



**SURFACE MICROHARDNESS AND ROUGHNESS PROPERTIES OF BIODENTINE  
FOLLOWING TREATMENT WITH VARIOUS ENDODONTIC IRRIGANTS**

**ÇEŞİTLİ ENDODONTİK İRRİGANLARLA TEDAVİYİ TAKİBEN BİODENTİNE'İN  
YÜZEY MİKROSERTLİĞİ VE PÜRÜZLÜLÜK ÖZELLİKLERİ**

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**Makale Kodu/Article code:** 4057  
**Makale Gönderilme tarihi:** 02.05.2019  
**Kabul Tarihi:** 11.12.2019  
**DOI :** 10.17567/ataunidfd.658069

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**ABSTRACT**

**Aim:** Endodontic irrigants may be used during a second-visit treatment or retreatment of root canals with perforations requiring repair biomaterials. After a final flushing with a chemical irrigant, some solution may remain in the root canal space, which can affect the surface of the biomaterial, altering its properties and roughness. The present study aimed to evaluate the effect of various irrigating solutions on surface microhardness and roughness of Biodentine (Septodont, Saint Maur des Fosses, France).

**Materials and Methods:** Fifty Biodentine specimens were prepared and randomly divided into five groups, with 10 samples in each group. The specimens were then stored in different solutions for 5 min: distilled water (control), 5.25% sodium hypochlorite (NaOCl), 17% ethylenediaminetetraacetic acid (EDTA) solution, 2% chlorhexidine (CHX), or ozonated water. Surface microhardness (Vickers hardness number [VHN]) and surface roughness were evaluated using 2-D profilometry. The data were analyzed using Kruskal-Wallis and Mann-Whitney U tests. The significance level was set at  $P < 0.05$ .

**Results:** The VHN of specimens exposed to NaOCl and CHX was significantly lower than the VHN of specimens exposed to distilled water and EDTA ( $p < 0.001$ ,  $p = 0.003$ ,  $p = 0.001$ , and  $p = 0.006$ , respectively). There was no significant difference in the mean VHN of the EDTA-treated specimens versus that of the control samples ( $p = 0.999$ ). Regarding the surface roughness of Biodentine, there were no significant differences between irrigation solutions ( $\chi^2 = 4.243$ ;  $p = 0.374$ ).

**Conclusions:** Exposure to all the irrigation solutions, except EDTA and ozonated water had an adverse effect on surface microhardness of Biodentine, whereas none of the irrigation solutions significantly changed surface roughness. Therefore, in clinical situations, such as perforation repair with Biodentine, use of EDTA and ozonated water may be preferred.

**Keywords:** Vickers surface microhardness, profilometry, irrigation solutions, Biodentine

**ÖZ**

**Amaç:** Biomateryallerle tamir gerektiren perforasyonlu kök kanallarının yeniden tedavisi veya ikinci-seans tedavisi esnasında endodontik irriğanlar kullanılabilir. Kimyasal bir irriğanla son yıkamadan sonra bir miktar solüsyon kök kanal boşluğunda kalabilir, bu da biyomateryalin sertlik ve yüzey özelliklerini değiştirebilir. Bu çalışma çeşitli irriğasyon solüsyonlarının Biodentine (Septodont, Saint Maur des Fosses, Fransa)'ın yüzey mikrosertliği ve pürüzlülüğü üzerine etkisini değerlendirmeyi amaçlamaktadır.

**Gereç ve Yöntemler:** Elli adet Biodentine örneği hazırlandı ve rastgele olarak her grup 10 örnek içerecek şekilde beş gruba ayrıldı. Sonrasında örnekler farklı solüsyonlarda 5 dakika bekletildi: distile su(kontrol), %5.25 sodyum hipoklorit (NaOCl), %17 etilendiamintetraasetik asit (EDTA) solüsyonu, %2 klorheksidin (CHX), veya ozonlu su. Yüzey mikrosertliği (Vickers sertlik değeri [VHN]) ve yüzey pürüzlülüğü 2-D profilometre kullanılarak değerlendirildi. Veriler Kruskal-Wallis ve Mann-Whitney U testleri kullanılarak değerlendirildi. İstatistiksel anlamlılık seviyesi  $P < 0.05$  olarak belirlendi.

**Bulgular:** NaOCl ve CHX'de bekletilen örneklerin VHN değerleri distile su ve EDTA'da bekletilenlere göre anlamlı derecede düşüktür ( $p < 0.001$ ,  $p = 0.003$ ,  $p = 0.001$ , ve  $p = 0.006$ , sırasıyla). EDTA-uygulanmış örneklerin ortalama VHN değeriyle kontrol örneklerinkinin arasında anlamlı fark bulunmamaktadır ( $p = 0.999$ ). Biodentine'in yüzey pürüzlülüğüne ilişkin ise irriğasyon solüsyonları arasında anlamlı fark bulunmamaktadır ( $\chi^2 = 4.243$ ;  $p = 0.374$ ).

**Sonuçlar:** EDTA ve ozonlu su haricindeki tüm irriğasyon solüsyonlarına maruz kalmanın Biodentine'in yüzey sertliği üzerine olumsuz etkisi bulunsa da, diğer taraftan irriğasyon solüsyonlarının hiçbir yüzey pürüzlülüğünü anlamlı derecede değiştirmemiştir. Bundan dolayı, Biodentine ile perforasyon tamiri gibi klinik durumlarda, EDTA ve ozonlu su kullanımı tercih edilebilir.

**Anahtar kelimeler:** Vickers yüzey mikrosertliği, profilometre, irriğasyon solüsyonları, Biodentine

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**Kaynaçça Bilgisi:** Erşahan Ş, Yıldırım C, Martı Akgün Ö, Özmen B, Başak F, Demir P, Tekeli S. Çeşitli Endodontik İrriganlarla Tedaviyi Takiben Biodentine'in Yüzey Mikrosertliği ve Pürüzlülük Özellikleri. Atatürk Üniv Diş Hek Fak Derg 2020; 30: 242-246.

**Citation Information:** Erşahan S, Yıldırım C, Martı Akgün Ö, Özmen B, Başak F, Demir P, Tekeli S. Surface Microhardness and Roughness Properties of Biodentine Following Treatment with Various Endodontic Irrigants. J Dent Fac Atatürk Uni 2020; 30: 242-246.



## INTRODUCTION

Root perforations are frequent complications during endodontic treatment. Such perforations result in openings into the periodontal ligament space and affect the prognosis of root canal treatment. For long-term success, perforations should be repaired as quickly as possible using a biocompatible material to prevent bacterial contamination.<sup>1</sup> An ideal material for perforation repair should adhere to the root canal wall and have sufficient sealing ability, in addition to having biocompatible or bioactive properties. Furthermore, the material should be dimensionally stable, insoluble in tissue fluids, nonresorbable, and radiopaque.<sup>2</sup> At present, mineral trioxide aggregate (MTA) is the preferred furcation repair material due to a unique combination of properties including dimensionally stability, insolubility in tissue fluids, and radioopaque. However, disadvantages include its handling properties, prolonged setting time, limited resistance to washout before setting, and potential staining of the tooth structure.<sup>3</sup> Therefore, new root repair materials with improved properties are continually being developed. Recently, a new root repair tricalcium silicate material, Biodentine (Septodont, Saint Maur des Fosses, France), has been introduced for use as a root-end filling material and for perforation repair. Biodentine is composed of tricalcium silicate, calcium carbonate, zirconium oxide, and a water-based liquid containing calcium chloride, which acts as a setting accelerator and water-reducing agent. Previous studies showed that Biodentine exhibited good sealing ability, high compressive strength, and a short setting time.<sup>4,5</sup> In addition, its biocompatibility was similar to that of MTA.<sup>6</sup>

Endodontic irrigants may be used during a second-visit treatment or retreatment of root canals with perforations requiring repair biomaterials. After a final flushing with a chemical irrigant, some solution may remain in the root canal space, which can affect the surface of the biomaterial, altering its properties and roughness.<sup>7</sup> A previous study of the effect of irrigating solutions on microhardness of MTA showed a change in microhardness values of MTA following contact with irrigation solutions.<sup>8</sup> No previous studies have explored the effect of different irrigants on surface properties of Biodentine, especially in perforation cases. Thus, the aim of this study was to evaluate the effect of different irrigants commonly used in endodontics (sodium hypochlorite [NaOCl],

ethylenediaminetetraacetic acid [EDTA], chlorhexidine gluconate [CHX], and ozonated water) on surface roughness and microhardness of Biodentine. The null hypothesis was that exposure to the irrigants would have no effect on surface properties of Biodentine.

## MATERIAL AND METHODS

Surface microhardness (Vickers hardness number [VHN]) and surface roughness were evaluated using 2-D profilometry. The material assessed was Biodentine.

### Sample preparation

Biodentine was mixed according to the manufacturer's instructions. The mixed material was packed incrementally into 50 cubic silicone molds (1 cm × 1 cm × 1 cm) and placed on a glass slab. The samples were then subjected to a constant vertical compaction force of 3.22 MPa applied for 1 min.<sup>9</sup> The extruded material was wiped away, and a wet cotton pellet was placed on top of the Biodentine. The samples were allowed to set for 10 min at room temperature. The upper surface of material was then wet polished at room temperature, using minimum hand pressure and silicon carbide grinding papers of 600-, 1000-, and 1200-grit. The polished specimens were rinsed in distilled water for 1 min and dried in oil-free air for 5 sec.

Fifty specimens were prepared and randomly divided into five groups, with 10 samples in each group. The specimens were then stored in either distilled water (control), 5.25% NaOCl (Wizard, Rehber Kimya, Istanbul, Turkey), 17% EDTA solution (pH 7.4) (Merck, Darmstadt, Germany), 2% CHX solution (IE Ulagay, Istanbul, Turkey), or ozonated water for 5 min at room temperature. All the samples were then posteriorly washed with distilled water for 5 min to remove any traces of the irrigating solution and left to dry for 48 h.

### Microhardness measurement

The VHN of each specimen was measured using a microhardness tester (Micromet 5114, Buehler Ltd., Lake Bluff, IL, USA) and a diamond indenter with a load of 500 g for 30 s.<sup>10</sup> The Vickers microhardness value was displayed on the digital read-out of the microhardness tester. The VHN was calculated based on the following formula:  $VHN = 1.854 \times (F/d^2)$ , where F was the load ( $kg^{-1}$ ) and d was the mean of the two diagonals produced by the indenter in millimeters.<sup>10</sup> The average of the results of three indentations was considered the representative

hardness value of each specimen. Three indentations were randomly made on the polished surface at separate sites, no closer than 1 mm to adjacent indentations or periphery of the specimen. Thus, in total, 30 indentations were made for each group. The mean and standard deviation of the microhardness values for each experimental group was calculated.

### Surface roughness testing

The surface roughness of the samples was measured using a 2-D profilometer (Perthometer M1, Mahr, Göttingen, Germany), with a cut-off length of 0.25 mm, tracing length of 2 mm, and tracing speed of 100 µm/s. For each sample, three measurements were taken at different locations and in different directions. The three roughness measurements obtained from each dentin specimen were averaged to obtain a single value for each sample.

### Statistical analysis

The data were subjected to statistical analysis using SPSS version 21.0 (SPSS Inc., Chicago, IL, USA). Kruskal–Wallis and  $\chi^2$  tests were performed to determine statistically significant between-group differences in surface microhardness and surface roughness. The statistical significance level was set at  $p < 0.05$ .

## RESULTS

Mean roughness and microhardness values are presented in Table 1 and 2. As shown in Table 1, there was a significant reduction in the mean VHN of the NaOCl- and CHX-treated samples as compared with that of the distilled water- and EDTA-treated samples ( $p < 0.001$ ,  $p = 0.003$ ,  $p = 0.00$ ,  $p = 0.006$ , respectively). However, there was no significant difference in the mean VHN of the ozonated water-treated samples versus distilled water-treated samples ( $p = 0.294$ ), EDTA-treated samples versus distilled water-treated samples ( $p = 0.999$ ), EDTA-treated samples versus ozonated water-treated samples ( $p = 0.991$ ), or NaOCl-treated samples versus CHX-treated samples ( $p = 0.999$ ) (Table 3).

Table 1 presents the 2-D optical profilometer analysis values. The Kruskal–Wallis test revealed no significant differences between the surface roughness values of the samples in the different groups ( $\chi^2 = 4.243$ ;  $p = 0.374$ ).

## DISCUSSION

The surface microhardness of a material provides some indication of its surface strength.<sup>11</sup> In the present study, the microhardness of Biodentine

Table 1. Comparison of surface roughness values of test groups

Group	Surface Roughness		
	Mean	Min; Max	Mean±SS
NaOCl	1.557 (1.216)	0.833; 3.356	1.759±0.898
CHX	1.481 (0.814)	0.696; 2.369	1.542±0.539
EDTA	1.339 (0.844)	0.670; 2.142	1.286±0.477
Ozonated water	1.414 (0.575)	0.940; 2.126	1.469±0.384
Distilled water	1.138 (0.448)	0.990; 1.800	1.239±0.310
<b>Statistics*</b>	$\chi^2 = 4.243$ ; $p = 0.374$		

\*Kruskal-Wallis test

Table 2. Comparison of microhardness values of test groups

Group	Microhardness		
	Mean	Min; Max	Mean±SS
NaOCl	47.6 (26.8)	28.6; 72.6	46.5±14.9
CHX	48.3 (15.1)	33.8; 71.7	48.3±11.4
EDTA	73.9 (13.1)	65.1; 83.8	74.0±6.9
Ozonated water	67.1 (23.9)	47.2; 77.4	63.1±11.6
Distilled water	75.3 (6.6)	63.4; 88.4	76.6±6.6
<b>Statistics*</b>	$\chi^2 = 29.323$ ; $p < 0.001$		

Table 3. Posthoc comparisons between groups for microhardness test

Grup	CHX p	EDTA P	Ozonated water p	Distilled water p
NaOCl	0.999	<b>0.003</b>	0.470	<b>&lt;0.001</b>
CHX		<b>0.006</b>	0.715	<b>0.001</b>
EDTA			0.991	0.999
Ozonated water				0.294

was assessed after storage in different irrigation solutions using a Vickers hardness tester. Previous studies demonstrated the usefulness of Vickers hardness as an indicator of the progress and quality of the hydration process during the setting reaction, as well as the strength of calcium silicate-based materials.<sup>9,11</sup> A few previous studies tested the effect of irrigating solutions on the microhardness of MTA.<sup>7,11</sup> These studies demonstrated low or decreased microhardness values of MTA following contact with irrigation solutions. To validate endodontic indications of Biodentine, especially in cases of root perforations, we evaluated the effect of immersion in different irrigation solutions on its mechanical properties. In an attempt to evaluate the effect of the irrigants on the material's microstructure, we conducted a profilometric evaluation.

The results of the present study demonstrated that immersion of Biodentine specimens in NaOCl and CHX for 5 min significantly reduced the surface microhardness of the material. In contrast, immersion in EDTA and ozonated water did not change its microhardness. The hardness values of EDTA-treated and ozonated water-treated specimens were similar to those of the specimens stored in distilled water (control) but higher than those of the specimens stored in NaOCl and CHX.



Only one previous study evaluated the microhardness of Biodentine, but the study examined only the effects of EDTA and acids on the microhardness of the material.<sup>12</sup> The study found that physical properties of Biodentine were weaker after EDTA treatment, which was in contrast to the findings in the present study. The discord between our results and those of the previous study may be related to differences in test methods (Vickers microhardness test versus reference point indentation). No previous studies have reported the effects of the irrigation solutions NaOCl, CHX, and ozonated water on surface microhardness of Biodentine samples. Therefore, the results of this study were compared with those of a previous study, which examined the effects of NaOCl, CHX, EDTA, and BioPure MTAD on surface microhardness of MTA.<sup>13</sup> The previous study showed that the mean VHN of EDTA and BioPure MTAD was lower than that of the other groups, which is not in agreement with our results. Moreover, a recent study showed that solutions with acidic PH (EDTA) weakens the microhardness of MTA.<sup>14</sup> The discord may be explained by different test methodologies (Knoop test versus Vickers test), different materials (Biodentine versus MTA), and different exposure times (5 min versus 7 days).<sup>13</sup> We selected a shorter exposure time to simulate clinical situations, in which only 5 min of irrigation is generally considered sufficient for post-treatment in the repair of root perforations.<sup>13</sup> Furthermore, in the present study, we used ozonated water as an irrigation solution rather than BioPure MTAD solution. As NaOCl and CHX compromised the microhardness of Biodentine samples when compared with that of controls, EDTA, and ozonated water, the use of NaOCl and CHX after perforation repair may need to be reconsidered.

In addition to testing surface microhardness, we examined the surface microstructure of Biodentine samples using a profilometer and found no significant difference in values between the tested irrigants. No previous studies have reported the effect of irrigation solutions on roughness of Biodentine samples. Therefore, the results of this study were compared with those of a previous study that examined the effects of irrigation solutions on roughness of MTA.<sup>8</sup> The aforementioned study evaluated surface characteristics and calcium depletion of white MTA in response to exposure to different irrigants (1.3% sodium hypochlorite, 17% EDTA, and Biopure MTAD). The authors reported that Biopure MTAD exerted the greatest effect on surface properties of MTA.<sup>8</sup> In the

present study, the effect of NaOCl and EDTA on surface roughness of Biodentine was not significantly different from that of the controls. The difference between our results and those of the previous study may be explained by differences in the chemical compositions of Biodentine and MTA (e.g., the absence of calcium aluminate and other components in Biodentine). Gandolfi *et al.* (2013) reported that the surface characteristics of Biodentine and ProRoot MTA differed due to variations in the chemical and physical properties of the materials.<sup>15</sup> As compared with MTA, they stated that Biodentine resulted in the release of higher levels of free calcium ion and that it had higher alkalizing capability. The same study reported that calcium phosphate deposit formation was reduced with Biodentine and that these deposits were sphere shaped ( $< 1 \mu\text{m}$ ) and coated the surface of the material. In contrast, MTA was characterized by the formation of rounded agglomerates 1–5  $\mu\text{m}$  in diameter.<sup>15</sup> Thus, the surface properties of Biodentine may be different from those of MTA. The high calcium ion release of Biodentine is due to the presence of a di- and tri-calcium silicate (in powder) and calcium chloride component (in liquid). Calcium ions released from Biodentine likely form a thin layer on the surface of the material. Therefore, in the present study, higher levels of free calcium ion and short exposure time (i.e. 5 min) may explain the similarity in the surface roughness values of all groups. According to the results of the present study, our hypothesis was partially accepted.

In the present study, we tried to simulate clinical conditions. Therefore, Biodentine was allowed to set for 10 mins. After setting, the samples were kept in the solutions for 5 min to mimic the clinical situation. In root perforations, Biodentine is favored over MTA due to its short setting time (10 min) and ease of manipulation. After allowing Biodentine to set, root canal procedures are then continued, and various irrigation solutions are used for less than 5 min.

In this study, differences in the microhardness values of Biodentine may be associated with the concentration, chemical properties, and pH of the irrigation solutions. The type of irrigation solution used may affect the amount of calcium ions released from Biodentine and its alkaline characteristics.<sup>16</sup> Thus, different irrigation solutions may have dissimilar effects on surface microhardness and roughness of Biodentine. In the present study, although the surface roughness of Biodentine was not affected by the irrigation solutions, surface microhardness was affected.

## CONCLUSION

Exposure to all the irrigation solutions, except EDTA and ozonated water had an adverse effect on surface microhardness of Biodentine, whereas none of the irrigation solutions induced significant changes in surface roughness. Therefore, in clinical situations, such as perforation repair with Biodentine, the use of EDTA and ozonated water may be favored over other irrigants.

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### Conflicts of interest statement

*The authors declare no conflict of interest.*

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