

PLAQUE REDUCTION AFTER DIFFERENT NUMBERS OF INTERDENTAL BRUSH STROKES - A SPLIT- MOUTH RANDOMISED CONTROLLED TRIAL



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OBJECTIVES. The aim of this study was to evaluate the number of strokes of an interdental brush required to remove interproximal plaque; zero, one, two, four, eight, and sixteen interdental brush strokes were compared.

MATERIALS AND METHODS. This was a split-mouth, single-centre, investigator-blinded randomised controlled trial. The mouths of 24 patients were virtually split into six segments, five of which were to be brushed with different numbers of strokes, and the other serving as a negative control (no brushing). Plaque was recorded before and after interdental brushing at six sites on each of the teeth in each segment (four interproximal plus buccal and lingual surfaces). Segment randomisation was computer-generated, and allocation concealment was performed via sequentially numbered opaque sealed envelopes.

The primary outcome of the study was plaque index reduction, measured dichotomously. Mixed modelling and Tukey's HSD were applied, using the difference in plaque index recorded between baseline and post-brushing as an outcome variable.

RESULTS. All 24 patients completed the study. With respect to baseline, interproximal plaque was removed from 3.6 interproximal sites in controls (no brushing), 7.4 sites after 1 stroke (forth and back), 9.8 sites after 2 strokes, 9.7 sites after 4 strokes, 10.9 sites after 8 strokes, and 13.3 sites after 16 strokes. The difference between stroke counts in terms of plaque removal was statistically significant ($P < 0.0001$).

In particular, a significant difference in plaque removal was detected between one stroke and no interdental brushing (difference: 3.9 sites, 95% CI from 0.8 to 7.0), while there was no statistically significant difference between 1, 2, and 4 strokes.

Likewise, there was no statistically significant difference between 8 and 16 strokes (difference: 2.4 sites, 95% CI from -0.6 to 5.4), whereas 8 strokes removed significantly more bacterial plaque than 1 stroke (difference: 3.5 sites, 95% CI from 0.4 to 6.5).

CONCLUSIONS. Each number of strokes removed significantly more plaque than controls, but eight strokes (forth and back, four in the buccal interdental space and four in the lingual interdental space) appears significantly more effective than one stroke (forth and back, in the buccal interdental space) in terms of plaque removal. Hence, eight interproximal brush strokes (four buccal and four lingual) should be performed in each interdental space to achieve considerable removal of dental plaque.

CONFLICT OF INTEREST STATEMENT. The authors declare that they completely self-funded the study, and that there are no conflicts of interest.

INTRODUCTION

An important factor for the prevention of gingivitis, periodontitis and dental caries is the control of pathogenic agents contained in the bacterial biofilm adjacent to the dental surfaces. Indeed, biofilm build-up is associated with clinical signs of inflammation at both implant and tooth sites¹. This can be minimized by correct daily use of a toothbrush at home². Numerous types of toothbrushes and motivational techniques have been proposed for removal of dental plaque³⁻⁶.

Nevertheless, toothbrushes do not efficiently reach into the interdental spaces between adjacent teeth, resulting in parts of the teeth remaining unclean^{7,8}. In fact, interproximal areas of the dentition are the most difficult areas to clean and keep clean⁹. For this reason, self-care recommendations for interdental cleaning are traditionally given to patients, and a recent systematic review showed that using floss or interdental brushes in addition to toothbrushing might reduce gingivitis or plaque, or both, to a greater extent than toothbrushing alone¹⁰.

Interdental brushes are tools frequently used for interdental cleaning, particularly in older patients¹¹. Several systematic reviews have compared the various types of interdental cleaning procedures, and the evidence suggests that cleaning with interdental brushes may be more effective than flossing for interdental plaque removal¹⁰⁻¹³. Additionally, another study has suggested that brushing interdental spaces might reduce the risk of new cardiovascular events in patients with coronary heart disease¹⁴.

However, despite the utmost importance of the effectiveness of devices used to clean interproximal areas between implants or teeth¹⁵, the literature lacks clear indications on how to use interproximal toothbrushes. Hence, it is difficult for clinicians to advise patients on how interdental brushes should be used. Traditionally, the advice is to move the interdental brush back and forth few times in the interdental space to remove biofilm¹⁶. However, precisely how many strokes are necessary to remove interdental plaque has not yet been ascertained. To our knowledge, no randomised controlled trial has yet investigated this aspect of interdental brushing.

Hence, we set out to evaluate the number of strokes of an interdental brush required to remove interproximal plaque, comparing the plaque indices before and after zero, one, two, four, eight, and sixteen interdental brush strokes in a split-mouth single session randomised controlled trial.

MATERIALS AND METHODS

Trial design

The study was designed, carried out and reported according to the Consort Statement extension checklist criteria for reporting within-person randomised trials¹⁷. This was a split-mouth single-blind randomised controlled trial. Patients' mouths were to be virtually split into six segments (sextants), five of which were to be brushed with a different number of strokes, and the other serving as a negative control (no brushing). The efficacy of the 5 tested methods of interdental brushing was evaluated in terms of plaque removal, recorded by a blinded investigator.

Eligibility criteria for participants

This investigation was carried out at a private clinic located in Campi Bisenzio, Florence, Italy, from June to October 2016. During this period, 24 subjects were consecutively recruited for the study.

The inclusion criteria used for selection were:

- Age >18 years;
- Presence of at least two natural teeth per sextants, or tooth crowns supported by natural teeth or dental implants;
- Baseline full-mouth plaque score (FMPS) >50%.

The exclusion criteria were:

- Subjects with manual inability to perform interdental brushing procedures;
- Pregnant women.

The presence of dental caries and periodontal diseases did not represent exclusion criteria. Informed written consent was obtained from all subjects to be entered in the study, during which the aims and methods of the research were explained.

Outcomes

The primary outcome of the study was plaque index reduction, measured dichotomously¹⁸, at four interproximal surface sites per tooth (mesial-buccal, mesial-lingual, distal-buccal, distal-lingual) in each of the six segments. The secondary outcome was plaque index reduction, measured dichotomously¹⁸, at six surface sites per tooth (buccal, lingual, mesial-buccal, mesial-lingual, distal-buccal, distal-lingual) in each segment.

Sample size

To calculate the sample size, a pilot study was conducted on 3 subjects. The six segments in subjects were randomly allocated to a different method of interdental brush strokes (0, 1, 2, 4, 8 or 16). The sample size was calculated using Bonferroni correction for multiple comparisons ($\alpha = 0.0033$), and the statistical power ($1-\beta$) = 80%. The root mean square error for variance was 2.93 sites. The minimal clinically significant difference was 3.5 sites with or without plaque. On the basis of these data, the number of participants necessary for the study was 22. However, the total number of participants recruited was increased to 24 in order to prevent imbalanced comparisons between the investigated regions of the mouth.

Training and calibration session

One investigator (MG) was required to attend a training and calibration session on 10 participants aimed at: i) instruction and calibration of the measurement of a dichotomous plaque index (presence/absence)^{3,18}, ii) instruction regarding the compilation of the data collection sheets, and iii) preliminary data recording session. The plaque index was measured twice in the six segments of the mouth, with an interval of 30 min. A total of 60 pairs of measurements were recorded and used to evaluate the investigator's reliability via the intraclass correlation coefficient.

Interventions

Each participant was instructed not to perform oral hygiene procedures after the evening meal preceding the study appointment. Patients filled in a questionnaire recording their smoking habits and the frequency of their interproximal device use (dental floss and/or interdental brushes).

The investigator (GM) recorded the baseline plaque index according to O'Leary (1967)¹⁸ at 6 sites per tooth (mesial-buccal, buccal, distal-buccal, mesial-palatal/lingual, palatal/lingual, distal-palatal/lingual), using a staining solution for dental plaque (GC Tri Plaque ID Gel™, Tokyo, Japan). After the registration, the investigator went out of the room.

Another operator (FF) instructed participants on the use of the angled interdental brush, illustrating the procedure using an anatomical model of the mouth; participants were instructed to insert the device at a slight angle (upwards in the lower arch and downwards in the upper arch), and explained the six virtual segments, namely:

- Segment 1: from the distal point of tooth 1.8 to the buccal/palatal point of the 1.3;
- Segment 2: from the buccal/palatal point of tooth 1.3 to the buccal/palatal of the 2.3;
- Segment 3: from the buccal/palatal point of tooth 2.3 to the distal point of the 2.8;
- Segment 4: from the distal point of tooth 3.8 to the buccal/lingual point of the 3.3;
- Segment 5: from the buccal/lingual point of tooth 3.3 to the buccal/lingual point of the 4.3;
- Segment 6: from the buccal/lingual point of the 4.3 to the distal point of the 4.8.

The buccal and the lingual/palatal points of the canines were excluded from the analysis because they represented a limiting zone between the different treated areas.

Only at this point, a sequentially numbered sealed opaque envelope containing the randomisation code was opened, and each segment was accordingly assigned to a different number of interproximal brush strokes. Participants were asked to clean each interdental space as instructed using an angled cylindrical interdental brush (TePe Munhygienprodukter, Malmö, Sweden) of the appropriate size.

The randomisation codes assigned the following treatments to each of the six segments:

- 0 strokes (negative control);
- 1 stroke (one single movement forth and back in the buccal interdental space);
- 2 strokes (two movements forth and back, one in the buccal interdental space and one in the lingual interdental space);
- 4 strokes (four movements forth and back, two in the buccal interdental space and two in the lingual interdental space);
- 8 strokes (eight movements forth and back, four in the buccal interdental space and four in the lingual interdental space);
- 16 strokes (sixteen movements forth and back, eight in the buccal interdental space and eight in the lingual interdental space).

Each participant was provided with a mirror for the purposes of observing and coordinating their brush strokes. The operator (FF) supervised the interdental brushing of the participants, indicating the number of brushstrokes to be performed in each segment, as dictated by the randomisation instructions. The operator also instructed the participants which size of angled interdental brushes to use based on the size of each interproximal space. Specifically, five colour-coded angled interdental brush sizes were available: pink 0.4 mm, red 0.5 mm, blue 0.6 mm, yellow 0.7 mm, green 0.8 mm. The widest interdental brush (green 0.8 mm) was used in the presence of very large interdental spaces (periodontal patients, diastema, missing adjacent tooth, etc.), while the narrowest (pink 0.4 mm) was used in very tight interproximal spaces. After brushing, the blinded investigator (MG) returned to the treatment room to perform a second assessment of the plaque index.

Randomisation and allocation concealment

In each participant, the six segments of the mouth were randomly assigned to be treated by each of five interdental cleaning procedures and in one segment no treatment was performed (negative control treatment). A computer-generated randomisation list assigned the six treatments (five test treatments and one no treatment as control) to the six segments of the mouth. Randomisation was performed in blocks, to avoid imbalanced comparison between the different treatments tested and the investigated regions of the mouth. Treatment allocation was noted on a registration and treatment assignment form kept by the central registrar (NM). Allocation concealment was performed using sequentially numbered opaque sealed envelopes, which were opened immediately before brushing.

Blinding (masking)

The investigator who measured the plaque index was blinded to the treatment allocated.

Statistical analysis

The null hypothesis of the study was that there would be no difference in plaque removal among the five tested treatments and control. Descriptive statistical analysis was performed using frequency and percentage for the qualitative variables, while means and standard deviations were computed for the quantitative variables. A restricted maximum likelihood (ReML) method for fitting a mixed model was calculated, using the difference in plaque index values recorded between baseline and post-brushing as an outcome variable, and the treatment as an explicative variable. Baseline plaque index was designated as a covariate, and the patient as a random effect. Tukey's post-hoc test for multiple comparisons was used to analyse the differences between the treatments in terms of plaque removal, taking into consideration the fact that the six segments examined were located within the patient.

RESULTS

A total of 24 subjects (12 females and 12 males) were enrolled in the study. Their mean age was 61.8 years (standard deviation 11.8) [minimum 25; maximum 77]. Three subjects were smokers. At baseline, the mean floss use across the sample was 0.5 times a day (0.6) [0; 2], the mean interdental brush use was 0.6 times a day (0.7) [0; 2], and the mean FMPS was 79% (12%) [51%; 96%]. No drop-outs were recorded during the experimental phase. Using plaque index as the outcome variable, the results of the investigator calibration showed a reliable intraclass correlation coefficient of 0.86. Descriptive statistics regarding the **plaque index at interproximal sites** are reported in **TABLE 1**. The adjusted plaque difference between pre- and post-brushing took into account the plaque recorded at baseline. Post-treatment differences in adjusted plaque score were found at 3.6 sites in controls (no brushing), 74 sites with 1 stroke (forth and back), 9.8 sites with 2 strokes, 9.7 sites with 4 strokes, 10.9 sites with 8 strokes, and 13.3 sites with 16 strokes. The difference between stroke counts was statistically significant ($P<0.0001$). In particular, Tukey's HSD revealed a significant difference between one stroke and no interdental brushing (difference: 3.9 sites, 95% CI from 0.8 to 7.0), while there was no statistically significant difference between 1, 2, and 4 strokes. Likewise, there was no statistically significant difference between 8 and 16 strokes (difference: 2.4 sites, 95% CI from -0.6 to 5.4), whereas 8

TABLE 1 PLAQUE INDEX AT INTERPROXIMAL SITES

	0 strokes	1 stroke	2 strokes	4 strokes	8 strokes	16 strokes
Number of interproximal sites	18.8 [2.6]	17.7 [3.7]	18.5 [3.2]	18.7 [2.9]	18.4 [2.6]	18.3 [2.7]
Plaque at baseline	17.7 [2.8]	15.0 [4.5]	16.9 [4.1]	16.4 [3.6]	16.7 [3.5]	15.9 [4.4]
Plaque after brushing	13.5 [4.0]	8.4 [4.9]	6.8 [4.1]	6.7 [4.6]	5.7 [4.1]	2.9 [3.0]
Plaque difference	4.2 [3.8]	6.7 [4.8]	10.1 [4.4]	9.7 [3.6]	11.0 [4.7]	13.0 [4.1]
Adjusted plaque difference	3.6	74	9.8	9.7	10.9	13.3
Statistical difference*						

*Tukey's HSD test: the values related to different interdental brushstrokes linked by a continuous bar were not significantly different.

strokes removed statistically more bacterial plaque than 1 stroke (difference: 3.5 sites, 95% CI from 0.4 to 6.5).

Descriptive statistics regarding the **plaque index at all sites** are reported in **TABLE 2**. After brushing, the interproximal plaque was lower than at baseline at 3.5 sites in controls (no brushing), 7.7 sites after 1 stroke, 10.4 sites after 2 strokes, 10.7 sites after 4 strokes, 11.0 sites after 8 strokes, and 13.8 sites after 16 strokes. The difference between stroke counts was statistically significant ($P < 0.0001$). Once again, Tukey's HSD revealed a significant difference between one stroke and no interdental brushing (difference: 4.1 sites, 95% CI from 0.8 to 7.5), but there was no statistically significant difference between 1, 2, and 4 strokes. As before, there was no statistically significant difference between 8 and 16 strokes (difference: 3.0 sites, 95% CI from -0.3 to 6.3), whereas 8 strokes removed significantly more bacterial plaque than 1 stroke (difference: 3.3 sites, 95% CI from 0.02 to 6.6).

DISCUSSION

The objective of the present study was to compare the relative effects on plaque removal of different numbers of interdental brush strokes, specifically zero, one, two, four, eight and sixteen, in a split-mouth single-session randomised controlled trial. To the authors' knowledge, no randomised studies on this issue have previously been conducted.

Our results showed that interdental brushing was able to remove a significant amount of bacterial plaque with respect to control (no brushing), irrespective of the number of strokes. In other words, it is sufficient to perform even one single interdental brush stroke forth and back in the buccal interdental space to achieve significant removal of interproximal plaque. Naturally, however, we also found that increasing the frequency of interdental brushing increases bacterial plaque removal. In particular, eight brush strokes forth and back, four in the buccal interdental space and four in the lingual interdental space, was significantly more effective in terms of plaque removal than one stroke. As there was no statistically significant difference in terms of plaque removal between eight and sixteen brushstrokes, it would appear that eight interdental brush strokes would be the strategy to recommend.

That being said, is interesting to observe that the plaque also decreased in the control segment and at the non-interproximal sites, neither of which were subjected to interproximal brushing. This could be due to the action exerted by the tongue or cheeks, or to bias on the

TABLE 2 PLAQUE INDEX AT ALL SITES

	0 strokes	1 strokes	2 strokes	4 strokes	8 strokes	16 strokes
Number of sites	27.0 [3.7]	25.2 [5.4]	26.4 [4.6]	26.7 [4.1]	26.4 [3.8]	26.2 [4.0]
Plaque at baseline	22.3 [3.4]	19.0 [6.3]	21.5 [5.7]	20.7 [5.6]	20.8 [4.9]	20.2 [6.1]
Plaque after brushing	18.2 [5.1]	12.0 [6.1]	10.8 [5.2]	10.1 [6.2]	9.8 [5.5]	6.5 [4.0]
Plaque difference	4.1 [4.5]	7.0 [4.7]	10.7 [5.0]	10.7 [3.9]	11.0 [4.6]	13.8 [4.9]
Adjusted plaque difference	3.5	7.7	10.4	10.7	11.0	14.0
Statistical difference*						

*Tukey's HSD test: the values related to different interdental brushstrokes linked by a continuous bar were not significantly different

part of the blinded investigator, who may have expected a reduction in bacterial plaque in each segment. Nonetheless, each number of brush strokes was able to remove significantly more bacterial plaque compared to the control areas.

The main limitation of this study was the absence of follow-up. It would be interesting to carry out a longer-term version of this experiment to determine whether the beneficial effect of interdental brushing seen here is maintained over time, or, conversely, whether regular interdental brushing damages the periodontal tissue over time. Moreover, our results cannot be easily extrapolated to other types or shapes of interproximal brush. Further studies should be conducted to explore these issues.

CONCLUSIONS

Although one stroke with an interdental brush seems to be sufficient to remove a considerable amount of plaque, eight interproximal brush strokes (forth and back, four buccal and four lingual) appears to be the optimal strategy among those tested in this single-session trial.

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