THE BURNISHING EFFECT OF SODIUM (SALINE) CHLORIDE ON ROOT DENTIN: AN IN VITRO STUDY

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I - INTRODUCTION

An objective of periodontal treatment is the predictable regeneration of the periodontium in areas previously affected by periodontal disease (1). Adequate removal of plaque, calculus and cytotoxic substances from the diseased root surface appears to be essential for periodontal regeneration (2). In addition, the dentin surface smear layer produced by most forms of root manipulation could potentially affect fibroblast adaptation during healing of the periodontal wound. Root surface conditioning by topical application of acidic solutions has been shown to remove the smear layer resulting from root instrumentation (3, 4). Attempts to expose more collagen fibril material by altering the application technique have yielded positive results. STERRETT and BAIN revealed a "shag carpet" appearance of deeply tufted fibrils, using a burnishing technique by rubbing the dentin surface with a cotton pellet soaked in citric acid, more intertubular fibrils were exposed and dentinal tubules widened to a greater extent compared to passive application of the acid (5). Recently, it has been shown that the burnishing of root dentin with sodium chloride (saline) produced a surface free of smear layer (6). However, there is little available information comparing the effects of the duration of saline burnishing on root dentin. The purpose of this SEM study was to study the effect of saline burnishing on dentin specimens, as well as the influence of the duration of such treatment.

II - MATERIALS AND METHODS

Dentin blocks were prepared from 14 freshly extracted human impacted molars. The teeth were immediately cleaned and rinsed in distilled water and then stored for 36 hours at 4°C in distilled water until ready for use. All teeth were free from caries, cervical restorations or erosions. A high-speed hand piece with copious water coolant was used to resect the crowns at the cemento-enamel junction (CEJ). Following this, each root surface was thoroughly planed and flattened with a fine diamond tapered bur to expose the underlying dentin. Each root was then sectioned longitudinally into 6 equal parts and, using a N°3 round bur in a high speed hand piece with copious water coolant, a horizontal shallow groove was made in an apico-coronal direction on the pulpal surface of every root for identification purposes. The dentin specimens measured about 3 x 4 x 2 mm. This yielded a total of 72 dentin specimens, 12 specimens aving been discarded for non-conformity.

III - TREATMENT

The solutions used for treatment of the root dentin specimens were sodium chloride - 0.9 % (saline pH 5.1) and citric acid (pH 1.1). The pH of each solution was tested with a hand held, battery operated pH meter Model 5941.00.1. The specimens were divided into two equal groups of 36 specimens each.

Group A: Thirty-six dentin blocks were randomly divided into 6 sub-groups, with each tooth contributing one block to each sub-group. The following conditions applied to each of the sub-groups: sub-groups I, II, III, IV and V were soaked in sodium chloride for 5s, 10s, 30s, 1 min. and 2 min respectively. The specimens in sub-group VI were soaked in citric acid for 3 min and served as controls. Group B: The remaining 36 blocks were assigned to sub-groups in exactly the same manner as for Group A, except that the specimens were subjected to burnishing with saline and citric acid (control) for the equivalent periods as for Group A.

SEM Preparation

All specimens were prepared for scanning electron microscopy (SEM). After fixation, dehydration was
done in a graded series of ethanol and with 100% acetone as a final step. Each of the sectioned pieces was mounted on aluminum stub, coated in gold with a sputter technique. The specimens were examined in the scanning electron microscope (2) operated at 25 KV and with a tilt angle of between 0° and 40°. Micrographs were taken at 2000-x magnification. The roots were examined with respect to presence or absence of smear layer and to exposed collagen fibers. Presence of open dental tubuli on the root surface was the primary criterion of the efficacy of the smear layer-removing capacity of the treatment.

IV - RESULTS

The responses of each of the specimens within particular conditioning regimens are presented in Table 1.

Table 1 : Effects by the conditioning agents on root dentin

<table>
<thead>
<tr>
<th>Groupe A : Soaked specimens</th>
<th>Citric acid 3 minutes</th>
<th>Saline 5 s.</th>
<th>10 s.</th>
<th>30 s.</th>
<th>1 min.</th>
<th>2 min.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Smear layer</td>
<td>No</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Cracks</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Exposed dentin</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Exposed dentinal tubules</td>
<td>Open</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
</tbody>
</table>

Groupe B : Burnished specimens

| Smear layer                | No                    | Yes         | Yes   | Yes   | No     | No     |
| Cracks                     | No                    | Yes         | Yes   | No    | No     | No     |
| Exposed dentin             | Yes                   | Yes         | Yes   | No    | Yes    | Yes    |
| Exposed dentinal tubules   | Open                  | No          | Yes   | No    | Yes    | Yes    |

The soaked saline specimens, irrespective of duration, exhibited an amorphous, irregular surface coating completely obscuring the dental tubules (Fig. 1). In no instance was the dentin surface or any patent dental tubuli visible. All the citric acid treated specimens (Group A and B, sub-group VI) showed a similar appearance. The dentin surface was readily visible in all specimens as evidenced by lack of smear layer and abundance of patent dentinal tubules (Fig. 2). All of the dentinal tubules were exposed and did not reveal any intensified peritubular dentin at the surface.

Figure 1: SEM photomicrograph of dentin surface soaked in saline for 1 minute. The surface is covered by remnants with an irregular pattern (smear layer). (Original magnification x 2000)

Fig. 2: SEM photomicrograph of dentin surface soaked in citric acid for 3 minutes. There is no smear layer and the presence of widely exposed dentinal tubules. (Original magnification x 2000).

In the burnished saline specimens (Group B), there were no differences in root surface texture for the different treatment times, expect for the 1 and 2 minutes exposure sub-groups (IV and V).

After saline burnishing for 5, 10 and 30 seconds, the root surface revealed a homogenous smear layer that plugs the dentinal tubuli openings. These plugs made it difficult to detect and count even partially plugged tubule openings (Fig. 3).
Figure 3 : SEM photomicrograph of dentin surface burnished with saline for 30 seconds. A homogenous layer of smear layer plugs the visible dentin tubuli openings (Original magnification x 2000)

Burnishing the root dentin for 1 and 2 minutes revealed distinct areas with patent dentinal tubules and an intertubular area of "matted" fibrillar matrix (Fig.4). Occasionally grooving appeared in this fibril surface, it should be noted that the peripheral areas of the control dentin sections exhibited a tufted fibrillar surface.

Figure 4 : SEM photomicrograph of dentin surface burnished with saline for 2 minutes. There is a removal of the smear layer and dentinal tubule orifices are visible. Note the presence of fibrillar collagen network (Original magnification x 2000)

V - DISCUSSION

As saline is not widely accepted as a root conditioner agent but mainly as a storage medium (8) it was decided to use a universally accepted root surface conditioner as control in this study. Citric acid used as a root conditioner for 3min either with the soaking technique(9) or the burnishing technique(10) has been shown to be effective in removing the smear layer and exposing dentinal tubules. After mechanical instrumentation of a root surface, a smear layer is usually present (11). It has been characterized as an amorphous structure obscuring the underlying dentin surface (3). Such a layer has been shown to vary in thickness from one location to the next, and may well comprise different percentages of organic and inorganic material ranging in size from less than 1 µm to more than 15 µm (12).

The detrimental role of such a surface covering in periodontal healing is now recognized (3). The burnishing technique is a marked deviation from former conventional demineralization techniques (13, 14).

MILLER suggested that one reason for his clinical success was that citric acid conditioning removed a "smear layer" from the root surface. In so doing, the dentinal root surface becomes more "hospitable" to cell attachment and subsequent new connective tissue attachment formation.

The purpose of the present study was to explore the possibility of obtaining an acceptable smear removing and collagen exposing effect using saline burnishing. None of the different timings (5, 10 and 30seconds) applied with the burnishing technique could completely remove the smear layer from the tubule openings. However, burnishing with saline for 1 and 2 minutes removed the superficial smear layer and exposed the dentinal tubules with evidence of collagen fibrils exposed on the dentinal surface. SEM photomicrographs of saline burnished samples at application times of 1min and 2min are evidence that smear layer removal is time dependent. It appears reasonable to assume that a longer exposure time would enhance the smear removing effect. This is in agreement with TROMBELLi et al who evaluated the effect of tetracycline hydrochloride 10 mg/ml and 100 mg/ml and found that the morphologic alterations produced were related to the duration of application rather than to the concentration applied.

STERRETT et al. found that effectiveness of TC-HCL>75 mg/ml was time dependent and concentration and concentrations at 3 and 5 minutes were more effective...
than 1 minute (16) a possible explanation for these varying results could be the frequency of solution application. Two independent studies concluded that optimal demineralization might be due to a number of factors including application technique and time (5, 17).

A recent investigation on dentin demineralization with the burnishing technique concluded that it is not time dependent at concentration of between 50 mg/ml and 150 mg/ml, neither it was time dependent at 1, 3 and 5 minutes (18). These results contrast with the findings of the present study when burnishing with saline was applied to the root dentin. The data indicated that 1 mn and 2 mn burnishing with saline produced significant results in relation to smear layer removal and collagen fibers exposure. In the burnishing technique, this study concurs that the application technique and the timing used are the most important factors that affects the results.

Studies by BLOMLÖF and LINDESKOG (19) and Bergenholz and Babay6 claimed that burnishing root dentin for 20 seconds with citric acid and for 10 seconds with saline were sufficient to remove the smear layer. In this study 5, 10 and 30 seconds of burnishing root dentin with saline were not sufficient to remove the smear layer (Fig.3). Differences in the burnishing technique (light versus heavy) may explain these contradictory results.

WEN et al have shown that excessive pressure, as with burnishing, may mask or flatten the exposed dentin matrix, creating a "self-induced" smear layer with obliterated tubules or peeling of the exposed fibrillar structure (20).

These observations are further supported by morphometric analyses of the surface area occupied by tubule orifices. Pashley et al studied the effect of burnishing by different pastes on dentin permeability, and concluded that burnishing alone created a partial smear layer occluding the orifices of dentinal tubules, thus producing a significant decrease in dentin permeability (21).

Thus differences in the topography of the root dentin in different studies may be attributable to factors such as application pressure and time of application. As the applied pressure in this study was not standardized, it is appropriate to consider the application pressure as an explanation factor. The non-removal of the smear layer by saline in Group B (subgroups I, II and III) may be explained by its astringent action. This astringent action combined with the application pressure may have contributed to its formation. It is conceivable that the 1 and 2 minutes applications may have been associated with lighter pressure, helping it remove the superficial portion of the smear layer and exposing the dentinal tubule. As advocated by Wen, it may be undesirable to apply excessive pressure irrespective of the conditioning agent (20). According to the results of the present study, it may be desirable to apply saline for at least 1 minute with light burnishing to maximize the tubular openings but whether this burnished surface is more or less acceptable than the one produced by the other methods is only a conjecture. Although smear layer removal appears to be time dependent with saline burnishing of the root dentin, the role of application pressure needs further investigations.

VI - CONCLUSIONS

Based on the findings of this in vitro study, the following conclusions can be drawn:

1. Soaking the root dentin in saline did not result in the disappearance of the smear layer irrespective of the time used.
2. Burnishing the root dentin with saline for up to 30 seconds did not remove the smear layer.
3. A one and two minutes saline burnishing application to the root dentin removed effectively the smear layer and exposed a fibrillar network of collagen fibers.
4. Citric acid application for 3 minutes on the root dentin irrespective of the application technique used, removed the smear layer completely.

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ABSTRACT
The purpose of this in vitro study was to evaluate the burnishing effect of saline application on dentin specimens (NaCl 0.9 %). Seventy-two dentin specimens (60 experimental and 12 controls) were equally divided into two groups. In Group A: sub-groups I, II, III, IV and V were respectively soaked with saline for 5s, 10s, 30s, 1min and 2min. Group VI was soaked in citric acid for 3min and served as control. Identical treatment was done in-group B with one exception, burnishing instead of soaking was applied to the root surface. Five, ten and thirty seconds burnishing with saline and all the different timings with the soaking technique did not remove the smear layer. This was in contrast to burnishing dentin surface for 1and 2min, which effectively removed the smear layer. It cannot be excluded that the smear layer removal is time dependent when the dentin is burnished with saline.

Key words: Burnishing, dentin, saline, SEM.

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RESUME
Le but de cette étude in vitro est d'évaluer l'effet polissant du sérum salé sur des échantillons de dentine (NaCl 0,9 %). 72 échantillons (60 expérimentaux et 12 témoins) ont été divisés à égalité en deux groupes. Dans le groupe A, les sous-groupes I, II, III, IV et V ont été trempés respectivement dans le sérum salé pendant 5s, 10s, 30s, 1 min et 2 min. Le groupe VI a été trempé dans l'acide citrique pendant 3 minutes et a servi de témoin. Le groupe B a reçu un traitement identique au groupe A, à l'exception du traitement des échantillons. Les échantillons ont été polis au lieu d'être trempés. Tous les différents temps d'utilisation quand les échantillons ont été trempés et aussi le polissage à 5s, 10s et 30s n'ont pu éliminer la couche de débris. Il a été conclu que l'enlèvement de la couche de débris dépend de l'utilisation quand les échantillons sont polis au sérum salé (NaCl 0,9%).

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