Interproximal Plaque Mass and Fluoride Retention after Brushing and Flossing – a Comparative Study of Powered Toothbrushing, Manual Toothbrushing and Flossing

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Purpose: To determine differences in interproximal plaque mass and fluoride retention with different modes of toothbrushing and flossing.

Materials and Methods: Forty-seven subjects in good health used four treatments in a four-period, randomized, crossover design: 1) manual brushing only; 2) manual brushing and daily flossing; 3) electric brushing using a rotational oscillation toothbrush; and 4) electric brushing using a sonic toothbrush. Subjects used a standard sodium fluoride dentifrice during the eight-day experimental periods and a fluoride-free dentifrice during the seven-day washout periods between treatments. Interproximal plaque samples were taken on Day 1 and on Day 8, weighed, and analyzed for fluoride content.

Results: The amount of interproximal plaque was lowest with sonic brushing, which left 43–65% less plaque than all other treatments. Manual brushing and flossing yielded less plaque than manual brushing alone and rotational oscillation brushing. Differences were statistically significant ($p < 0.05$) for treatment; there was no time effect on plaque mass. For fluoride retention, at Day 1 sonic brushing gave at least 54% more fluoride in the interproximal plaque than all other treatments, which was significant. All treatments demonstrated a significant increase in fluoride concentration with time except manual brushing and flossing, which showed a significant decrease. At Day 8, the fluoride concentration was significantly higher for sonic brushing than for manual brushing or rotational oscillation brushing by over 40%, and all treatments exhibited significantly greater fluoride than the manual brushing and flossing combination.

Conclusion: The mode of toothbrushing may impact the amount of plaque retained interproximally and its fluoride concentration.

Key words: Plaque mass, fluoride, retention, interproximal, electric toothbrush, sonic toothbrush

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Toothbrushing with a dentifrice (toothpaste) is the most widespread oral hygiene habit in the industrialized world (König and Navia, 1995; Marinho et al, 2003). The role of the modern dentifrice is not only to facilitate mechanical plaque removal but also to serve as a vehicle for delivery of anti-plaque and anti-caries agents (Holt and Murray, 1997). Daily brushing with a fluoride-containing dentifrice

is now an accepted measure for caries prevention. The major thrust of research in this field has for many years been dentifrice formulation and the optimal concentration of fluoride in the dentifrice. However, there is increasing recognition that there are two other major determinants of the anti-caries effect of fluoride dentifrices: accessibility to sites at risk and retention of fluoride at these sites (Sjögren, 2001).

Accessibility to Sites at Risk

The proximal surfaces of the premolar and molar teeth are the smooth surfaces most susceptible to caries (Haak et al, 2002) and least accessible for mechanical cleaning by conventional toothbrushing. Daily home care routines for removal of posterior interproximal plaque do not seem to find ready acceptance among patients (Wilson, 1998). Conventionally, floss is recommended for this purpose and many patients appear to lack the manual dexterity or the perseverance necessary to floss effectively and atraumatically (Wilson, 1998). In order to motivate patients, there is a need for less demanding home care procedures to remove interproximal plaque.

Advances in toothbrush design have contributed to progress in this field. Electric brushes, while commercially available for many years, are under constant development due to the highly competitive nature of the toothbrush market. Typical of these are rotational oscillations brushes such as the Braun Oral-B brushes (Braun GmbH, Kronberg/Ts., Germany), which have a round brush head and a 70° rotational oscillation movement. Comparative studies of manual and electric toothbrushes have reported not only more effective plaque control but also important motivational benefits for powered brushes (Warren and Chater, 1996; Stålnacke et al, 1995).

The sonic toothbrush, the so-called third generation of electric toothbrushes, combines dynamic fluid pressure and shear forces created by high frequency sweeping vibration of the bristle tips to disrupt and disperse bacterial plaque. A study by Tritten and Armitage (1996) demonstrated improved interproximal cleaning by a Sonicare sonic toothbrush (Philips Oral Healthcare, Inc., Snoqualmie, Washington, USA) compared to a manual toothbrush, the suggested mechanism being the ability to disrupt plaque at a distance from the bristle tips, i.e. the area can be cleaned without insertion of the cleaning device into the interproximal space.

Post-brushing Retention of Fluoride

As mentioned above, post-brushing retention of fluoride in the oral fluids is also an important determinant of the anti-caries effect of brushing with a fluoride dentifrice. In this context, a major advance is the recommended change, based on recent research, from conventional vigorous post-brushing rinsing with copious amounts of water, to swishing with the remaining toothpaste slurry and a small quantity of water (Sjögren, 1995; Sjögren and Melin, 2001). In an experimental *in situ* study, Sjögren et al (1995b) showed that toothbrushing followed by active rinsing with the toothpaste slurry, compared to copious rinsing with water, retarded the demineralization of enamel and dentin at approximal sites. In a clinical study in preschool children, Sjögren et al (1995a) reported that the same method reduced the incidence of approximal caries by 26%. These studies illustrated the positive effect of restricting rinsing with water after brushing with a fluoride dentifrice.

While the powered toothbrushes have undergone numerous comparative studies with manual brushes with respect to effect on interproximal plaque and inflammation (see, for example, the review by Heasman and McCracken, 1999), there appear to be no published reports comparing post-brushing interproximal fluoride retention. In a single-blind crossover pilot study of six healthy subjects, Sjögren (1998) compared fluoride retention in dental plaque after use of a manual toothbrush, the same manual brush and dental floss, and a sonic brush. A fluoride dentifrice containing 1500 ppm fluoride ion and minimal post-brushing rinsing as described above were used throughout. Each experimental period lasted seven days. The concentration of fluoride retained in the interproximal plaque was 1.4 mM after use of the sonic brush, 0.9 mM after use of the manual brush, and only 0.2 mM after manual brushing and daily flossing. The differences were statistically significant ($p < 0.05$). On the basis of these results, further testing in a larger study was warranted.

Thus the aim of the present study was to evaluate the retention of fluoride in interproximal plaque after various oral hygiene routines using a fluoride dentifrice and minimal post-brushing rin-

sing throughout. Simultaneously the amount of plaque accumulated in this space was also to be measured.

MATERIALS AND METHODS

Study Population

Forty-seven persons participated in this study. Participants were recruited from the dental faculty and students at Göteborg University. All volunteers were in good health with at least 24 natural teeth and a DMFS score of less than 21. A normal salivary secretion rate, defined as a paraffin-stimulated salivary flow of greater than 1 mL/min, was also required for inclusion in the study. Age ranged from 22 to 38 with a mean age of 25. The subject population comprised 19 men and 28 women.

Study Design

Four regimens were evaluated in a four-period, crossover design:

1. Manual brushing only, using a Jordan Active Tip toothbrush (Jordan a/s, Oslo, Norway)
2. Manual brushing after flossing, using a Jordan Active Tip toothbrush and waxed floss (Johnson & Johnson Consumer Products, Skillman, New Jersey, USA)
3. Electric brushing using a rotational oscillation brush, the Braun Oral-B Ultra Plaque Remover (D9), and
4. Electric brushing using a sonic toothbrush, the Sonicare Plus (now renamed Sonicare Advance).

Subjects followed each of the above regimens twice daily for an experimental period of eight days. A washout period of seven days preceded each experimental period. No interproximal cleaning was permitted during the study except when subjects were following the manual brushing and flossing regimen. No professional cleaning was carried out either at the start or at the end of each 8-day treatment period. Treatment order was randomized across all subjects. The examiner (ABL) was blinded to the order of treatments.

Subjects were provided with a sodium fluoride dentifrice (1500 ppm F^-, Pepsodent Superfluor, Elida Fabergé, Stockholm, Sweden) to use in the experimental periods and a fluoride-free dentifrice (Tandkräm utan fluor, Coop, Stockholm, Sweden) to use in the washout periods. At each brushing session, subjects applied 1 g dentifrice to the brush and brushing was performed for two minutes. Following brushing, subjects rinsed by swishing in the mouth 5 mL of added water with the foam induced by the brushing. No additional rinsing was permitted. Subjects were provided with timers to help ensure adherence to the protocol.

In order to standardize the participants' brushing and flossing techniques, one of the authors (KS) carefully instructed and trained each participant. The subjects were instructed not to eat or drink anything for at least two hours after brushing. They were to refrain from cleaning or other intervention, except as instructed, in the premolar and first molar interproximal areas during the experimental periods.

Collection of Plaque Samples

On Day 1 (after the first hygiene measure) and Day 8 (after 19 treatments totally) of each experimental period, interproximal plaque was collected from the premolars and first molars, according to the method described by Sjögren et al (1996). Briefly, a length of fluoride-free, waxed dental floss (Johnson & Johnson) was carefully drawn through the interproximal space. The plaque collected was then dislodged by passing the floss through a slit in the lid of a 0.5-mL Eppendorf plastic tube (Eppendorf PCR, Sarstedt, Nümbrecht, Germany). The Eppendorf tube was weighed before and after the addition of plaque and the weight of plaque determined to the nearest 0.1 mg. An aliquot of 330 µL TISAB III buffer (Orion Research, Inc., Boston, Massachusetts, USA) and distilled water (1:10) was added to the sample and the tube sealed and centrifuged before storage at –80°C until analyzed.

For analysis, the plaque suspensions were thawed and then homogenized by sonication for 5 s. The fluoride concentration of each suspension was analyzed using an ion-selective fluoride electrode (Orion 96-09, Orion Research, Inc.), again following the method described by Sjögren et al (1996).

Analysis of Data

The data were analyzed using a model of two-way analysis of variance (ANOVA). The subjects repre-

Table 1 Wet weight interproximal plaque collected (mg)

Treatment	Day 1	Day 8	Day 1 & Day 8 (combined)			Reduction *vs.* manual brush
	mean (SD)	mean (SD)	mean (SD)	Homogenous groups*		
Manual brush	17.1 (13.5)	16.0 (11.7)	16.5 (10.6)	A		–
Manual brush + floss	10.7 (9.9)	9.9 (9.0)	10.3 (8.4)		B	38%
Rotational oscillation brush	14.8 (11.5)	17.2 (10.8)	16.0 (8.8)	A		3%
Sonic brush	5.7 (2.5)	6.0 (2.6)	5.9 (1.8)		C	64%

* Groups with the same letter are statistically not different. Differences between Day 1 and Day 8 are statistically not different for each treatment.

sent blocks in which each of the four experimental methods was evaluated. This model accounts for every subject using each experimental method by comparing the methods within subjects, thus eliminating inter-subject variation. Separate analyses were undertaken for Day 1 and Day 8. A significance level of $p < 0.05$ was chosen.

RESULTS

Wet Weight

The mean wet weight of interproximal plaque collected at the start and end of each treatment period is presented in Table 1. ANOVA showed no significant difference in the amount of plaque collected between Day 1 and Day 8 for any specific treatment, including manual brushing and daily flossing. However, significant differences were found among the treatments on Day 1 and Day 8, including manual brushing and daily flossing *versus* manual brushing alone and rotational oscillation brushing and sonic brushing *versus* each other treatment. For Day 1 and Day 8 combined, there was 38% less plaque collected after manual brushing and flossing and 36% less after rotational oscillation brushing than after manual brushing alone. When comparing the reduction of interproximal wet weight of collected plaque, the manual brushing combined with flossing resulted in a 38% reduction compared to manual brushing alone. The rotational oscillating brush resulted in 3% reduction, and the sonic toothbrush in a 64% plaque reduction compared to manual brushing alone.

Fluoride Concentration

The mean concentration of fluoride in the interproximal plaque is shown in Table 2. For all treatments except for manual brushing plus flossing there was a significant increase in the fluoride concentration from Day 1 to Day 8; for manual brushing plus flossing, there was a significant decrease in the fluoride concentration. Considering only the Day 1 data, the sonic toothbrush treatment gave a fluoride concentration in the residual interproximal plaque significantly higher than any of the other treatments. At Day 8, the fluoride concentration after sonic brushing was significantly higher than that after manual brushing alone or that after rotational oscillation brushing, both which were significantly higher than that achieved by manual brushing and flossing combined.

DISCUSSION

There was no significant change in the amount of plaque collected from the start of a treatment period (Day 1) to its end (Day 8). One might expect a decrease in interproximal plaque over a longer period of time, since decreases in plaque have been reported for other regions of the dentition over longer periods. Tritten and Armitage (1996), for example, observed significant decreases in the plaque index after 12 weeks of regular brushing with the Sonicare toothbrush or with a manual toothbrush. However, it is not surprising to see no time dependency for plaque quantity in this short-term study.

Table 2 Concentration of Fluoride in Interproximal Plaque Collected (ng/mg)

Treatment	Day 1		Day 8	
	mean (SD)	Homogenous groups*	mean (SD)	Homogenous groups*
Manual brush	3.6 (2.3)	D	5.7 (4.4)	G
Manual brush + floss	3.4 (2.4)	D	2.6 (1.8)	F
Rotational oscillation brush	3.7 (2.9)	D	5.2 (3.6)	G
Sonic brush	5.7 (3.4)	E	8.0 (5.9)	H

* Groups with the same letter are statistically not different. Differences between Day 1 and Day 8 for each treatment are significant.

The differences in plaque mass between treatments are noteworthy in two respects. Firstly, the addition of daily flossing to manual brushing produced a significant decrease in the amount of interproximal plaque. Secondly, the sonic brushing treatment shows an advantage for reduction in interproximal plaque over all other treatments. This includes brushing with the rotational oscillation toothbrush and also the combination of manual brushing and daily flossing.

A recent meta-analysis of comparative studies on plaque reduction (Heanue et al, 2003) concluded that only powered toothbrushes with rotational oscillation motion offer a significant advantage, albeit slight, in plaque reduction compared with manual brushes. That analysis, however, did not consider studies of the interproximal plaque as we have examined it here. There is reason to expect that the sonic toothbrush would leave less interproximal plaque than the rotational oscillation toothbrush. Hope et al (2003) showed the Sonicare toothbrush removed four times more plaque biofilm than another Braun Oral-B rotational oscillation toothbrush with similar motion (Excel Plaque Remover (D15)) in an *in vitro* model of the interproximal space between two molars.

The superiority of the sonic toothbrush over a manual toothbrush and floss we attribute to poor flossing technique, which is surprising since the subjects would be expected to be highly competent at flossing. Better flossing by the subjects – or daily professional flossing – might have been expected to yield as little or less interproximal plaque than brushing with the sonic toothbrush. Although it was not tested here, we speculate that combining the sonic toothbrush with daily flossing may result in the least amount of interproximal plaque.

Brushing with the sonic toothbrush appears to deliver significantly higher concentrations of fluoride to interproximal plaque. Thus, at Day 1, the fluoride retained in the interproximal plaque was at least 54% greater for sonic brushing than for any other treatment. One week later, the fluoride retained with the sonic brushing had increased significantly and was still more than 40% greater than with manual brushing alone or with rotational oscillation brushing and two times greater than that retained with manual brushing and flossing combined. We suspect the different fluid dynamics of the sonic brush are responsible for the higher fluoride levels. Hope and Wilson (2002) noted that another sonic toothbrush, the Sonicare Elite, appeared to generate greater fluid flow than the rotational oscillation Braun Oral-B D15 brush in their *in vitro*, interproximal model.

A similar phenomenon for the brushes used in this study may have helped the sonic brush both to deliver more fluoride to the surface of the interproximal plaque biofilm and to amplify the diffusion of fluoride into that biofilm. The impact is noticed immediately at Day 1. By Day 8, through continuation of a treatment, the amount of fluoride within the interproximal plaque was increased for all treatments except manual brushing with daily flossing. The increase may be attributed to a build-up of fluoride with repeated exposure and slow diffusion of that fluoride to the interior of the biofilm. In the case of manual brushing with daily flossing, we hypothesize the interproximal biofilm is smeared as floss passes over and through it. This smearing disrupts the natural porosity of the biofilm, filling or covering over the gaps and channels in the original biofilm structure. The residual biofilm after flossing thus presents less surface area for transfer of fluoride

from the dentifrice during brushing. Fluoride transfer is reduced in proportion with the decrease in surface area and thus the interproximal fluoride concentration was reduced.

Determination of total fluoride content of plaque using an ion specific electrode cannot be performed, due to formation of CaF2-like substances not measured. However, in a double blind cross over designed trial, the fluoride measured can be compared as an effect of treatment, even if the total of fluoride present in plaque, and at the tooth surface is not determined.

CONCLUSION

To conclude, the results from this study show that the mode of toothbrushing influences both the interproximal plaque amount and the fluoride concentration. In particular, the sonic brush yielded significantly less interproximal plaque and significantly greater fluoride retention in that plaque than all other treatments.

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