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Influence of Different Irrigation Solutions on the Push-Out Bond Strength of Fiber Posts to Root Dentin and **Root Canal Dentin Microhardness: Methodological Study**

Farklı İrrigasyon Solüsyonlarının Fiber Postların Kök Kanal Dentinine Bağlanma Mukavemeti ve Dentin Mikrosertliği Üzerindeki Etkisi: Metadolojik Çalışma

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ABSTRACT Objective: This study aimed to compare the effect of sodium hypochlorite (NaOCl), chlorhexidine (CHX), ethylene diamine tetra acetic acid (EDTA) and etidronic acid (HEBP) solutions and their combinations on a push-out bond strength of fiber posts to root canal dentin and on root dentin microhardness. Material and Methods: Sixty human maxillary central incisors were sectioned below the cementoenamel junction, the roots were endodontically treated and were divided into 6 groups (n=10) according to the final irrigation protocol: (1) Distilled water, (2) CHX, (3) HEBP-NaOCl combination, (4) HEBP, (5) 2.5 % NaOCl, (6) 17% EDTA. The fiber posts were then cemented with self-etching resin cement. Bonded specimens were cut (1-mm-thick), the first slice of the coronal 3rd was used for dentin microhardness analysis, and other slices were used for the push-out bond strength test. The data were statistically analyzed by using Shapiro-Wilk, Oneway ANOVA, and the Games-Howell test. Results: The lowest microhardness was observed in HEBP-NaOCl and NaOCl (p<0.05). CHX, HEBP, and EDTA resulted in less reduction in microhardness as compared to other groups (p<0.05). HEBP-NaOCl and NaOCl showed significantly higher mean bond strength to root dentin in the cervical thirds than in other groups (p<0.05). Conclusion: With regard to the push-out bond strength of the fiber post to dentin, NaOCl and HEBP-NaOCl increased bond strength compared to other groups. NaOCl and HEBP-NaOCl have lower microhardness of dentin. Compared to HEBP, EDTA increases microhardness and does not make a significant difference in push-out bond strength. Therefore, it is recommended to use HEBP alone.

ve etidronik asit (HEBP) solüsyonlarının fiber postların kök kanalına dentinine bağlanma mukavemeti ve kök dentin mikrosertliğine etkisini değerlendirmektir. Gereç ve Yöntemler: Altmış adet insan üst çene santral kesici dişi mine sement sınırının altından kesilerek, kökler endodontik olarak tedavi edildikten sonra irrigasyon protokolüne göre 6 gruba (n=10) ayrılmıştır: (1) Distile su, (2) CHX, (3) HEBP-NaOCl kombinasyonu, (4) HEBP, (5) %2,5 NaOCl, (6) %17 EDTA. Fiber postlar self-etch rezin siman ile dişlere simante edilmiştir ve örneklerden 1 mm kalınlığında kesitler alınarak, koronal üçlüdeki ilk kesit dentin mikrosertliği analizi için ve diğer kesitler push-out bağlanma gücü testi için kullanılmıştır. Veriler, Shapiro-Wilk, One-way ANOVA, Games-Howell testi kullanılarak istatistiksel olarak analiz edilmiştir. Bulgular: En düşük mikrosertlik HEBP-NaOCl ve NaOCl'de gözlenmiştir (p<0,05). CHX, HEBP ve EDTA, diğer gruplara kıyasla mikrosertlikte daha az azalma ile sonuçlanmıştır (p<0,05). Servikal bölgede HEBP-NaOCl ve NaOCl, diğer gruplara göre anlamlı derecede daha yüksek bağlanma kuvveti göstermiştir (p<0,05). Sonuçlar: Fiber postun dentine bağlanma kuvvetiyle ilgili olarak, NaOCl ve HEBP-NaOCl, diğer gruplara kıyasla bağlanma gücünü artırmıştır. Çalışmanın sonuçları değerlendirildiğinde, NaOCl ve HEBP-NaOCl, daha düşük dentin mikrosertliğine sahip olduğu görülmüştür. HEBP ile karşılaştırıldığında, EDTA kullanımı mikrosertliği artırmış ve push-out bağlanma gücünde önemli bir fark oluşturmamıştır. Bu nedenle HEBP'nin tek başına kullanılması tavsiye edilir.

ÖZET Amaç: Bu çalışmanın amacı, sodyum hipoklorit (NaOCl), klor-

heksidin [chlorhexidine (CHX)], etilen diamin tetra asetik asit (EDTA)

Keywords: Etidronic acid; root canal treatment; sodium hypochlorite

Anahtar Kelimeler: Etidronik asit; kök kanal tedavisi; sodyum hipoklorit

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Endodontically treated teeth are not similar to non-restored vital teeth structurally and they necessitate specialized restorative treatment to assist in direct or indirect coronal restoration.^{1,2} When treating these teeth, the use of intraradicular posts, especially fiber posts, is recommended to ensure longevity of the upper restoration.³ Fiber posts consist of carbon, quartz, glass, or silica fibers embedded in an epoxy or methacrylate resin matrix. Fiber posts have led to great advancements in the restoration of endodontically treated teeth, especially regarding mechanical properties, such as a high flexural strength and elasticity modulus similar to that of dentin, minimizing the transmission of stresses on the root walls and decreasing the possibility of fractures.⁴ When clinical success is evaluated, the longevity of fiber posts depends on their mechanical properties such as minimal transmission of stresses on the root walls and effective bonding between the post, dentin, and adhesive resin cement.^{1,4-6} Various factors can be the reason for the debonding of fiber posts, for example, the irrigant used in the arrangement of root canal, thick smear section, and configuration factor (C-factor), described as the rate between the bonded and unbonded faces in posts arrangement.⁷⁻¹⁰ The presence of a smear layer may limit the effectiveness of disinfectants in the dentinal tubules and affect the adaptation of root canal filling materials to the canal walls. There is evidence that resin-based cements bond to thicker smear layers with lower adhesion values.² High C-factor inside the root canal can threaten the achievable bond with the radicular dentin and adhesive cement. Unbonded surface during polymerization moves and flows, thus reducing shrinkage stresses. However, because a root canal has little unbonded surface area, insufficient stress relief will occur or it will be more likely to be pulled or separated from more bonded areas. Failure of fiber posts for the most part is the result of separation that mainly happens throughout the adhesive interface between post and dentin.3

The strength of the bond between the fiber post and dentin plays an important role in the clinical success of endodontically treated teeth. Push-out bondstrength test has been usually used in measuring the adhesive strength of fiber posts, because the test is easy to reproduce and interpret, and it provides a realistic assessment of bond strength to dentin, even at low levels.⁵ Endodontic irrigants used have effect on the microhardness of root canal dentin, which in turn might have an influence on adhesion of sealer to root canal dentin has been reported in endodontic literature by using Vickers diamond testing machine.⁵

The utilization of disinfecting irrigation solutions before bonding procedures can interfere with the adhesion process, changing the properties of some hydrophilic resins.^{11,12} Residues of these substances may also impede the deep penetration of the bonding agent and its intimate contact with dentin, also affecting the adhesion process.^{11,12} It is recommended to use sodium hypochlorite (NaOCl) as the final irrigation solution to remove the inorganic components of the smear layer.¹³ Ethylene diamine tetraacetic acid (EDTA), is often used for chelating and applied followed by NaOCl to remove the smear layer completely. However, EDTA treatment weakens dentin and affects the mechanical integrity of the posts.¹⁴ To resolve these problems, a new chelation agent, Alhydroxyethylidene-1, l-bisphosphonate (HEBP), also called etidronic acid was introduced as a substitute to EDTA.¹⁵ It affects the structure of dentin less and, can be mixed with NaOCl solution without impeding its antimicrobial features.¹⁶⁻¹⁸ Moreover, when compared with the conventional treatment methods such as EDTA treatment after NaOCl, a mixture of NaOCl and HEBP solutions may reduce the formation of a smear layer during root canal instrumentation. The HEBP-NaOCl solution is also less aggressive on root dentine compared with EDTA.19

Despite its clinical use, there is a lack of knowledge about how HEBP affects the push-out bond strength (PBS) of fiber posts to the root canal dentin (RCD). To eliminate this deficiency, the present study aimed to compare the effect of different root dentin treatment protocols using HEBP, NaOCl, chlorhexidine (CHX), EDTA solutions, and HEBP-NaOCl on PBS of fiber posts and the microhardness of RCD. The null hypothesis established in this study is that "none of the canal irrigation solutions used during treatment affect the attachment of fiber posts to RCD."

MATERIAL AND METHODS

This study was prepared and implemented according to the principles of the Declaration of Helsinki. This study was approved by İstanbul Medipol University Non-Invasive Clinical Research Ethics Committee (date: May 4, 2018, no: 10840098-604.01.01-E.12779). The patients were informed about the subject of the study. Sixty human maxillary central incisors with similar root anatomy previously extracted for periodontal reasons were selected for use in this study. These teeth were stored in 0.1% thymol solution at 4°C for 30 days. Teeth were checked for their suitability for study (light microscope at x20 magnification).

SPECIMEN PREPARATION

The teeth were decoronated with a root length of 16 mm. After the working lengths were determined and root canals were enlarged with rotary instrument files (Dentsply Maillefer, Ballaigues, Switzerland) to #50 size, 0.05 taper (F5). After each file, the root canals were rinsed with 5 mL of 2.5% NaOCl between instruments. The canals were dried with paper points and then obturated with gutta-percha cones (Dentsply-Maillefer) and sealer (AH-Plus; Dentsply DeTrey GmbH, Konstanz, Germany) using a lateral condensation technique. After the completion of endodontic treatment, cervical root canal openings were filled with a provisional restorative material (Cavit-G; 3M ESPE AG, Seefeld, Germany). The samples were then stored in distilled water-soaked gauze for 1 week at 37°C/100% humidity. A 10 mm deep post cavity leaving an apical plug of 4 to 5 mm gutta-percha in the canal was prepared using a #2 peeso reamer (Cytec[™] Blanco Post, Hahnenkratt, Königsbach-Stein, Germany). The samples were then randomly divided into 5 experimental groups with a control group (CG). The post spaces were rinsed with each irrigation solution for a period of 60 s.

Group (n=10) Treatment Solution

- 1 CG Distilled water (DW)
- 2 CHX 2.5% NaOCl+DW+2% CHX

3 HEBP-NaOCl 2.5% NaOCl+18% HEBP-NaOCl combination 1:1+DW

- 4 HEBP 2.5% NaOCl+18% HEBP+DW
- 5 NaOCl 2.5% NaOCl+DW
- 6 EDTA 2.5% NaOCl+17% EDTA+DW

External surface of the #2 fiber posts was cleaned with 70% alcohol and DW. Then the specimens were left for air drying. The etching procedure of root canals was done with 37% phosphoric acid (3M ESPE) for 15s, then rinsing occurred in water, and drying was done gently with paper points. Then, the adhesive system (Single Bond 2, 3M ESPE) was performed concerning the manufacturer's instructions. All posts were cemented with a dual cement (Rely X Unicem). Light curing was then performed using a conventional quartz-tungsten-halogen light in standard mode (1,200 mW/cm² output; Guilin Woodpecker Medical Instrument, Guilin, Guangxi, China) by placing the light tip perpendicularly through the post for 40 seconds. Samples were incubated in DW at 37°C for 7 days.

PBS TEST

Precision saw cross sections were taken from the teeth (IsoMet 2000, Buehler, USA). Teeth were cut 1 mm away from the enamel-cement junction to examine dentin microhardness and 1.0 mm thick specimens were obtained. Five cervical slices were obtained from each tooth. The first slice of the coronal root third was used for dentin microhardness analysis. Remaining four slices were obtained from the cervical thirds of each root and they were used for PBS test (Figure 1). A cylindrical plunger tip (1 mm diameter) was attached to the test device and placed on the sample, only in contact with the fiber pole. The samples were placed on a metal base. Samples were pushed out test at 0.5 mm/min crosshead speed using a universal testing machine (Special Edition, Yıldız Technical University, İstanbul, Türkiye). The adhesion region of each section was calculated by the specified equation: $A = [\pi (r + R) *h2 + (R-r)2],$ where r and R are the smallest and the largest radius of the fiber pole. Final data are given in Newtons, results in MPa are obtained.

MICROSCOPIC EVALUATION

The fracture types of the samples were determined with a stereomicroscope at 40x magnification. Adhe-

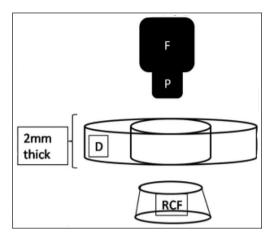


FIGURE 1: Scheme of the specimen positioned on the apparatus for load application in the universal testing machine (F: Force; P: Plugger; RCF: Root canal filling; D: Diameter).

sive type: Between the post and the cement. Cohesive type: Between the cement and dentin. Mixed defect (post, cement and dentin) (Olympus SZ61; Olympus Optical Co., Tokyo, Japan). Electron microscopy was used to examine fracture types (Jeol JSM 6360LV, Tokyo, Japan).

DENTIN MICROHARDNESS ANALYSIS

The apical sections of the samples were placed vertically in auto polymerized acrylic resin and the coronal parts became smoother polishing was done with #400, #500, and #600-grit silicone carbide papers under distilled water. Then, microhardness tests of samples were performed with a Vickers microhardness tester (Microhardness tester, Bulut Makine, Türkiye). Three different indentations were performed on each sample at depths of $200 \,\mu\text{m}$ from the side of the canal lumen using a 200-g load and a 20-sec dwell period.²⁰ These indentations were obtained for each specimen and were measured and converted into Vickers hardness.

STATISTICAL ANALYSIS

Statistical Number Cruncher was done with the Statistics System program. Whether the data were suitable for normal distribution was examined using the Shapiro-Wilk test. Oneway ANOVA and Games-Howell test were used for post hoc evaluations. Friedman and Dunn's test was used for non-normally distributed variables. p<0.05 was considered significant for the significance level of the study.

RESULTS

The PBS measurements of HEBP-NaOCl cases were found to be significantly higher than CG, CHX, HEBP, and EDTA (p<0,05). The PBS measurements of the cases in the NaOCl were found to be significantly higher than those in the CG, CHX, HEBP, and EDTA (p<0.05) (Table 1, Figure 2, Figure 3).

It was revealed that there was a statistically significant difference between the microhardness value measurements of the samples compared to the groups (p<0.01). Dunn's test was performed to determine the difference and according to its results; microhardness values of HEBP-NaOCl cases were found to be significantly lower than CHX, HEBP, and CG (p<0.05). The microhardness values of the cases in the NaOCl

TABLE 1: Evaluation of push-out bond strength.						
	Push-out b					
	Ort±SS	Median (Minimum-maximum)	p value	Post-hoc		
^a Control (n=10)	10.75±6.29	8.4 (3.75-21.90)	0.001**	a-b-d-f <c-e< td=""></c-e<>		
^b CHX (n=10)	12.72±5.09	12.7 (3.99-20.32)				
°HEBP-NaOCI (n=10)	17.67±3.07	18.1 (10.87-21.66)				
dHEBP (n=10)	9.98±3.68	9.40 (5.59-16.14)				
⁰NaOCI (n=10)	17.69±4.67	17.6 (11.01-31.47)				
^f EDTA (n=10)	12.29±4.71	12.8 (4.79-19.66)				
Total (n=60)	13.73±5.55	14.6 (3.75-31.47)				

Oneway ANOVA & post-hoc Games Howell test; **p<0.01; SS: Standard deviation; CHX: Chlorhexidine; HEBP: Etidronic acid; NaOCI: Sodium hypochlorite; EDTA: Ethylene diamine tetra acetic acid.

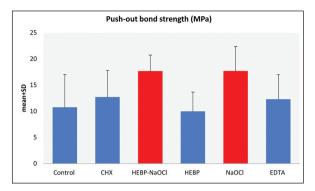


FIGURE 2: Evaluation of push-out bond strength between groups. SD: Standard deviation; CHX: Chlorhexidine; HEBP: Etidronic acid; NaOCI: Sodium hypochlorite; EDTA: Ethylene diamine tetra acetic acid.

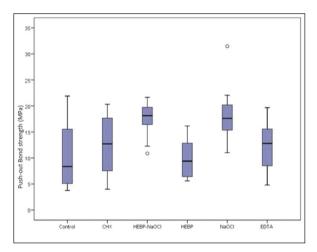


FIGURE 3: Evaluation of push-out bond strength between groups. CHX: Chlorhexidine; HEBP: Etidronic acid; NaOCI: Sodium hypochlorite; EDTA: Ethylene diamine tetra acetic acid.

were found to be significantly lower than the cases in the CHX and HEBP (p < 0.05) (Table 2, Figure 4).

DISCUSSIONS

After the root canal preparation and the arrangement of the post space, a thicker smear layer is formed in the canal.²¹ Therefore, the cementation of fiber posts is adversely affected. To prevent this, it is very important to clean the RCD surfaces after arranging the mechanical post space for ideal post retention, especially when resin cement is preferred.²² The chemicals used for cleaning cause deterioration in the chemical and mechanical properties of dentin. In addition, weakening of dentin microhardness may impair the adhesion and sealing of root canal sealers to the root dentin walls. The present study revealed that microhardness values of the NaOCl and HEBP-NaOCl cases were found to be significantly lower than others. the reduction of microhardness in NaOCl and HEBP-NaOCl might also be explained by the strong demineralizing effects of the mixture, which may, in turn, cause a reduction in hardness of dentin and denaturation of collagen fibrils.^{14,23,24} On the other hand, CHX and EDTA did not affect microhardness and, showed similar values to the CG. Akcav and Sen investigated the effect of EDTA solution on microhardness of root dentin and found that EDTA reduces microhardness of root dentin.²⁵ EDTA has strong demineralizing effects, which may in turn cause softening of the dentin and denaturation of collagen fibrils.²⁴ Dineshkumar et al. also evaluated root dentin microhardness using 18% HEBP as the last rinsing parameter and they found that HEBP-treated root dentin has the highest microhardness values.²⁶ Similarly, our results showed that HEBP irrigation in-

TABLE 2: The means and standard deviations of the root dentin microhardness values in cervical dentin segments for en- dodontic irrigation solution treatment groups and control group.						
Microhardness						
	Ort±SS	Median (minimum-maximum)	p value	Post-hoc		
°Control	51.84±11.11	52.81 (38.6-63.2)	0,004**	c <b,d,a< td=""></b,d,a<>		
♭CHX	59.22±6.92	61.01 (49.5-65.3)		e <b,d< td=""></b,d<>		
°HEBP-NaOCI	36.19±7.31	32.90 (29.4-48.1)				
dHEBP	65.00±11.56	61.06 (56.0-81.9)				
^e NaOCI	39.66±3.07	39.87 (35.8-43.1)				
^f EDTA	45.30±2.84	44.88 (42.4-81.9)				

Oneway ANOVA & post-hoc Games Howell test; **p<0.01; SS: Standard deviation; CHX: Chlorhexidine; HEBP: Etidronic acid; NaOCI: Sodium hypochlorite; EDTA: Ethylene diamine tetra acetic acid.

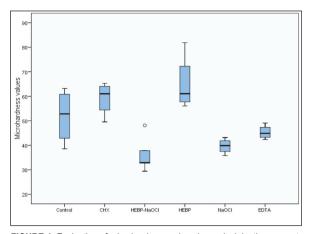


FIGURE 4: Evaluation of microhardness values in cervical dentin segments between groups. CHX: Chlorhexidine; HEBP: Etidronic acid; NaOCI: Sodium hypochlorite; EDTA: Ethylene diamine tetra acetic acid.

creased the microhardness of the dentin. As CHX, EDTA and HEBP did not reduce the microhardness of root dentin, these irrigants can be suggested as good candidates for irrigation. However, one limitation of our study was the measurement of microhardness values regardless of different regions. Therefore, the relation between microhardness and PBS values was not analyzed.

HEBP-NaOCl and NaOCl significantly higher PBS values were obtained in the cervical part compared to CHX, EDTA, HEBP, and CG. This finding suggest that the use of irrigation solution solutions as the final irrigation material has a significant effect on the PBS of the fiber post. This finding can be explained by the fact that NaOCl disrupts the collagen structure of dentin in the cervical region, which reveals the PBS.²⁷ There were no major discrepancies among the PBS amounts of the coronal in CHX, EDTA, and HEBP. This finding suggests that using these solutions as a final irrigant shows no remarkable effect on the PBS of the fiber post. No meaningful difference was recorded between the PBS of the coronal regions in HEBP-NaOCl and NaOCl. Moreover, a higher PBS value was detected in the coronal region in HEBP-NaOCl, according to compare NaOCl. This finding demonstrates that when

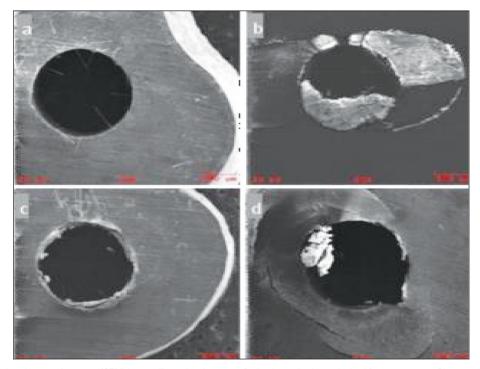


FIGURE 5: Scanning electron microscopy (SEM) images illustrating the types of adhesive, cohesive and mixed fracture patterns. Photographs of a failed specimens after push-out test. a) The specimen with the type of adhesive failure between root dentin and the luting agent (original magnification X20). b) The specimen with adhesive fracture type, between post and cement (value of original magnification X20). c) The specimen with cohesive fracture within the post (value of original magnification X20). d) The specimen with mixed failure (original magnification). Remnants of the luting agents are visible (*).

NaOCl is used in combination with HEBP, its penetration to the cervical region increases and therefore it causes the same adhesion values in the coronal region.

NaOCl also removes mineralization inhibitors while dissolving organic tissue while releasing smear tissue.²⁷⁻³⁰ And this may explain the significantly lower microhardness of NaOCl. In addition, it was observed that the use of 2.5% NaOCl as an irrigation solution significantly increased the Ca/P ratio in dentin.³¹ It has a bactericidal effect due to its capability to precipitate and coagulate bacterial intracellular ingredients.³² Moreover, several solutions have been advised to take out the inorganic tissue, e.g. EDTA and HEBP solutions, which are strongly acidic.³³ Regarding the cervical level, the highest mean PBS was measured for NaOCl and HEBP-NaOCl. It is not surprising that NaOCl demonstrated the highest PBS because the NaOCl is the most effective irrigant on organic substances. When CHX, EDTA, and HEBP were used, the PBS stayed similar to that of CG. Although HEBP decreased the PBS in the cervical field, this decrease did not make a statistically major difference. NaOCl and HEBP-NaOCl statistically enhanced the PBS in the cervical region. Since the number of organic compounds increased in the cervical region, NaOCl interacted with the collagen structures in this region and increased the PBS. When inorganic/organic component ratios are considered, inorganic phase density (hydroxyapatite/HA) decreases as cervical. The demineralization effect of the solution depends on several factors such as the acidity of the solution and the degree of penetration. Moreover, the strength of the push-out depends on the binding strength of the resin with the surrounding tissues. Regarding PBS to dentin, irrigation protocols affected the regions differently. The null hypothesis of "There is no measurable difference in PBS between groups" was rejected. The present results showed that the PBS for the cervical root dentin in NaOCl and HEBP-NaOCl have bigger values than in the other groups. When push-out values are evaluated the presented results suggest the use of NaOCl and HEBP-NaOCl mixture treatment as cleaning and chelating agents instead of using the

CHX, EDTA, and HEBP. In terms of microhardness, HEBP solution more effective solution for irrigation solutions.

According to the observed higher PBS values of the CHX, NaOCl, and HEBP-NaOCl combination groups, mixed failure type was predominantly observed (Figure 5). The reduction in adhesive failures indicates the high quality of the bond when NaOCl and HEBP-NaOCl were applied. On the other hand, more adhesive failure was seen in the EDTA and HEBP than in other groups. Maybe, pretreatment with silane before bonding would be better to avoid the adhesive type failure model.

A methodological issue to be noted in connection with the current study has to do with the parts of root canals where specimens used for testing. We preferred to use cervical root portions in the present study, instead of apical one having sclerotic dentin effecting the bonding negatively.³⁴ Some previous studies have recommended the use of midroot portions for bond strength testing, especially in oval shaped root canals.³⁵ Because oval shaped root canals have untouched and uncleaned surfaces in the coronal portion.³⁵ However, as we selected incisor teeth with straight root canals, we obtained specimens from coronal root portions.

CONCLUSIONS

Use of HEBF alone yielded good bond strength of fiber post to root canal dentin and had no negative effects on the microhardness of root canal dentin. Therefore, its use alone is recommended, instead of its combination use with 1:1 NaOCl.

Source of Finance

During this study, no financial or spiritual support was received neither from any pharmaceutical company that has a direct connection with the research subject, nor from a company that provides or produces medical instruments and materials which may negatively affect the evaluation process of this study.

Conflict of Interest

No conflicts of interest between the authors and / or family members of the scientific and medical committee members or members of the potential conflicts of interest, counseling, expertise, working conditions, share holding and similar situations in any firm.

Authorship Contributions

Idea/Concept: Şeyda Erşahan; Design: Yelda Erdem Hepşenoğlu, Burcu Yılmaz, Aysu Aydınoğlu; Control/Supervision: Afife Binnaz Hazar Yoruç; Data Collection and/or Processing: Kadir Sağır, Burcu Yılmaz, Aysu Aydınoğlu, Yelda Erdem Hepşenoğlu; Analysis and/or Interpretation: Aysu Aydınoğlu,Kadir Sağır; Literature Review: Şeyda Erşahan; Writing the Article: Şeyda Erşahan, Yelda Erdem Hepşenoğlu, Aysu Aydınoğlu; Critical Review: Burcu Yılmaz, Aysu Aydınoğlu; References and Fundings: Afife Binnaz Hazar Yoruç; Materials: Yelda Erdem Hepşenoğlu, Burcu Yılmaz.

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