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Dental stain prevention by abrasive toothpastes: A new *in vitro* test and its correlation with clinical observations

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Synopsis

A new *in vitro* test based on the removal of a model film from polished hydroxyapatite discs has been developed to predict more precisely the stain prevention properties of toothpastes *in vivo.* It is argued that dental stain prevention, as opposed to removal, is the predominant function provided by abrasive pastes suitable for unrestricted home use. It is necessary, therefore, to focus on abrasive systems capable of efficient removal of the relatively soft and invisible up-to-24-hour-old salivary pellicle formed between brushings, rather than on harder visible mature stain. A cleaning model of this film has therefore been developed. The abrasive effect on the model film of a typical silica xerogel has been examined, and the data compared with clinical observation of stain build-up. A good correlation was apparent. Stain prevention increased dramatically from zero to 4% w/w silica, after which higher levels had little or no beneficial effect, despite a continuing increase in potential dentine damage.

INTRODUCTION

It is well known (1) that the chemical nature of many of the foods and beverages that we consume can create a hostile environment in the mouth unless some form of regular cleaning, beyond that of natural salivary irrigation, is practiced. Acids and sugars, via bacterial plaque growth, promote physiological damage to teeth and gums in addition to tainting the breath, and residual colored compounds will cause unsightly discoloration of the tooth surfaces (2). These effects can be mitigated to a large extent via daily brushing with a dentifrice (3).

The rapid removal of plaque and food residues during brushing is believed to be predominantly a mechanical bristle function, and thus related mainly to brush design and operator skill (4).

The control of dental stain, however, requires, in addition, the presence of an abrasive agent (5). Since Roman times a wide variety of finely ground minerals have been essential ingredients of tooth-cleaning preparations (6). Today, finely divided amorphous silicas (SiO_2), precipitated chalks ($CaCO_3$), and calcium phosphates (e.g., dicalcium phosphate dihydrate) are normally used in toothpastes (7).

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These minerals, of Moh's hardness 2.5 to 3.5, are softer than tooth enamel (Moh's hardness about 5), thus ensuring that their potential for serious tooth wear in extended use is acceptably low. This is not the case, however, in relation to dentine surfaces commonly exposed when gum recession has taken place. Dentine surfaces can be eroded by these abrasives during brushing, and therefore it is very important to control this property in formulated toothpastes. Products designed for unrestricted home use are necessarily constrained to moderate-to-low dentine abrasion rates defined and measured currently by standard radiotracer techniques (8,9).

Fifty years ago, Kitchen and Robinson (5) established the strong positive correlation between the abrasive stain removal properties of dentifrices and the rate of dentine abrasion. The apparent similarity in the mechanical properties of dentine and mature stained pellicle has therefore made it difficult to dissociate dentifrice abrasive performance from its damage potential.

A widely used method of obtaining subjects with stained teeth for clinical studies is to make them brush only with water (5) or an abrasive-free paste (10) for a month or so. Comparative paste performance tests can then be carried out either under controlled brushing conditions provided by a hygienist or within the panelist's own regime. Satisfactory clinical methods for visual assessment of stain and its reduction have been established (11), and an *in vitro* stain removal screening test using artificially stained bovine teeth has been devised by Stookey *et al.* (12). This widely recognized method for comparing potential dental abrasive cleaning power is claimed to correlate well with clinical observations of mature stain removal. Accordingly, the performance of a range of pastes tested for us by the Indiana University Oral Health Research Institute using the Stookey method closely followed their respective dentine abrasion rates.

For the reasons discussed above, it is not surprising to find in most clinical performance studies that regular toothpaste abrasive systems are not powerful enough to remove mature stain instantly or completely, but are able only to reduce it somewhat (11,13). Moreover, this test protocol hardly represents the reality of everyday brushing, in which up to 24 hours is insufficient time to acquire any visible deposit under normal conditions.

It is likely, therefore, that the primary abrasive function in dentifrices is not stain removal but stain prevention. The acquired pellicle, composed mainly of salivary glycoproteins, is known to form readily on tooth enamel surfaces and to thicken and toughen with time (13–15). Undisturbed, it can grow to 10 microns in thickness and become colored (5) over a period of weeks. However, if the young (up to 24-hour) pellicle is substantially removed during each brushing, no build-up of potentially visible films can occur.

Inefficient toothbrushing, for whatever reason, will therefore lead to stain. This usually forms in areas not exposed to the regular abrasive action of foods and toothbrush bristles, particularly in the gingival crevice and interproximal regions of the dentition. Under habitual poor brushing conditions, therefore, the abrasive function of the paste is virtually redundant in these areas, and neither stain removal nor prevention is likely to be achieved however powerful the paste abrasive system. In the current absence of safe and effective chemical treatments, persistent stainers have the choice of remaining so, visiting the dental hygienist for professional cleaning at regular intervals, or improving their brushing technique.

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This paper discusses a new evaluation technique for examining abrasive stain *prevention* by dentifrices, and demonstrates its predictive function in relation to stain prevention studies *in vivo.* The principal object of the new model was to simulate relatively immature 12-to-24-hour-old pellicle films on smooth hydroxyapatite surfaces.

MATERIALS AND METHODS

LABORATORY STUDIES

Substrate. In order to ensure maximum reproducibility consistent with reality within the test, a model hard substrate was employed. Thus highly polished sintered 17-mm hydroxyapatite (HAP) discs (Calcitek Inc, Carlsbad, California) were used to simulate enamel tooth surfaces. This was achieved by lapping the discs using a rotary grinder/polisher (Buehler UK Limited, Coventry, UK) on wet P600, then P1200 metallographic grinding papers to give a mirror finish. Typical degrees of roughness (Ra) measured profilometrically were of the order 0.1–0.2 microns, similar to that of teeth. Care was taken not to touch these surfaces after polishing.

Soil. A self-indicating model organic film intended to simulate the gross mechanical properties of up-to-24-hour pellicle was formed by precipitating an iron (III) complex with tannic acid from aqueous solution directly onto the polished HAP discs.

Freshly mixed solutions (0.1% w/w) of diammonium iron (II) sulphate 6-hydrate and tannic acid (GPR grades, BDH, Poole, UK) are initially colorless, but form a dark colloidal iron (III) tannic acid complex ("ferric tannate") on contact with air. The fresh mixture is thus painted onto HAP surfaces with a fine squirrel-hair brush and gently dried with a warm hairdryer. Three thin applications were found to provide a colored organic film of thickness about one micron, which is believed to be of the correct order of thickness to simulate the immature pellicle of interest (14). This film on HAP discs gives a darkness measurement of $L^* = 50 \pm 5$ (CIE 1976 L*a*b* system) as determined by a Minolta Chromameter® (Minolta Limited, Milton Keynes, UK).

Although it is conceded that a ferric tannate film cannot simulate the chemical properties of stain-precursive dental pellicle, it displayed upon inspection many of the key physical properties of such a layer. Thus it proved resistant to brushing with nylon filaments in the presence of all common toothpaste ingredients except abrasive agents. This is consistent with the findings of Kitchin and Robinson (5) and others (11) that regular brushing with abrasiveless systems cannot prevent long-term stain formation.

The model soil/substrate additionally proved to be stable, rapidly and easily prepared, and progressively and reproducibly sensitive to different dental abrasive types and slurry concentrations under mechanical brushing conditions. This prompted an investigation of the abrasive performance of a series of paste systems with a view to determining to what extent this might mirror clinical stain prevention data.

Mechanical brushing regime. The ferric tannate (FT) stained hydroxyapatite discs were mounted horizontally in the bottom of a trough containing the aqueous toothpaste slurries under test, and weighted toothbrush heads oscillated over the disc surface by way of a mechanical scrubbing machine. A modified Martindale abrasion tester (Goodbrand-Jeffreys Ltd., Stockport, UK) was found to be particularly suitable. In this apparatus the brush heads describe Lissajous' figures, which combine linear and elliptical motions to

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ensure comprehensive coverage of the brushed sample. An oscillation rate of 150 cycles per minute and a brushing force of 4.4 N was chosen to typify observed brushing practice (4). Cleaning performance was found to be relatively insensitive to brush head forces in the typical range. The force was applied to conventional 34-tuft flat-trim 0.2-mm bristle nylon toothbrush heads (Mentadent P Professional®, soft; Elida Gibbs Dental Division, London, UK) via weights loaded onto vertical spindles mounted in linear ball bearings. Soil removal after 50, 100, and 150 oscillations was monitored. A control paste containing 10% silica xerogel abrasive was included in each determination, done in duplicate. The test was deemed valid when the control paste performance remained within ± 5% of its running mean value.

Photometric measurements. The Minolta Chromameter® CR-300 (8-mm aperture) was used to measure the whiteness (L*) of the discs before soiling (clean), after soiling (soiled), and after cleaning (cleaned). Removal of the FT film was given by the equation:

$$\%\ \text{Removal} = \frac{L^*\ (\text{cleaned}) - L^*\ (\text{soiled})}{L^*\ (\text{clean}) - L^*\ (\text{soiled})} \times 100$$

CLINICAL STAIN-PREVENTION STUDIES

Protocol. A panel of 45 adults (predominantly female, aged 18–50 years) was used to determine the stain-prevention properties of the test pastes *in vivo.* Particular consideration was given to the subjects' rights and personal convenience. Informed consent was gained for all participants. Four pastes were tested, blind, for six weeks each, end to end, in a fully randomized crossover design.

At the beginning of each six-week period the labial aspect of the eight incisors of the subjects were scaled and polished by a dental hygienist. The subjects were given a standard soft-nylon flat-trim toothbrush (Mentadent P Professional®) and a supply of paste to use at home in their normal manner. Panelists complaining of unsightly stain levels on their teeth during the test periods were called in for examination and scoring by the clinician. Following a temporary cleanup, they were given the most abrasive paste to use for the remainder of the six-week period. This modification to the protocol was felt to be necessary in retaining the goodwill and compliance of the panelists over the whole 24-week trial period. It was exercised by only a small number of panelists using the least abrasive paste.

Method of assessment. The degree of extrinsic stain after each six-week period was determined by two assessors (J.E.W., T.M.) using substantially the method of Lobene (1968) (11). Thus Lobene's four-point intensity criteria were applied to the three regions of each tooth: gingival, interproximal, and body, viz.,

0: no stain
1: light stain (visible only under magnification)
2: moderate stain (visible to naked eye)
3: heavy stain

The assessors were at no time directly aware of which paste each subject had been using during the trial. Agreement between, and reproducibility of, the assessors was excellent.

Data. The proportion of subjects exhibiting visible stain (scores 2 or 3) on at least one site within each six-week period was determined for each paste. Mean total stain scores

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associated with each paste were also recorded. However, owing to exercise of the panelist-withdrawal option during use of the low-abrasivity paste, comparable total stain scores for all the pastes were not possible.

Pastes. Four pastes were tested, representing a range of abrasivity from substantially zero to that typically found in regular toothpastes on the market. The abrasive performance of the pastes was determined by the new *in vitro* method at product concentrations of 33% by weight, slurried in water.

The pastes were formulated to contain base ingredients typical of currently marketed "silica" formulations (sorbitol, sodium lauryl sulphate, and flavor). Thus the principal water-insoluble components were both thickening and abrasive-type amorphous silicas and opacifying levels of titanium dioxide. Of these, only the abrasive silicas exhibit any significant abrasive function. Experimental pastes were made containing zero, 2%, 4%, and 10% by weight of a typical commercially available abrasive xerogel silica (Gasil® 200TP, Crosfield Chemicals Ltd., Warrington, UK). This abrasive is representative of the efficient, high-strength, angular dental silicas of 5 to 10 microns average particle size used extensively in modern toothpastes.

The unoccupied formulation space in the pastes containing less than 10% abrasive silica was filled with sorbitol and thickening silica in proportions necessary to maintain the viscosity and flavor of the 10% variant.

RESULTS

IN VITRO ABRASIVE EFFICIENCY

The performance of the four pastes is shown graphically in Figure 1. Abrasive removal of the FT film from the polished HAP discs is plotted against the number of brush head oscillations applied.

In this method it will be seen that the most efficient pastes reach a cleaning plateau at about 100 oscillations, and that the same order of effectiveness between pastes is observed at 50, 100, and 150 oscillations. Equally, rates of cleaning differentiate the pastes in the same order. A convenient simple expression of comparative abrasive performance was taken to be the percentage clean at 100 oscillations (FT-100 score). Under these conditions, mean differences of >5 percentage units are required for significance ($p < 0.01$).

Thus the *in vitro* performance of the pastes containing 10% and 4% abrasive silica was not substantially different, but a marked reduction was apparent for the 2% abrasive silica paste, with only slight abrasive cleaning provided by the zero-abrasive variant. No soil removal was observed after brushing with typical water-soluble components of pastes (e.g., humectant, surfactant), suggesting that the added titanium dioxide opacifier may be providing a small degree of abrasion.

IN VIVO STAIN-PREVENTION EFFICIENCY

In Table I below performance is expressed both as the percentage of the panel exhibiting visible stain (2,3 scores) on at least one site and as the mean total-stain score per panelist

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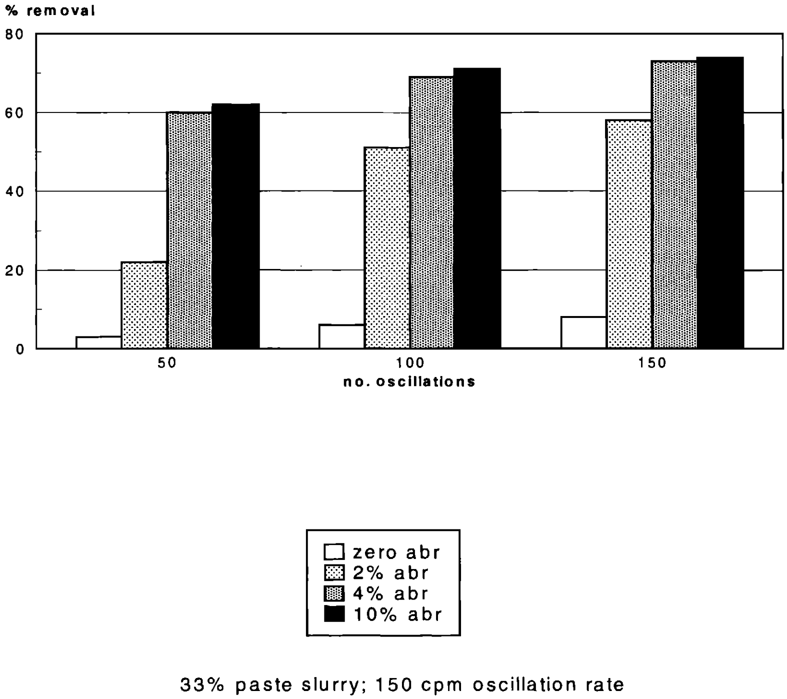

Figure 1. Film removal as a function of abrasive silica content of paste.

following six weeks' use. Corresponding British Standard Dentine Abrasion Values (DAV) (8) are also given.

The stain-prevention performances of the 10% and 4% abrasive silica pastes were similar (Mann Whitney U test) and significantly more effective ($p < 0.001$) than the 2% and zero-abrasive versions. The virtually identical stain-prevention action of the pastes containing 10% and 4% w/w abrasive silica is achieved despite a substantial difference in measured dentine aggression.

Comparison of the data with that of Figure 1 indicates a remarkable degree of coincidence of effect. This is illustrated in Figure 2.

DISCUSSION

Formulators of abrasive toothpastes for general use should aim to provide the maximum

Table I
Six-Week Clinical Stain Formation as a Function of Abrasive Silica Content and Dentine Abrasion Potential of Four Pastes

% Abrasion silica	% Visible stainers	Mean total-stain score (SE)	DAV
10	19	4.2 (0.33)	55
4	19	4.7 (0.35)	30
2	32	9.5 (0.34)	17
0	65	>11.2*	5

* Incomplete six-week score owing to exercise of cosmetic withdrawal option.

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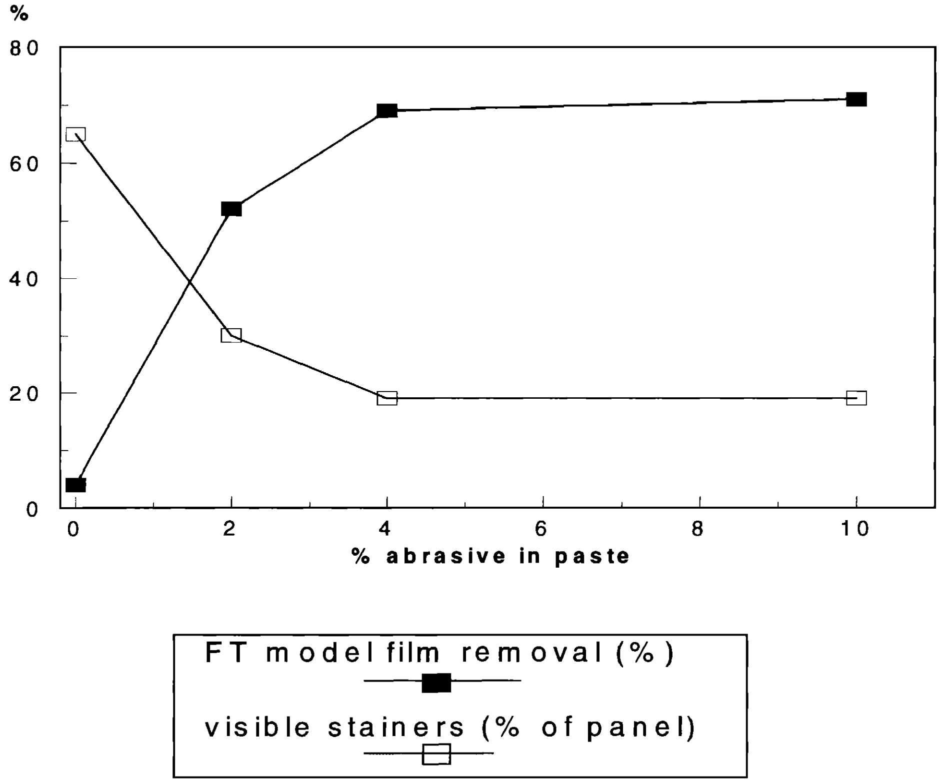

Figure 2. Comparison of *in vitro* and clinical stain-prevention data for pastes of different abrasive content.

stain-control function consistent with the minimum dentine abrasion potential. Most marketed toothpastes implicitly claim to have this balance despite containing a wide range of levels and types of abrasive. It is unlikely, however, that all demonstrate the optimum balance of care and efficiency, particularly in view of the widely held mechanistic hypothesis that the abrasive is included for stain *removal.* If, however, one recognizes that the primary realistic function of the abrasive in toothpastes is that of stain-*prevention,* it becomes apparent that different conditions may apply and that an *in vitro* test more closely simulating the removal characteristics of immature pellicle is required.

The data above suggest a strong clinical predictive function for the *in vitro* FT test in relation to abrasive stain prevention within a normal domestic regime of one to two brushings a day. It thus becomes a valuable tool for abrasive screening and optimization within the science of dentifrice formulation.

Toothpastes for general sale and use are formulated to satisfy the requirements of a majority of consumers, and thus the human panel used represented a typical range of age and personal habits. It became clear during clinical pilot studies that the rate and extent of stain formation following professional cleaning was strongly influenced by the subject; within subjects, however, behavior appeared relatively invariable. This is consistent with the belief that toothbrushing is a ritual activity, and that skill and diligence vary considerably between subjects. Long-term parallel test designs would require careful habit screening and balancing to obtain panels that are sensitive only to paste cleaning

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performance. The authors preferred a full crossover design in order to eliminate habit-related effects and to keep the panel size manageable. The constraint of shorter exposure periods using this regime was not felt to weaken the exercise, as pilot surveys of stain build-up over three months had shown that after six weeks no substantial change in the performance ranking of different pastes occurs.

Thus our six-week results indicated that about half the panel were sensitive to the abrasive level in the product. Two distinct sub-groups, however, revealed themselves less so.

One group, making up about 20% of the panel, produced visible stain within six weeks whatever paste they were given. This was not related to their declared usage of tea, coffee, red wine, or tobacco. Infrequent brushing, poor technique, or irregular tooth geometry could, however, be contributory factors.

The other group, about 30% of the total, developed no visible stain with any of the pastes, including the zero-abrasive paste, during the trial periods. Once again, there was little reason to connect this with drinking or smoking habits, or lack of them. However, it was apparent that these panellists were almost exclusively females in their teens and early twenties.

In conclusion, it should be noted that, by directing attention primarily to the concept of dental stain control via prevention instead of removal, a separation of the abrasive efficiency and damage functions becomes possible. Using this test regime, the scope to manipulate abrasive parameters to provide dentifrices exhibiting high physical stain control with low dentine damage potential is greatly increased.

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REFERENCES

(1) M. Pader, "Surfactants in Oral Hygiene Products," in *Surfactants in Cosmetics,* M. M. Rieger, Ed. (Marcel Dekker, New York, 1985), pp. 295–296.
(2) R. S. Manly, A structureless recurrent deposit on teeth. *J. Dent. Res.,* 22, 479–486 (1943).
(3) A. Frandsen, "Mechanical Oral Hygiene Practices," in *Dental Plaque Control Measures and Oral Hygiene Practices,* H. Loe and D. V. Kleinman, Eds. (IRL Press, Oxford, 1986), pp. 93–116.
(4) B. R. Pugh, Toothbrush wear, brushing forces and cleaning performance, *J. Soc. Cosmet. Chem.,* **29**, 423–431 (1978).
(5) P. C. Kitchin and H. B. G. Robinson, How abrasive need a toothpaste be? *J. Dent. Res.,* **27**, 501–506 (1948).
(6) W. B. Davis, Cleaning and polishing of teeth by brushing. *Community Dent. Oral Epidemiol.,* **8**, 237–243 (1980).
(7) M. Pader, *Oral Hygiene Products and Practice* (Marcel Dekker, New York, 1988), pp. 233–239.
(8) British Standard Institution, London, *Specification for Toothpastes,* BS 5136:1981.
(9) J. H. Hefferren, A laboratory method for assessment of dentifrice abrasivity, *J. Dent. Res.,* **55**, 563–573 (1976).

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(10) D. J. Lamb, R. A. Howell, and G. Constable, Removal of plaque and stain from natural teeth by a low abrasivity toothpaste, *Br. Dent. J.*, **157**, 125–127 (1984).
(11) R. R. Lobene, Effect of dentifrices on tooth stains with controlled brushing, *J.A.D.A.*, **77**, 849–855 (1968).
(12) G. K. Stookey, T. A. Burkhard, and B. R. Schemehorn, *In vitro* removal of stain with dentifrices, *J. Dent. Res.*, **61**, 1236–1239 (1982).
(13) C. A. Saxton, The effects of dentifrices on the appearance of the tooth surface observed with the scanning electron microscope, *J. Periodontal Res.*, **11**, 74–85 (1976).
(14) A. H. Meckel, The formation and properties of organic films on teeth, *Arch. Oral Biol.*, **10**, 585–597 (1965).
(15) W. G. Armstrong, Origin and nature of the acquired pellicle, *Proc. Roy. Soc. Med.*, **61**, 923–930 (1968).

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