

## EFFECT OF FLUORIDE-RELEASING MATERIALS ON THE PREVENTION OF ENAMEL EROSION: A MICROHARDNESS AND SCANNING ELECTRON MICROSCOPIC EVALUATION

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**ABSTRACT:** The aim of the study was to evaluate the preventive effect of different bioactive restorative materials on the neighbouring enamel under erosive conditions. Fifty-two intact human incisors were collected. Standard Class V cavities were prepared and the specimens were randomly divided into 4 groups (n=13) according to the restorative materials used: (i) control group: composite resin (CR) group (Harmonize/KERR), (ii) resin modified glass ionomer (RMGIC) group (RivaLight Cure/SDI), (iii) glass carbomer (GC) group (GlassFill/GCP-Dental), and (iv) high viscosity glass ionomer (HVGIC) group (EQUIA/GC). After polishing with aluminum oxide discs, the microhardness values of the restorative materials and the neighbouring enamel were measured with a Vickers hardness device. The specimens were then subjected to an erosive procedure. The final microhardness measurements were performed and the data were subjected to statistical analysis. The surface topographies of the specimens from each group were evaluated, before and after erosive challenge, with a scanning electron microscope (SEM). Significant decreases were observed in the microhardness values of the neighbouring enamel for all the materials after the erosive challenge when compared to the baseline. However, the decreases in the microhardness of the neighbouring enamel were significantly lower in the GC and HVGIC groups than in the RMGIC and CR groups. The SEM findings were in accordance with the microhardness test results. The bioactive HVGIC and GC materials might be better tooth-colored restoration options for preventing neighbouring enamel demineralization in patients who are at risk of dental erosion.

Keywords: Bioactive materials; Dental erosion; Fluoride release; Minimal invasive dentistry; Remineralization.

### INTRODUCTION

Dental erosion, or as is more current biocorrosion, is defined as the irreversible loss of tooth tissue due to a chemical dissolution process caused by the exposition of intrinsic or extrinsic acids and has become an increasing oral health problem in recent decades.<sup>1,2</sup> The primary exogenous sources of acids are usually drinks or food and, as the lifestyle and dietary habits change, the consumption of beverages, like soft drinks, energy drinks, and coffee, has increased dramatically.<sup>3,4</sup> Accordingly, with the increased exposure to an acidic environment, erosion has become the most common cause of tooth wear.<sup>5</sup>

The struggle with erosion is usually carried out first as prevention and in later phases, after tissue loss has occurred, as restoration. The prevention of erosion may be provided by changing dietary habits, topical fluoride (F), or adhesive agent applications in the early phases.<sup>6</sup> However, when a vast amount of hard tissue loss occurs, erosive lesions may cause sensitivity and also esthetic and/or functional problems.<sup>7</sup> In this situation, to rehabilitate the tooth contour and to prevent further substance loss, the restoration of these lesions is necessary.<sup>8</sup> Although, ideally, restorative procedures would be applied after the elimination of the etiological

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factors that induced the erosion, it is a fact of life that patients only rarely give up their erosive habits.<sup>9</sup> Consequently, the erosive conditions usually persist even if the erosive lesions are restored. As acidic conditions have the potential to impair the quality of the restorations, it is vital to know the behavior of dental materials under erosive challenge for predicting the longevity of restorations and their effects on the neighbouring hard tooth tissues.

The “minimal invasive approach” is based upon the philosophy of not only restoring the teeth, but also of preserving the sound tooth tissues from further loss.<sup>10</sup> In this context, chasing the idea of a material that can both restore the erosive lesions and strengthen the tooth tissue against acidic attacks is the best option. By having the capability to elicit a response in living tissues of regeneration and repair, such as inducing the formation of hydroxyapatite, the use of bioactive materials is one of the favourite approaches in conservative dentistry.<sup>11</sup> Glass ionomer cements, which are members of the bioactive dental materials family, fit in with this concept due to their remineralizing abilities. However, because of their relatively high solubility, they are more vulnerable to acid attack than other direct restorative materials. In an *in vitro* study, Yu et al. reported that glass ionomer cements showed a higher wear rate than compomer and composite resins under erosive conditions.<sup>12</sup> However, in recent years, with the developments in glass ionomer materials, more durable restoratives have been launched on the market. Among these newer materials, glass carbomer cement, was launched with the claim of inducing “new dentin production” in addition to the benefit of F release. Another new reinforced glass ionomer material (Equia), is a restorative system combining a high viscosity glass ionomer and a nanofilled resin coating agent. This restorative system, with increased wear resistance and F release, has been shown to be highly successful in clinical trials.<sup>13-15</sup>

There is not sufficient information about the protective effect of the glass ionomer materials on human enamel, under erosive conditions. Francisconi et al.<sup>16</sup> reported that no significant differences were observed in the protective effect of glass ionomers (resin-modified glass ionomer and conventional glass ionomer) on the neighbouring enamel (wear and percentage of surface microhardness change) compared the protective effect of resin-composite and amalgam. However, research is still continuing of the protective effects of novel glass ionomer materials, like glass carbomer and high viscosity glass ionomers. Therefore the aim of the present study was to evaluate the effects of the currently available bioactive restorative materials on the neighbouring enamel, by investigating the microhardness and surface topography changes after erosive pH cycling.

The null hypothesis being tested was there would be no difference in the microhardness of the enamel tissue neighbouring the tested restorative materials after erosive cycling.

#### MATERIAL AND METHODS

The study was approved by the local ethics committee of Hacettepe University. Fifty-two intact human mandibular incisors, extracted for periodontal reasons, were collected. The remained soft tissue, calculus, and plaque were removed from the tooth surfaces by hand instruments, a rubber cap, and a pumice slurry. The cleaned teeth were examined under a stereomicroscope (Dino-Lite Pro, Anmo Electronics Corp, Taiwan) at 10× magnification to discard those with caries, cracks, and

developmental and structural defects in the enamel structure. The teeth, that were found suitable, were stored in 0.5% chloramine-T solution for disinfection for one week and then stored in distilled water until the experiment at room temperature. For the sample preparation, the teeth were embedded into acrylic blocks with the buccal surfaces facing upwards. The surfaces of the specimens were ground using 400, 600, and 800 grid SiC papers and then standard class V preparations with the dimensions of 3×3×1.5 mm were performed on the gingival third of each specimen. The specimens were randomly divided into 4 groups and each group was restored with one of the test materials according to manufacturer’s instructions (Table 1):

**Table 1. Materials used in the study**

Material	Manufacturer	Composition
Glass carbomer (GCP Glass Fill)	GCP Dental/Leiden, Netherlands	Fluoroaluminosilicate glass, apatite, polyacids
GCP gloss	GCP Dental/Leiden, Netherlands	Modified polysiloxanes
Equia	GC Corp GC Europe Leuven, Belgium	Strontium fluoroaluminosilicate glass, aqueous polyacrylic acid
Equia Coat	GC Corp GC Europe Leuven, Belgium	Methyl methacrylate, colloidal silica, camphorquinone, urethane methacrylate, phosphoric ester monomer
Resin modified glass ionomer (Riva Light Cure)	SDI Ltd./ Victoria, Australia	Fluoroaluminosilicate glass, polyacrylic acid, tartaric acid, 2-hydroxyethyl methacrylate, Dimethacrylate cross-linker, acidic monomer
OptiBond FL	KERR, Orange, CA, USA	Acid: 37.5% phosphoric acid. Primer: HEMA, 2-[2-(methacryloxy)ethoxycarbonyl] benzoic acid, GPDM, ethanol, water, photo-initiator Bond: HEMA, 3-trimethoxysilylpropyl methacrylate, 2-hydroxy-1,3-propanediyl bismethacrylate, alkaline fluorosilicates (Na), photoinitiator
Nanohybrid universal composite (Hamonize)	KERR, Orange, CA, USA	2 2'-ethylenedioxyethyl dimethacrylate, 3-trimethoxysilylpropyl methacrylate, Poly (oxy-1,2-ethanediyl), α, α'- [(1-methylethylidene) di-4,1-phenylene] bis [endi - [(2-methyl-1-oxo-2-propen-1-yl) oxy].

*Composite resin (CR) group:* The enamel and dentin surfaces were etched for 15 sec with 37.5% orthophosphoric acid (Gel Etchant, KERR, Orange, CA, USA) and rinsed for 30 sec. After gently air drying, the adhesive system (Optibond FL; KERR, Orange, CA, USA) consisting of Optibond FL Prime (15 sec) and Optibond FL

Adhesive (15 sec) was applied, gently air dried (5 sec) and light cured (20 sec). For photoactivation, a LED curing unit (Henry Schein, HS-LED Light 1200, NY, USA) was used in all the groups. The preparations were filled with a nanohybrid universal composite resin (Harmonize, KERR, Orange, CA, USA) and covered with a polyester strip. A glass slide was placed on the strip with light pressure allowing the excess material to extrude. Then the glass slide was removed and the material was light cured for 40 sec under a polyester strip. The restorations were finished and polished using polishing discs (Optidisc, KERR, Orange, CA, USA).

*Resin modified glass ionomer (RMGIC) group:* A resin modified glass ionomer (Riva Light Cure, SDI, SDI Ltd./ Victoria, Australia), was mixed according to the manufacturer's instructions and placed on the preparations with a hand instrument. The material was light cured for 20 sec as described for the CR Group. The restorations were polished using the same polishing discs as mentioned above.

*Glass carbomer (GC) group:* Prior to mixing, each glass carbomer capsule was inserted into a universal capsule gun (GCP Dental, Leiden, Netherlands) and standardized according to the manufacturer's instructions. Each capsule was mixed for 7 sec in a high frequency amalgamator (Carbomix, GCP Dental, Leiden, Netherlands). The pin from the nozzle was removed after mixing and it was inserted into the capsule gun and the lever was pulled twice to prime the material which was then extruded onto the preparation. The excess material was removed as described for the previous groups and the material was thermo-cured for 90 sec using an LED unit with an output of 1,400 mW/cm<sup>2</sup> (CarboLED, GCP Dental, Leiden, Netherlands). The restoration was covered with GCP gloss, light-cured, and polished using polishing discs. After polishing the coating agent was reapplied.

*High viscosity glass ionomer (HVGIC) Group:* Before application each capsule of the high viscosity glass ionomer (Equia, GC Europe, Leuven, Belgium) was shaken and the plunger was depressed. After that, the capsule was inserted on a capsule applier and clicked once to activate. The capsule was then inserted into the mixer and mixed for 10 sec. After mixing, the capsule was re-inserted into the capsule applier and the material was applied on the preparations as previously described. The excess material was removed and the restoration covered with Equia Coat (Equia, GC Europe, Leuven, Belgium). After polishing with polishing discs, the coating agent was reapplied.

*Microhardness measurements:* Following the restorative procedures, the specimens were stored in distilled water for 24 hr. One specimen from each group was kept as a the control for further evaluation with a scanning electron microscopic (SEM). The remaining 12 specimens from each group were air dried for 30 sec and the initial microhardness values of the restorative materials and the neighbouring enamel tissue were measured using a Vickers microhardness tester (Shimadzu, Tokyo, Japan). The enamel measurements were performed at the distance of 50 µm from the external restoration margin.

A vertical load of 50 g was applied for 15 sec, and the indentation length was photographed and measured using a microscope and software. The test was performed at 3 different points on each specimen and the mean value was calculated for each specimen.

*Erosive challenge:* After the microhardness values were measured, the specimens were subjected to an erosive cycling model. Following this model, the specimens were immersed in an acidic drink (cola) for 15 min., 3 times a day, for 7 days, washed with water for 15–20 sec, and kept in artificial saliva in the intervals. The final microhardness measurements were performed. The data were subjected to statistical analysis with the Paired samples t-test, One-way Anova, and the Tamhane tests ( $p=0.05$ ).

*SEM evaluation:* The surface topography of the previously reserved control specimens and one randomly selected specimen from each group subjected to the erosive challenge was evaluated under a SEM (FEI, Nova NanoSEM 430, Czech Republic) to monitor the changes on the enamel and the restorative materials. For this purpose, the representative specimens were rinsed with distilled water, mounted on metal stubs, sputter coated with gold under pressure, and examined with 250× and 500× magnifications.

## RESULTS

*Microhardness results for materials:* All materials showed significantly different microhardness values from each other at baseline. The baseline measurements showed that the HVGIC group had the highest microhardness values followed by the RMGIC, CR, and GC groups, respectively ( $p<0.05$ ). After the erosive cycling, all the materials showed lower microhardness values than their baseline values ( $p<0.05$ ). Following the erosive challenge, the RMGIC and HVGIC groups showed higher microhardness values than the CR and GC groups ( $p<0.05$ ). The differences between the microhardness values of the RMGIC group versus the HVGIC group, and the CR group versus the GC group, were not significant ( $p>0.05$ ) (Table 2).

**Table 2.** Microhardness of the enamel and the restorative material before and after the erosive cycling. (Values are mean±standard deviation)

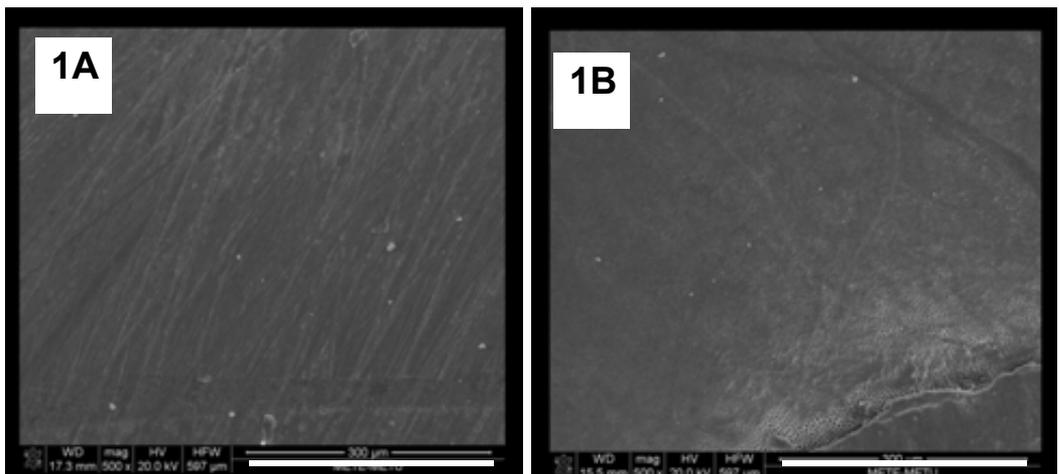
Group	Time period			
	Before erosive cycling		After erosive cycling	
	Enamel (Mean±SD)	Restorative material (Mean±SD)	Enamel (Mean±SD)	Restorative material (Mean±SD)
CR	403.17±32.26	84.25±7.97	195.56±31.94 <sup>a</sup>	55.26±8.39
RMGIC	400.48±70.67	100.56±5.89	192.88±79.22 <sup>a</sup>	72.74±26.37
GC	410.53±71.37	55.76±7.46	281.69±75.02 <sup>b</sup>	48.45±20.38
HVGIC	409.45±64.81	119.86±17.74	272.51±67.15 <sup>b</sup>	72.11±10.57

Different superscripted lower case letters within the same column indicate a statistically significant difference ( $p<0.05$ ). The initial microhardness values of the enamel in all the groups were standard. After the erosive cycling all the materials and the neighbouring enamel showed a decrease in the microhardness values. The microhardness loss in the neighbouring enamel after the erosive cycle was significantly higher in the samples which were restored with RMGIC and CR compared to GC and HVGIC. The differences in the microhardness loss between the CR and the RMGIC groups and between the GC and HVGIC groups were not statistically significant. This signifies that the GC and HVGIC restorations have shown a protective effect on the enamel surrounding the restoration.

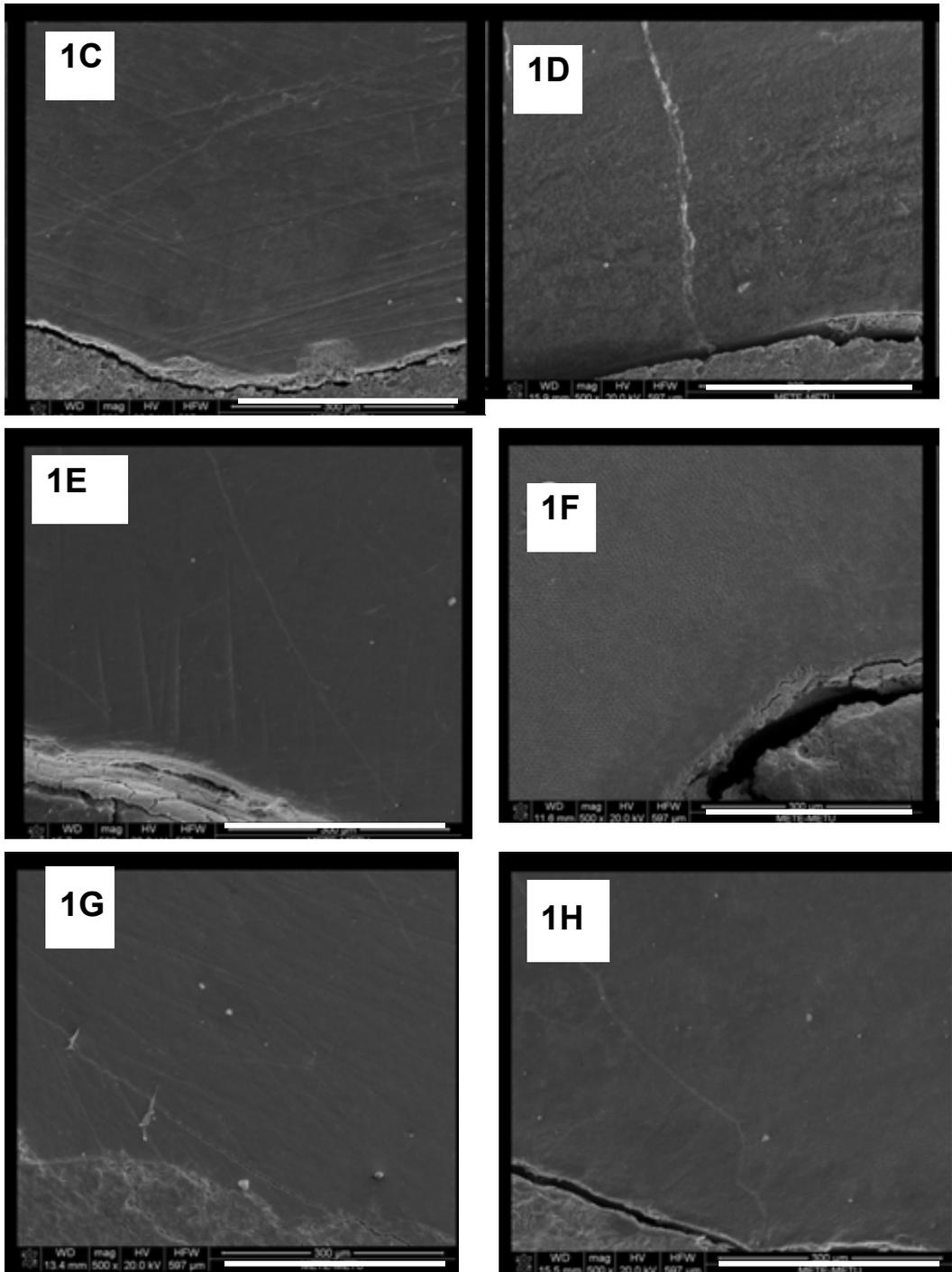
*Microhardness results for enamel:* At baseline, the microhardness values of neighbouring enamel was statistically homogeneous for all the test groups ( $p>0.05$ ). However, significant decreases were observed in the microhardness values of the enamel for all the groups after the erosive challenge compared to the baseline values ( $p<0.05$ ). The post erosive microhardness values of the neighboring enamel were significantly higher in the HVGIC and GC groups compared to the CR and RMGIC groups ( $p<0.05$ ) (Table 2).

*SEM evaluations:* Baseline SEM evaluations of the tested restorative materials exhibited apparent variations from each other. A well-integrated adhesive layer and a durable interaction between the composite resin, the adhesive resin, and the enamel tissue was seen in the CR group. Mild crack lines and a porous surface topography were detected in the RMGIC group. Additionally, a loose gap formation was observed at the enamel-restorative material interface. In the GC group, large crack lines were detected within the restorative material and a vulnerable interaction between the enamel and the restoration was noted. The HVGIC group exhibited slight crack lines within the material and a partial and mild gap formation was observed at the interface.

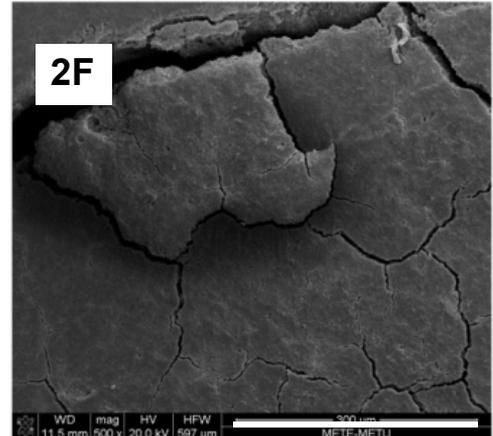
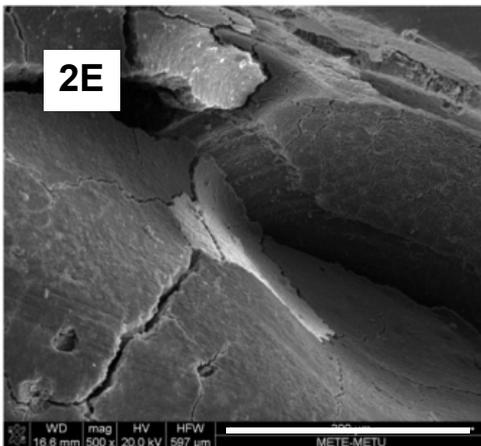
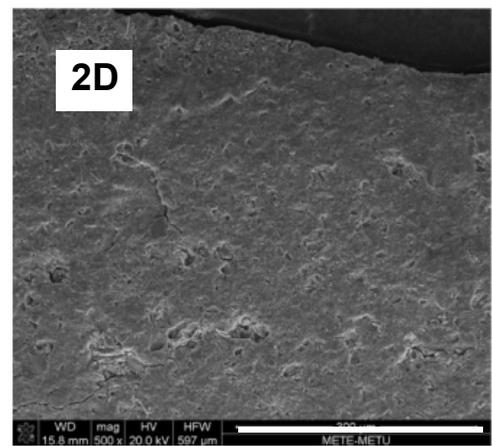
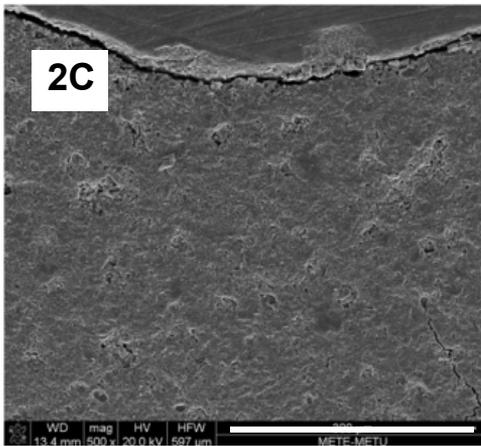
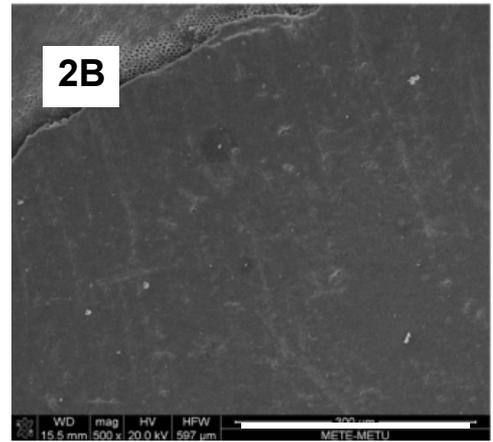
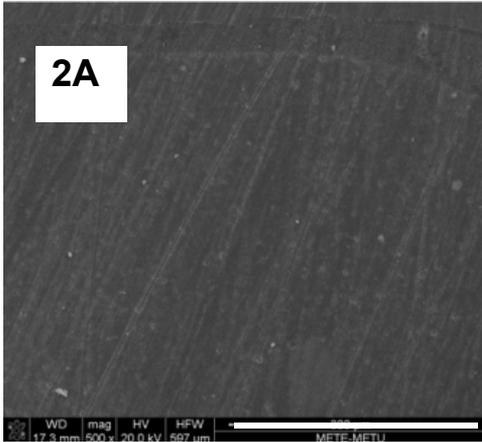
After the erosive cycling, a tenuous spacing between the resin composite and the tooth tissue was observed in the CR group. The RMGIC, HVGIC, and GC groups all showed gap formation at the tooth and material interface. However, in the GC group, a significant spacing between the tooth tissue and material was observed along with distinct crack lines across the material. Although in the RMGIC and HVGIC groups there was a slightly increased porosity observed compared to the baseline, the crack lines and gap formation was not as radical as with the GC group. The tooth tissue surrounding the CR showed a severe demineralization following the erosive procedure while the tissue surrounding the RMGIC seemed to have less demineralization than with the CR group. The GC and HVGIC groups showed the most protective effect against erosion. However, the protected area in the GC group seemed to be a limited zone around the restoration whereas in the HVGIC group the protective effect was broader (Figures 1A–H and 2A–H).



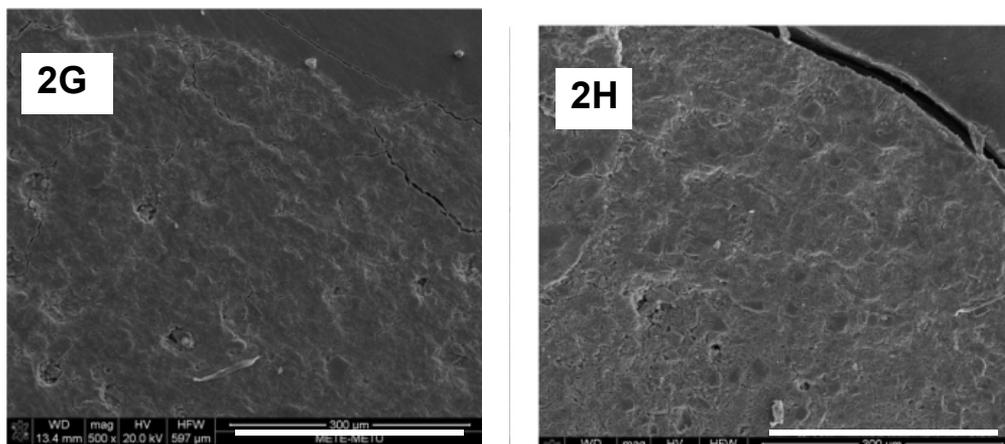
**Figures 1A and 1B.** The SEM images of the neighbouring enamel tissue of the tested materials before and after the erosive challenge (magnification 500×, marker length=300 µm). 1A: Neighbouring enamel tissue of CR at baseline before the erosive challenge (control); 1B: Neighbouring enamel tissue of CR after the erosive challenge.



**Figures 1C–H.** The SEM images of neighbouring enamel tissue of the tested materials before and after the erosive challenge (magnification 500×, marker length=300 µm). 1C: Neighbouring enamel tissue of RMGIC at baseline before the erosive challenge (control); 1D: Neighbouring enamel tissue of RMGIC after the erosive challenge; 1E: Neighbouring enamel tissue of GC at baseline before the erosive challenge (control); 1F: Neighbouring enamel tissue of GC after the erosive challenge; 1G: Neighbouring enamel tissue of HVGIC at baseline before the erosive challenge (control); and 1H: Neighbouring enamel tissue of HVGIC after the erosive challenge



**Figures 2A–F.** The SEM images of the tested materials before and after the erosive challenge (magnification 500×, marker length=300 µm). 2A: CR material at baseline before the erosive challenge (control); 2B: CR material after the erosive challenge (control); 2C: RMGIC material at baseline before the erosive challenge (control); 2D: RMGIC material after erosive challenge; 2E: GC material at baseline before the erosive challenge (control); and 2F: GC material after the erosive challenge.



**Figures 2G and 2H.** The SEM images of the tested materials before and after the erosive challenge (magnification 500 $\times$ , marker length=300  $\mu$ m). 2G: HVGIC material at baseline before the erosive challenge (control); and 2H: HVGIC material after the erosive challenge.

## DISCUSSION

Today, the increased prevalence of dental erosion hints at the necessity of understanding the behaviour of dental materials and also their interactions with the surrounding tooth tissue, under erosive circumstances. There are various methods to evaluate the degradation of the material or the mineral loss in dental tissue such as the measurement of microhardness,<sup>17</sup> weight change,<sup>18</sup> and surface roughness.<sup>17</sup> In the current study, the microhardness test was used for evaluating the alterations in the bioactive restorative materials and their preventive effects on the neighbouring enamel tissue under erosive circumstances.

The findings of this study revealed that all the tested materials and the neighbouring enamel tissues showed a decrease in the Vickers hardness values after the erosive challenge. However, the groups restored with GC cement and HVGIC, showed a significantly lower decrease in the microhardness values compared to the groups restored with RMGIC and CR. Therefore the null hypothesis, that there would be no difference in the microhardness of the enamel tissue neighbouring the tested restorative materials after erosive cycling, has to be rejected. In contrast to the current results, a previous study by Salas et al.,<sup>19</sup> reported that glass ionomers showed no protective effect on the enamel after erosion. Likewise, an *in situ* study by Rios et al. reported that GIC's showed no protective effect over enamel under erosive challenge.<sup>20</sup> The glass carbomer cement used in this study, is a novel glass ionomer based material with enhanced bioactivity and hydroxyapatite fillers that can form an enamel like structure on the tooth-material interface.<sup>21</sup> The nanohydroxyapatite and nanofluoroapatite particles in the glass carbomer cement, allegedly gained this material a powerful remineralizing ability and also the capability of inducing the formation of a dentin/enamel-like tissue. This may explain the observed protective effect provided by the glass carbomer in the present study.

In this study, a protective effect was also observed on the neighbouring enamel with HVGIC- Equia. In an *in vitro* study, evaluating the F release of different glass ionomer based materials, high viscosity GIC-Equia, was reported by Lopes et al. to

show a higher F release than resin modified GIC or glass carbomer cement.<sup>22</sup> Therefore, the higher F release rate of Equia might be responsible for this protective effect, since it would lead to a higher resistance to acidic dissolution.

In the present study the SEM findings pointed to the protective effect of HVGIC being expanded to a wider area in contrast to the situation with GC which affected only a narrow zone surrounding the restoration. Bueno et al.<sup>23</sup> reported that the acid erosion has a positive correlation with the F release. Since greater amounts of dissolution from the cement may lead to a higher F release, it may be hypothesized that the low solubility of glass carbomer, as presented in a previous study,<sup>24</sup> may explain the limited anti-erosive effect of this material. It may be speculated that a higher solubility rate would result in a higher F ion release which would have the potential to create a protective effect over a larger area. In keeping with its lower solubility, the GC material tested in the present study revealed that its protection zone on the tooth surface was limited to that area that it directly contacted.

In the current study, the tested HVGIC showed the highest microhardness values at baseline followed by RMGIC, CR, and GC respectively. These results are in accordance with the previous studies. Arslanoğlu et al.<sup>25</sup> evaluated the microhardness of four tooth-colored materials and found that Equia showed the highest microhardness followed by Fuji II LC, Riva light cure, and glass carbomer, respectively. Evaluating the surface topography of the restorative materials, they also reported internal cracks occurred in all the glass ionomer materials which is also consistent with the results of the present study. After the erosive procedure, RMGIC and HVGIC showed higher microhardness values than CR and GC. However these findings are not applicable since these are different materials with different physical and mechanical properties, and to compare their after-erosion hardness would be contraindicated.

Based on the SEM findings of the present study, the GC material showed severe internal cracks, marginal disintegration, and gap formation in the tooth-restoration interface. In contrast, the resin composite showed none of these unfavorable signs. Similar to the current study's results, Chen et al.<sup>26</sup> also observed fracture lines within the glass carbomer material in their *in vitro* study where they evaluated the marginal leakage of Ketac Molar Easymix and glass carbomer. Although, in the present study, glass carbomer restorations provided superior prevention from erosion, the observation of the severe fractures, even in the control specimens which were spared from erosive cycling, may raise doubts regarding the material's suitability for clinical use. In a previous *in vitro* study by Cehreli et al., the microleakage of the coated and uncoated glass carbomer was compared with glass ionomer and compomer materials.<sup>27</sup> The authors reported that although they observed crack lines within the uncoated specimens, the coated specimens were found to be intact. In the present study, a coating agent was applied on the glass carbomer restorations as instructed by the manufacturer. Even although the erosive cycling might result in the removal of the surface gloss, the control specimens in the GC group, which were not subjected to an erosive procedure, also showed similar defects. Likewise, in another *in vitro* study by Meral and Baseren, similar crack lines and disintegration were reported in artificially aged GC specimens even though the coating agent was applied.<sup>28</sup>

Therefore, the advantages and disadvantages of this material should be carefully interpreted when deciding on its clinical use.

It is important to note, that this is only an *in vitro* study which has certain limitations in its ability to mimic the oral conditions. Therefore these results require to be validated with clinical studies to achieve more comprehensive data.

### CONCLUSIONS

1. The high viscosity glass ionomer and glass carbomer materials tested in this study revealed a superior protective effect on neighbouring enamel after erosive challenge than the current resin-based materials.
2. The use of the current high viscosity glass ionomer restorative might be beneficial for the prevention of neighbouring enamel tissue demineralization in patients at risk of dental erosion.
3. Although the tested glass carbomer restorative might be advantageous for prevention of enamel from erosion, the imperfections of the material and the intense gap formation between the tooth tissue and the restorative material has to be taken in consideration in clinical use.

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