

EFFECT OF SODIUM HYPOCHLORITE AND ETHYLENE DIAMINE TETRA-ACETIC ACID ON THE PUSH-OUT BOND STRENGTH AND SEALING ABILITY OF DIFFERENT BIO-CERAMIC ROOT REPAIR MATERIALS. (IN VITRO STUDY)

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ABSTRACT

This study aimed to evaluate the effect of sodium hypochlorite (NaOCl) and ethylene diamine tetra-acetic acid (EDTA) on the bonding and sealing ability of different root repair materials.

Methodology: for the push-out bond strength test, ninety dentine discs with 2 mm thickness and a central opening of 1.5 mm were used. The discs were randomly distributed into 3 groups (n=30) based on the repair material used (white MTA Angelus, Biodentine, and Well-Root putty). The materials were condensed inside each disc and allowed to be initially set for 10 minutes. Then each group was further subdivided into 3 subgroups (n=10) based on the irrigant used (NaOCl, EDTA, and no irrigation was used in the 3rd control group) and immersed into the corresponding irrigant for 30 minutes. After complete setting for 72 hours, the bond strength was evaluated with a universal testing machine. For the sealing ability, the same methodology was followed but instead of using dentine discs, we used human-extracted lower molar teeth (n=72) with a standard furcation perforation. After 72 hours, the specimens were subjected to the dye penetration test to assess the sealing ability.

Results: Well-Root putty showed the highest bond strength and Biodentine showed the highest sealing ability.

Conclusion: NaOCl had a positive effect on the bond strength and sealing ability of both Biodentine and Well-Root putty while the EDTA had a negative effect on the properties of the three tested repair materials.

KEYWORDS: MTA, Biodentine, Well-Root putty, push-out bond strength, Dye penetration,

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INTRODUCTION

The success of root canal treatment depends mainly on proper chemomechanical preparation and three-dimensional obturation of the root canal system, so our main target during and after the endodontic therapy is to achieve what is called a “hermetic seal” that ensures complete isolation of the root canal system.

Root Perforation is a direct communication between the root canal system and the periodontium. Perforation prognosis depends on the size, site, time of occurrence, time of repair, and material of repair. So ideally it is important to repair any perforation as soon as possible after occurrence to achieve better tissue healing ⁽¹⁾.

The ideal perforation repair material should be biocompatible, dimensionally stable, antibacterial, easy to manipulate, radiopaque, and unaffected by blood contamination, tissue fluid, and different root canal irrigation solutions ⁽²⁾.

Nowadays many different materials are used for perforation repair with more ease of manipulation properties that enable them to compete with MTA and Biodentine, these materials include Bioceramic putty which comes in the form of paste consistency providing a very ease of use in comparison to MTA.

Therefore, conducting a study to evaluate the effect of the most common irrigant solutions (NaOCl and EDTA) on different perforation repair materials was thought to be of value.

Our Hypothesis is that NaOCl and EDTA have no effect on the tested root repair materials.

MATERIALS AND METHODS

Sample preparation and grouping

For the push-out test, ninety extracted human single-rooted teeth were used to prepare dentine discs (n=90). The sample size was calculated based on a previous study ⁽³⁾.

The mid-root of each tooth was sectioned horizontally into slices with a thickness of (2 ± 0.01) using a diamond disc and mandrel. The canal space of each slice was enlarged with a complete pass of Gates Glidden burs sizes 2, 3, 4, 5, and 6 (Dentsply Maillefer, Switzerland) to achieve a diameter of 1.5 mm. Then the samples were randomly divided into 3 groups (n=30) based on the repair material used (MTA, Biodentine, and Well-Root putty).

After mixing the materials according to the manufacturer’s instructions, the repair materials were condensed inside the canal space with an endodontic plugger and allowed to be initially set for 10 minutes at 37°C and 100% humidity, then each group was subdivided into 3 subgroups (n=10) based on the irrigant used (NaOCl, EDTA, and no irrigant in the control group). The samples were immersed in the irrigants for 30 minutes.

After rinsing the samples of the two experimental groups, all samples were wrapped with moist cotton pellets and allowed to be completely set for 72 hours at 37°C and 100% humidity. After that, Loading was performed by a universal testing machine at a speed of 0.5 mm/min until deboning occurs.

The bond strength value in megapascals (Mpa) was computed by dividing the maximum load needed to dislodge the filling material in Newtons (N) by the bonded interfacial area (mm^2)

$$\text{Bond strength} = \frac{\text{Maximum load at dislodgement (N)}}{\text{Bonding surface area (mm}^2\text{)}}$$

For the sealing ability test, seventy-two extracted human lower molar teeth were used and the sample size was calculated based on a previous study ⁽⁴⁾. After the access cavity preparation, the molar teeth were decoronated 3 mm above the cervical line and the roots were amputated 3 mm apical to the furcation to facilitate the sample standardization. A standard perforation was made in the furcation area using a round bur size 2.

Then each molar was covered with 2 layers of nail polish including the access cavity walls except 2 mm around the perforation and the orifices of canals were blocked by the flowable composite. The samples then were divided into 3 groups (n=24) based on the repair material used (MTA, Biodentine, and Well-Root putty).

After repairing the perforation, the repair materials were allowed to be initially set for 10 minutes then each group was subdivided into 3 subgroups (n=8) according to the irrigant used (NaOCl, EDTA, and no irrigant in the control group). Each access was filled with the corresponding irrigant for 30 minutes except for the control group that wasn't exposed to any irrigant. After rinsing the two experimental groups, all samples were allowed to be completely set for 24 hours at 37°C and 100% humidity.

After the complete setting, the access cavities were filled with 2 % Methylene blue dye for 72 hours. After that, the dye was rinsed properly for 30 minutes, each tooth was sectioned longitudinally in a buccolingual direction and the highest amount of dye penetration in millimeters (mm) was measured using computer software on the image captured by a digital camera mounted on the stereomicroscope with a magnification of 16x.

Statistical analysis

Categorical data were presented as frequency and percentage values and were analyzed using fisher's exact test. Numerical data were presented as mean and standard deviation (SD) values.

They were explored for normality by checking the data distribution and using Kolmogorov-Smirnov and Shapiro-Wilk tests. Data showed parametric distribution so one-way ANOVA followed by Tukey's post hoc test was used for intergroup comparisons and repeated measures ANOVA followed by Bonferroni post hoc test was used for intragroup comparisons. The significance level was set at $p \leq 0.05$. Statistical analysis was performed with R statistical analysis software version 4.1.2 for Windows⁽⁵⁾.

RESULTS

For the push-out test

For the **MTA group**, there was a negative significant effect of both NaOCl and EDTA on the push-out strength of MTA. With the highest value for control (53.02 ± 11.83) and the lowest value for NaOCl (17.02 ± 6.27) ($p < 0.05$).

For the **Biodentine group**, while NaOCl had a positive significant effect on the push-out strength of Biodentine, EDTA had no significant effect on push out of Biodentine. With the highest value for NaOCl (26.33 ± 11.22) and the lowest value for EDTA (5.37 ± 2.60) ($p < 0.05$).

For the **Putty group**, while NaOCl had a positive significant effect on the push-out of Putty, EDTA had a negative significant effect on push-out of Putty. With the highest value for NaOCl (72.28 ± 17.54) and the lowest value for EDTA (47.11 ± 13.70) ($p = 0.097$).

TABLE (1) Mean and standard deviation values (mm) of the dye penetration for intergroup comparison

	Control	NaOCl	EDTA	P-value
MTA	0.0745 \pm 0.1521 ^a	0.5453 \pm 0.1814 ^b	0.1026 \pm 0.1443 ^a	<0.05
Biodentine	0.0760 \pm 0.1123 ^a	0.0955 \pm 0.1843 ^a	0.0904 \pm 0.1404 ^a	0.964
Putty	0.199 \pm 0.340 ^a	0.0999 \pm 0.1397 ^a	0.8518 \pm 0.0817 ^b	<0.05

*Different superscript letters indicate a statistically significant difference within the same horizontal row *; significant ($p \leq 0.05$) ns; non-significant ($p > 0.05$)*

For the sealing ability test

As shown in (table 1)

For the **MTA group**, while NaOCl had a negative significant effect on the sealing ability of MTA, EDTA had no significant effect on the sealing ability of MTA. With the lowest value for control (0.0745 ± 0.1521) and the highest value for NaOCl (0.5453 ± 0.1814) ($p < 0.05$).

For the **Biodentine group**, there was no significant effect of both NaOCl and EDTA on the sealing ability of Biodentine. With the highest value for control (0.0760 ± 0.1123) and the lowest value for NaOCl (0.0955 ± 0.1843) ($p = 0.964$).

For the **Putty group**, while NaOCl had no significant effect on the sealing ability of MTA, EDTA had a negative significant effect on the sealing ability of putty. With the lowest value for NaOCl (0.0999 ± 0.1397) and the highest value for EDTA (0.8518 ± 0.0817) ($p < 0.05$).

DISCUSSION

Within the limitation of this study, NaOCl had a positive significant effect on the push-out strength of both Biodentine and Well-Root putty. But had a significant negative effect on the MTA. According to **Uyanik et al.** ⁽⁶⁾, its positive effect can be justified by the high pH environment of NaOCl (10.5-12.9) ⁽⁷⁾ which has a positive influence on the setting reaction of the bioceramic materials and enhances numerous physical and chemical properties of them.

These results are in agreement with previous studies ^(8,9,10,11,12) that also found an increase in the push-out bond strength of the tested materials after exposure to NaOCl.

In our study, the negative effect of the NaOCl on the white MTA Angelus group can be explained by the study of **Zapf AM et al.** ⁽¹³⁾ who found that mixing the MTA with NaOCl significantly reduces the Ca(OH)_2 formation. And another study by **Yamashita, Kozue, et al.** ⁽¹⁴⁾ reported that NaOCl immersion inhibited the formation of Ca(OH)_2 on

the surfaces of set white MTA discs which can have a detrimental effect on the properties of the MTA including the push-out bond strength and sealing ability. Also, NaOCl inhibited WMTA-induced mineralized nodule formation. Therefore, NaOCl should be firmly removed from root canals that are repaired by white MTA after root canal treatment to maintain its ability to stimulate hard tissue formation.

Regarding the effect of EDTA on the tested materials, EDTA had a significant negative effect on the MTA and putty groups and an insignificant effect on the Biodentine group. These results can be justified by that EDTA has a negative influence on the hydration properties of calcium silicate cements because of its chelating action ⁽¹⁵⁾. This causes the dissolution of the binding phase of the cement, which in turn, inhibits its adhesion to the dentine. EDTA chelates the calcium ions released from the tricalcium silicate cement during hydration, thereby disturbing the formation of hydrated products ⁽¹⁶⁾.

Regarding the effect of NaOCl on the perforation repair materials sealing ability, we found that NaOCl enhanced the sealing ability of Biodentine and putty but on the other hand decreased the sealing ability of MTA significantly.

This effect can be justified by that NaOCl is a halogenated compound that can cause mineral accumulation in human root dentin and expose inorganic material which unlike EDTA may prevent dentin dissolution or may leave a smear layer of mineralized tissue that could increase the Ca/P ratio of the dentin surface intensifying the hydration process of bioceramic materials that is responsible for the strength, barrier properties, and sealing ability of these materials ⁽¹⁷⁾.

These results are in agreement with previous studies ^(18,19) except for the negative effect of NaOCl on the MTA group which can be explained for the same reasons as its negative effect on the MTA group in the push-out bond strength test that we mentioned before.

Regarding the effect of EDTA on the sealing ability of the tested materials, in our study, we found that EDTA insignificantly decreased the sealing ability of Biodentine and MTA but significantly decreased that of the putty group

These results can be explained by the chelating action of EDTA that can lead to an increase in the solubility of the repair materials which, in turn, negatively affects their sealing abilities. Moreover, calcium-depleting irrigants such as EDTA are capable of dissolving the smear layer rapidly and infiltrate into the interfacial layer; where they can interfere with the chemical adhesion between repair materials and dentin allowing the dye leakage through these micro-gaps^(18,20).

CONCLUSION

Based on the present results, within the circumstances of this in-vitro study, it can be concluded that the early exposure to NaOCl had a positive effect on the Biodentine and Well-Root putty bond strength, sealing ability and vice versa for the white MTA Angelus and the early exposure to EDTA had a negative effect on the bond strength, sealing ability of the whole tested materials.

So Caution should be taken to avoid exposing the perforation repair materials in their early stages of setting to EDTA and in particular to NaOCl in case of white MTA Angelus in a single visit treatment, and perhaps postpone the root canal treatment to the next visit to ensure the complete setting of the repair materials.

REFERENCES

- Hegde M, Varghese L, Malhotra S. Tooth root perforation repair—A review. *Oral Health Dent Manage*. 2017; 16(2):1-4.
- von Loetzen SC, Hülsmann M. Root perforation repair concepts and materials: A review. *ENDO (Lond Engl)*. 2018 Jun 1;12(2):87-100.
- Alsubait SA. Effect of Sodium Hypochlorite on Push-out Bond Strength of Four Calcium Silicate-based Endodontic Materials when used for repairing Perforations on Human Dentin: An in vitro Evaluation. *The Journal of Contemporary Dental Practice*. 2017;18(4):289-94.
- Yahya MM. Sealing ability of Biodentine as a root Perforation Treatment Material (An in Vitro Study). *Tikrit J Dent Sci*. 2015;1:62-8.
- R. C. Team, "A language and environment for statistical computing," *Computing*, vol. 1. Jan. 01, 2006.
- Uyanik MO, Nagas E, Sahin C, Dagli F, Cehreli ZC. Effects of different irrigation regimens on the sealing properties of repaired furcal perforations. *Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontology*. 2009 Mar 1;107(3):e91-5.
- Torabinejad M, Hong CU, McDonald F, Ford TP. Physical and chemical properties of a new root-end filling material. *Journal of endodontics*. 1995 Jul 1;21(7):349-53
- Hong ST, Bae KS, Baek SH, et al. Effects of root canal irrigants on the push-out strength and hydration behavior of accelerated mineral trioxide aggregate in its early setting phase. *J Endod* 2010; 36:1995-9.
- Paulson L, Ballal NV, Bhagat A. Effect of root dentin conditioning on the pushout bond strength of biodentine. *Journal of Endodontics*. 2018 Jul 1;44(7):1186-90.
- Sariyilmaz E, Sivas Yilmaz Ö, Keskin C, Keleş A. Effect of sodium hypochlorite and chlorhexidine irrigating solutions and their inactivating agents on the push-out bond strength of mineral trioxide aggregate. *Bio-Medical Materials and Engineering*. 2019 Jan 1;30(3):279-85.
- Tiwari N, Borkar AC, Tandale A, Nighot N, Ghare S, Maral S. Comparative evaluation of the effect of various endodontic irrigants on the push-out bond strength of endosequence, Biodentine™, and MTA Plus™ root repair materials: An in vitro study. *Journal of the International Clinical Dental Research Organization*. 2019 Jan 1;11(1):9.
- Reboloso de Barrio E, Gancedo-Caravia L, García-Barbero E, Pérez-Higueras JJ. Effect of exposure to root canal irrigants on the push-out bond strength of calcium silicate-based cements. *Clinical Oral Investigations*. 2021 May;25(5):3267-74.
- Zapf AM, Chedella SC, Berzins DW. Effect of additives on mineral trioxide aggregate setting reaction product formation. *Journal of Endodontics*. 2015 Jan 1;41(1):88-91.
- Yamashita K, Tomokiyo A, Ono T, Ipposhi K, Alhasan MA, Tsuchiya A, Hamano S, Sugii H, Yoshida S, Itoyama T,

- Maeda H. Mineral trioxide aggregate immersed in sodium hypochlorite reduce the osteoblastic differentiation of human periodontal ligament stem cells. *Scientific reports*. 2021 Nov 11;11(1):1-0.
15. Ballal NV, Sona M, Tay FR. Effects of smear layer removal agents on the physical properties and microstructure of mineral trioxide aggregate cement. *Journal of dentistry*. 2017 Nov 1;66:32-6.
 16. Lee YL, Lin FH, Wang WH, Ritchie HH, Lan WH, Lin C-P. Effects of EDTA on the hydration mechanism of mineral trioxide aggregate. *J Dent Res* 2007; 86:534-8.
 17. Inaba D, Ruben J, Takagi O, Arends J. Effect of sodium hypochlorite treatment on remineralization of human root dentine in vitro. *Caries Res* 1996;30:218-24.
 18. Uyanik MO, Nagas E, Sahin C, Dagli F, Cehreli ZC. Effects of different irrigation regimens on the sealing properties of repaired furcal perforations. *Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontology*. 2009 Mar 1;107(3):e91-5.
 19. Al-Azzawi AK, Al-Zubaidi AK. The effect of various endodontic irrigants on the sealing ability of biodentine and other root perforation repair materials: in vitro study. *J. Baghdad Coll. Dent*. 2014;26:1-8.
 20. Roy CO, Jeansonne BG, Gerrets TF. Effect of an acid environment on leakage of root-end filling materials. *Journal of endodontics*. 2001 Jan 1;27(1):7-8.