EFFECT OF SODIUM HYPOCHLORITE ON THE BOND STRENGTH OF AN ADHESIVE SYSTEM TO SUPERFICIAL AND DEEP DENTIN

EFEITO DO HIPOCLORITO DE SÓDIO NA RESISTÊNCIA DE UNIÃO DE UM SISTEMA ADESIVO NA DENTINA SUPERFICIAL E PROFUNDA

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The objective of this study was to evaluate the bond strength to superficial (SU) and deep (D) dentin, accessed via apical (DA) or occlusal (DO), using One-Step adhesive system applied according to the manufacturer's instructions (C) or following deproteinization with 10% sodium hypochlorite (H) for 60s, after acid etching. Three sound extracted human molars were prepared for each experimental condition. Restorations were performed using Z100 in 2mm increments, each one being light-cured for 40s. Teeth were longitudinally sectioned to obtain stick-shaped specimens with a cross-sectional area of 0.8mm², which were submitted to bond strength test (0.6mm/min). Results obtained after statistical analysis using a two-way ANOVA (substrate vs. surface treatment) and Tukey's test were: SU (35.4 ± 12.3), DO (26.5 ± 8.5), DA (26.1 ± 10.2) following conventional surface treatment, and SU (28.82 ± 12.7), DO (24.3 ± 8.3) and DA (23.5 ± 8.5), after surface treatment using sodium hypochlorite. The interaction of the factors was not significant ($p>0.05$). However, the main factors were significant ($p<0.05$). Mean bond strength values in superficial dentin were statistically superior to deep dentin ($p<0.05$), although no statistically significant difference was observed between the two via of access to deep dentin (SU>DO=DA). The conventional surface treatment resulted in higher bond strength values than the sodium hypochlorite treatment ($p<0.05$). It was concluded: 1) bond strength values were higher in superficial dentin, 2) no difference was found between the two deep substrate preparations, and 3) the application of sodium hypochlorite following dentin acid etching may reduce bond strengths.

UNITERMS: Depth dentin; Sodium hypochlorite; Adhesive system.

INTRODUCTION

The dentin acid etching was firstly proposed by Fusayama, et al.[8] (1979) and allowed the complete elimination of smear layer and smear layer plugs. This approach facilitated the penetration of adhesive systems into inter and intratubular dentin, resulting in the formation of a hybrid layer and resin tag formation[15]. High microtensile bond strength values have been reported in the literature, and enhanced retention of class V resin composite restorations was mainly attributed to hybrid layer formation[20,26,28].

It has been already demonstrated that dehydration of collagen fibrils exposed by acid etching determines an abrupt reduction of dentin matrix permeability, compromising infiltration of hydrophilic resin monomers[3,4] and the subsequent hybrid layer formation. Therefore it is essential to preserve the dentin moist in order to preserve the collagen fibrils expanded prior to adhesive application[14]. However, no objective criteria about how to keep the ideal moisture, in which maximum infiltration of resin monomers can occur is yet to be addressed. It seems that there is a strong relationship between the composition of each employed bonding agent and the ideal moisture degree on dentin surface. As a result, a different degree of surface moisture is necessary for each adhesive system, making the establishment of such specification difficult due to the great variety of materials present in the market[21,22].

It can be noticed that the maintenance of the collagen network permeability after acid etching is critical. In face of that, some studies have proposed the removal of exposed collagen using deproteinizing agents[29,12,25]. Some researchers state that the elimination of exposed collagen using sodium hypochlorite not only reduces technique sensitivity, but also allows the achievement of a more porous surface, which would be responsible for adhesive interfaces with similar bond strength values to those obtained in enamel after etching with phosphoric acid[29]. Other authors speculate that these high bond strength values obtained after deproteinization of conditioned dentin would be mainly due to the formation of larger resin tags and innumerous lateral branches between them, since a hybrid layer can not be formed when exposed collagen is removed[25,18].

Based on this concept, it could be speculated that the effect of dentin deproteinization would be more evident in deep substrate areas, as in these regions the area of intertubular dentin is smaller[17] and therefore the contribution of resin tags and lateral branches on the resin retention would be more significant. Consequently, it is expected that the use of sodium hypochlorite to remove collagen fibrils from areas of deep dentin should guarantee increased bond strength compared to superficial dentin areas, since in superficial areas the tag formation is lower and the retention of resin composite is mainly due to hybrid layer formation[17].

Hence, the objective of this *in vitro* study was to evaluate the microtensile bond strength of a one-bottle adhesive system to superficial and deep dentin, after removing the exposed collagen following acid etching of the substrate.

MATERIAL AND METHODS

This study was approved by the Institutional Ethics Committee under protocol number 86/02 process 55/02. Eighteen sound recently extracted human molars were obtained and stored in 0.5 % chloramine solution[5].

The occlusal surfaces of twelve teeth were ground flat using 220-grit sandpaper under copious water cooling. For 6 teeth (superficial dentin group - SU), grinding was performed until the elimination of occlusal enamel (Figure 1A). For the other 6 teeth (deep dentin via occlusal - DO), superficial dentin was ground until exposure of deep substrate without reaching pulp horns (Figure 1B).

The remaining 6 teeth (deep dentin via apical group - DA) had their roots sectioned parallel to the occlusal surface, at the height of pulp horns (Figure 1C). This surface was then flattened (#220-grit sandpaper) until complete elimination of any remaining pulp. After that, all dentin surfaces were polished with 600-grit sandpaper to standardize smear layer formation.

Teeth of SU, DO, DA groups were again divided. Half of each group (n=3) was restored using One-Step adhesive system (Bisco, USA) applied according to the conventional method (C), and strictly following the manufacturer's instructions (Table 1). The other half was acid etched with phosphoric acid, rinsed and

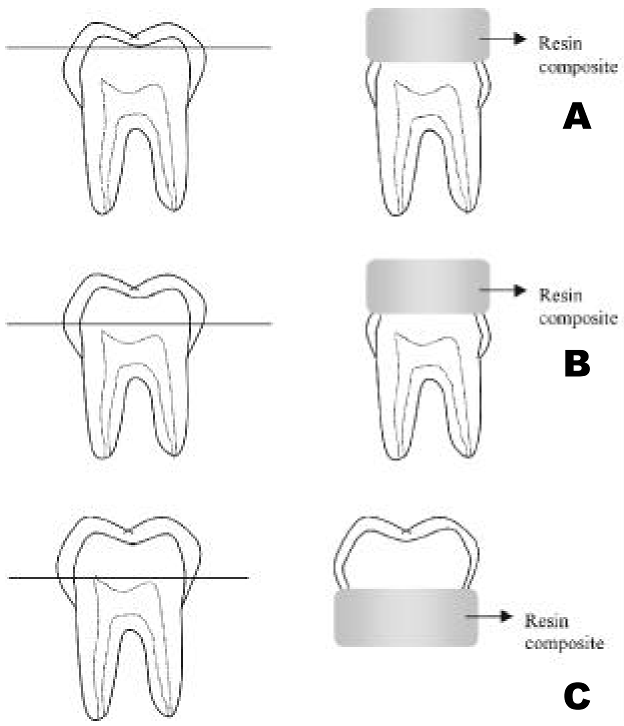

FIGURE 1 - (A) superficial dentin preparation; (B) deep dentin preparation achieved via occlusal access, and (C) deep dentin preparation achieved via apical access.

then submitted to surface treatment with 10% sodium hypochlorite solution for 60 s (H). The dentin surfaces were washed thoroughly (15 s) and completely dried for both groups. The moisture standard were kept the same for both groups, as the same adhesive system was employed. An amount of 3,5 µl of water was applied on conditioned surface by means of a micropipette (Pipetman, Gilson[22]) before the application of One Step adhesive system (Table 1).

Two layers of Z100 resin composite (3M, USA), approximately 2 mm high, were placed on the prepared dentin surfaces. Each layer was light-cured for 40 s using a VIP light-curing unit (Bisco, USA), with light intensity of 600 mW/cm^2. After storage in water at 37^{o}C for 24 h, specimens were sectioned using a diamond saw mounted in a Labcut 1010 machine (Extec, USA). Sequential perpendicular sections to the bonding interface were made, so that stick-shaped specimens were obtained with a cross-sectional area of approximately 0.8 mm^2. The precise measurement for each specimen was recorded using a digital caliper (Absolute Digimatic, Mitutoyo, Japan). Each specimen was then fixed to a jig consisting of a modified caliper[1] using cyanoacrylate glue (Zapit, DVA, USA), and then adapted to a Kratos universal testing machine (Kratos Dinamômetros, Brazil). Specimens were submitted to microtensile bond strength test at a crosshead speed of 0.6 mm/min. The mode of fracture for each specimen was confirmed using a light stereoscopic with X2 magnification and classified according to the following criteria: adhesive (A), cohesive (C) and mixed adhesive (M/A) fracture.

The specimens that presented “A” fractures were subjected to statistical analysis using two-way ANOVA (substrate, surface treatment and interaction). Tukey’s test was applied to contrast the means values. The cross-sectional areas of stick-shaped specimens were compared among groups using one-way ANOVA (mm^2) in order to verify the data homogeneity, since it is already known that the bond strength values may be influenced by adhesive areas dimension[2].

RESULTS

No statistically significant difference was observed among the mean cross-sectional areas ($p>0.05$) (Table 2).

The interaction of the main factors were not statistically significant ($p>0.05$). The main factors were statistically significant ($p<0.05$). Microtensile bond strength results are demonstrated in Table 3. The mean microtensile bond strength value obtained for superficial dentin (SU) was statistically superior to both deep dentin preparations ($p<0.05$). Statistically similar results were observed between mean values obtained in both deep preparations ($p>0.05$), indicating that the access via (apical - A or occlusal - O) had no influence on microtensile bond strength results as noted in table 3. It was also observed, that specimens prepared according to the conventional method (C) presented significantly higher mean values than those reported for specimens submitted to sodium hypochlorite (H) surface treatment ($p<0.05$).

DISCUSSION

Superior bond strength values to those obtained for the conventional hybridization technique were

TABLE 1- Material, batch number, composition and mode of application of each material

Material/ Batch#	Composition	Mode of application
One Step (Bisco) CE0459	1. Uni-Etch –32% phosphoric acid 2. Adesivo – Bis-GMA; BPDM; HEMA, iniciator, acethone	a, b, c, d, e, f, g
Z-100 (3M ESPE) FL29	1. Inorganic matrix: 71% of zircon/silica filler volume with particle size of 0.19 to 3.3 µm 2. Organic matrix: Bis-GMA, TEGDMA	h

a – acid etching (15 s); b – washing using air-water spray (15 s); c – air-dry d – dentin rewetting with 3,5 ml of water e – application of 2 coats of adhesive under agitation for 10 s each; f – drying with air-blow for 10 s at a distance of 20 cm; g – light-curing (450 mW/cm^2 for 10 s); h – light-curing (450 mW/cm^2 for 40 s, each layer).

reported in some studies in which sodium hypochlorite was applied as deproteinizing agent on the demineralized dentin[29]. One possible explanation for such finding, is based on the supposition that deproteinization using sodium hypochlorite transforms a demineralized substrate, full of collagen fibrils, in a much more porous, irregular and highly mineralized tissue[13,18], with larger dentinal tubule openings[25], and hence with greater chances to establish more resistant bonding interfaces. Therefore, these high bond strength values would be mainly attributed to the retention of resin composite to the dentinal tubules and their lateral branches[25,18].

If this hypothesis was true, it could be expected that the depth of the dentinal substrate would have a significant influence on the retention of composite materials bonded to dentin treated with sodium hypochlorite, since tubule distribution and density vary considerably in different dentin depths[9]. However, results obtained in the present experimental design showed that the mean bond strength values were significantly lower for the groups submitted to surface treatment with sodium hypochlorite, regardless of dentin depth.

Several factors can be responsible for the decreased bond strength results observed for the groups in which the demineralized dentin surfaces were further deproteineized before adhesive application. Some studies speculate that partial dissolution of collagen fibrils and/or desestabilization of molecules that compose the dentin structure occur during deproteinization, a factor that might compromise the formation of a reliable bonding interface[17]. Perdigão, et al.[19] (2000) did not observe the characteristic periodic bands of the collagen fibrils when previous treatment with sodium hypochlorite was performed before adhesive application. Oxygen released by sodium hypochlorite molecules is another factor that might justify the decreased bond strength values, as it may inhibit adhesive polymerization and hence compromise the mechanical performance of the obtained bonding interfaces.

It is clear that the use of sodium hypochlorite is still controversial[15], especially because bond strength results are dependent on the employed adhesive system. In practical terms, retention of some adhesive systems after application of 10% sodium hypochlorite is variable: it can be inferior[7,19], superior[29] or even similar[12,25] to those values obtained for the same materials applied to only demineralized dentin. Moreover marginal adaptation and microleakage studies, in demineralized dentin substrates were treated with sodium hypochlorite also shows controversial results. They can demonstrate similar results to non treated surfaces[23] (without sodium hypochlorite application) or inferior marginal sealing and adaptation when sodium hypochlorite was applied to demineralized dentin[6,24,7].

The present study confirmed that the substrate play an important role on the adhesion of current adhesive systems. Despite the employed surface treatment, lower bond strength values were obtained in deep dentin, probably due to the smaller amount of intertubular dentin in this substrate. Other studies also reported similar results[27,11,10].

Actually superficial dentin has few dentinal tubules (about 1% of the surface area) and is composed largely of intertubular dentin[9]. Deep dentin, near the pulp, especially after acid etching is composed mainly of largely funnel shaped dentinal tubules with much less intertubular dentin. Although hybrid layer refers to resin-infiltrated demineralized intertubular dentin, the hybrid layer is usually penetrated by resin tags in each dentinal tubule. These tags, more frequent is deep dentin, represent a minor fraction of the bonding for the dentin substrate, since they are not firmly bonded to the walls of the tubules[17].

Regarding the access used to achieve the deep

TABLE 2- Mean values of cross-sectional area (mm^2) and standard deviations obtained for the experimental group

Groups	Area (mm^2)
SUC	0.82 ± 0.12
SUH	0.83 ± 0.08
DAC	0.79 ± 0.07
DAH	0.82 ± 0.08
DOC	0.81 ± 0.60
DOH	0.83 ± 0.60

TABLE 3- Mean bond strength values (MPa), number of sticks (n), and standard deviations obtained for the experimental groups.

Substrate	Surface treatment	
	C [a]	**H [b]**
SU [a]	35.44 ± 12.3 (65)	28.82 ± 12.7 (49)
DO [b]	26.48 ± 8.50 (64)	24.27 ± 8.30 (56)
DA [b]	26.13 ± 10.2 (68)	23.52 ± 8.50 (78)

* Different letters mean statistical different means for main factors.

dentin, this study proposed an alternative method: the deep substrate can be obtained either via occlusal or apical. As no statistically significant difference was detected between the microtensile bond strength for the two different via, the bonding efficacy can be once more credited to the presence of intertubular dentin, as tubule exposed via occlusal are more retentive due to their increased diameter towards the pulp chamber. The method of achieving deep dentin via apical is easier to standardize and allows greater reproducibility of results. It can therefore be employed as an alternative for studies that evaluate techniques and materials in deep dentin substrates.

CONCLUSIONS

The depth of the substrate influenced the microtensile bond strength results obtained in this study, and was always higher for superficial dentin, despite the surface treatment performed. The surface treatment influenced the microtensile bond strength results, so that higher values were achieved when the extra deproteinization step was not performed.

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RESUMO

O objetivo foi avaliar a resistência de união (RU) de dentina superficial (SU) e profunda (P), obtida por acesso apical (PA) ou oclusal (PO), ao adesivo One-Step com (H) ou sem aplicação de hipoclorito de sódio a 10% (C) após o condicionamento ácido. Para os 6 grupos foram utilizados seis molares hígidos. Nos grupos SUC, PAC e POC, o adesivo e a resina Z100 foram aplicados. Nos grupos SUH, PAH e POH, após condicionamento, aplicou-se H por 60s, adesivo e Z100. Os dentes foram fatiados em cortes paralelos e perpendiculares para a obtenção de cp de 0,8mm² e testados sob tração (0,6mm/min). Os valores de RU foram submetidos a ANOVA de dois fatores. Não se detectou diferença para a interação (p=0,209), porém os fatores principais foram diferentes (p<0.05). Os resultados do tratamento C para os diferentes substratos foram: SU (35,4 ± 12,3), PO (26,5 ± 8,5), PA (26,1 ± 10,2) e para o tratamento H foram: SU (28,82 ± 12,7), PO (24,3 ± 8,3) e PA (23,5 ± 8,5). Os valores de RU em dentina SU foram estatisticamente superiores do que na dentina profunda (p<0,05), não foi observado diferença estatística nas duas vias de acesso para dentina profunda (SU>PO=PA). Os valores de RU com o tratamento C foram superiores aos obtidos quando utilizou-se o H. Conclui-se que: 1) os valores de RU foram maiores em dentina SU; 2) não houve diferença entre os dois substratos de dentina profunda; 3) aplicação do hipoclorito de sódio após o condicionamento diminuiu a RU.

UNITERMOS: Profundidade dentinária; Hipoclorito de sódio; Sistema adesivo.

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