

## SEM STUDY OF DIFFERENT TREATMENTS OF THE SMEAR LAYER ON DIFFERENT CAVITY WALLS

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**KEYWORDS:** Smear layer, treatment of cavity walls, dentin adhesion, cavity cleanness, smear layer treatment.

**MOTS CLES:** Enduit pariétal, traitement de las murailles cavitaire, adhésion dentinaire, nettoyage de le cavitaire, traitement de le enduit pariétal

### ABSTRACT

The present study evaluated the effects of slightly demineralizing treatments on dentinal cavity walls, since some recently developed adhesive procedures are applied over the smear layer. Ten experimental treatments — mechanical, chemical or mechanical/chemical — were applied on MOD cavity walls prepared in vitro with diamond burs. The dentinal surface of a lateral and of a pulpal wall of each cavity was evaluated through scanning electron microscopy, and the effects of the treatments were compared. All treatments except air/water spray removed some of the smear layer, and slight differences were observed regarding the studied cavity walls: on dentin from pulpal walls the enamel hatchet associated with tannic acid produced a better effect than the other treatments, and on dentin from lateral walls the biological detergent rubbed with cotton pellets was a little more effective than the other treatments. The effect of the smear layer treatment on dentin is different according to the wall and to the applied treatment

### RESUME

Cette étude a évalué les effets des traitements légèrement déminéralisants des parois des cavités dentinaires, vu que quelques techniques d'adhérence récemment développées ont été appliquées sur l' enduit pariétal. Dix traitements expérimentaux – mécaniques, chimiques ou mécaniques/chimiques - ont été appliqués aux parois des cavités MOD préparées in vitro grâce à des fraises à pointe de diamant. La surface dentinaire d'une paroi latérale et d'une paroi pulpaire de chaque cavité a été examinée au microscope à scansion électronique, et les effets des traitements ont été comparés. Tous les traitements, à l'exception des vaporisations d'air/eau ont enlevé un peu de enduit pariétal et de légères différences ont été observées à propos des parois des cavités étudiées: sur la dentine à partir des parois pulaires, le *hatchet* de l'émail associé à l'acide tannique a donné un meilleur résultat que tout autre traitement; sur la dentine à partir des parois latérales, le détergent biologique frotté avec des tampons d'ouate s'avéra un peu plus efficace que les autres traitements. L'effet du traitement de la couche tachée sur la dentine varie selon la paroi et le traitement appliqué

### INTRODUCTION

Investigations have shown that the penetration of monomers into the intertubular dentin, creating the hybrid layer, may be more important than chemical adhesion or formation of resin tags within the dentinal tubules (Nakabayashi,1985; Wang et al.,1991; Swift et al.,1995). However, excessive demineralization of dentin may produce denaturation of collagen, and resins may not penetrate into the matrix as deeply as do acidic conditioners. Thus, a weak zone may be created, with unprotected collagen, causing the failure of dentinal bonding (Nakabayashi et al., 1996; Pashley et al., 1997). These facts led to the development of adhesives with

acidic monomers, i.e., the smear layer may be slightly demineralized due to the association of acids with hydrophilic/hydrophobic monomers allowing a concomitant diffusion through the smear layer into the underlying dentin, creating a hybrid layer of unaltered collagen fibers enveloped by resin (Nakabayashi,1985; Wang et al.,1991; Nakabayashi et al., 1996; Van Meerbeek et al., 1993; Van Meerbeek et al.,1998).

Since the smear layer contains contaminants it is conceivable that a pre-treatment of the cavity walls may minimize the smear layer and also remove, at least partially, its contaminants (White et al.,1989; Prati et al.,1990). For this reason, we examined the effect of some non- or slightly demineralizing treatments, alone

and in combination with a mechanical agent, through the SEM appearance of the smear layer on dentinal cavity walls prepared in vitro in human teeth.

## MATERIAL AND METHODS

Thirty fresh, non-carious, unerupted human third molars, obtained from patients between 16 and 27 years of age (mean age: 21 years), were studied. All patients were fully informed about the procedures of this study. Roots were removed, and mesio-occlusodistal (MOD) cavities were prepared with new cylindrical diamond burs under water-cooling. Immediately after cavity preparation, each one of the ten treatments listed in Table 1 was carried out on three teeth, on each cavity wall. The enamel hatchet (treatment 6) was applied for 30 seconds, and when associated with the chemical substances (treatments 7-10) an additional 15 seconds period of concomitant application was performed (Table 1). Air/water spray was applied for 5 seconds before and after all of the treatments.

SUBSTANCES	APPLICATION METHOD	TIME
(1) air/water spray (negative control)	---	15
(2) 5% tannic acid	Cotton pellets	15 s
(3) biological detergent "Tergensol"* (0.2% lauryl sodium sulfate)	Cotton pellets with rubbing	15 s
(4) 0.5% sodium hypochlorite	Cotton pellets with rubbing	15 s
(5) enamel hatchet	---	30 s
(6) enamel hatchet with 5% tannic acid	---	45 s
(7) enamel hatchet with biological detergent	---	45 s
(8) enamel hatchet with 0.5% sodium hypochlorite	---	45 s
(9) enamel hatchet with 37% phosphoric acid	---	45 s
(10) 37% phosphoric acid (positive control)	Cotton pellets	15 s

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**Table 1.** Treatments of cavity walls

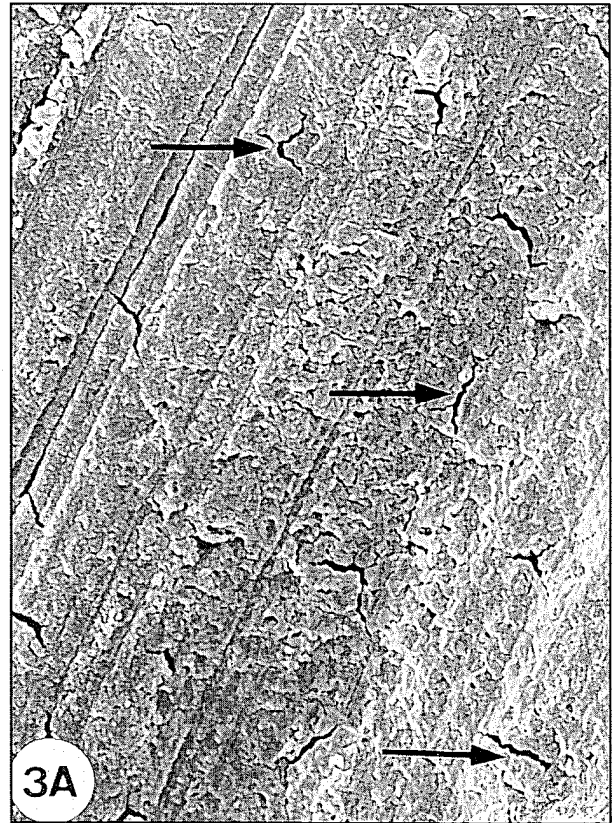
The teeth were longitudinally fractured into two halves in mesiodistal orientation with a chisel. Then, one of the fragments was processed for scanning electron microscopy. Specimens were air-dried and mounted on aluminum stubs. After sputtering with a 40 nm layer of gold in a Balzers SCD 050 apparatus, the treated cavity wall surfaces were examined in a JEOL 6100 scanning electron microscope operating at 10-15 kV. Electron micrographs were analyzed and compared with each other with regard to the degree of removal of the smear layer and the consequent visualization of the underlying dentin (Brannstrom et al., 1979).

## RESULTS

Cavity walls treated with air/water spray (negative control) exhibited variable amounts of smear layer covering them, with superficial particles not firmly attached to the underlying smear layer. Grinding marks produced by the diamond particles of the instrument were evident on the surface of pulpal walls (Fig. 1). Additionally, these marks appeared as parallel lines on the surface of lateral walls of the cavities (not illustrated). In general the depth of the marks roughly indicated the amount of smear layer that was created.



**Fig. 1** - Air/water spray (negative control). Electron micrograph showing all of the pulpal wall surface covered by smear layer; grinding marks of the diamond bur can be observed (460X).

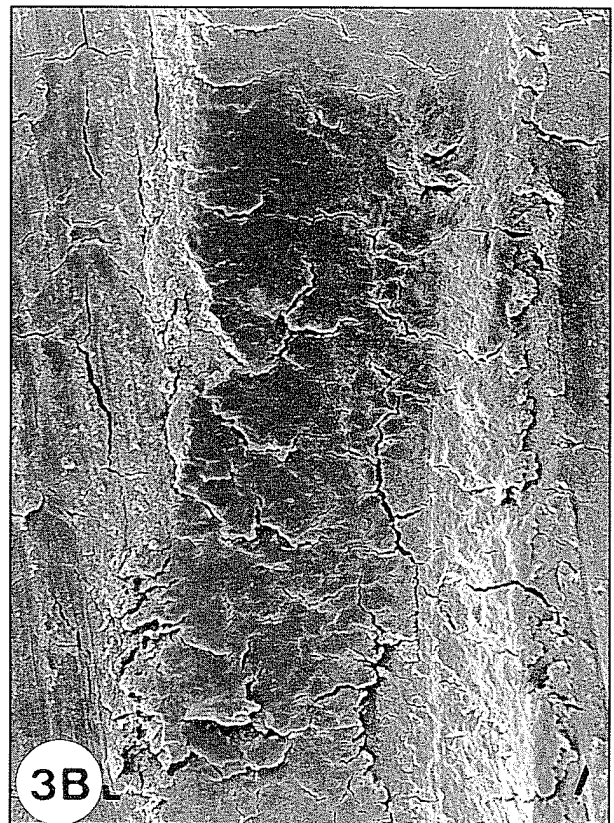


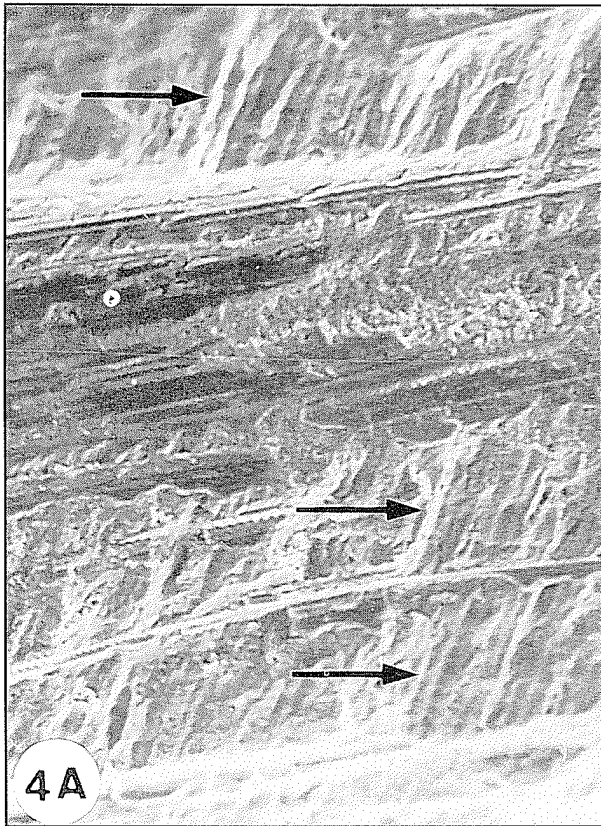
**Fig.2** - 5% tannic acid. Electron micrograph showing a pulpal wall with a smear layer covering all dentinal structures. The diamond bur grinding marks are evident all over the surface (400X).

**Fig.3** - Enamel hatchet combined with 5% tannic acid. Electron micrograph showing, in A, the smear layer on a pulpal wall with a smooth appearance; arrows indicate transversally cut dentinal tubules (2100X). In B, a lateral wall showing the smear layer covering the dentinal structure (440X).

**Fig.4** - Biological detergent. Electron micrograph showing, in A, a lateral wall with an apparently thin smear layer, because some profiles of dentinal tubules (arrows) appear cut in longitudinal sections (700X). In B, a pulpal wall in which the smear layer is covering the underlying dentinal structures. The diamond bur grinding marks are evident all over the surface (450X).

**Fig.5** - Enamel hatchet combined with biological detergent. Electron micrograph showing a pulpal wall totally covered with smear layer masking the dentinal surface. Some smooth areas (arrows) produced by the enamel hatchet are seen but no dentinal structure can be identified (400X).



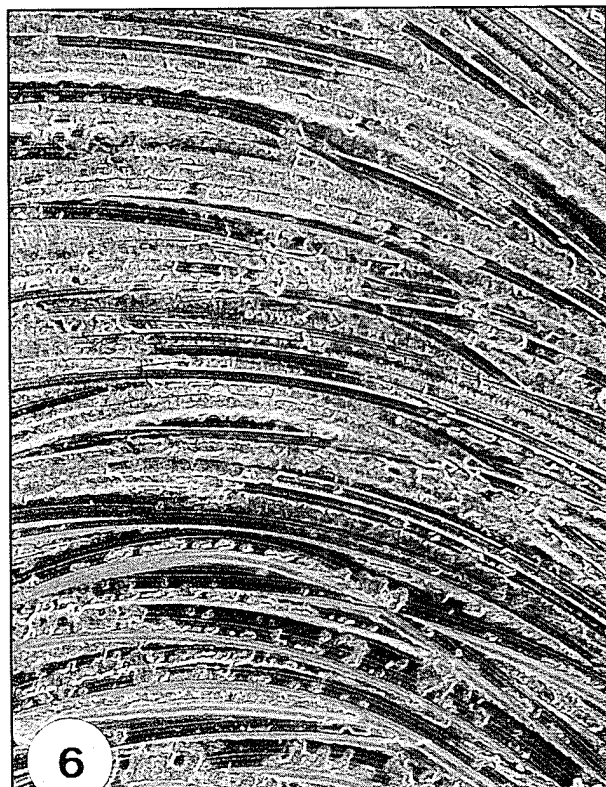


Treatment with 5% tannic acid produced a very slight removal of the smear layer on the dentinal surface of pulpal or lateral walls, rarely showing the underlying structure (Fig. 2). When associated with the use of an enamel hatchet, tannic acid produced a smear layer with a smooth dentinal surface on pulpal walls. The location of the dentinal tubules could be identified by cracks in the smear layer over their lumina (Fig. 3-A). In contrast, on lateral cavity walls this treatment produced a very subtle removal of the smear layer, with no evidence of the dentinal structure (Fig. 3-B).

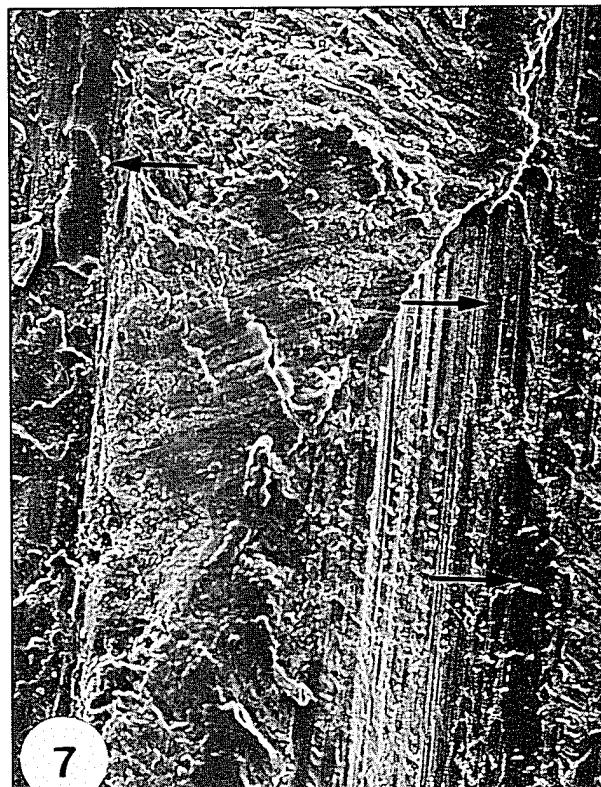
The biological detergent treatment produced a moderate removal of the smear layer on the dentin of lateral cavity walls, allowing the visualization of longitudinal sectioned dentinal tubules. Apparently, a thin smear layer persisted on these walls, although the removal of superficial particles was evident (Fig. 4-A). However, this aspect could not be observed on pulpal walls where only a slight removal of the smear layer occurred. Thus, a rough superficial smear layer persisted, with strong evidence of the action of the rotary instrument used to create the smear layer (Fig. 4-B). When the detergent was applied in combination with the enamel hatchet, it produced a very slight removal of the smear layer (Fig. 5).







**Fig.6** - 0.5% sodium hypochlorite. Electron micrograph of a pulpal wall showing the smear layer totally covering the underlying dentinal surface. The grinding marks of the diamond bur are still very evident (400X).



**Fig.7** - Enamel hatchet combined with 0.5% sodium hypochlorite. Electron micrograph of a lateral wall showing the smear layer covering all the dentinal surface. It is not possible to identify any dentinal structure. Some small smooth areas (arrows) may identify the enamel hatchet action that was not so evident as it was on pulpal walls (400X).

The treatments with sodium hypochlorite produced a very subtle removal of the most superficial portion of the smear layer on cavity walls, and free smear particles could still be observed on the surfaces (Figs. 6 and 7). The results obtained with 37% phosphoric acid (positive control) showed a complete removal of the smear layer, revealing the tubular structure of dentin. Thus, the tubules appeared opened and enlarged, showing a funnel-shaped aspect (not illustrated). The intertubular dentin exhibited a clean and smooth aspect (not illustrated). When phosphoric acid was applied in combination with the use of an enamel hatchet, the removal of the smear layer was similar to that observed with phosphoric acid alone. Some areas with occluded tubules could be observed, although no smear layer or smear particles were present in the intertubular dentin.

## DISCUSSION

The present study shows that all of the non- or

slightly demineralizing treatments of cavity walls produced some removal of the dentinal smear layer. In general, the effects of the non- or slightly demineralizing treatments were similar, with very subtle differences between them. As expected, the performance of phosphoric acid applied as a positive control treatment exceeded that of the others, completely removing the smear layer. The treatment of cavity walls with phosphoric acid has been extensively studied in adhesive restorative procedures. As the hazardous effects of phosphoric acid on dentin were demonstrated both in dentinal structure and in dentinal permeability (Swift et al.,1995 Pashley,1992), self-etching adhesive primers were developed to minimize dentin etching (Wang and Nakabayashi,1991; Nakabayashi and Saimi,1996). However, application of these self-etching primers to smear layer contaminated by blood, saliva or bacteria (Brannstrom et al.,1974) may compromise bonding. Our study intended to evaluate the removal of the superficial layer of the smear layer as a means of improving it as a bonding substrate.

The methods of application of the substances were carried out based on the literature: acid solutions in general were applied passively with cotton pellets without rubbing, and organic solvents or detergents, with rubbing (Brannstrom and Johnson,1974; Bitter,1989; Bitter,1990; Takahashy et al.,1993). Moreover, the study of treatments on the walls of MOD cavities allowed the observation of their effects on different cavity walls, rather than on flat dentinal surfaces, thereby making the study more clinically relevant. In addition, the absence of proximal walls in the MOD cavity model facilitated the coronal fracture after treatment, in order to prepare the teeth for SEM observation.

When 5% tannic acid was applied alone, similar results were obtained on both lateral and pulpal dentinal walls. However, when tannic acid was used in combination with an enamel hatchet, we observed slightly better results on pulpal walls than on lateral walls. The smaller removal of the smear layer on lateral walls could be due to the cavity design. The use of the enamel hatchet on pulpal walls may have left a thinner smear layer, thereby increasing the effect of the tannic acid on these walls. The effect of the biological detergent was satisfactory and it seems to have had a better effect on lateral walls. Since this treatment was applied with rubbing using cotton pellets, the cavity design might also have facilitated this procedure, as the simple friction of cotton pellets with water may result in some removal of the smear layer (Brannstrom and Johnson,1974). The advantage of using detergents is their cleaning action, which facilitates the removal of smear layer contaminants, mainly rotor lubricants, and also superficial smear particles. The 0.5% sodium hypochlorite produced a very subtle removal of the smear layer, which is in accordance with previous studies. Sodium hypochlorite acts as an antiseptic agent, and it may increase the mineral concentration of the dentinal surface, as it also removes organic material (Cameron,1987; Baumgartner and Cuenin,1992). This fact may facilitate the union between glass ionomer cements or polycarboxylate cements and dentin or enamel (Prati et al.,1989;Van Dijken,1990;Tanaka and Nakai,1993).

## CONCLUSIONS

Non- or slightly demineralizing treatments produced little removal of dentinal smear layer. Their effects varied according to the effectiveness of the mechanical agents employed on the different cavity walls.

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