Comparative evaluation of tooth substance loss and its correlation with the abrasivity and chemical composition of different dentifrices

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ABSTRACT

Context: In India, teeth cleaning with tooth powder is common in rural and semi-urban areas. These dentifrices may contain low-quality abrasives, which may have a deleterious effect on dental hard tissues.

Aims: This study aims to evaluate the tooth substance loss caused by different dentifrices and to correlate it with chemical composition, size, and shape of abrasives used.

Settings and Design: An indigenously made automated machine was used for brushing the specimens.

Materials and Methods: Sixty-four freshly extracted premolars were allocated to eight groups (n = 8). Colgate toothpaste was used as the control group. Each specimen was brushed in a vertical motion for 2½ h at 200 strokes/min with a constant applied load of 200 g corresponding to 6-month brushing. The difference in weight (pre- and post-brushing) was determined by an analytical weighing machine. Chemical analysis was done to determine the presence of iron oxide by Inductively Coupled Plasma Mass Spectrometry method. Shape and size of the abrasive particles was evaluated under scanning electron microscopy (SEM).

Statistical Analysis Used: One-way analysis of variance and Paired t-test were used to analyze the data.

Results: Tooth substance loss was maximum in the group brushed with red tooth powder, which was shown to contain the highest amount of iron oxide and also exhibited large, irregularly shaped abrasive particles under SEM.

Conclusions: Tooth substance loss was documented to be correlated with chemical composition (iron oxide) and the size and shape of abrasive particles used in dentifrices.

Key words: Abrasion, dentifrices, iron oxide, tooth brushing machine, tooth powder

Dentifrices (toothpaste and toothpowders) are complex formulations, and it is necessary to achieve a fine balance to provide cosmetic and oral health benefits, while limiting chemical and/or physical damage to teeth.\(^1\) Toothpowder is the most common form of oral hygiene practice in semi-urban and rural areas of India for economic reasons as well as due to misconception that these indigenous herbal products may be beneficial for dental and gingival health.

Abrasives are the insoluble components added to dentifrices to aid the physical removal of stains, plaque, and food debris. The most commonly used abrasives are silica and calcium carbonate.\(^2\) A high-quality dentifrice contains silica,\(^3-5\) but its use increases the cost and hence low-quality calcium carbonate, iron oxide, etc., are used to bring down the cost.

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Toothpowders, in general, are known to be 5 times more abrasive than toothpastes due to the quantity of abrasives used (95%) and their particle size.\textsuperscript{[6]} Hence, concern has been expressed about its detrimental effects on tooth substance which pose an important oral health problem. The chemical composition of most of the tooth powders is not known, but they may contain chemicals of low pH, which could cause softening of the dental hard tissues.

The aim of the present study was to comparatively evaluate tooth substance loss caused by commonly used dentifrices and to determine the agents responsible for the abrasivity. Composition, pH, particle size, and shape of different dentifrices were evaluated and compared.

**MATERIALS AND METHODS**

Sixty-four permanent maxillary and mandibular premolars freshly extracted for orthodontic reasons from patients (age group, 12–20 years) were used in the study. The specimens were allocated to eight groups of eight teeth each to test the abrasivity of the seven tooth powders (selected on the basis of a pilot screening of persons with significant tooth wear and their history of dentifrice used) and the control group of one of the most commonly used toothpaste as follows:

- Group 1: Colgate Tooth Paste (White)
- Group 2: Baidyanath Tooth Powder (Red)
- Group 3: MDH Tooth Powder (Brown)
- Group 4: Lal Dant Manjan (Red)
- Group 5: Vicco Tooth Powder (Brown)
- Group 6: Divya Dant Manjan (Brown)
- Group 7: Monkey Brand Tooth Powder (Black)
- Group 8: Colgate Tooth Powder (White).

**Specimen preparation**

The extracted teeth were preserved in saline thymol solution. Adherent tissue was removed by gentle scraping and immersed in absolute ethanol for 5 min to remove superficial moisture. Then each specimen was dried with absorbent paper and weighed thrice in an electronic analytical balance accurate to 0.1 mg, and the mean reading was taken as the weight of the specimen.

**Tooth brush abrasion technique**

The testing was done by subjecting the specimens to simulated toothbrushing in an automated brushing machine, which was designed, indigenously made and pretested for number of strokes and the force applied (Pramod workshop, Faridabad, Haryana, India). The design of the machine was based on the model described by Young et al. (Equilabor, SP, Brazil). This brushing machine had four brushing arms; each one holding two brushing heads which ran over eight receptacles that housed the specimen (Figure 1). The tooth specimen was positioned on the specimen holder (Figure 2a and b) with the help of an adhesive (Figure 3), and a drop of a blue dye was added to the adhesive so that after brushing the adhesive can be easily scrapped off from the specimen surface. The slurry of dentifrice was contained...
over the specimen holder by passing it through a syringe of 5 ml, which was placed above the brush head (a hole, large enough to allow continuous flow of tooth powder slurry over the brush head was made in the syringe head). The slurry was prepared by mixing toothpowder with distilled water in the ratio of 1 mg/0.75 ml respectively in a rubber bowl. The head of the toothbrush (medium sized nylon bristles) was 32 mm long and 9 mm wide. The specimen was placed parallel to the brush head over the specimen holder so that brushing strokes were vertical.

In the present study, the specimens were subjected to a linear toothbrush abrasion movement with a rate of 200 brush strokes (back and forth) per minute, for 2½ h, totaling 30,000 brush strokes for each specimen (which is equivalent to 2 years of manual tooth brushing). The constant load applied was 200 g provided by metallic weights. After brushing was completed, the adhesive on the specimens was carefully scrapped off, and the specimens were rinsed with distilled water and gently dried with absorbent paper. The specimens were again weighed thrice in the analytical balance, and the mean reading was taken as the weight of the specimen postbrushing. The difference between weight of each specimen before and after brushing was taken as the amount of tooth substance loss (tooth wear).

**Evaluation of pH of different dentifrices**

A pH tester (Eutech Instruments waterproof pH Tester 30, National Analytical Corporation, Mumbai, India), which was calibrated in a pH meter standard solutions (pH 4, 7, 10.01) was used to measure the pH of all test specimens, both in artificial saliva and distilled water. One gram of test specimen was mixed in 5 ml (20% weight/volume) and 10 ml (10% weight/volume) of artificial saliva (pH of 6.8) and distilled water, respectively.

The specimens to be evaluated for pH were divided into four groups as follows:

- Group A - 1 g of specimen in 10 ml (10% weight/volume) of artificial saliva
- Group B - 1 g of specimen in 5 ml (20% weight/volume) of artificial saliva
- Group C - 1 g of specimen in 10 ml (10% weight/volume) of distilled water
- Group D - 1 g of specimen in 5 ml (20% weight/volume) of distilled water.

Samples were taken in centrifuge tube (Falcon, Discovery Labware, Tewksbury, USA) and placed in a vortex shaker (Yorco Corporate Towers, New Delhi, India) for 5 min so that all the constituents were properly mixed in their respective solutions. After vortex shaking, the tubes were placed in test tube holder so that the undissolved ingredients settled down at the bottom of the tube and the supernatant portion of the tube was collected by Eppendorf tube (Eppendorf India Limited, New Delhi, India, 100–1000 µl) into another centrifuge tube and was measured to check the pH change using pH tester.

**Detection of iron oxide and charcoal in different dentifrices**

The detection of Iron oxide was done by Inductively Coupled Plasma Mass Spectrometry (ICP-MS) (Perkin-Elmer-Élan DRC-E, Massachusetts, USA) method and charcoal was detected by physical method.

1 ppm of Stroke iron standard (Sigma-Aldrich Limited, India) was diluted to 2, 5, 10, 20 parts per billion (ppb) with milli-Q (HPLC grade) water to prepare different standards. 100 ml of sample was taken in a 100 ml volumetric flask and to that 5 ml ultrapure nitric acid (Sigma-Aldrich Limited, India) and 100 ml milli-Q (HPLC grade) water was mixed and placed inside ICP-MS for evaluation.

Inside the ICP, samples to be analyzed were introduced into the central channel, usually as a mist of liquid, formed by passing the liquid sample into a nebulizer. As droplets of nebulized sample entered the central channel of the ICP, it evaporated, and any solids that were dissolved in the liquid vaporized and were broken down into atoms. These dissociated ions were separated on the basis of their mass-to-charge ratio. Here, the standard was iron oxide. The detected ions gave a response in digits (area). The printout of the concentration of sample and acid blank coming from ICP-MS was taken and calculated to obtain the result in ppb.

**Calculation**

\[
\text{Metal content} = \frac{(\text{Reading of sample} - \text{Acid blank}) \times 100 \times 1.43}{\text{Weight of sample} \times 1000}
\]

where, \((\text{Reading of sample} - \text{Acid blank}) = \text{Sample response}\)

\(1.43 = \text{Factor of Iron Oxide}\)

**Physical method for detection of charcoal**

In this method, 3 g of sample (dentifrice) was taken in a Petri dish, and the suspension was made. The suspension was then diluted with suitable solvent (water 70% and methanol 30%). Then the solution was kept still for 10 min so that the un-dissolved particles got settled down at the bottom of the Petri dish. Then the Petri dish was kept over white paper background to be observed for black carbon particles, seen with magnifying lens.

**RESULTS**

The data collected was subjected to statistical tests (One-way analysis of variance [ANOVA] and Paired t-test) using the Stata 11.0 software (Stata Corp., College Station, Texas, USA). One-way ANOVA was used for multiple group comparison.
Paired t-test was used to compare tooth structure loss with different dentifrices. The data were presented as numbers, mean ± standard deviation or median (minimum–maximum). The p value was predetermined at 0.05.

The data of difference in weight (in grams) pre- and post-brushing has been listed in Table 1 as an indicator for abrasivity of all the dentifrices tested in this study. The data obtained were statistically significant. Among the groups brushed by different dentifrices, Lal Dant Manjan showed the maximum and Divya Dant Tooth Powder showed the least weight loss after the control group of toothpaste.

Comparative evaluation of tooth substance loss (in grams) between control group (Group 1) (Colgate toothpaste) and different test groups (Groups 2–8) of different tooth powders was done [Table 2]. All the groups showed statistically significant difference, except for Group 6 (Divya Dant toothpowder) when compared with the control group of toothpaste.

The pH of various dentifrices was evaluated. Vicco Tooth Powder showed the lowest pH value of 4.34 and 2.98 and Colgate toothpaste showed the highest pH of 8.48 and 7.67 in 10 ml of distilled water and artificial saliva, respectively [Table 3].

The presence of iron oxide and charcoal is shown in Table 4. The maximum iron oxide content was seen in Lal Dant Manjan and Baidyanath Tooth Powder. Charcoal was present only in Monkey brand toothpowder, among all the samples tested.

Analysis of shape and size of abrasive particles (iron oxide and calcium carbonate) in different dentifrices was done by scanning electron microscopy [Figures 4-11]. The size of iron oxide in Baidyanath Tooth Powder and Lal Dant Manjan was the largest, in the range of 20–25 µm, suggesting the coarse nature of iron oxide and high abrasivity compared to all the other tested tooth powders [Table 5].

**DISCUSSION**

In recent years, extensive tooth substance loss is seen in clinical practice and is found to be associated with toothpowder use. Some of the indigenous methods of teeth cleaning materials claim to have several beneficial effects.
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such as controlling bleeding from gums, preventing dental caries, reduction of sensitivity, and causing tooth whitening. However, in contrast to the claims made, extensive tooth substance loss, leading to severe hypersensitivity, exposure of dental pulp and total crippling of the natural dentition has been observed.[7-9]

It has been seen that oral hygiene practices across the country vary largely. People in semi-urban and rural areas use various types of toothpowders such as white, black, or red powders, bark of neem or mango trees, various Ayurvedic toothpowders, even charcoal, and tobacco powders as they are cheaper and easily available.

Table 3: Evaluation of pH of various test samples

| Samples                    | pH of 1 g/10 ml of distilled water | pH of 1 g/5 ml of distilled water | pH of 1 g/10 ml of artificial saliva | pH of 1 g/5 ml of artificial saliva |
|----------------------------|------------------------------------|------------------------------------|--------------------------------------|-------------------------------------|
| Colgate Tooth Powder       | 8.07                               | 8.46                               | 7.74                                 | 8.31                                |
| Baidyanath Tooth Powder    | 6.91                               | 6.85                               | 6.60                                 | 6.35                                |
| MDH Tooth Powder           | 6.71                               | 6.52                               | 6.79                                 | 6.71                                |
| Lal Dant Manjan            | 5.98                               | 5.88                               | 5.87                                 | 5.77                                |
| Vicco Tooth Powder         | 4.34                               | 4.39                               | 2.98                                 | 3.87                                |
| Divya Dant Tooth Powder    | 4.02                               | 5.41                               | 3.02                                 | 3.44                                |
| Monkey Brand Tooth Powder | 5.58                               | 6.00                               | 6.13                                 | 5.82                                |
| Colgate Tooth Paste        | 8.48                               | 8.80                               | 7.67                                 | 8.38                                |

Figure 5: Sample 2 - Monkey Brand Tooth Powder iron oxide shape – rhomboidal-shaped size - 4–7 µm calcium carbonate shape - irregular shaped size - 5–14 µm

Figure 6: Sample 3 - Divya Dant Tooth Powder iron oxide shape - cuboidal shaped size - 6–10 µm calcium carbonate shape - irregular shaped size - 5–12 µm

Figure 7: Sample 4 - MDH Tooth Powder iron oxide shape – cuboidal-shaped size - 2–4 µm calcium carbonate shape – irregular-shaped size - 15–20 µm

Figure 8: Sample 5 - Colgate Tooth Powder iron oxide shape - rod-shaped size - 15–20 µm calcium carbonate shape – irregular-shaped size - 4–8 µm
Various methods have been used to study abrasivity of dentifrices. These comprise measuring weight changes of the test object,\cite{18} surface profile measurements,\cite{11,12} electron microscopy,\cite{13} and radioactivity measurements.\cite{14,15} In this study, the weighing method was used to evaluate the tooth substance loss, as it allowed comparison of products under standardized conditions, at a reasonable cost and without too many technical difficulties.

The tooth brushing machine was indigenously fabricated, based on the model described by Young \textit{et al.} and pretested for number of strokes (equivalent to 2 years of manual brushing) and the force applied.\cite{16} A study on tooth wear evaluation using similar tooth brushing model was reported by De Freitas and Paranhos Hde.\cite{17} However, stroke/min was higher, 365 strokes/min (it was 200 strokes/min in the present study) but the time of brushing was lower, i.e., 100 min (it was 150 min in the present study). Probably, the increased frequency was used to compensate for decreased time of brushing.

Among all the products tested in this study, Lal Dant Manjan had the highest abrasivity, which was followed by Baidyanath Tooth Powder. The high abrasivity of red colored dentifrices could possibly be attributed to iron oxide as compared to other dentifrices. The red color could be imparted to dentifrices by red ochre.\cite{18} It is documented to be harshly abrasive to tooth enamel.

Abrasivity should be sufficient to remove surface deposits including dental plaque, but it should not damage enamel. Typically, this requires that particle size and shape of abrasive agents should be in a desirable range (i.e., 1–20 µm or 5–15 µm) and should not be sharp or angular.\cite{19} Crude red ochre, which typically contains clay minerals and/or other impurities in addition to the red iron oxide, may not be suitable for the purpose. Charcoal was found in Monkey brand toothpowder, which may cause permanent tooth discoloration and excessive abrasion.

The samples were also tested by adding them in artificial saliva to simulate the oral conditions during brushing, as saliva contains buffer to resist changes in pH and also provides a constant supply of ions to tooth surface. It also favors lubrication, hence, it may also control the abrasive wear to some extent. In this study, Vicco Tooth Powder (pH 4.34 and 2.98) and Divya Dant Tooth Powder (pH 4.02 and 3.02) had pH

\begin{table}
\centering
\caption{Iron oxide and charcoal content in different dentifrices}
\begin{tabular}{|l|c|c|}
\hline
\textbf{Samples} & \textbf{Iron oxide (w/w), %} & \textbf{Presence of charcoal} \\
\hline
Colgate Tooth Powder & 0.31 & Absent \\
Baidyanath Tooth Powder & 1.46 & Absent \\
MDH Tooth Powder & 0.48 & Absent \\
Lal Dant Manjan & 1.84 & Absent \\
Vicco Tooth Powder & 0.42 & Absent \\
Divya Dant Tooth Powder & 0.22 & Absent \\
Monkey Brand Tooth Powder & 1.21 & Present \\
Colgate Tooth Paste & 0.53 & Absent \\
\hline
\end{tabular}
\end{table}

\begin{figure}
\centering
\includegraphics[width=\textwidth]{Figure9.png}
\caption{Sample 6-Baidyanath Tooth Powder iron oxide shape - rod-shaped size - 20–25 µm calcium carbonate shape – irregular-shaped size - 4–10 µm}
\end{figure}

\begin{figure}
\centering
\includegraphics[width=\textwidth]{Figure10.png}
\caption{Sample 7 - Lal Dant Manjan iron oxide shape – rod-shaped size - 20–22 µm calcium carbonate shape - irregular shape size - 3–8 µm}
\end{figure}

\begin{figure}
\centering
\includegraphics[width=\textwidth]{Figure11.png}
\caption{Sample 8 - Colgate Tooth Paste iron oxide shape – rhomboidal-shaped Size - 12–14 µm calcium carbonate shape – irregular-shaped size - 4–10 µm}
\end{figure}
values <5.5. Thus they were more acidic as compared to other samples, which may cause more dissolution and softening of the dental hard tissues. The softened enamel then gets easily worn off by abrasive action of any external body like a toothbrush or normal occlusal forces. Although the pH of both Vicco and Divya Dant Powder was acidic, they showed lower abrasive values. Various factors which affect abrasivity include particle shape, size, hardness, distribution, and concentration. It has been shown that the abrasive wear rate increases linearly as the particle size and concentration is increased to a critical size.\(^{[20]}\) The particle size and concentration was found to be in desired range in both the aforesaid tooth powders accounting for their less abrasive behavior.

**CONCLUSIONS**

The result of the present study did support the above conclusions, as we found that the presence of iron oxide (high quantity) in Lal Dant Manjan and Baidyanath Tooth Powder, both of them showed the highest tooth structure loss when brushed with tooth brushing machine. This study tested eight different dentifrices with the same standard toothbrushes showing different tooth structure loss. The result demonstrated that toothbrushes, *per se*, were not responsible for tooth wear, but the dentifrices have a direct effect.

The present *in vitro* work was done on a limited sample size, simulating 2 years of tooth brushing. Further, longitudinal studies with larger sample size and longer duration of brushing cycles are required to positively substantiate the results of the present study.

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**Conflicts of interest**

There are no conflicts of interest.

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