Persistency of different Desensitizing agents to Tooth-Brushing Abrasion: Fluorescence Stereo Zoom Microscope Study

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ABSTRACT
The purpose of this study was to compare the persistency of selected desensitizing agents after tooth brushing using fluorescence stereo zoom microscope**.** Forty 5 x 5 x 5-mm dentin discs were prepared from freshly extracted non-carious human first and second premolars and then were randomly divided into four groups (n = 10). Dentin desensitizing agents (All-Bond Universal, Gluma Desensitizer, and BisBlock) were applied with a fluorescent red dye (Rhodamine B). The persistency of the desensitizers was analyzed by fluorescence stereo zoom microscope (AxioZoom) over a period of eight weeks of tooth brushing. The mean florescence intensity (MFI; pixel) values were calculated for each specimen. The results were analyzed using Kruskal–Wallis, Mann–Whitney U, and Wilcoxon signed-rank tests with a significance threshold of $p < 0.05$. The BisBlock group showed statistically significant higher MFI values than the All-Bond Universal and Gluma groups in all time stages (baseline, 1st, 2nd, 6th, and 8th weeks) ($p < 0.05$), whereas there were no statistically significant differences between the Gluma and All-Bond Universal groups ($p > 0.05$). However, the MFI values showed a reduction during the eighth week in all groups ($p < 0.05$). Tooth brushing caused various abrasion levels with the three desensitizing agents used in this study in proportion to time.

Keywords: Dentin-desensitizer, persistency, tooth-brushing, abrasion, stereo-zoom-microscope.

Persistencia de diferentes agentes Desensibilizantes a la abrasión por Cepillado de Dientes: Estudio con Microscopio de Zoom Estéreo de Fluorescencia

RESUMEN
El propósito de este estudio fue comparar la persistencia de agentes desensibilizantes seleccionados después del cepillado de dientes usando un microscopio de zoom estéreo de fluorescencia. Se prepararon cuarenta discos de dentina de 5 x 5 x 5 mm a partir de primeros y segundos premolares humanos no cariados recién extraídos y luego se dividieron aleatoriamente en cuatro grupos (n = 10). Se aplicaron agentes desensibilizantes de dentina (All-Bond Universal, Gluma Desensitizer y BisBlock) con un tinte rojo fluorescente (Rhodamine B). La persistencia de los desensibilizadores se analizó mediante microscopio de fluorescencia estero (AxioZoom) durante un período de ocho semanas de cepillado de dientes. Se calcularon los valores de intensidad de fluorescencia media (MFI; píxel) para cada muestra. Los resultados se analizaron mediante las pruebas de rango con signo de Kruskal-Wallis, Mann-Whitney U y Wilcoxon con un umbral de significación de $p < 0,05$. El grupo BisBlock mostró valores MFI estadísticamente significativos más que los grupos All-Bond Universal y Gluma en todas las etapas de tiempo (línea base, 1.a, 2.a, 6.a y 8.a semanas) ($p < 0.05$), mientras que no hubo diferencias estadísticamente significativas entre las Grupos Gluma y All-Bond Universal ($p > 0,05$). Sin embargo, los valores de MFI mostraron una reducción durante la octava semana en todos los grupos ($p < 0.05$). El cepillado de los dientes provocó varios niveles de abrasión con los tres agentes desensibilizantes utilizados en este estudio en proporción al tiempo.

Palabras clave: Desensibilizante, persistencia, cepillado-de-dientes, abrasión, microscopio-estereoscópico-con-zoom.

INTRODUCTION
Dentin hypersensitivity (DH) is a common clinical condition that is characterized by short, sharp pain arising from exposed dentin [1, 2]. Dentin exposure mostly arises as a result of enamel loss due to abfraction, abrasion, erosion, or stripping of the root surface caused by gingival recession or periodontal treatment [1].

The treatment of DH is based on the prevention or elimination of the possible causes of pain and can be classified into the following groups: anti-inflammatory agents [3], invasive methods (gingivectomy, pulpectomy, laser) [1], and professional applications such as adhesives and dentin desensitizers [4]. Several dentin desensitizers with different ingredients have been used to produce tubule occlusion and reduce hypersensitivity [5]. Professional application products include chemical agents, such as fluoride [6], oxalates [7], calcium compounds [8], potassium nitrate [9], strontium salts [10], glutaraldehyde [11], adhesive materials [12], and arginine-containing desensitizers [13].

Glutaraldehyde-containing resin material occludes the dentinal tubules, probably through the coagulation of plasma proteins in the dentinal fluid. Due to the effect of the precipitation of plasma proteins, it inhibits the flow of fluid through the tubules that causes sensitivity [3]. However, oxalate-containing forms oxalate crystals within the tubules, blocking fluid movement and, thus, relieving pain. Dentin bonding agents can also be used to reduce DH [14]. New generation methacryloyloxydecyl dihydrogen phosphate (MDP) included in bonding agents can form non-soluble Ca^2 salts on dentin surfaces. Thus, it can be suggested that these agents promote long-term strong adhesion [15].

Many desensitizing agents to alleviate DH have proven promising, with reduced levels of sensitivity reported [2, 16]. However, the desensitizing effects of many of these materials can fail over time by refraining from the type of food and drink ingested, medical conditions (stomach ulcer and reflux), and daily tooth brushing. Therefore, the success of long-term treatment of DH is thought to depend on occluding and penetrating the dentinal tubules to resist acid attacks as well as on tooth brushing [17]. Abnormal oral hygiene habits and tooth brushing appear to cause increased wear. It has been demonstrated that softer toothbrushes lead to more tooth wear than harder toothbrushes [18], while sonic toothbrushes cause significantly less wear than manual toothbrushes on eroded enamel and dentine [19]. However, a number of *in vitro* and *in situ* studies have focused on the effect of brushing forces on dentine wear, and the impact of toothbrush abrasion on DH is less clear-cut. In addition, currently, little is known about the persistency of dentin desensitizing agents against tooth brushing.

Stereo microscopes are a technology that uses a combination of macroscopic visualization and a camera with high sensitivity to fluorescent signals [20]. With its large working distance, a single objective and continuous zoom, it allows a natural visual perception of depth and dimensionality with a greater magnification.

AxioZoom stereo microscopes was used in the present study to analyze the abrasion level of the dentin desensitizing agents after tooth brushing because this system can detect 3D viewing of seamlessly from large specimens such as dentin and enamel to the smallest detail thus detecting fluorescent dyes like Rhodamine B. In addition, this system can obtain quantitative and objective results by processing images and analyzing intensity (pixel) differences efficiently.

Based on this information, the aim of this study was to comparatively evaluate the persistency of professionally applied dentin desensitizing agents used for the reduction of DH after tooth brushing over a period of eight weeks using AxioZoom. The null hypothesis of this study was that there would be no significant persistency differences between the desensitizing agents after eight weeks of tooth brushing.

MATERIALS AND METHODS

This study was approved by the Istanbul Medipol University Ethics Committee for the use and access of non-invasive clinical research (project no:192, 2016). Forty freshly extracted non-carious human first and second premolars were used for this study. The teeth were cleaned using an ultrasonic scaler and slurry of pumice and water with a slowly rotating rubber cup.

Desensitizing agents

Desensitizing agents (All-Bond Universal, Gluma Desensitizer, BisBlock) with different mechanisms of action were used in this study. The desensitizers were applied to the dentin according to the manufacturers' instructions; the details and application steps of these materials are shown in table 1.

Sample preparations

The roots of the teeth were cut approximately 2 mm below the cementoenamel junction using a diamond disc (Diamond Wafering Blade; Buehler, Lake Bluff, IL, U.S.A.) with a precision cutting machine (Isomet 1000, Buehler) under water cooling to attain two equal halves. Then, the samples were prepared to a size of 5 x 5 x 5-mm (height, width, and length, respectively). Thereafter, a standard smear layer was created on the dentin samples using a rotating polishing machine (MetaServ 250, Buehler) with 600-grit silicon carbide paper under constant water irrigation. The smear plugs were removed with 17% ethylenediamine tetra-acetic acid for two minutes and rinsed with distilled water. The dentin samples were examined under a stereomicroscope (Olympus SZ61, Munster, Germany) at ×30 magnification, and only samples with exposed dentin tubules were used. The exposed dentin surface of the samples was signed, and the dentin samples were randomly assigned to four treatment groups, with 10 specimens in each group (n = 10):

Table 1. The desensitizing agents, compositions, and application procedure used in the study according to manufactures' data sheets.

Desensitizing agents	Active components	Manufacturer	Application steps as recommended by the manufacturer
All Bond Universal	10 MDP, dimethacrylate resins, HEMA, ethanol, water, and initiators	Bisco Inc. Schamburg, USA	Apply two separate coats of all-bond universal, scrubbing the preparation with a microbrush for 10–15 s per coat. Evaporate excess solvent by thoroughly air-drying with an air syringe for at least 10 s. Light cure for 10 s.
Gluma Desensitizer	Glutaraldehyde (5%) and hydroxyethylmet hacrylate (HEMA, 35%)	Heraeus Kulzer, Hanau, Germany	Apply on dried dentin and leave for 30–60 s. Dry then spray with water
BisBlock	Oxalic acid, potassium salt, and water	Bisco Inc. Schamburg, USA	Etch for 15 s, rinse, and gently air-dry for two to three seconds. BisBlock applied and dwelled for 30 s. Apply for at least 30 s on the acid etched dentin, rinse, and leave moist for bonding.

1) No treatment was applied to the dentin surfaces; this group served as the negative control.
2) Two separate coats of All-Bond Universal were applied on the dentin surface with a microbrush for 10–15 s per coat by scrubbing. Then, the excess solvent was gently evaporated with an air syringe for at least 10 s and light-cured for 10 s.
3) The dentin surfaces were treated with Gluma Desensitizer for 30–60 s and gently air-dried for 5 s.
4) The dentin surfaces were etched for 15 s, rinsed, and dried for 2–3 s. BisBlock was applied for 30 s, then rinsed. Adhesive was applied to seal the crystals and tubules.

The desensitizing agents were applied following the manufacturers' instructions (table 1). All-Bond Universal and Gluma Desensitizer were mixed with a fluorescent agent (Rhodamine B; Sigma-Aldrich Corp., St. Louis, MO, U.S.A.) directly in the supplied bottle, whereas a One-step adhesive system was mixed with the dye in the BisBlock group to ensure fluorescent marking. The dye was added in advance to allow for complete dissolution with the tested concentration (0.16 mg/ml) [21].

For the test groups, the exposed surface of each specimen was brushed twice per day using a tooth-brushing machine (Oral-B Triumph 5000™, Procter Gamble, Cincinnati, OH) with a loading mass of 200 g, and 293 oscillations/rotations of brushing speed were applied with a slurry of fluoride-free toothpaste (Paradontax, GlaxoSmithKline, Istanbul, Turkey). The samples were then rinsed thoroughly in distilled water and put through imaging. For the duration of the experiment, the samples were stored in distillated water at 4°C in a dark environment.

Sample imaging

The fluorescently labeled test samples and negative control samples were imaged at each stage (baseline [before tooth brushing], first, second, sixth, and eighth weeks after toothbrush abrasion) using AxioZoom V16 Microscopy (Carl Zeiss, Oberkochen, Germany). All samples (test and control groups) were gently air-dried to remove the excess liquid at the surface and then placed on the AxioZoom stage. The surface images were captured with a PlanNeoFluar Z 1.0X/0.25 falling weight deflectometer (or FWD) 56 × 20 mm objective and an AxioCam HR R3 camera in conjunction with the Zen modules software (Zeiss Technology Ltd., Germany). Throughout the testing procedure, all samples were imaged at the same location with the same objective and exposure time.

Processing of images

The mean florescence intensity (MFI; pixel) of the dentin surface in each image was quantitatively determined using the Image J 1X Fiji program (National Institutes of Health, U.S.A.), and an intensity (pixel) difference analysis was performed after the weekly average value of each sample was determined.

Statistical analysis

The statistical analysis was performed using the IBM SPSS Statistics 22 (IBM SPSS, Turkey) program. A Shapiro–Wilks normality test was performed to assess the normal distribution of the data. For intergroup comparisons, a Kruskal–Wallis test and post-hoc analysis with a Mann–Whitney U test were used. Friedman and Wilcoxon signed-rank tests were used for the intragroup comparisons of the parameters. The significance was evaluated at the level of $p < 0.05$.

RESULTS

The MFI values of the dentin surfaces and statistically significant differences between the time stages (baseline, baseline, 1st, 2nd, 6th, and 8th weeks) by group are presented in table 2.

For the All-Bond Universal group, the findings of the Friedman tests demonstrated that the MFI showed a statistically significant decrease at all time stages (first, second, sixth, and eighth weeks) compared to the baseline ($p < 0.05$). There were significant differences between the first week and sixth and eighth weeks ($p < 0.05$), second week and sixth and eighth weeks ($p < 0.05$), and sixth week and eighth week ($p < 0.05$). There was no statistically significant difference between the first and second weeks ($p > 0.05$). Regarding the comparison of all-time stages for Gluma Desensitizer, there was a statistically significant decrease in the MFI between the baseline and the other time stages ($p < 0.05$). There were significant differences between the first week and sixth and eighth weeks ($p < 0.05$) and the second and eighth weeks ($p < 0.05$), but the differences between the first and second, second and sixth, and sixth and eight weeks were not statistically significant ($p > 0.05$).

In the BisBlock group, the findings of the Friedman tests demonstrated that the MFI showed a statistically significant decrease at all time stages (first, second, sixth, and eighth weeks) compared to the baseline ($p < 0.05$). Furthermore, the findings of the Wilcoxon signed-rank tests demonstrated a statistical decrease between the first week and second, sixth, and eighth weeks ($p < 0.05$), second week and sixth and eighth weeks ($p < 0.05$), and sixth week and eighth week ($p < 0.05$). The BisBlock MFI decrease showed consistency in all time stages.When evaluating the percent decrease in MFI between the time stages for the All-Bond Universal group, the highest percent decrease was observed between the sixth and eighth weeks (40%), whereas it was lowest between the

Table 2. MFI Values, standard deviations (SD) of groups and significant differences between them.

Groups	All Bond	Gluma	Bis-Block	Neg. Control	†p
	Mean±SD(median)	Mean±SD(median)	Mean±SD(median)	Mean±SD(median)	
Baseline	13.73±2.44 (14.1)	14±5.42 (13.8)	33.67±17.43 (31.4)	1.35±0.12 (1.4)	**0.001***
1st week	10.6±2.07 (10.8)	11.08±3.69 (11.7)	17.97±5.21 (17.9)	1.35±0.12 (1.4)	**0.001***
2nd week	8.54±2.56 (7.8)	8.96±2.03 (9.2)	14.27±3.27 (15.8)	1.35±0.12 (1.4)	**0.001***
6th week	5.6±1.89 (5.9)	6.89±3.31 (6.8)	11.8±3.61 (10.9)	1.35±0.12 (1.4)	**0.001***
8th week	3.57±1.37 (3.5)	3.84±1.54 (4.6)	8.18±2.26 (8.3)	1.35±0.12 (1.4)	**0.001***
§p	**0.001***	**0.001***	**0.001***	-	-
Baseline 1st wk ¶p	0.018*	0.018*	0.028*	-	-
Baseline 2nd wk ¶p	0.018*	0.043*	0.018*	-	-
Baseline 6th wk ¶p	0.018*	0.018*	0.018*	-	-
Baseline 8th wk ¶p	0.018*	0.018*	0.018*	-	-
1st wk – 2nd wk ¶p	0.128	0.176	0.018*	-	-
1st wk – 6th wk ¶p	0.018*	0.018*	0.018*	-	-
1st wk – 8th wk ¶p	0.018*	0.018*	0.018*	-	-
2nd wk – 6th wk ¶p	0.028*	0.128	0.018*	-	-
2nd wk – 8th wk ¶p	0.018*	0.018*	0.018*	-	-
6th wk – 8th wk ¶p	0.018*	0.091	0.018*	-	-

† Kruskal Wallis test, § Friedman test, ¶ Wilcoxon sign test,* $p < 0.05$.

baseline and first week (23%). Gluma Desensitizer displayed the highest percent decrease between the second and sixth weeks (35%), whereas the lowest was between the baseline and first week (15%). For BisBlock, the highest percent decrease was seen between the baseline and first week (43%), whereas the lowest was between the first and second weeks (11%). Overall, the percent decreases between the baseline and eighth week for the All-Bond Universal, Gluma Desensitizer, and BisBlock groups were 75%, 66%, and 74%, respectively.

The MFI values as determined by the AxioZoom of all groups at each time stage are shown in figure 1.

According to the AxioZoom values for the baseline, the negative control group showed less intensity compared to all dentin desensitizing agent groups. Furthermore, the difference was statistically significant (p: 0.002; $p < 0.05$). BisBlock displayed statistically significant higher MFI values than All-Bond Universal (p: 0.002) and Gluma Desensitizer (p: 0.006; $p < 0.05$). However, there were no significant differences between the All-Bond Universal and Gluma Desensitizer groups (p: 0.749; $p > 0.05$).

For the first, second, sixth, and eighth weeks, similar to the baseline results, BisBlock showed higher MFI values than the All-Bond and Gluma groups, and the differences were statistically significant ($p < 0.05$). However, there were no statistically significant differences in the MFI between Gluma Desensitizer and All-Bond Universal ($p > 0.05$). These results are reflected in both table 2 and figure 1.

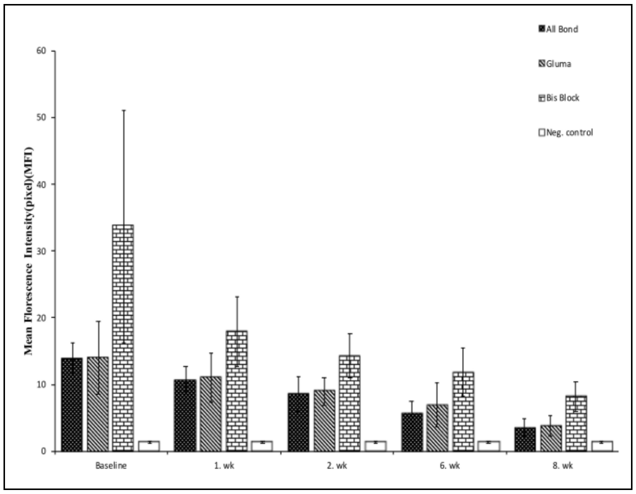

Fig. 1. MFI values of dentin samples of all groups at each time stages.

Figure 2 shows the AxioZoom images taken of the surfaces of the dentine for the control and test groups (baseline, first, second, sixth, and eighth weeks).

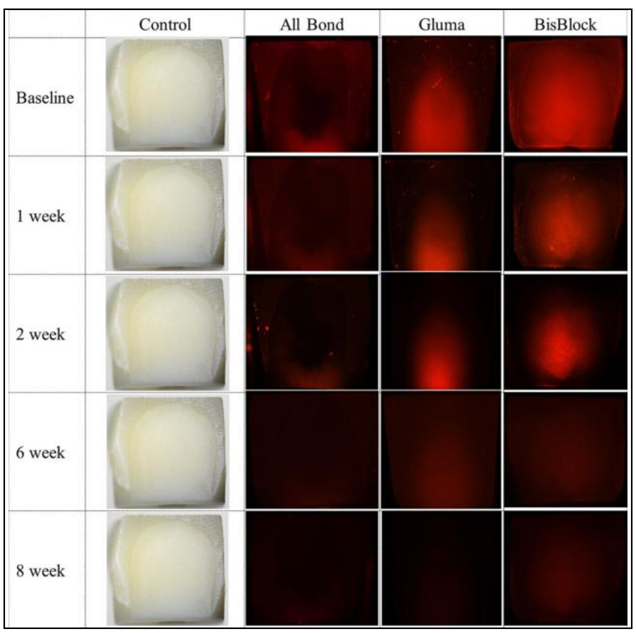

Fig. 2. AxioZoom stereo zoom microscope images of the dentine surfaces at each experimental stages of all groups.

In figure 2, a reduction of fluorescence (Rhodamine B) can be clearly observed from the baseline to the eighth week for all dentin desensitizers. However, BisBlock presented high fluorescence immersion compared to All-Bond Universal and Gluma Desensitizer, and this difference was significant ($p < 0.05$). The control tooth with no dye immersion was observed at the dentin and enamel.

DISCUSSION

This comparative *in vitro* study investigated the eight-week abrasion effect of tooth brushing on dentin desensitizers, which have important roles in preventing DH. To the best of the author's knowledge, this is the first study to investigate the persistency of dentin desensitizing agents against tooth brushing using AxioZoom, which supplies quantitative and objective results.

This experimental study was designed to demonstrate the usual interrelation between the clinical treatment of DH and daily tooth-brushing habits. In this study, the MFI values as well as wear rates of the dentin adhesives/desensitizers decreased consistently from the baseline to eighth week in all groups as a demonstration of the abrasive effect of tooth brushing on professionally applied dentin desensitizing agents used in the reduction of DH.

According to our results, as the brushing time increased, the amount of applied dentin desensitizing agents decreased for all agents' groups. The eight-week persistency after tooth brushing abrasion for BisBlock in the study was statistically higher than that of All-Bond Universal and Gluma Desensitizer from baseline to the eighth week, whereas there was no significant difference between All-Bond Universal and Gluma Desensitizer when their MFI values were compared. Therefore, the null hypothesis that there would be no significant persistency differences between the desensitizing agents after eight weeks of tooth brushing was rejected.

Tooth brushing is an obligation for oral health maintenance and is generally considered safe. Clinical studies have shown that there is a strong correlation between tooth-brushing force, brush type, and tooth wear loss on both enamel and dentin [22-24].

Dentin hypersensitivity is a frequent problem that irritates patients and has a negative effect on their quality of life.

Although the treatment of DH comprises different kinds of methods, dentin desensitizers such as BisBlock, All-Bond Universal, and Gluma Desensitizer can be used to cover the tubules with an acceptable level of desensitization, as in this study.

All-Bond Universal is a multi-mode ethanol/water-based single-bottle adhesive that combines the acid, primer, and bond in one bottle and may be used as a total-etch, self-etch, or selective-etch technique. This adhesive incorporates 10-MDP as a functional monomer in its composition, in which the phosphate group of this monomer interacts with the calcium in hydroxyapatite and contributes to the long-term durable chemical adhesion at the resin–dentin interface [25]. In the present study, All-Bond Universal was used as a desensitizer agent in self-etch mode and showed the highest percentage of wear (75%) with the significantly lowest MFI values after tooth brushing compared to BisBlock over the course of the study period. In addition, according to the AxioZoom findings, this adhesive/desensitizer almost completely disappeared on the dentin surface after two weeks of tooth brushing, which corroborated the MFI findings. One possible reason for this finding might have been due to the high pH (3.2) of the adhesive system since this acidity is not strong enough to create higher bond strength on the dentin surface. It has been previously reported that an acid-etching application prior to universal adhesive on the dentin surface improves the bond strength [26, 27]. Nevertheless, All-Bond Universal was used in self-etch mode in the present study and might have created a relatively weaker bond strength on the dentin surface, which may have affected the wear resistance to tooth-brushing strokes compared to BisBlock. It has been stated that the residual water in water-, acetone-, and alcohol-based primers cannot completely evaporate on dentin surfaces because of the high surface tension of water and that this remaining water disrupts the complete polymerization of adhesives and results in water trees within the adhesive layer [28, 29]. Fu et al. [30] reported that dentin adhesives/desensitizers could block fluid permeation through the dentin tubules when applied on dentin surfaces but cannot completely stop the water/ion diffusion into and out of the tubules, which could result in the degradation of the adhesive material mechanical properties over time. Considering these reports, the adhesive layer of All-Bond Universal on top of the dentin surface when used as a desensitizer might have showed hydrolysis when stored in water, and the mechanical properties of this adhesive might have become more susceptible to tooth-brushing abrasion compared to the other desensitizers over the course of the study period. Furthermore, according to the manufacturer's information, this adhesive system does not contain silica filler particles in its composition, which can increase the film thickness; hence, it did not perform better resistance to tooth-brushing abrasion.

Gluma Desensitizer is a biological fixative containing an aqueous solution of 5% glutaraldehyde and 35% 2-hydroxyethyl methacrylate (HEMA). It has been reported that Gluma Desensitizer applied onto hypersensitive dentin surfaces forms deposits by the reaction of glutaraldehyde and HEMA between the dentinal fluid proteins, thereby resulting in occlusion of open peripheral dentin tubules [31]. In the present study, Gluma Desensitizer exhibited the lowest wear due to tooth-brushing abrasion (66%) among the tested desensitizers and demonstrated the lowest MFI values compared to BisBlock, with no significant difference from All-Bond Universal. Although there were no significant differences in the MFI values between Gluma Desensitizer and All-Bond Universal, the AxioZoom images clearly showed that Gluma Desensitizer showed visible persistency to tooth-brushing abrasion for up to six weeks. Considering these results, two coats of Gluma Desensitizer precipitated glutaraldehyde and HEMA deep within the dentinal tubules and may have maintained the persistency to tooth-brushing abrasion under the conditions of this study. Choi et al. [32] reported that multiple layers of protein septa precipitated deep within the tubules and maintained the sealing ability of Gluma after acid erosion or tooth-brushing abrasion. However, a visible covering layer could not be observed at the eight-week period, and it was assumed that this material could not withstand tooth-brushing abrasion and was completely removed from the surface.

BisBlock is an oxalate-containing desensitizer consisting of low concentrations of oxalic acid that obstructs the dentin tubules. The application of oxalate to exposed dentin surfaces results in occlusion of open dentin tubules; it reacts with calcium ions on the dentin to form insoluble calcium oxalate crystals [33, 34]. Contrary to the application procedure of other oxalate-containing desensitizers, BisBlock incorporates the total-etching procedure prior to the oxalate and bonding agent application, and this specific application procedure could provide long-lasting effects as compared to other oxalate-containing desensitizers [35, 36]. In the present study, although BisBlock showed quite a bit of wear in percentage (74%) between the baseline and eighth week, it demonstrated the highest MFI over the course of the study period compared to the other desensitizers. It has been reported that acid etching of the dentin surface decreases calcium ions and leads to increased penetration of oxalate ions into the dentin tubules, which forms calcium oxalate crystals deep into the dentin. Forming these crystals decreases the dentin permeability and does not affect the bonding performance of adhesive systems [37]. Likewise, Yiu et al. [38] also reported that oxalate-containing desensitizers do not negatively affect the bonding performance of selected total-etch adhesives. Therefore, the application of two coats of a total-etch adhesive may have formed a more resistant adhesive layer to the frequency of tooth brushing on the dentin surface and resulted in the highest MFI for BisBlock among the tested desensitizers at each of the evaluation periods. It should be remembered that the effects of oxalates in reducing DH decrease over time [39], even if two coats of dentin adhesive are applied, as shown in the present study. However, a DH evaluation was not performed in this study.

To the best of the authors' knowledge, there have been no reported investigations specifically on the persistency of dentin desensitizers to tooth-brushing abrasion. In addition, in many of the dentin adhesive studies using dentin desensitizers, different evaluation periods have been used to evaluate the effectiveness. However, in the present study, an eight-week study period was chosen in a randomized clinical study in which two professional desensitizing agents were used [40].

There were some limitations in the present study. Based on the results of the study, the persistency of the dentin adhesives/desensitizers was evaluated using AxioZoom after tooth-brushing abrasion, but the occluding effect of these materials was not evaluated. For this purpose, an alternative method such as SEM analysis could be used to illustrate the occluding effect of these desensitizing materials; unfortunately, this was not available for this study. It should be emphasized that the laboratory conditions differed from oral complex conditions, and the erosive/abrasive condition, temperature, protective effect of the pellicle, and salivary buffer capacity might have directly affected these results. Therefore, further *in vivo* and/or *in situ* research is needed to understand the persistency as well as occluding effect of these dentin adhesives/desensitizers for more valid and reliable results.

CONCLUSIONS

Within the limitations of the present study, all dentin adhesives/desensitizers showed a gradual decrease in MFI values and surface wear after tooth-brushing abrasion over the study period. The different MFI values confirmed that the dentin adhesives/desensitizers used under tooth-brushing abrasion varied according to product type. Although BisBlock had one of the highest MFI values represented at the end of the study period and may be considered a resistant treatment for tooth-brushing abrasion, all materials should be reapplied after eight weeks to produce a sealing ability to maintain the reduced DH.

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CONFLICTS OF INTEREST

The authors declare no conflict of interest.

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