴.

Despite STB being highly recommended by periodontal specialists, there are few studies¹⁵⁻¹⁷ demonstrating the effectiveness of such tools at controlling dental biofilm at crevicular sites. The aim of this study was to compare the effectiveness of single-tuft brushes against the gold standard, namely conventional toothbrushes, at controlling newly formed biofilm at the dentogingival area of healthy individuals.

Material and methods

Sample selection

This study was approved by the Ethics Committee of the São Leopoldo Mandic Dental School, Campinas / SP, protocol #356827/2013. Sample size was based on previously published studies of similar design^{18,19} and consisted of 20 dental students from the São Leopoldo Mandic Dental School.

The inclusion criteria were: systemically and periodontally healthy subjects (probing depth \leq 3mm and no gingival bleeding)²⁰⁻²¹, aged between 18 and 30 years with a minimum of 20 remaining teeth, who agreed to participate in the study. Exclusion criteria were: presence of cervical restorations, antimicrobial therapy for any medical or dental condition within 6 months prior to the trial, use of drugs known to affect the periodontal environment (anti-inflammatories, pain-killers, contraceptives, anticonvulsants, immunosuppressants, cyclosporin, anticoagulants and calcium channel blockers) also within 6 months prior to the baseline periodontal examination, orthodontic treatment or devices, pregnant women and breastfeeding mothers.

Study design

Two types of toothbrushes were compared: a single-tuft brush (Bitufo® - Hypermarcas, Senador Canedo - GO, Brazil) and a conventional toothbrush (Bitufo® - Hypermarcas, Senador Canedo - GO, Brazil). 0.12% chlorhexidine mouthwash (Bitufo®,

Senador Canedo - GO, Brazil) was used every 12 h as a positive control, whereas a placebo solution (Bitufo®- Hypermarcas, Senador Canedo - GO, Brazil) with similar features as the chlorhexidine mouthwash, but without the active ingredient, was used as a negative control. The groups were defined as follows: STB – single-tuft brushes (test), n=20; CT - conventional toothbrush (gold standard), n=20; CHX - chlorhexidine mouthwash (0.12% - positive control), n=20; and PS - placebo solution (negative control), n=20. During each experimental phase, the subjects were instructed to use solely the method designated to their group, excluding any other additional cleaning strategy. Each experimental phase lasted 72 h with a 7-day washout period in between, in order to avoid a possible residual (carryover) effect of the previous treatment method. During this washout period, all volunteers used a standard toothbrush and toothpaste provided by the researchers.

The mouthwash solutions were packed and coded in order to prevent identification of the used product. The codes were revealed only when the study was complete.

Clinical Experimental Phase

Following patient selection, a clinical oral examination was performed by a single examiner (IH), trained and calibrated to obtain the following initial clinical parameters: visible plaque index (PI), disclosed plaque index (DPI) and gingival bleeding index (GBI), according to Ainamo and Bay²², as shown in Figure 1 (A, B and C, respectively). In addition, periodontal evaluation was performed, which included probing depth (PD), gingival recession and clinical attachment level, in order to assure absence of gingivitis clinically. Subsequently, professional biofilm removal was performed on each volunteer using a rubber cup and prophylaxis paste. Personalized instructions for toothbrushing were given individually and verbally by another researcher (CV), according to brush (conventional and STB) and solution (placebo and chlorhexidine). Only during the washout period were the subjects encouraged to apply other conventional oral hygiene methods, such as dental floss or tape.

Following the trial phase, a second professional prophylaxis session was performed. The volunteers were then randomly assigned (using a computer list) to their respective sequence of oral hygiene methods, observing the 7-day washout period²⁰. PI, DPI and GBI were recorded both at the beginning and at the end of each trial period.

Statistical Analysis

Only the subjects who completed the study (n=18) were considered for statistical purposes. Prior to the analysis, the Kolmogorov-Smirnov test was applied to assess normality. For intra-group analysis (between periods) of the data (PI, DPI and GBI), Student's t test was used. For inter-group analysis (between treatments), ANOVA/Tukey tests were applied. BioEstat 5.0 (Sustainable Mamirauá Institute, Belém, PA, Brazil) software was used for statistical calculations. For all analyzes, the significance level was set at 5%.



Fig.1. Intra-oral examination for the following clinical parameters: (A) visible Plaque Index (PI); (B) Disclosed Plaque Index (DPI), and (C) Gingival Bleeding Index (GBI).

Results

Twenty volunteers were selected from March to June 2013. The participants were aged between 18 and 30 years (mean age 21.1 years), 13 females and 7 males. Among the 20 initially selected individuals, 18 completed the study and two were lost to follow-up.

Intergroup analysis at the early experimental stage (T-0) revealed no statistically significant difference between treatments (STB, CT, CHX, PS) for any of the evaluated parameters (PI, DPI, GBI), demonstrating homogeneity between the groups (Figure 2A). After 72 h (T-72) (Figure 2B), a significant difference was observed ($p < 0.05$) only for PS in terms of PI and DPI, but not GBI.

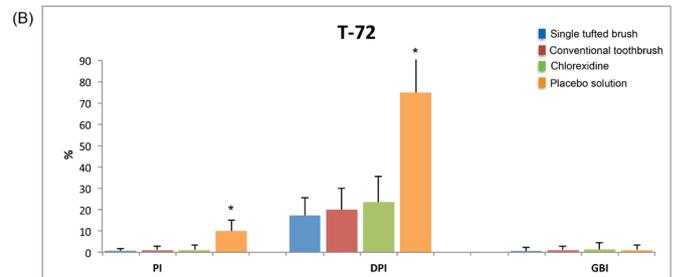
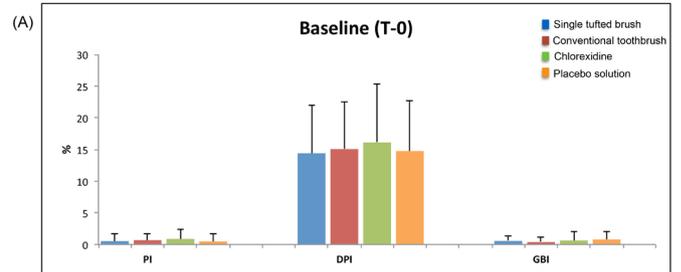


Fig.2. Mean (\pm SD) for the clinical parameters at baseline (T-0) (A), and after 72 h (B) for single-tuft brush (STB), conventional toothbrush (CT), chlorhexidine (CHX) and placebo solution (PS). * indicate significant intragroup differences for the clinical parameters evaluated, by Anova and Tukey test ($p < 0.05$).

In the intra-group comparison between T-0 and T-72, the percentage of accumulated visible plaque (PI) (Figure 3) increased significantly only in the PS group ($p < 0.05$). As shown in Figure 4, assessing the percentage of disclosed plaque (DPI), a significant difference was observed in CT, CHX and PS, while the STB group showed similar results between T-0 and T-72 ($p < 0.005$).

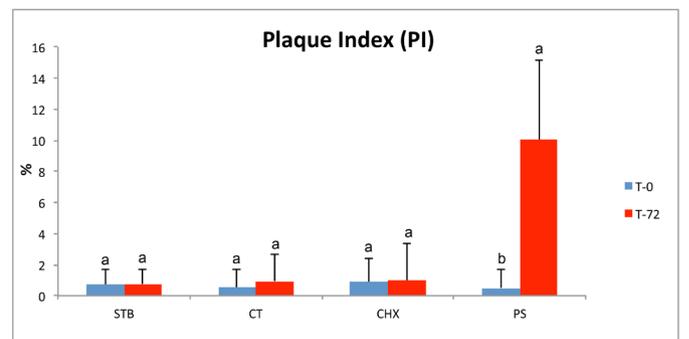


Fig.3. Mean (\pm SD) for visible Plaque Index (PI), for the treatments, at baseline (T-0) and after 72 h (T-72) to single-tuft brush (STB), conventional toothbrush (CT), chlorhexidine (CHX) and placebo solution (PS). Different lowercase letters indicate significant intragroup differences over time, by Student *t* test ($p < 0.05$).

Regarding GBI (Figure 5), no significant differences were observed between T-0 and T-72 for all treatments.

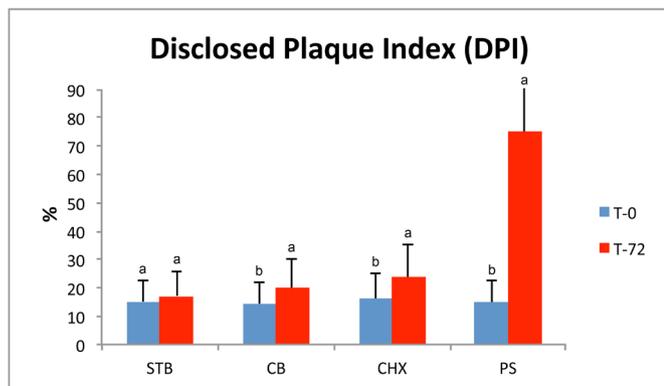


Fig. 4. Mean (\pm SD) Disclosed Plaque Index (DPI) for the treatments at baseline (T-0) and after 72 h (T-72) for single-tuft brush (STB), conventional toothbrush (CT), chlorhexidine (CHX) and placebo solution (SP).

Different lowercase letters indicate significant intragroup differences over time, by Student *t* test ($p < 0.05$).

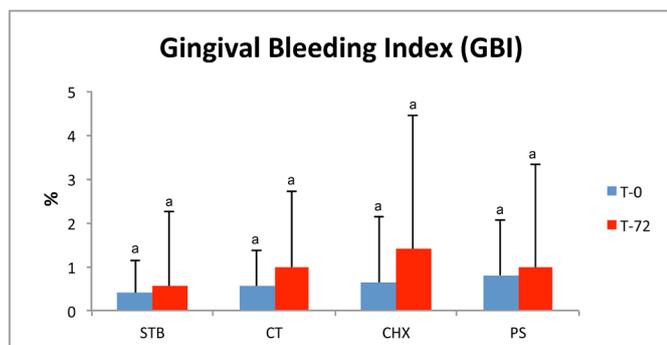


Fig. 5. Mean (\pm SD) Gingival Bleeding Scores (DPI) for the treatments at baseline (T-0) and after 72 h (T-72) to single-tuft brush (STB), conventional toothbrush (CT), chlorhexidine (CHX) and placebo solution (SP).

Different lowercase letters indicate significant intragroup differences over time, by Student *t* test ($p < 0.05$).

Discussion

Faced with the limitations of conventional hygiene methods, new types of brushes have been developed, including electric toothbrushes, single-tuft and interdental brushes^{7,8,23}. Tooth brushing per se is often insufficient to remove dental biofilm, particularly from interproximal and dentogingival areas. In turn, such scenario will demand complementary strategies to tackle biofilm disruption, including the use of dental floss/tape, interdental brushes, mouthwashes, etc. Only a handful of methodologically sound studies have focused on the effectiveness of such methods, especially in the intra-sulcular area. In this context, the present cross-sectional and prospective study aimed to evaluate the effectiveness of single-tuft brushes to control the new formation of bacterial biofilm at this particular site.

The methodological design of the present study allowed for testing all volunteers for all evaluated methods, thus reducing possible biases. Additionally, the inclusion of washout periods minimized the residual effects of the methods used before each new treatment²⁴⁻²⁶. Such study design may counterbalance carryover interferences and provide an estimate of treatment effect with

minimal increase in variance, even if carryover is included in the model.

At baseline (T-0), no difference was observed between treatments for any of the parameters evaluated, demonstrating sample homogeneity between groups. At T-72, however, differences were observed in the placebo group for PI, DPI, but not for the GBI. These results reflect the relatively low hygiene withdrawal period in this sample of periodontally healthy individuals. At 72 h, no clinically evident signs of gingival inflammation could be detected³⁻²⁷, corroborated in the present study by the GBI values.

Additionally, scanning electron microscopy studies evaluating the initial stages of supragingival plaque formation confirmed the presence of a microbial deposit-free zone located between the biofilm layer and the gingival margin²⁸. Bergström²⁹ and Quirynen and coworkers³⁰, when analyzing the initial stages of biofilm formation by sequential photographic records registered clinically this event. In their study, the biofilm-free zone was not stained by plaque-disclosing solutions. Subsequently, studies have shown that a supragingival plaque-free zone persists for up to 96 h^{31,32}.

In this study, both disclosed and visible plaque indices were used to assess plaque scores and biofilm new formation. Visible plaque index (PI) was used to demonstrate gross plaque accumulation, whereas DPI was used to detect low amounts of plaque, as it is a much more sensitive method than PI. Although the use of disclosing solutions in the management of biofilm control can be somehow discouraging for some patients, when combined with index scales, they enable comparisons between new and existing oral hygiene products³³. The choice for disclosing tablets over disclosing solutions was based on the fact that the former is widely used and is likely to be less disturbing of the biofilm, since the latter involves mechanical application of the solution with a cotton swab, which in turn may disrupt biofilm and risk false negatives³⁴.

When confronting the results between T-0 and T-72, only the placebo solution showed a significant difference to PI. It is important to stress again that this index requires a greater accumulation of biofilm on the tooth surface for clinical detection. As for DPI, greater biofilm accumulation for the conventional brush, placebo solution and chlorhexidine solution groups was present, which was not observed in the group that used the single-tuft brush. This may reflect the macrostructural characteristics of the single-tuft brush, which has a small head with bristles directed towards the area to be cleaned and must necessarily be used on a single surface of the tooth at a time, thus resulting in thorough, slower and more rational brushing. These findings agree with those by Ferraz et al.¹⁵, who compared mechanical biofilm control with conventional and single-tuft brushes and concluded that the single-tuft brush group had a lower PI than the conventional brush groups after a 4-week period. The results obtained in the study by Lee and Moon²⁶, which evaluated the effectiveness of single-tuft brushes on the buccal and lingual surfaces of molars also corroborate the findings of the present study. They concluded that difficult access areas could be best reached using this type of brush.

In the present study, chlorhexidine was used as a positive control, as it is independent from an individual's manual

dexterity and is regarded as a gold standard for chemical plaque control due to its bactericidal and bacteriostatic properties³⁵⁻³⁷. A concentration of 0.12% was selected based on a previous study³⁸, which demonstrated that a lower concentration of CHX was just as effective at reducing gingivitis as the traditional 0.20%. Rinsing is easier than either brushing or flossing and takes less time, therefore requiring a shorter attention span. Patients also tend to be more concerned with a "fresh breath" than with plaque and gingivitis levels; consequently, patient adherence to rinsing may be higher in this case than to adequate brushing and flossing (or other cleaning dispositive)³⁹.

Rapp and coworkers¹⁶ compared the Bass technique (conventional brushes) using single-tuft brushes alone or in combination with dental flossing in interproximal areas. Their results showed that, histomorphometrically, the Bass technique and the combination of single-tuft brushes with floss yielded very similar results and slightly better than the Bass-floss combination, while the use of single-tuft brushes without dental flossing showed poorer results. The findings from Rapp et al.¹⁶ do not corroborate those from the present study. It is important to highlight that their analysis involved a histomorphometric evaluation of biopsies from interproximal areas after 28 days. Such different methodological approaches are not directly comparable and any loose parallels can only be established based on extrapolation.

Franceschi and Oppeman¹⁷ evaluated the interproximal cleaning capacity of dental flossing and toothpicking with that of a single-tuft brush and found that both were able to maintain adequate levels of hygiene and gingival health. Under special circumstances, whenever the use of dental floss is not applicable, other methods can be applied due to their popularity, which can make plaque control more acceptable. As there was a combination of single-tuft brushes with toothpicks, the effectiveness of the former on its own cannot be verified, though it may be suggested that their results corroborate those from the present study, since the use of toothpicks alone is generally regarded as inefficient.

In a randomized, single-blinded, controlled clinical trial¹⁰ performed with orthodontic patients, subjects wearing lingual fixed appliances were asked to brush with a triple head or an orthodontic toothbrush alone for one month. Subsequently, they were instructed to brush in conjunction with a single-tufted toothbrush for an additional month. Their teeth were professionally cleaned at baseline and one month later. Similarly to the present study, the authors observed a positive effect of the single-tuft brush: when used alone, the triple-headed toothbrush seemed to have removed dental biofilm more effectively than the orthodontic toothbrush. The addition of a single-tuft brush, however, eliminated differences between groups.

As far as the authors are aware, there have only been a few studies comparing single-tuft brushing with conventional brushing at the dentogingival areas. Caution must be taken with the interpretation of the results from this study, to prevent a hasty notion that single-tuft brushes should be indicated as a sole method for oral hygiene. The present study did not aim to directly influence clinicians into recommending STB as a substitute to conventional mechanical biofilm control methods. Additionally, Lee & Moon²⁶ reported that participants in their study complained that using single-tuft brushes was rather tiresome. Such drawbacks suggest

that single-tuft brushes should be used as an additional tool and not as single method of oral hygiene. Some limitations of this study include the short-term nature of the collected data, making it difficult to forecast long-term results. In addition, the age of the volunteers varied from 18 to 30 years, with occasional differences in motivational levels and possible inherent differences in the anatomy of their dentition, which may, to some extent, interfere with the results. Longer follow-up studies should be performed to evaluate the longitudinal effects of the tested methods.

In general, this study was able to demonstrate the short-term effectiveness of single-tuft brushes, though it must be stressed that they should only be used as an adjuvant strategy to conventional brushing to tackle crevicular areas of buccal and lingual surfaces in the same way as dental flossing is combined to conventional brushing to tackle interdental areas.

In conclusion, the single-tuft brush tested in this study was effective at controlling short-term dental biofilm new formation at the dentogingival area.

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