



UvA-DARE (Digital Academic Repository)

Prevention and therapy of periodontal diseases and oral malodour

van der Sluijs, E.

[Link to publication](#)

Citation for published version (APA):

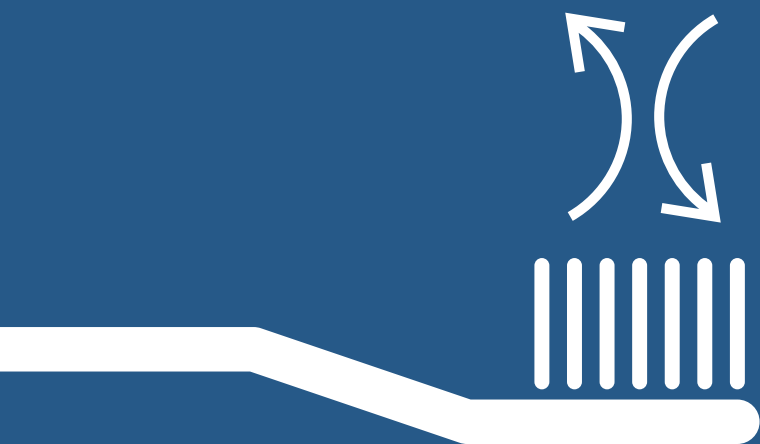
van der Sluijs, E. (2017). Prevention and therapy of periodontal diseases and oral malodour: Brush, rinse and cool DIDES

General rights

It is not permitted to download or to forward/distribute the text or part of it without the consent of the author(s) and/or copyright holder(s), other than for strictly personal, individual use, unless the work is under an open content license (like Creative Commons).

Disclaimer/Complaints regulations

If you believe that digital publication of certain material infringes any of your rights or (privacy) interests, please let the Library know, stating your reasons. In case of a legitimate complaint, the Library will make the material inaccessible and/or remove it from the website. Please Ask the Library: <http://uba.uva.nl/en/contact>, or a letter to: Library of the University of Amsterdam, Secretariat, Singel 425, 1012 WP Amsterdam, The Netherlands. You will be contacted as soon as possible.



2

A specific brushing sequence and plaque removal efficacy: a randomized split-mouth design

E. van der Sluijs

D.E. Slot

N.L. Hennequin-Hoenderdos

G.A. van der Weijden

Abstract

Aim

It has been propagated by dental care professionals to start toothbrushing the lingual aspect of teeth first. In general it is assumed that these surfaces of teeth are more difficult to clean. The evidence to support this recommendation is sparse.

Methods

In this randomized controlled clinical trial using a split mouth design, 46 students were included. Before the visit, participants were requested to refrain from any oral hygiene procedure for 48 hours. First, the plaque index score (PI) was assessed full mouth. Two randomly chosen contra-lateral quadrants were used to start brushing from the lingual aspect first. The opposing two quadrants were used to start brushing from the buccal aspect. After the brushing exercise was completed, full mouth PI was scored again. Sub-analyses were performed for the buccal, lingual, and approximal surfaces.

Results

At baseline, there was no statistically significant difference between the two sets of contra-lateral quadrants ($P = 0.770$). Starting at the lingual aspect of the lower jaw resulted in a 55% reduction of plaque scores in comparison to 58% when the brushing exercise was started buccally. The difference in mean plaque scores between brushing orders was 0.04, which was not significant ($P = 0.219$). None of the sub-analyses revealed any significant differences for the isolated surfaces.

Conclusion

Using a manual toothbrush reduced plaque scores between 55%-58% with no difference between brushing from either the lingual or buccal aspect first. Within the limitations of the present study a recommendation to start toothbrushing the lingual aspect is not supported by the outcome in this young student population.

Introduction

It is well recognized that supra-gingival plaque accumulation, which involves an established bacterial colonization of the dental tissues, is a determinant in the initiation of gingivitis (1). For the control of this complex microbial biofilm, routine oral hygiene is important (2). Toothbrushing is a simple and effective means of removing plaque and therefore has a useful role in the prevention and control of periodontal disease (3). The American Dental Association recommends to brush at least twice a day, for 2 min (4). On average, a 42% plaque score reduction is obtained after toothbrushing with manual toothbrushes (5). Improvements have been made in the design of brushes. During the last decades, different shapes and locations for the handle and bristles have been designed to increase plaque removing efficiency specifically in hard-to-reach places (6).

Instructions on how to brush the teeth focus primarily on the relationship of the bristles to the teeth and gingival tissues as well as on the direction of the brushing stroke (7). Usually, it is not pointed out where to start brushing or in which order the teeth should be brushed. Bass already in 1948 (8) recommended that: 'All the surfaces of all the teeth to which the brush can be applied, should be brushed. A good system is to brush the buccal and labial surfaces of all teeth first, then the occlusal and lingual surfaces of the grinders in all four quadrants and finally the lingual surfaces of the anterior teeth'. A patient-preferred sequence of toothbrushing was evaluated in 1975 by filming adults' brushing their teeth. The upper right buccal area was the most frequent starting position and the most natural sequence was then across the upper anterior to upper left, returning via the lower arch in the opposite direction (9). Macgregor & Rugg-Gunn (10) showed also that the brushing procedure, buccal surfaces first and subsequently lingual surfaces, is a commonly used brushing sequence by both children and adults.

Today, the most common brushing method recommended is the Modified Bass Technique (11), in which it is instructed that the head of the brush is positioned in an oblique direction towards the apex. Filament tips are directed into the sulcus at an angle of approximately 45° to the long axis of the tooth. The brush is moved in a back-and-forth direction using short strokes without disengaging the tips of the filaments from the sulci. On the lingual surfaces of the anterior teeth, the brush head is kept in the vertical direction (12). Closer attention to the sequence of brushing surfaces does seem indicated in view of the common fault of omitting to clean certain area such as lingual and palatal surfaces. O'Hehir & Suvan (7) concluded that apart from greater accumulations of both hard and soft deposits, mandibular lingual surfaces demonstrate more bleeding on probing than other areas of the mouth. Previously in 1994 Axelsson, Lang and Karring (13) concluded that the toothbrush should therefore first be applied to the lingual of the mandibular posterior teeth. Some dental care professionals have dealt with this problem by instructing patients to brush the inside of the bottom teeth first (7). After the biofilm had been removed, they were instructed to add toothpaste to the toothbrush and brush once more around the mouth (14). Twenty-nine private-practice dental hygienists across the United States tested this idea with a total of 126 patients during recall appointments. Reduced bleeding scores and less calculus were found upon re-examination. However, notes from the examiners also indicated that

patients reported brushing longer than usual with this new approach which could be a confounder for the observed improvements (7).

To address the limited evidence for commencing tooth-brushing with lingual surfaces first, the aim of this study was to compare the efficacy of brushing while starting at the lingual aspect of the lower jaw as opposed to starting at the buccal side of the upper jaw.

Materials and methods

Ethical procedures

The study followed instructions based on the Helsinki principles (2013), approximating good clinical practice guidelines, and held the approval of the medical ethical committee of the Amsterdam Medical Center #2014_118. The study was also registered at the Dutch Trial Register (#NTR4604). Before enrolment, all volunteers were given verbal and written information with respect to the aim, rationale and duration of the study. The investigator explained the details of the trial and potential risk involved. All eligible subjects who agreed to participate signed an informed consent form prior to the study procedures.

Sample size

Sample size calculations were performed using the PS Power and Sample Size Programme (15). Earlier data indicated that the difference in response to the primary outcome in matched pairs is normally distributed with standard deviation 0.40 (new toothbrush without dentifrice Rosema 2013) (16). If the true difference in the mean response of matched pairs is 0.18, then 40 participants were needed to be able to reject the null hypothesis that the response difference is zero with a probability (power) of 0.8. The Type I error probability associated with this test of the null hypothesis was 0.05. Additional participants were entered to compensate for potential dropout.

Screening and inclusion

The participants were non-dental students from different universities and colleges in and around Amsterdam. The study took place in the period from June until September 2014 at the Academic Centre for Dentistry of Amsterdam (ACTA), the Netherlands, Department of Periodontology. The study participants were identical to the population of a previous study involving a single brushing exercise (17). The participants had been screened by a dental hygienist (MPCL). To qualify for inclusion, the subjects were required to be ≥ 18 years old, classified as systemically healthy as assessed by the medical questionnaire, periodontally healthy as assessed by scoring the DPSI ≤ 3 minus (18,19), ≥ 5 teeth per quadrant and right-handed brushers. Excluded were those who presented with an orthodontic appliance or a removable (partial) denture, overt caries, any pathological alterations of the oral mucosa, pregnancy and use of medications within 2 weeks before the appointment including antibiotics or chronic use of non-steroidal anti-inflammatory drugs (excluding birth control pills).

Design

This study had a single-(examiner) blind design using a split-mouth model where contra-lateral quadrants were randomly assigned to treatment (20). The study coordinator (EVDS) supervised the assigned brushing procedures.

Randomization and blinding

Every participant received a unique subject identification number. No stratification was applied. Randomization was performed using true random numbers, which were generated by sampling and processing a source of atmospheric noise (21). The randomization code was kept in a sealed envelope in the investigator site file and was only accessible by the coordinator who was therefore responsible for allocation concealment. Participants were instructed not to reveal their intervention in any way to the clinical examiner. Records of the earlier examination were not available to the examiner at the time of re-examination.

Primary outcomes

The main study parameter was the level of dental plaque as measured by Silness & Løe (22) Plaque Index (PI) as modified and described by Van der Weijden et al. (23), where 0 = no plaque, 1 = a film of plaque adhering to the free gingival margin and adjacent area of the tooth, 2 = moderate accumulation of soft deposits on the tooth and gingival margin that can be seen with the naked eye, and 3 = abundance of soft matter on the tooth and gingival margin. Throughout the study examinations were carried out by one and the same experienced examiner (NLHH) under the same conditions.

After the assessment, the participant completed a questionnaire designed to evaluate their opinion and their perception concerning the 48-h non-brushing period, the toothbrush and procedure used. The VAS scoreline had two extremes at either end of the line with the negative on the left side. The participants placed a vertical mark on a 10-cm-long uncalibrated line. The investigator measured the distance along the line from the left extreme to the marking made by the participant (24).

Study procedure

A SMS (Short Message Service) was sent to remind each participant before the visit. A 48-h non-brushing period was used to accumulate an adequate amount of plaque to assess the incremental effect on plaque scores of a single brushing exercise (25). First, a full-mouth dental plaque score was performed. Subsequently, before the brushing exercise a short verbal instruction was given to the participant. The brushing procedure of this study was carefully standardized and controlled. Two randomly chosen contra-lateral quadrants were used to start brushing from the lingual aspect of the lower jaw first (Oral-B indicator P35 soft) followed by brushing lingually of the upper jaw. The participant brushed without dentifrice with a new and dry toothbrush buccal of the lower jaw and buccal of the upper jaw. The procedures were performed under supervision and the participants were guided through the randomized set of contra-lateral quadrants. Each side of the (buccal or lingual) quadrants was brushed for 15 s, in total 30 s per quadrant as controlled by the use of a timer. No mirror was available during the brushing procedure. If necessary, the par-

ticipants were able to expectorate. Rinsing with water afterwards was not allowed. Subsequently the opposing two quadrants were brushed (starting buccally in the upper jaw, followed by buccally in the lower jaw finishing by brushing the lingual of the upper and lower jaws) again using a new dry toothbrush and full-mouth plaque scores were assessed. Finally, the participant completed a questionnaire designed to evaluate their opinion and their perception concerning the 48- hours non-brushing period, the toothbrush and procedure used. Upon completion of the study, participants received monetary compensation for their contribution.

Statistical analysis

The statistical analysis was performed using a statistical package (IBM Corp. Released 2012. IBM SPSS Statistics for Windows, version 21.0. Armonk, NY, USA: IBM Corp) before the allocation was revealed. The primary parameter was the overall dental plaque score. The mean and standard deviation of all readings were calculated for each participant and in addition the mean incremental difference and proportional change were computed. For a subanalysis, all vestibular sites, all lingual sites and all approximal sites were calculated. The central incisors were excluded from the measurement to avoid cross-over effects of brushing the adjacent contra-lateral quadrants. The total mean and variation of the dental plaque scores was analysed by each intervention sequence. Data were tested for normal distribution by the Kolmogorov–Smirnov test and found to correspond with the normal distribution assumption. A paired T-test was applied and values of $P < 0.05$ were accepted as statistically significant. The mean scores and standard deviation regarding the VAS questions were calculated. As the VAS questions were descriptive and did not differentiate between intervention sequences, no statistical analysis was performed concerning these data.

Results

As proposed by CONSORT (26), the study flow is shown in Fig. 1. In total, 50 participants were assessed for eligibility of which 46 participants were included. The mean age of these was 22.5 (2.86) with a range of 20–34. There were more female participants in this clinical trial ($N = 27, 59\%$) than men ($N = 19, 41\%$). No adverse events were reported.

Primary outcome

Table 1 shows the overall mean plaque scores and standard deviation for pre- and post-brushing plaque scores. The split- mouth design implicates that each participant contributed with two baseline and two post-brushing values. At baseline, the plaque scores when starting to brush from the lingual aspect showed a mean of 1.75 which was not significantly different ($P = 0.770$) when compared to start brushing from the buccal aspect. The post-brushing scores, when starting from the lingual, were also not different compared to starting from the buccal ($P = 0.094$). The incremental change in plaque scores was 0.94 when starting from the lingual aspect as compared to 0.98 when starting from the buccal ($P = 0.219$).

Figure 1 Flow chart depicting subject enrollment and measurement (26).

Q = Quadrant

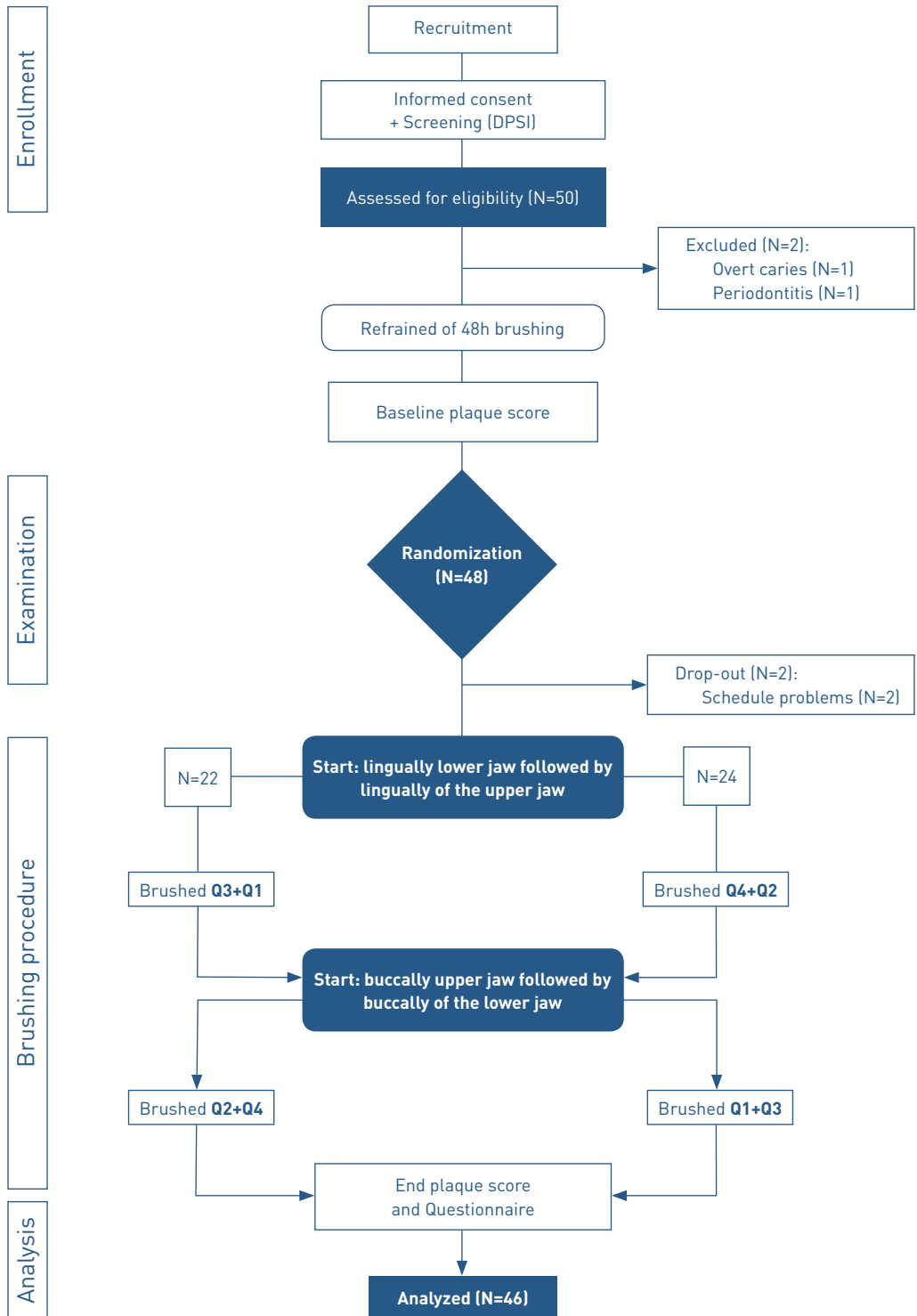


Table 1 Overall dental plaque scores on baseline and end presented by mean and SD (standard deviation) in relation to a specific sequence in brushing order.

N=46	Start Lingual	Start Buccal	Statistical analysis*
Base	1.75 (0.28)	1.74 (0.30)	0.770
End	0.80 (0.38)	0.76 (0.38)	0.094
Incremental difference	0.94 (0.30)	0.98 (0.31)	0.219
% Reduction	55% (19)	58% (18)	

* Paired sample T-test

Subanalysis

Table 2 provides the mean and standard deviations of the sub-analysis at the buccal, lingual and approximal sites separately. None of the comparisons was significantly different between starting from the lingual as compared to starting from the buccal aspect of the quadrants. The plaque score reduction of the lingual surfaces, a particular area of interest for this study, showed an incremental change of 0.94 (0.30) when starting brushing from the lingual aspect, while starting from the buccal aspect the change was 0.98 (0.31). This difference was not significant ($P = 0.219$).

Secondary outcome

Table 3 presents the questionnaire data where self-perception was measured. The participants felt the toothbrush was cleaning their teeth well enough 6.29 (1.93). Bristles' stiffness received a mean VAS score of 5.04 (1.73) (neither too soft nor too rigid). A brushing exercise without the use of dentifrice was judged as unpleasant 3.21 (2.33). The participants judged dry brushing most unpleasant 2.81 (1.58).

Discussion

The ideal brushing technique is the one that allows sufficient plaque removal in the least possible time, without causing any damage to the tissues (27). Different toothbrushing methods have been recommended over time, but many have also been abandoned. There probably is no single oral hygiene method that is correct for all patients. If a patient prefers a specific oral hygiene strategy, the clinician can evaluate this and modify the technique to maximize effectiveness, rather than changing it (12).

In most cases, small changes in the patient's own method of toothbrushing will suffice, always bearing in mind that more important than the selection of a certain method of toothbrushing is the willingness and thoroughness on the part of the patients to effectively clean their teeth (12).

Aim and findings

Many patients spend too little time brushing, or they brush haphazardly especially at the end of the toothbrushing session when lacking of concentration (12). The purpose of this

Table 2 Sub-analysis evaluating dental plaque scores for the buccal, lingual and approximal sites separately. Data are presented by mean and SD (standard deviation) in relation to a specific sequence in brushing order.

Sites	N=46	Start Lingual	Start Buccal	Statistical analysis*
Buccal	Baseline	1.56 (0.54)	1.59 (0.51)	0.199
	End	0.30 (0.30)	0.29 (0.26)	0.720
	Incremental difference	1.26 (0.48)	1.31 (0.41)	0.143
	% Reduction	83% (14)	82% (16)	
Lingual	Baseline	1.55 (0.50)	1.51 (0.52)	0.176
	End	0.45 (0.38)	0.49 (0.33)	0.395
	Incremental difference	1.10 (0.46)	1.02 (0.42)	0.143
	% Reduction	73% (23)	67% (28)	
Approximal	Baseline	1.83 (0.26)	1.84 (0.24)	0.614
	End	0.95 (0.44)	1.01 (0.47)	0.066
	Incremental difference	0.88 (0.36)	0.83 (0.38)	0.178
	% Reduction	49% (21)	46% (23)	

* Paired sample T-test

clinical trial was to evaluate a specific sequence in toothbrushing order with respect to plaque removal while using a manual toothbrush. This particular sequence has been propagated in the dental literature but has never been sufficiently evaluated. The proposition is to start brushing at the lingual aspect of the lower teeth which is suggested to be more effective than starting at the buccal aspect. Primary attention is then directed towards the tooth surfaces that are insufficiently cleaned by most patients. Irrespective of the specific brushing order, dental plaque scores reduced in the present study by at least 55%. The results of this study show no statistically significant difference of change in mean plaque scores ($P = 0.551$) between starting to brush either from the lingual (55% reduction) or buccal aspect (58% reduction).

Dentifrice use

Brushing without the use of toothpaste, also called ‘dry brushing’, was used in this experiment (7). This supposedly would allow the patient to feel the layer of bacterial biofilm before and after brushing more easily. Due to the flavour and wetting agents, toothpaste would make the mouthfeel clean when it is not clean yet (14). Based on the questionnaire, which was completed by the participants after the single brushing exercise (Table 3), it is apparent that the participants judged brushing without the use of dentifrice as unpleasant (mean score 3.21; on a scale of zero to ten where ‘5’ is considered as neutral). Today it is common practice to combine a toothbrush with a dentifrice. The dentifrice provides the fresh mouthfeel and pleasant taste that makes brushing an acceptable or even pleasant experience for some individuals (28). The cleaning capacity of toothbrushing is, however, not enhanced by the use of a dentifrice as has clearly been shown in a recent systematic review (29). Future research is needed concerning the psychosocial factors that might in-

fluence attitude and performance in oral hygiene in relation to dentifrice use (30). Also the potential contribution of adjuvant chemical agents relative to the treatment of malodour, staining, caries, gingivitis, dental plaque, dental calculus, demineralization and dentinal hypersensitivity deserves further studies (31).

Power and clinical relevance

In 1979, MacGregor & Rugg-Gunn (10) reported that of an average 50 s brushing time only 10% of that time was spent on brushing the lingual surfaces. The mandibular lingual section is often the area brushed last (7). Substantial differences are present in the amounts of supra-gingival plaque colonizing the different regions of the dentition. While most individuals have repeatable patterns of plaque on their dentition, a variety of factors influence the development of supra-gingival plaque formation such as saliva composition, diet, tooth position, and tooth surface morphology (2). Plaque scores tend to be greatest in the lingual part of the posterior teeth. In the present comparison, starting to brush from either the buccal or lingual aspect showed that the difference in plaque score reduction was 1% for the buccal surfaces (82–83%). The lingual surfaces, which were a particular area of interest, for this study, showed that when starting from the lingual, plaque scores reduced by 73% as compared to a reduction of 67% when starting from the buccal. Corresponding mean incremental differences were 1.10 and 1.02, respectively, and not statistically significant different ($P = 0.143$).

The *a priori* sample size calculation based on earlier data (16) showed that 40 participants were needed to reject the null hypothesis. Based on the data of the lingual surfaces, the 'post hoc' power calculation showed that with a difference of 0.08 and a pooled standard deviation of 0.44 at least 239 subjects would have been needed to make this difference significant at an alpha level of 0.05. In determining both the statistical significance and the clinical relevance, an assessment of study power is essential and has serious implications for any conclusions that can be drawn. An excessively large sample may show statistical significance even when there is no clinical practicality (32). It is also essential to consider the magnitude of the observed differences between the two treatment groups. Small differences are not likely to be clinically relevant.

Plaque index and brushing duration

In a systematic review by Slot et al. (5), all available studies evaluating a single brushing exercise were evaluated to determine the effect of manual toothbrushing on the plaque score reduction. A total of 59 papers with 212 brushing experiments were evaluated including a total of 10806 participants. The outcome analysed from each of these experiments was the reduction in plaque scores in terms of percentage calculated from the difference before and after brushing. The overall estimate was that a manual toothbrush provides a 42% reduction in plaque scores irrespective of the plaque index used. Considering this, the plaque score reduction in the present study was on average 13–15% higher. However, several indices are available for the assessment of the presence, location and quantity of clinically evident bacterial plaque. These indices are used as a marker by clinicians to monitor the oral hygiene performance and compliance of patients (1). Separating the results

Table 3 Questionnaire related to the self-perception of the participants using a Visual Analogue Scale (VAS) presented as mean and standard deviation (SD) with negative extremes on the left and positive extremes on the right (from 0 to 10).

N=66		Extreme		Mean (SD)
		From	To	
1	To refrain from brushing for 48 hours was...	Uncomfortable	Comfortable	2.59 (1.73)
2	I think the toothbrush cleaned my teeth well enough...	Not agree	Totally agree	6.29 (1.93)
3	I think the stiffness of the toothbrush filament...	Very rigid	Very soft	5.04 (1.73)
4	Brushing without toothpaste was...	Unpleasant	Pleasant	3.21 (2.33)
5	Brushing with a dry toothbrush was...	Unpleasant	Pleasant	2.81 (1.58)

from the systematic review by plaque index score showed that the reduction was 30% when evaluated with the Q&H Plaque Index (33) and 53% with the Navy Plaque Index (34). In a similar analysis of powered toothbrushes (25), Rosema et al. (25) showed that numerically the highest plaque score reduction was apparent with the Sillness & Løe Plaque Index (22) as used in the present study. In the systematic review (5), Slot et al. (2012) also performed a subanalysis with respect to brushing duration, the estimated efficacy as the weighted mean plaque score reduction was 27% after 1 min and 41% after 2 min of brushing assessed according to the Q&H Plaque Index (33). Subsequently for the present study for all participants 2-min toothbrushing duration was selected. This may have done injustice to what O’Hehir & Suvan (7) observed, namely that without toothpaste people tend to brush longer and more evenly around the mouth. Due to the supervised brushing protocol and the restricted brushing time, this latter aspect could not be evaluated.

Limitation

Single-use toothbrushing studies are routinely used for screening the efficacy of toothbrushes to evaluate to which extent an intervention can produce a beneficial outcome under ideal circumstances (36). Although the results of these trials do not provide definitive proof of clinical superiority (37), the reasons for using the single-use protocol are its cost-effectiveness and ethical acceptability. In addition, no existing disease could worsen during the 1-day plaque accumulation (6,35). Despite the apparent benefits of the model, it may be influenced by the ‘Hawthorne effect’. This phenomenon may have modified the behaviour of the study participants purely as a result of them knowing that they are contributing to an experiment (38). This could have affected the plaque score reductions during a single brushing exercise in this study. This study was carried out in a clinical setting in which the participants may be more concentrated than usual besides their brushing was supervised. This study indicated only right-handed brushers. It might be that the outcome is different in left-handed participants which could be a subject of future research.

Conclusion

Using a manual toothbrush reduced plaque scores between 55% and 58% with no difference between brushing from either the lingual or the buccal aspect first. Within the limitations of this study a recommendation to start toothbrushing the lingual aspect is not supported by the outcome in this young student population.

Acknowledgements

The authors fully acknowledge Martijn van Leeuwen (MPCL), who was responsible for the screening of the participants. The study was sponsored by an unrestricted educational grant from the Procter & Gamble Company. The manual toothbrushes were also provided free of charge by Procter & Gamble.

Conflict of interest

The authors have stated explicitly that there is no conflict of interest in connection with this article. Van der Weijden, Slot and their research team at ACTA have previously received either external advisor fees, lecturer fees or research grants from toothbrush and dentifrice manufacturers. Those manufacturers included Braun, Colgate, Dentaïd, GABA, Lactona, Meda Pharma, Oral-B, Procter & Gamble, Sara Lee, Sunstar and Unilever.

Authors contributions

Conception or design of the study: EVDS, DES, NLHH, GAW

Analysis and/or interpretation of the data: EVDS, DES, GAW

Drafted the manuscript: EVDS

Critically revised the manuscript: DES, NLHH, GAW

All authors gave their final approval and agreed to be accountable for all aspects of the work

References

1. DeVore CH, Beck FM, Horton JE. Plaque score changes based primarily on patient performance at specific time intervals. *J Periodontol* 1990; 61: 343–346.
2. Prasad KV, Sreenivasan PK, Patil S, Chhabra KG, Javali SB, DeVizio W. Removal of dental plaque from different regions of the mouth after a 1-minute episode of mechanical oral hygiene. *Am J Dent* 2011; 24: 60–64.
3. Løe H, Theilade E, Jensen SB. Experimental. *J Periodontol* 1965; 36: 177–187.
4. www.mouthhealthy.org (accessed 19 April 2016).
5. Slot DE, Wiggelinkhuizen L, Rosema NA, Van der Weijden GA. The efficacy of manual toothbrushes following a brushing exercise: a systematic review. *Int J Dent Hyg* 2012; 10: 187–197.
6. Lee DW, Moon IS. The plaque-removing efficacy of a single-tufted brush on the lingual and buccal surfaces of the molars. *J Periodontal Implant Sci* 2011; 41: 131–134.
7. O’Hehir TE, Suvan JE. Dry brushing lingual surfaces first. *J Am Dent Assoc* 1998; 129: 614.
8. Bass CC. The necessary personal oral hygiene for prevention of caries and periodontoclasia. *New Orleans Med Surg J* 1948; 101: 52–70.
9. Galinsky M. Ergonomics of toothbrushing. *J Dent Res* 1975; 54: 115.
10. Macgregor ID, Rugg-Gunn AJ. A survey of toothbrushing sequence in children and young adults. *J Periodontal Res* 1979; 14: 225–230.
11. Wainwright J, Sheiham A. An analysis of methods of toothbrushing recommended by dental associations, toothpaste and toothbrush companies and in dental texts. *Br Dent J* 2014; 217: 1–4.
12. Lang NP, Lindhe J. Clinical periodontology and implant dentistry. In: Van der Weijden F, Slot DE, Echeverria JJ, Lindhe J, eds. *Mechanical Supragingival Plaque Control*, 6th edn, Chapter 36. UK, Wiley-Blackwell, 2015, pp. 677–716.
13. Lang NP, Karring T. European Workshop on Periodontology, 1st Proceeding. Axelsson P, Session III Mechanical Plaque Control. London, Quintessence Publishing Co, 1994.
14. O’Hehir TE. The new standard for prevention. *Auxiliary* 2011; 21: 20.
15. Dupont WD, Plummer WD. PS power and sample size program available for free on the internet. *Control Clin Trials* 1997; 18: 274.
16. Rosema NA, Hennequin-Hoenderdos NL, Versteeg PA, van Palenstein Helderma WH, van der Velden U, van der Weijden GA. Plaque-removing efficacy of new and used manual toothbrushes—a professional brushing study. *Int J Dent Hyg* 2013; 11: 237–243.
17. Van der Sluijs E, Slot DE, Hennequin-Hoenderdos NL, Van Leeuwen M, Van der Weijden GA. Prebrushing rinse with water on plaque removal: a split-mouth design. *Int J Dent Hyg* 2016; Mar 28. doi: 10.1111/idh.12216. [Epub ahead of print]
18. Gómez SM, Danser MM, Sipos PM, Rowshani B, van der Weijden GA. Tongue coating and salivary bacterial counts in healthy/gingivitis subjects and periodontitis patients. *J Clin Periodontol* 2001; 28: 970–978.
19. Van der Velden U. The Dutch periodontal screening index validation and its application in The Netherlands. *J Clin Periodontol* 2009; 36: 1018–1024.
20. Bentley CD, Disney JA. A comparison of partial and full mouth scoring of plaque and gingivitis in oral hygiene studies. *J Clin Periodontol* 1995; 22: 131–135.
21. Randomness and Integrity Services Ltd. Dublin, Ireland. Available at: www.random.org (accessed 19 April 2016).
22. Silness J, Løe H. Periodontal disease in pregnancy (II). Correlation between oral hygiene and periodontal condition. *Acta Odontol Scand* 1964; 22: 121–135.
23. Van der Weijden GA, Danser MM, Nijboer A, Timmerman MF, Van der Velden U. The plaque removing efficacy of an oscillating/rotating toothbrush. A short term study. *J Clin Periodontol* 1993; 20: 273–278.
24. Mottola CA. Measurement strategies: the visual analogue scale. *Decubitus* 1993; 6: 56–58.

25. Rosema N, Slot DE, van Palenstein Helderma WH, Wiggelinkhuizen L, Van der Weijden GA. The efficacy of powered toothbrushes following a brushing exercise: a systematic review. *Int J Dent Hyg* 2016; 14: 29–41.
26. CONSORT Group. The CONSORT statement 2010. Available at: <http://www.consort-statement.org> (accessed on 19 April 2016).
27. Hansen F, Gjerme P. The plaque-removing effect of four toothbrushing methods. *Scand J Dent Res* 1971; 79: 502–506.
28. Pader M. *Oral Hygiene Products and Practice*. New York, Marcel Dekker Inc., 1988.
29. Valkenburg C, Slot DE, Bakker EWP, Van der Weijden GA. What is the contribution on efficacy of plaque removal. A systematic review. *J Clin Periodontol* 2016; Aug 11. doi:10.1111/jcpe.12615. [Epub ahead of print]
30. Moore C, Addy M, Moran J. Toothpaste detergents: a potential source of oral soft tissue damage? *Int J Dent Hyg* 2008; 6: 193–198.
31. Paraskevas S, Timmerman MF, van der Velden U, van der Weijden GA. Additional effect of dentifrices on the instant efficacy of toothbrushing. *J Periodontol* 2006; 9: 1522–1527.
32. Bhardwaj SS, Camacho F, Derrow A, Fleischer AB Jr, Feldman SR. Statistical significance and clinical relevance: the importance of power in clinical trials in dermatology. *Arch Dermatol* 2004; 140: 1520–1523.
33. Quigley GA, Hein JW. Comparative cleansing efficiency of manual and power brushing. *J Am Dent Assoc* 1962; 65: 26–29.
34. Rustogi KN, Curtis JP, Volpe AR, Kemp JH, McCool JJ, Korn LR. Refinement of the Modified Navy Plaque Index to increase plaque scoring efficiency in gum line and interproximal tooth areas. *J Clin Dent* 1992; 3: 9–12.
35. Kawamura M. Dental behavioral science. The relationship between perceptions of oral health and oral status in adults. *Hiroshima Daigaku Shigaku Zasshi* 1988; 20: 273–286.
36. Suvan JE. Effectiveness of mechanical nonsurgical pocket therapy. *Periodontol* 2000 2005; 37: 48–71.
37. Claydon N, Addy M. Comparative single-use plaque removal by toothbrushes of different designs. *J Clin Periodontol* 1996; 23: 1112–1116.
38. Adair J, Sharpe D, Huynh CL. Hawthorne control procedures in educational experiments: a reconsideration of their use and effectiveness. *Rev Educ Res* 1989; 59: 215–228.