

Evaluation of Bond Strength and Dentinal Tubule Penetration of the Post Luting Cements

Post Yapıştırma Simanlarının Bağlanma Dayanımı ve Dentin Tübül Penetrasyonunun Değerlendirilmesi

ABSTRACT

Aim: The purpose of this study was to investigate the influence of residual endodontic sealers on the adhesion of post luting cements. For hence, dentinal tubule penetration and bonding ability of three different self-adhesive resin cements was compared. (Panavia SA; Kuraray, Tokyo, Japan. Relyx U200; 3M ESPE, Seefeld, Germany. Maxcem Elite; Kerr, CA, USA)

Material and Method: 180 mandibular second premolar teeth roots were used. 45 roots which were instrumented and obturated with gutta-percha and AH Plus sealer which were labeled with 0.1% fluorescein dye. Forty-five roots were only instrumented with ProTaper, but left not-filled. The teeth were divided into two main groups: root canal filling(FR) and no root canal filling (NFR). Then, these roots were cemented with three different self-adhesive resin cements. The specimens were divided into 6 groups.

Group 1: Filled root (FR)/ Panavia SA

Group 2: Not-filled root (NFR)/ Panavia SA;

Group 3:FR/RelyX U200

Group 4: NFT/RelyX U200

Group 5: FR/Maxcem Elite

Group 6: NFR/Maxcem Elite.

Before the insertion of the posts, cement was labeled with Rhodamin B. After cementation process of the fiber posts, the specimens were cross sectioned and the slices were submitted to the push-out test and dentin tubule penetration evaluation of the cements using confocal laser scanning microscopy.

Results: FR/Maxcem Elite cement group presented the lowest bond strength value to dentin in the cervical third ($P < .05$). The dentinal tubule penetration area was higher in the NFR specimens compared to the FR in the Panavia SA and Rely X U 200 groups ($p=0.024$, $p=0.047$). Panavia SA group yielded the highest dentinal tubule penetration percentage compared to the other groups. There was no statistically significant difference between the three cements regarding penetration depth ($p>0.05$).

Conclusion: The NFR groups showed better dentin tubule penetration values than FR groups.

Key words: Confocal Laser, Scanning Microscopy, Dental Bonding, Resin cements.

ÖZ

Amaç: Bu çalışmanın amacı, rezidüel kök kanal patının post yapıştırma simanlarının adezyonu üzerindeki etkisini araştırmaktır. Bu amaçla post

Yelda PALTUN

Sis DARENDELİLER YAMAN

Gazi University, Faculty of Dentistry,
Department of Endodontics,
Ankara, Turkey



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Corresponding Adress/İletişim Adresi:

Yelda PALTUN

Gazi Üniversitesi, Diş Hekimliği Fakültesi,
Endodonti Anabilim Dalı,
Ankara, Turkey

E-mail/E-posta: dt.yeldapaltun@gmail.com

yapıştırılmasında kullanılan üç farklı self adeziv rezin simanın (Panavia SA; Kuraray, Japonya. Relyx U200; 3M ESPE, Almanya. Maxcem Elite; Kerr, ABD) dentin tübül penetrasyonu ve push out testi yapılarak dentine bağlanma yeteneği karşılaştırılmıştır.

Gereç ve Yöntem: 180 adet alt çene küçük azı diş kökü kullanıldı. 45 kök enstrümanle edildi ve % 0,1 fluoesein boya ile etiketlenmiş AH Plus ve gutta perka ile dolduruldu. 45 adet kök ProTaper Universal ile prepare edildi. Dişler öncelikle kanal dolgusu yapılan (KD+) ve kanal dolgusu yapılmayan(KD-) diye 2 ana gruba ayrıldı. Daha sonra bu köklere üç farklı self adeziv rezin siman ile post uygulandı. Örnekler 6 gruba ayrıldı.

Grup 1: Kanal Dolgusu yapılan(KD+)/ Panavia SA rezin siman

Grup 2:Kanal Dolgusu yapılmayan(KD-)/ Panavia SA rezin siman

Grup 3: KD+/ RelyX U200 rezin siman

Grup 4: KD-/ RelyX U200 rezin siman

Grup 5: KD+/ Maxcem Elite rezin siman

Grup 6: KD-/ Maxcem Elite rezin siman

Postların yerleştirilmesinden önce simanlar Rhodamin B ile boyandı. Fiber postların simantasyon işleminden sonra, örnekler kesitlere ayrıldı ve elde edilen kesitlere push-out testi uygulandı ve simanların dentin tübül penetrasyon değerlendirmesi için de konfokal lazer tarama mikroskobu kullanıldı.

Bulgular: Push out testi değerlendirildiğinde gruplar içinde sadece KD+/ Maxcem Elite grubu servikal bölgede en düşük bağlanma değerlerini gösterdi($p<.05$). Dentin tübül penetrasyonu değerlendirildiğinde, dentin Dentin tübül penetrasyon alanı açısından ise KD- grubu KD+ grubu ile karşılaştırıldığında Panavia SA ve RelyX U200 grupları istatistiksel olarak anlamlı sonuçlar gösterdi ($p=0.024$, $p=0.047$). Panavia SA grubu diğer gruplara göre en yüksek dentin tübül penetrasyon yüzdesini verdi ($p < .05$). Penetrasyon derinliğine göre üç siman arasında istatistiksel olarak anlamlı bir fark bulunmadı ($p> 0.05$).

Sonuç: KD- grup, KD+ gruba göre daha iyi dentin tübül penetrasyon değerleri gösterdi.

Anahtar sözcükler: Konfokal Lazer, Tarama Mikroskobu, Dental Bağlanma, Rezin simanlar.

INTRODUCTION

Fiber posts are widely used in the restoration of non-vital teeth due to their mechanical properties which are compliant with those of the dentine and in the aim to prevent possible root fractures (1). Cementation of the fiber post is an important step to achieve a successful outcome. Use of resin-based cements are generally recommended for the cementation of fiber posts (2,3). Conventional resin-based cements require surface treatments, such as acid etching and dentine bonding, which complicates the luting procedure (4-6). Self-adhesive resin luting cements have been introduced to the market in order to overcome this problem. They do not require any surface treatment process of the teeth or restorations prior to the application. In addition to that, the self-adhesive resin luting cements are easier to handle and have stronger bond strength characteristics compared to the conventional ones (7,8).

Strong adhesion of a cement used for cementation of the post systems to the root canal wall is a desired property. The deeper penetration of the cements into the dentinal

tubules is generally believed to increase their bonding capacity, although there is no direct correlation between them (2). The push-out bond strength test is commonly used as a practical method that allows the bond strength evaluations of adhesive materials to root canal dentin (9). However, this method alone cannot measure the dentinal tubule penetration of the root canal materials into dentine tubules. Scanning electron microscopy (SEM) and confocal laser scanning microscopy (CLSM) visualize the micromorphological characteristics of the dentin-adhesive interface (10) and they can evaluate of the penetration of the cements into dentine tubules (11,12). Furthermore, the CLSM provides more valuable data compared with the SEM, regarding the penetration and distribution of the resin luting cements (8).

An hermetic filling of the root canal space is generally recommended prior to the application of posts. Intracanal preparation of the space for a fiber post requires a partial removal of root canal filling. However, residual root canal sealers may block the dentine tubules and lead to a reduction in the bond strength between the post and root canal dentine (13).

Thus, the aim of this study was two folded: (1) to evaluate the effect of root canal sealer remnants on the adhesion and dentinal tubule penetration of the post luting cements, (2) to compare the bond strength and dentinal tubule penetration of three different self adhesive resin cements (Panavia SA; Kuraray, Tokyo, Japan. Relyx U200; 3M ESPE, Seefeld, Germany. Maxcem Elite; Kerr, CA, USA) which were used for luting the fiber posts, using push-out bond strength test and CLSM, respectively.

MATERIALS AND METHODS

This study has been approved by the local ethical committee of Ankara University, Faculty of Dentistry (Clinical Research Ethics, no.36290600/08, date.18/02/15). Single rooted human mandibular premolars with straight root canals which had been extracted for reasons unrelated to this study were obtained from Department of Oral and Maxillofacial Surgery, Gazi University. Teeth having immature roots, root cracks, calcifications, curvatures, caries, and more than one canal were eliminated. One-hundred-eighty teeth meeting the criteria were stored in 0.1% thymol solution prior to the experiment. The dental crowns were removed about 14 mm from the root apex with using a diamond disc under water-cooling.

The root canal orifices were enlarged using a round diamond bur. The working length was established 1 mm from the root apex using a size of 10 K-file. Root canals were prepared by using the ProTaper Universal rotary instruments (Dentsply Maillefer, Ballaigues, Switzerland) to an apical size of F3. 2 mL of 1% sodium hypochloride was used after each change of instrument. Final irrigation was performed using 5 mL of 17% EDTA for 1 min. Then the canals were finally washed with 5 mL of saline solution and dried with paper points.

Push-Out Bond Strength Test

Ninety instrumented specimens were randomly divided into two groups. The 45 root canals in the first group were filled with resin based sealer AH Plus (Dentsply De Trey, Konstanz, Germany) and gutta-percha cones using the lateral compaction technique. Other 45 root canals were left instrumented and not obturated. The obturated roots were stored at 37°C in distilled water for 48 hours in order to allow setting of the sealers. Then, 90 specimens were prepared to ensure a standardized space for post insertion. The each root canal space was enlarged with a #2 Gates-Glidden bur (VDW, Munich, Germany) providing an access for #2 post drill to a depth of 9 mm. Then the sample were enlarged using a drill provided by manufacturer of the selected post system (Fiber post drill

size:2 Reforpost, Angelus, Brazil). The specimens were divided into six experimental groups (n=15) according to the cement types used for luting the posts and root canal filling: Filled root (FR), Panavia SA and not-filled root (NFR), Panavia SA; FR/RelyX U200, NFR/RelyX U200; FR/Maxcem Elite, NFR/Maxcem Elite.

The adhesive cements were prepared according to the manufacturer's instructions. Glass fiber prefabricated posts with a diameter of 1.3 mm and a length of 15 mm (Reforpost; Angelus, Brazil) were inserted into the root canals. The excess resin cement was removed and a halogen curing-light unit was performed for 30 seconds to provide polymerization.

After cementation process, the specimens were stored in saline solution at 37°C for 24 h and then embedded in methacrylate resin blocks (Meliodent, Heraus Kulzer GmbH, Germany). The samples were sectioned horizontally with a low-speed diamond saw under constant water cooling (Mecatome T201, Presi, France). Three slices were obtained per each root, these slices were containing cross-sections of coronal, middle and apical part of the bonded fiber posts were obtained by sectioning. The thickness of each sample slices were 2.0 ± 0.1 mm. Each sample was marked on its coronal surface with an indelible marker. The exact thickness of each slice was measured using a digital caliper. A cylindrical plunger tip with a diameter of 1mm was selected and positioned to contact the post only, without touching the surrounding root canal walls, in an apical-to-coronal direction. A compressive load at a crosshead speed of 0.5 mm/min was applied to the apical aspect of the slice via the plunger that was mounted on a universal testing machine (LRX, Lloyd Instruments, Fareham, England). The maximum failure load when the post was dislodged was recorded in Newtons (N) and calculated in megapascals (MPa) using the formula:

Debonding stress(MPa)= Debonding force(N)/ A(mm²)

where A=area of the post-dentin surface as follows:
 $A = \pi(r_1 + r_2) \sqrt{(r_1 - r_2)^2 + h^2}$.

After the push-out testing, specimens were examined with a stereomicroscope in order to determine whether the failure mode was adhesive between post and cement, or cohesive within the cement or mixed.

Confocal Laser Scanning Microscopy Evaluation

Ninety instrumented specimens were randomly divided into two groups of 45 teeth. Instrumented 45 root canals in the first group, were not obturated. Other 45 root canals were obturated with AH Plus which was

previously labeled with 0.1% sodium fluorescein (FNa; Sigma Aldrich, Steinheim, Germany) and gutta-percha cones using the lateral compaction technique.

Ninety specimens were randomly divided into six experimental groups (n=15) according to the adhesive cement types used for luting the posts: Filled root (FR), Panavia SA and not-filled root (NFR), Panavia SA; FR/RelyX U200, NFR/RelyX U200; FR/Maxcem Elite, NFR/Maxcem Elite.

All the adhesive cements were labeled with with %0.1 rhodamine-isothiocyanate (RITC; Sigma Aldrich, St Louis, MO, USA) prior to cementation. The other procedures used for post space preparation, and cementation were the same as in the push-out bond strength assessment.

Each sample was sectioned perpendicular to its long axis at 8 and 10 mm from the anatomic apex to obtain one specimen with a thickness of 2 mm. These specimens were observed using a confocal laser scanning microscopy (Zeiss lsm 510, Zeiss, Germany) under dual fluorescence mode at 10 µm below the surface of the sample with ×4 magnification. The sections were displayed under ×40 (oil immersion) magnification to discriminate the root canal perimeter and sealer/cement penetration in the dentinal tubules.

Digital images were then recorded and analyzed using IMAGE J 1.4 software (National Institutes of Health, Bethesda, MD). The root canal circumference and the cement penetration circumference areas were outlined and measured. These two measurements ratio was calculated as the percentage of cement covering the root canal wall. The maximum penetration depth was measured from the canal wall to the deepest point of cement penetration. The penetration area of the cement was also calculated. The area of the canal and area of the cement penetrated to tubule were measured, and the difference was calculated. Figures 1A-C show the dentin tubule penetration measurement parameters.

Statistical Analysis

Statistical analysis was performed in IBM SPSS programme for Windows Version 21.0 software package. Differences between the groups were determined by analysis of variance or two-way analysis of variance for repeated measures. Bonferroni test was used for pairwise comparisons. The significance level was set at $p < 0.05$.

RESULTS

Table 1 represents bond strength values achieved upon dislodging the posts from each level of the root canal in each cement group. The higher push-out bond strength values were obtained from the coronal thirds of the

Table 1: Push-out bond strength of the test groups (MPa)

	n	Filled Root Groups (FR)			Not-filled Root Groups (NFR)		
		Apical	Middle	Coronal	Apical	Middle	Coronal
Maxcem Elite	15	3,986±3,0 ^a	4,746±2,1	7,668±2,8 ^{*a}	6,038±2,4	6,748±1,9	8,938±3,0
Panavia SA	15	8,158±6,5	6,540±4,0	10,875±3,4 [*]	7,631±2,7	8,306±2,6	8,614±1,5
RelyX U200	15	7,634±5,6	5,826±1,7	10,954±5,8 [*]	6,764±3,9	7,661±5,2	9,330±4,3

^a Show differences within the same column, ^{*} Show differences within the same row ($p < 0.05$).

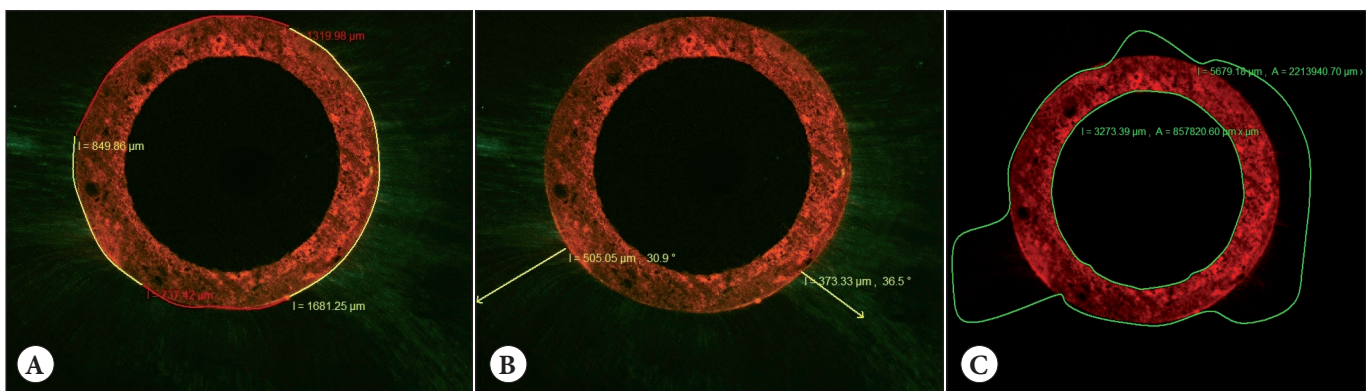


Figure 1: Measurement of dentine tubule percentage (A), maximum depth of cement penetration (B) and cement penetration area (C) using LSM Image Examiner Software (Carl Zeiss)

specimens compare with apical third in the all groups. The FR/Maxcem Elite group was the only group that presented the lowest value in the cervical third ($P < 0.05$). There is no statistically significant difference between the values of the groups in the FR and NFR groups in terms bond strength ($p > 0.05$).

Table 2 represents the dentine penetration area, maximum penetration depth and penetration percentage of the test groups. FR/Maxcem Elite group showed the lowest values of dentine tubule penetration area, although it was not significant. The dentinal tubule penetration area was

higher in the NFR specimens compared to the FR in the Panavia SA and Rely X U 200 groups ($p = 0.024$, $p = 0.047$). There was no statistically significant difference between the three cements regarding penetration depth ($p > 0.05$). NFR/Panavia SA and NFR/RelyX U200 groups showed higher penetration depth than those of FR/Panavia SA and FR/RelyX U200 ($p < 0.001$). Figure 2A-C shows the dentinal penetration pattern of RelyX U200.

Panavia SA group yielded the highest dentinal tubule penetration percentage compared to the other groups and NFR/Panavia SA group showed higher value compared

Table 2: Dentinal tubule penetration area ($\mu\text{m} \times \mu\text{m}$), depth (μm), and percentage (%) of the test groups

		Maxcem elite	Panavia SA	Rely X U 200	p
Dentinal tubule penetration area ($\mu\text{m} \times \mu\text{m}$)	FR	1046768,9 \pm 404742,8	1335013,7 \pm 673873,3	1114580,3 \pm 377053,8	0,741
	NFR	1418586,5 \pm 306962,5	2231660,2 \pm 2352735,2	1897069,6 \pm 638876,5	0,116
	p	0,341	0,024	0,047	
Dentinal tubule penetration depth (μm)	FR	416,9 \pm 196,9	486,7 \pm 263,7	428,2 \pm 215,4	0,680
	NFR	719,3 \pm 193,1	812 \pm 227,2	814 \pm 286,1	0,449
	p	0,001	<0,001	<0,001	
Dentinal tubule penetration percentage(%)	FR	46 \pm 21,5	58,2 \pm 12,9	53 \pm 10,8	0,091
	NFR	52,2 \pm 18,6	71,3 \pm 10	61,9 \pm 12,9	0,003
	p	0,265	0,019	0,110	

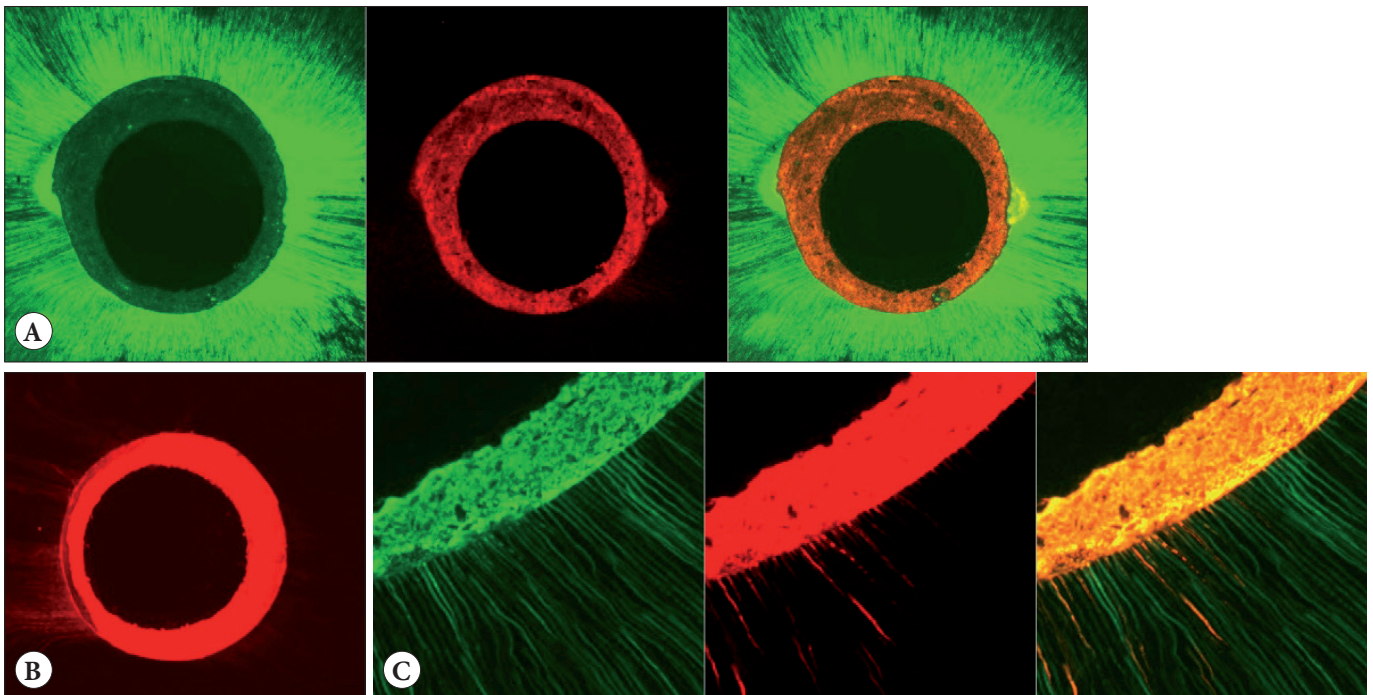
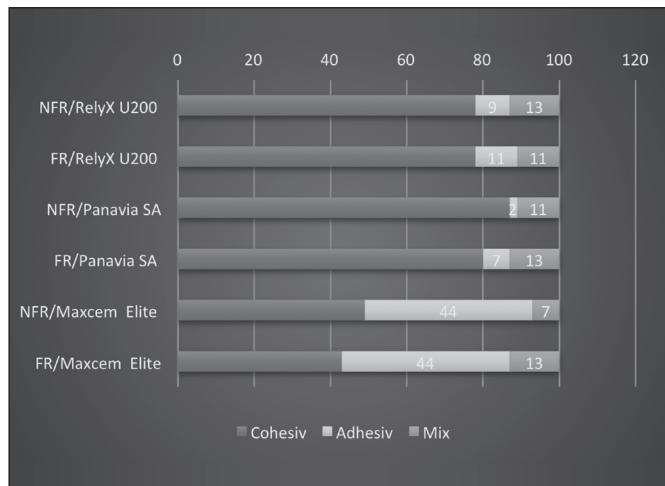


Figure 2: A) A representative confocal laser scanning microscopy image of FR/RelyX U200- remaining of AH Plus (green) in root canal walls and adhesive resin cement tags (red). B) A representative confocal laser scanning microscopy image of NFR/RelyX U200. C) A representative confocal laser scanning microscopy image of RelyX U200 40 \times (oil immersion) magnification - AH Plus tags (green), adhesive resin cement (red) and some yellow tags indicating the mixture of the two material.

to FR/Panavia SA group ($p < 0.05$). Figure 3A-C shows the dentinal penetration of Panavia SA groups.

Graphic 1 presents the percentage of the fracture failures of the specimens. Most of the specimens fractured at the cohesive interface. FR/Maxcem Elite and NFR/Maxcem Elite group showed more adhesive failure when compared with the other cements. Figure 4A-C representative CLSM images of Maxcem Elite groups.



Graphic 1: Type of failure mode percentage (%).

DISCUSSION

The push-out test results of the present study have revealed that the teeth with FR and with NFR groups did not show any statistically significant differences in terms of the bonding strength. Results of the present study is in collaboration with the findings of Burns et al. (14 and dowel spaces were prepared to a 7 mm depth. Dowel space walls were acid etched and primed, and prefabricated stainless steel dowels were luted using resin cement. After 24 hours, the load required to dislodge each dowel along a path parallel to its long axis was measured and recorded. A one-way analysis of variance ($\alpha = 0.05$). However, the dentine tubule penetration values of the luting cements including area, depth, and percentage were higher in the NFR group compared to the FR group. These results may support the idea that there is no direct correlation between dentinal tubule penetration quality and adhesion capacity of the cements. There may be another factors different from dentinal tubule penetration such as that have an influence on the bonding ability of the luting cements. The poorer penetration results of the FR group may be attributed to the possible remnants of the root canal sealers which may prevent the luting cements' penetration into the dentinal tubules.

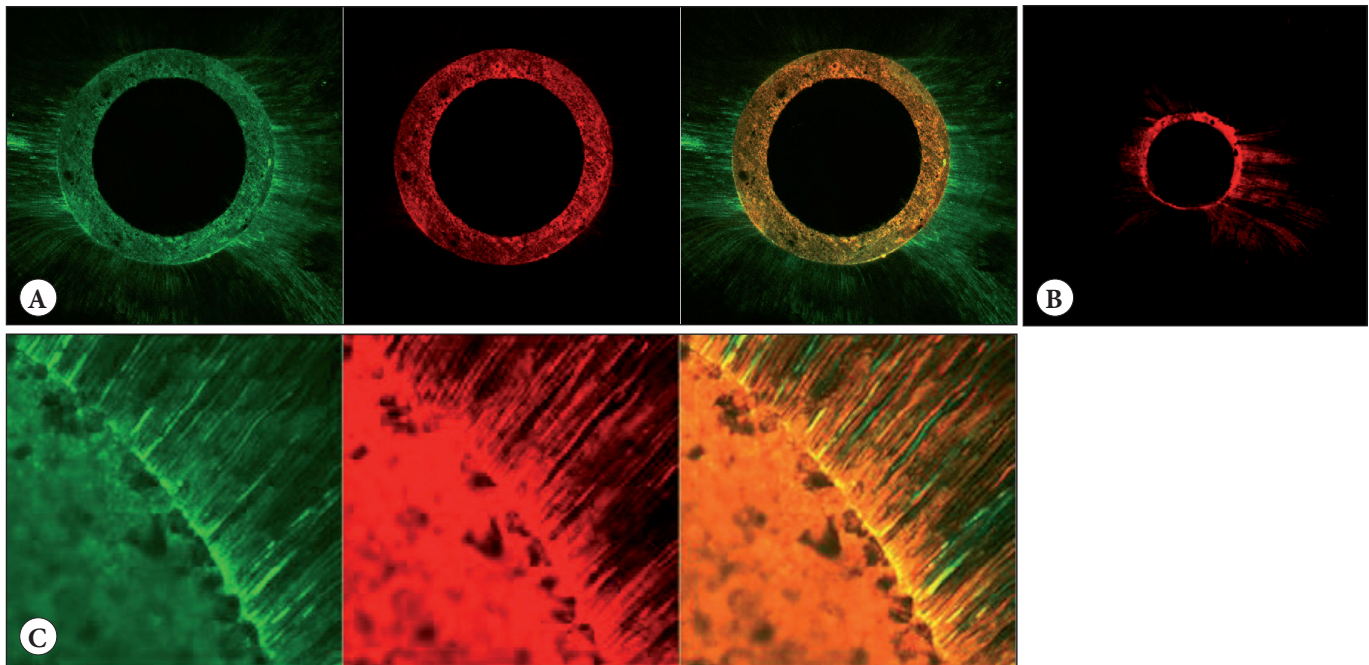


Figure 3: **A)** A representative confocal laser scanning microscopy image of FR/Panavia SA- remaining of AH Plus (green) in root canal walls and adhesive resin cement tags (red). **B)** A representative confocal laser scanning microscopy image of NFR/Panavia SA. **C)** A representative confocal laser scanning microscopy image of Panavia SA 40×(oil immersion) magnification - AH Plus tags (green), adhesive resin cement (red) and some yellow tags indicating the mixture of the two material.

The type of the sealer used for obturation of root canals may also has an effect on the adhesion of luting cements. In the present study, a resin based sealer was used with gutta-percha for obturation of root canals prior to the post application. The resin-based sealers have previously been demonstrated to provide deeper dentinal tubule penetration (15). Therefore the residual resin-based sealer in the dentinal tubules might reduce the penetration values of the luting cements (13). In addition the chemical compositions of the sealers may inhibit the polymerization of the luting cements. Eugenol-based sealers were reported to have a negative impact on the composite resin polymerization (16,17). Muniz & Mathias et al. (18). reported that the resin-based endodontic sealer allowed for greater compatibility with the adhesive system used for post cementation. As it was not always possible to completely remove the sealer from the canal walls (19).

In the present study, the bonding strength values at the apical thirds of all groups were lower compared to the coronal thirds. This result is in agreement with those of others who found stronger bond strength at the coronal intraradicular dentine than the deeper root regions (20,21). This may be caused from the decreased polymerization rate of the luting cements in the apical

root thirds, where the distance from the light source increases. Another reason for the low bond strength values in the apical sections is based on the root dentin morphology has a reduction in dentinal tubule density from coronal to apical (22).

When the results of this present study have been evaluated for cements it has been determined that the lowest bonding strength among the FR samples were in Maxcem Elite applications. In another study RelyX U200 was compared with Maxcem Elite and it was reported that Maxcem Elite also provided lower bonding strength (23). This was attributed to the acidic group monomers and their concentrations. Hence those monomers were argued to cause different etching patterns, different wetting features, different chemical adhesion capacities to the dentin (24). Bitter et al. (8). reported that RelyX U200 yielded high push-out values which was attributed to the chemical interaction of RelyX U200 acidic monomers with hydroxylapatite.

In the present study, the higher push-out bond strength and dentinal penetration values were obtained from the Panavia SA group. This may be attributed to the phosphate based monomer (10-MDP) of Panavia SA which may increase the material's bonding capacity. 10-MDP was reported to acidify the enamel and dentine,

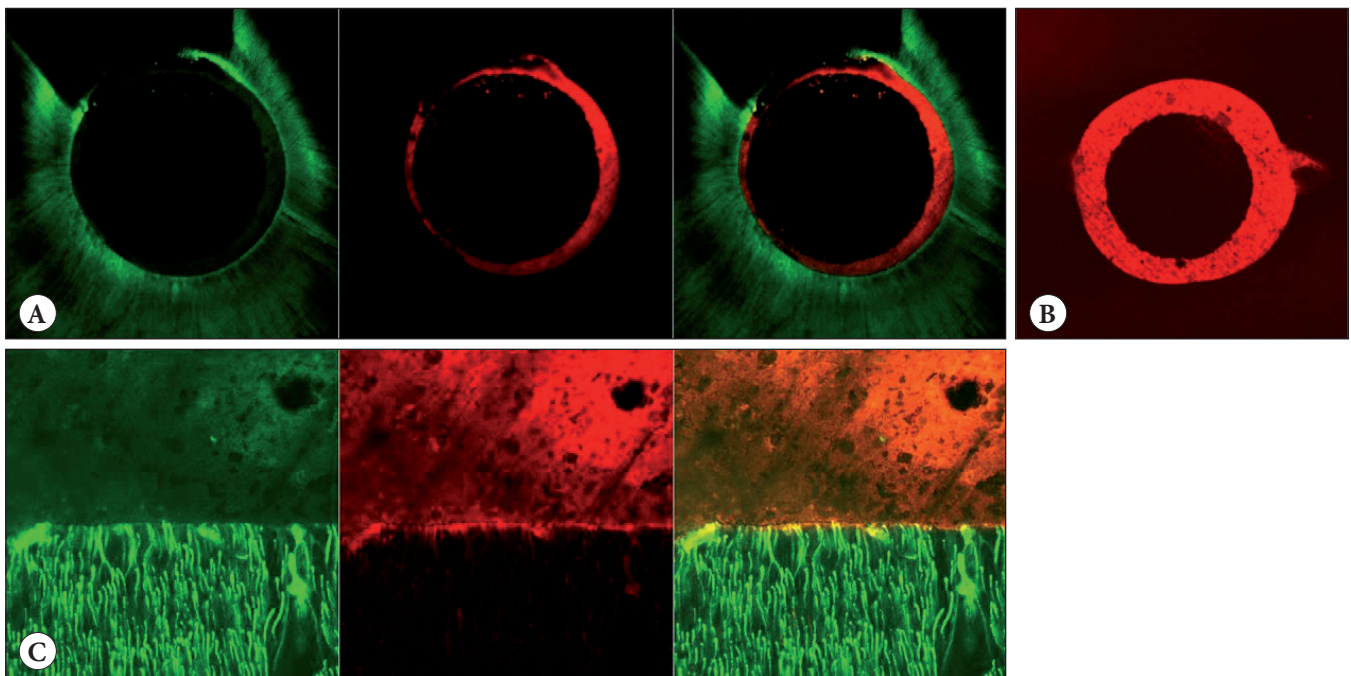


Figure 4: A) A representative confocal laser scanning microscopy image of FR/Maxcem Elite- remaining of AH Plus (green) in root canal walls and adhesive resin cement tags (red). B) A representative confocal laser scanning microscopy image of NFR/Maxcem Elite. C) A representative confocal laser scanning microscopy image of Maxcem Elite 40× (oil immersion) magnification - AH Plus tags (green), adhesive resin cement (red) and some yellow tags indicating the mixture of the two material.

chemically interact with hydroxiapatite, attach to calcium and phosphate ions, and replace with phosphate and hydroxyl ions in hydroxiapatite (24). High bonding strength values achieved in the studies conducted with Panavia SA were attributed to these chemical bonding properties in addition to the micromechanical bonding features (25).

Viscosity and pH of the cements (7,26) can be counted as other factors that can affect the penetration and bonding ability (25,27). The high viscosity of cements may inhibit wettability of the dentine surface and infiltration of the cement into dentinal tubules (28). On the contrary, the materials which have lower viscosity was suggested to show deeper penetration into the dentinal tubules. Therefore, the successful penetration results obtained from Panavia SA may be because of its lower particle size which lead in low viscosity (7). The poorer bond strength results obtained from Maxcem Elite may also be attributed to its low pH that remains acidic for longer periods (29). Long-term exposure of dentin surface to this acidic pH may disrupt the bonding characteristics by adversely affecting the micromechanical retention between the cement and tooth structure (30).

When the fractures occurred after push-out bond strength test were evaluated, higher percentage of adhesive failure type was observed in the Maxcem Elite group compared to the Panavia SA and RelyX U200 groups. This result also supports the more successful adhesion of Panavia SA and RelyX U200 to both root canal dentine and post surface than the Maxcem Elite.

In the present study, push-out bond strength test was used for determining the adhesion capacity of the luting cements. As this test gives limited information about this subject, the dentinal tubule penetration ability of the used materials were observed using CLSM. As it was previously reported that the fluorescence dyes used for CLSM evaluation can reduce the bonding capacity of the materials, we prepared and used different samples for the CLSM and push-out bond strength tests (31).

The variations in the structure and the composition of dentine are very important in terms of bonding. It is known that the localization and the orientation of dentine tubules affect the demineralization depth (32). The diffusion of adhesive resin into the collagen network is also known to be important for retention and hybridization (32). However, the actual contribution of resin tags to bonding strength is not clear yet (33). Therefore further study is required for evaluating the correlation between bond strength and resin tag.

CONCLUSIONS

In this present study NFR groups showed better dentine tubule penetration values when compared with FR. Dentine tubule penetration of cement has limited effect on push-out bond strength of the self-adhesive resin cement. There was no statistically difference between self adhesive resin cements.

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