Evaluation of shear bond strength, penetration ability, microleakage and remineralisation capacity of glass ionomer-based fissure sealants

**ABSTRACT**

**Aim** The aim of this study was to evaluate the bond strength, penetration ability, microleakage, and remineralisation capacity of glass ionomer-based fissure sealant materials.

**Methods** In this study, three glass ionomer-based fissure sealants were evaluated (Fuji Triage, Fuji VII EP, and GCP Glass Seal). A dye-penetration test was performed to evaluate microleakage under a stereomicroscope. The materials were applied to occlusal tooth surfaces, and bucco-lingual sections (1 mm width) were assessed to evaluate the penetration ability. Shear bond strength of tested materials was assessed using a universal testing machine. Finally, the remineralisation capacities of the materials were evaluated with EDS.

**Results** The Fuji Triage exhibited the lowest microleakage and unfilled area proportion (p<0.05). The highest shear bond strength was calculated with Fuji VII EP (p<0.05). The fluoride content for all treatment groups was significantly different when remineralisation values were compared to demineralisation (p<0.05).

**Conclusion** Both the Fuji Triage and Fuji VII EP yielded compatible and satisfactory results and all fissure sealants used in this study are sufficient as anti-caries agents.

**Keywords** Glass ionomer fissure sealant; Microleakage; Penetration; Remineralisation; Shear bond strength.

**Introduction**

The prevalence and incidence of dental caries has significantly declined in the last few decades, however, dental caries is still prevalent worldwide [Meja‘re et al., 2004; Kantovitz et al., 2013]. In primary and permanent teeth, the fissure region of the occlusal surface of the molars is the most susceptible caries site, because of the specific anatomy of the molars and inability to adequately remove plaque from those teeth [Meja‘re et al., 2004; Rohr et al., 1991]. The use of systemic and local fluorides, the application of fissure sealants, education in oral hygiene and proper diet are common preventive strategies for caries progression [Newburn, 1991; Borges et al., 2010].

Fluoride application is a useful method for controlling caries on smooth and approximal surfaces; however, fluorides are not equally effective in protecting occlusal pits and fissures [Newburn, 1991]. Sealant placement is considered to be the most effective method for preventing or arresting the development of occlusal caries [Borges et al., 2010]. There is currently a wide variety of sealing materials available, including glass ionomer cements (GICs) and unfilled or filled resins, with/without fluoride release [Sanders et al., 2011; Shimazu et al., 2012; Alsaffar et al., 2011]. GICs are primarily recommended for pits and fissure sealing for two reasons: (i) glass ionomer materials are less susceptible to moisture, which allows them to be used in uncooperative children or in partially erupted teeth where isolation could be a problem and (ii) glass ionomers may potentially act as a fluoride reservoir, making enamel more resistant to demineralisation [Simonsen et al., 1996]. Recently, newer technologies, such as glass ionomer materials containing casein phosphopeptide-amorphous calcium phosphate (CPP-ACP) and nano-fluorapatite/hydroxyapatite, were developed. Studies evaluating the addition of CPP-ACP to GICs indicated that CPP-ACP is stable in the presence of fluoride, improves calcium and phosphate ion release in lactic acid, and works synergistically with fluoride [Al Zraikat et al., 2011; Zalizniak et al., 2013]. The addition of nano-sized hydroxyapatite and fluorapatite particles were also reported to enhance the mechanical properties of GICs, including the strength, wear resistance, and remineralisation capacity of the restorative materials [Lucas et al., 2003; Moshaverinia et al., 2008].
For fissure sealant treatment to be beneficial for caries prevention, the sealant must thoroughly fill pits and fissures and remain completely intact and bonded to the enamel for a long time. Thus, the success of the treatment primarily depends on the marginal adaptation between the tooth and fissure sealants and the bond strength of the materials [Borges et al., 2010]. However, fissure sealants that provide fluoride are important, not only because they act as a physical barrier between the tooth and oral environment, but also because they function as active caries preventive, cariostatic, and remineralisation agents [Guler and Yilmaz, 2013; Ripa, 1991]. Although many studies have investigated the mechanical properties of different fissure sealant materials, no comparative studies have been performed comparing glass ionomer-based fissure sealants that were used in the present study. In addition, the current literature provides limited information on their physical characteristics and remineralisation capacity. Therefore, the purpose of this study was to determine the bond strength, penetration ability, microleakage, and remineralisation capacity of three different glass ionomer-based fissure sealant materials. The tested null hypothesis was that there were no statistically significant differences in the evaluated parameters for the different materials tested.

Materials and methods

Three glass ionomer-based fissure sealants (Fuji Triage, Fuji VII EP, and GCP Glass Seal) were used in this study. The chemical composition, manufacturer, batch numbers, and application procedures of the tested materials are shown Table 1. The present study was approved by the Medical Ethics Committee of Izmir Katip Celebi University, under report No. 2013/073.

**Microleakage: sample preparation**

A total of 120 sound, caries-free, human molars, extracted due to periodontal problems, were chosen for sealant application. The teeth were cleaned with fluoride-free pumice and a rubber cup. To examine the occlusal fissure morphology, teeth were cleaned using a bristled brush and pumice slurry, washed with water for 15 s, and then dried using an air syringe for 10 s. The teeth were stored in 0.1% thymol solution at 4°C and used within 1 month following extraction. Prior to sealant application, all teeth were subsequently washed again under tap water to remove the fluoride-free pumice from their surfaces, and then dried with an air syringe for 10 s. Following this procedure, all specimens were randomly divided into 3 groups (40 teeth each), with one group for each of the tested materials. The groups were designated as follows.

- Fuji Triage (Control) (FT): Gel Etchant (20% polyacrylic acid)/Fuji Triage/Fuji Coat LC.
- Fuji VII EP (FEP): Gel Etchant (20% polyacrylic acid)/Fuji VII EP/Fuji Coat LC.
- GCP Glass Seal (GS): Ethylenediaminetetraacetic acid (EDTA)/GCP Glass Seal/GCP Gloss.

### Materials and methods

#### Chemical composition

<table>
<thead>
<tr>
<th>Material</th>
<th>Chemical composition</th>
</tr>
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<tbody>
<tr>
<td>Fuji Triage (#1202091, GC Int., Tokyo, Japan)</td>
<td>Glass ionomer, aluminofluorosilicate glass, polyacrylic acid, distilled water, pigment, polybase carboxylic acid</td>
</tr>
<tr>
<td>Fuji VII EP (#1204041, GC Int., Tokyo, Japan)</td>
<td>Fluoroaluminosilicate glass, CPP-ACP, pigment, distilled water, polyacrylic acid, polybase carboxylic acid</td>
</tr>
<tr>
<td>GCP Glass Seal (#7301088, GCP Dental, Vianen, the Netherlands)</td>
<td>Nanoparticles glass, nanofluorozirconic acid, hydroxyapatite, liquid silica</td>
</tr>
<tr>
<td>GCP Gloss (#1307076, GCP Dental, Vianen, the Netherlands)</td>
<td>Modified polysiloxanes</td>
</tr>
<tr>
<td>Fuji Coat LC (#1310241, GC Int., Tokyo, Japan)</td>
<td>Multifunctional urethane methacrylate, aliphatic dimethacrylate, methyl methacrylate, tertiary amine</td>
</tr>
<tr>
<td>Gel Etchant 20% polyacrylic acid (#1305271, GC, Tokyo, Japan)</td>
<td>20% polyacrylic acid, distilled water, aluminum chloride hydrate</td>
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</table>

#### Application Methods

<table>
<thead>
<tr>
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<th>Application Methods</th>
</tr>
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<tbody>
<tr>
<td>Fuji Triage (#1202091, GC Int., Tokyo, Japan)</td>
<td>Etch for 20 s; gently air dry, shake the capsule, mix the capsule 10 seconds at high speed, remove from the mixer and make two clicks to prime the capsule then syringe. Extrude the mixture on the tooth surface, use a brush to spread a thin film. Dispense GCP Glass Seal/GCP Gloss.</td>
</tr>
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</tr>
<tr>
<td>GCP Glass Seal (#7301088, GCP Dental, Vianen, the Netherlands)</td>
<td>Clean the tooth surface with EDTA, gently air dry, shake the capsule, mix the capsule 10-15 seconds with high frequency mixers and make two clicks to prime the capsule then syringe. Extrude the mixture on the tooth surface, use a brush to spread a thin film. Dispense GCP Glass Seal/GCP Gloss.</td>
</tr>
<tr>
<td>GCP Gloss (#1307076, GCP Dental, Vianen, the Netherlands)</td>
<td>Clean the tooth surface with EDTA, gently air dry, shake the capsule, mix the capsule 10-15 seconds with high frequency mixers and make two clicks to prime the capsule then syringe. Extrude the mixture on the tooth surface, use a brush to spread a thin film. Dispense GCP Glass Seal/GCP Gloss.</td>
</tr>
<tr>
<td>Fuji Coat LC (#1310241, GC Int., Tokyo, Japan)</td>
<td>Dispense some drops of GCP Gloss onto a mixing pad and apply with a disposable brush, a thin layer of the coat to the fully built-up filling or sealant. Heat up each surface of the filling for 60-90 seconds.</td>
</tr>
<tr>
<td>Gel Etchant 20% polyacrylic acid (#1305271, GC, Tokyo, Japan)</td>
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</tr>
</tbody>
</table>

**TABLE 1** Chemical composition, application procedures and batch numbers of the tested materials.
The fissure sealant materials were handled and applied by one calibrated operator, according to the manufacturer’s instructions, as described in Table 1. All sealant materials were light-cured with an LED curing unit (Valo, Ultradent Products Inc., South Jordan, UT, USA) set at 1200 mW/cm². After curing, the margins of the sealants were checked under a stereomicroscope (Olympus SZ61, Olympus Optical Co, Tokyo, Japan) for any failure of sealant retention and application. After sealing, the teeth were kept in distilled water at 37ºC for 24 hours before being thermocycled to allow the glass-ionomer based materials to set.

**Thermocycling**

After applying the sealants, the specimens were subjected to thermocycling (Nova Co., Konya, Turkey) for a total of 1000 cycles at 5ºC and 55ºC, with a incubation time of 30 s in each bath and a transfer time of 15 s.

**Microleakage assessment**

After the thermocycling process, the root apices of the teeth were sealed with composite resin to prevent the infiltration of the dye solution through that area. The surface of each tooth was covered with two layers of nail varnish leaving a 1-mm window around the sealant. The sealant specimens were immersed in 0.5% basic fuchsin solution (Wako Pure Chemical Industry, Osaka, Japan) for 24 hours at room temperature. Following this procedure, the teeth were washed under running tap water for 30 s to remove any excess solution. The specimens were then sectioned parallel to the long axis, bucco-lingually, with a slow-speed diamond saw (Buehler Inc., Lake, Bluff, IL, USA), under water, to obtain 3–4 sections (1 mm width). The sections of each tooth were digitally photographed using a stereomicroscope at 20× magnification and transmitted to the computer for analysis of microleakage. For each tooth, the section showing the highest microleakage value was separated for further analysis with Olympus LabSens image analysis software (Olympus, Center Valley, PA, USA). In each sample, the microleakage proportion was expressed as the length of the dye penetration (mm) divided by the length of the sealant/tooth interface (mm) (Fig. 1).

**Shear bond strength: sample preparation**

A total of 60 human sound, caries-free molars, extracted for periodontal problems, were included in the study. The roots of the teeth were sectioned off 1 mm below the cemento-enamel junction with a diamond saw. Then, the tooth was embedded in acrylic resin to facilitate handling, keeping the buccal surface exposed. The surfaces were kept intact in a flat position, maintaining a sufficient area to build up the tested sealant materials. The buccal surface of each tooth was polished with 600-, 800-, and 1000-grit silicon carbide paper for 5 s each to obtain a flat and smooth enamel surface. The samples were randomly divided into three groups of 20. For all specimens, the surface treatments were applied according to the
manufacturer’s instructions. A Teflon mold (3 mm diameter, 2 mm height) was placed on the polished surfaces. The tested materials were placed in the molds to form a cylindrical button. Once the materials were light-cured, the specimens were stored in distilled water at 37°C for 24 hours to avoid dehydration.

**Shear bond strength test**

After 24 hours, the embedded specimens were attached to the testing device, and each sealant cylinder was tested using a universal testing machine (Shimadzu, Model AGS-X 5kN, Shimadzu Corporation, Kyoto, Japan). A shear load was applied to the base of each cylinder, at a crosshead speed of 0.5 mm/min, until failure. The force, in Newton (N), required to displace the restoration was recorded, and then, the bond strength (Megapascal, MPa) was calculated by dividing the shear force (N) by the area of the adhesion. Shear bond strength was calculated and expressed in MPa.

The fractured specimens were observed on a stereomicroscope at 20× magnification to evaluate the fracture patterns. The failure modes were classified as 3 types: (1) adhesive failure between the sealant material and enamel, (2) cohesive failure within the enamel or in the sealant material, and (3) mixed fracture.

**Remineralisation: specimen preparation**

A total of 15 human molar teeth, which had no evidence of physical damage, such as discoloration, surface texturing, or cracks, were selected for the study. The roots of the teeth were removed by sectioning approximately 1 mm below the cemento-enamel junction and perpendicular to the long axis, using a water-cooled diamond disc. In addition, the teeth were sectioned mesio-distally to obtain buccal surfaces of the teeth. The samples were randomly divided into three groups that contain 5 samples. The specimens were ground and polished using 1200-, 2000-, and 3200-grit silicon carbide abrasive paper, lubricated with water, to flatten the outer enamel surface for 5 s each. Each specimen was coated with a clear nail varnish, leaving 4 enamel window areas (2 mm × 2 mm) exposed on the enamel surfaces of the teeth.

**Preparation of the enamel erosion model**

Following sample preparation, the specimens were placed into a demineralising solution in an incubator for 48 hours to create artificial caries-like lesions. The demineralising solution contained 2.2 mM CaCl₂, 2.2 Mm NaH₂PO₄, and 50 mM acetic acid, adjusted to a pH of 4.8. After demineralisation, the specimens were carefully washed with deionised water and dried at room temperature.

**Remineralisation procedure**

Fifteen demineralised specimens were prepared and divided into 3 groups (1) Fuji Triage, (2) Fuji VII EP, and (3) GCP Glass Seal. For each specimen, a control window was painted with nail varnish following demineralisation, while the other three windows were covered with the corresponding dental material to remineralise the eroded enamel. The enamel of the three windows was remineralised for 2 weeks, 4 weeks, and 6 weeks in an incubator at 37°C with 5% CO₂. The dental material on one window was removed every 2 weeks, and then, the exposed window was painted with nail varnish. Following the 6-week test period, the nail varnish was removed from all of the specimens using acetone. After completing the remineralisation process, the samples were rinsed with distilled water and left to air dry at room temperature for 24 hours.

**Energy Dispersive X-ray Spectroscopy characterisation (EDS) analysis**

Before EDS examination, the teeth were sputter-coated using gold-palladium (EMITECH K550X Sputter Coater, Emitech Ltd., Ashford, UK), and the samples were placed in the vacuum chamber. The spot size was set at 3.0, and the voltage was set at 10 kV. EDS examination was performed using a scanning electron microscope (SEM) equipped with an EDS detector to assess mineral content, and the data were recorded.

**Microleakage and unfilled area proportions were analysed using a multivariate analysis of variance (MANOVA). Multiple comparisons were performed using a Bonferroni adjustment with the significance level set at 0.05. Shear Bond Strength results were analysed using one-way ANOVA and Bonferroni adjustment at a preset alpha of 0.05. The mean Ca/P ratio and the fluoride content values were analyzed using repeated measures ANOVA. Multiple comparisons were performed using a Bonferroni adjustment with the significance level set at 0.05. SPSS for Windows, Version 21.0 (SPSS Inc., Chicago, IL, USA) was used to conduct statistical analysis.**

**Results**

**Microleakage**

The microleakage scores for the tested sealant materials are shown in Table 2. Significant differences were observed between all of the fissure sealant materials in the following order: GCP Glass Seal > Fuji VII EP > Fuji Triage (p < 0.05).

**Penetration ability**

The proportions of unfilled area for the tested sealant materials are shown in Table 2. The Fuji Triage exhibited the lowest unfilled area proportion, while the GCP Glass Seal showed the highest. Although no significant differences were found between the Fuji VII EP and the other two materials (p > 0.05), there was a significant
difference observed between the Fuji Triage and the GCP Glass Seal (p < 0.05).

**Shear bond strength**

The shear bond strength values for the tested sealants are shown in Table 2. The Fuji VII EP exhibited the highest shear bond strength score, which was significantly different from that of the Fuji Triage and the GCP Glass Seal (p < 0.05). Although the Fuji Triage showed a higher shear bond strength value when compared to the GCP Glass Seal, no significant differences were found between those two materials (p > 0.05). The fracture modes for the Fuji Triage materials were adhesive (95.0%) and mixed fracture (5.0%); for the Fuji VII EP the fractures were adhesive (90.0%) and mixed fracture (10.0%); and for the GCP Glass Seal the fractures were all adhesive fractures (100.0%).

**Remineralisation**

The relationships between the mean Ca/P ratio values, with standard deviation and remineralisation times, are shown in Table 3. No significant differences were observed in the Ca/P ratio values between the tested materials (p > 0.05). The mean fluoride content values, with standard deviation and remineralisation times, are shown in Table 4. For all treatment groups, the fluoride content was statistically different after remineralisation when compared to demineralisation values (p < 0.05). In addition, the fluoride content in the GCP Glass Seal group was significantly higher than the Fuji Triage group (p < 0.05) after 6 weeks of remineralisation. However, there were no significant differences for the remineralisation times in the treatment groups (p > 0.05).

**Discussion**

Pit and fissure sealants have been considered an outstanding adjunct to oral health care preventive strategies in the decrease of occlusal caries onset and/or progression [Abou et al., 1991]. Over time, several modifications have been made to sealant formulations to produce sufficient materials that are success for clinical application. The latest innovations are the development of newly available glass ionomer based fissure sealants based on nanotechnology and addition of remineralisation agents in the contents of the materials [Zalizniak et al., 2013; Lucas et al., 2003; Zainuddin et al., 2012]. Before these materials may be

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### Table 2

<table>
<thead>
<tr>
<th></th>
<th>Microleakage proportion</th>
<th>Unfilled area proportion</th>
<th>Shear bond strength</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fuji Triage</td>
<td>0.000 (0.000) *</td>
<td>0.058 (0.064) *</td>
<td>3.474 (0.760) *</td>
</tr>
<tr>
<td>Fuji VII EP</td>
<td>0.129 (0.286) b</td>
<td>0.081 (0.111) c</td>
<td>4.984 (1.730) b</td>
</tr>
<tr>
<td>GCP Glass Seal</td>
<td>0.546 (0.277) c</td>
<td>0.150 (0.224) c</td>
<td>2.908 (0.235) c</td>
</tr>
</tbody>
</table>

*SD: Standard Deviation

*Equal letters indicate statistical significance

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### Table 3

<table>
<thead>
<tr>
<th></th>
<th>Mean (SD)</th>
<th>Mean (SD)</th>
<th>Mean (SD)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fuji Triage</td>
<td>1.86 (0.11) a, A</td>
<td>1.85 (0.14) a, A</td>
<td>1.93 (0.13) a, A</td>
</tr>
<tr>
<td>2 Weeks</td>
<td>1.92 (0.11) a, A</td>
<td>1.92 (0.12) a, A</td>
<td>1.95 (0.15) a, A</td>
</tr>
<tr>
<td>4 Weeks</td>
<td>1.91 (0.11) a, A</td>
<td>2.00 (0.19) a, A</td>
<td>1.98 (0.08) a, A</td>
</tr>
<tr>
<td>6 Weeks</td>
<td>2.00 (0.10) a, A</td>
<td>1.99 (0.16) a, A</td>
<td>1.97 (0.11) a, A</td>
</tr>
</tbody>
</table>

*Lowercase letters label statistically significance differences between the remineralization times (p < 0.05).

*Capital letters indicate statistically significance differences between the tested materials (p < 0.05).
tested in clinical studies, it is essential to evaluate the performance of these newly produced fissure sealants in the laboratory. Therefore, the primary objective of this study was to evaluate the bond strength, penetration ability, microleakage, and remineralisation capacity of three different glass ionomer fissure sealant materials. Based on the results of this study, our null hypothesis was rejected since differences in the microleakage, penetration ability, shear bond strength, and remineralisation capacity were observed among the different materials tested.

Microleakage is an important property that has been used in assessing the success of restorative materials used for tooth restorations. The most widely accepted method for evaluating the microleakage of restorations is the dye-penetration test. Common dyes for this test include basic fuchsin (0.5–2%), methylene blue (0.2–2%), silver nitrate (50%), crystal violet (0.05%), erythocyn (2%), and Rhodamine B (0.2%) [Feizier et al., 1987; Fortin et al., 1994; Tay et al., 2005]. In this study, a basic fuchsin dye solution was used to assess microleakage, as it provides a better correlation with SEM quantitative marginal analysis than methylene blue does [Tay et al., 2005]. To simulate oral conditions, the teeth were subjected to thermocycling following the restorative procedures used in this study [Koyuturk et al., 2008]. Of the various methods that exist, the most commonly used technique for measuring microleakage values is to evaluate sections of restored teeth. In the present study, 3–4 sections were made through each restoration to increase the reliability of measurements, and the sample with the highest microleakage score was selected for further analysis. This technique was combined with image analysis methods to obtain quantitative results instead of just conventional subjective scoring. Based on the results of the present study, the GCP Glass Seal had the highest microleakage scores when compared to the other fissure sealant materials. The reason for higher leakage produced by the GCP Seal may be explained by two facts: (a) inadequate micromechanical retention between the restoration and tooth structures caused by the lower etching capacity of surface treatment, and (b) possible lower flowability of the GCP Glass Seal compared to the other materials. In addition, besides the microleakage along the tooth-restoration interface, internal and surface crack lines were observed in many of the samples of the GCP Glass Seal group. In addition to the microleakage along the fissure sealant and enamel surface, dye penetration was evident within the crack lines, suggesting the severity of the loss of integrity.

The penetration depth is a very important parameter that may affect the sealant retention. Several factors, such as the material properties and the fissure morphology, have been suggested to have an influence on the penetration ability [Al-Jobair, 1987; Markovic et al., 2011]. The penetration ability in the present study was evaluated by measuring the proportion of the unfilled area, rather than using a qualitative method (rating score systems) conventionally used in comparable studies. The present study revealed a significant difference in the penetration depth between the tested materials. The Fuji Triage exhibited the lowest proportion of unfilled area, while the GCP Glass Seal showed the highest. It is known that less viscous sealants exhibit better flow, and thus, are able to penetrate more deeply into the fissures [Markovic et al., 2011; Khogli et al., 2013]. For this reason, the possible lower flowability of the GCP Glass Seal caused the higher proportion of unfilled area.

Another factor directly related to the clinical success of pit and fissure sealants is the materials’ capacity of bonding to occlusal pits and fissures and shear bond strength test has been used elsewhere to test sealant adhesion [Borsatto et al., 2010; Corona et al., 2005]. For bond strength evaluation, the buccal surfaces of all teeth were considered as this surface allowed the shearing force to be exactly perpendicular to the bonded specimen. Since bonding strength is directly related to the surface area between the material and the tooth surface, for all specimens, Teflon molds of a standard size were used to obtain the samples. In the shear bond strength test, the speed of the force applied to samples may vary; in the present study, the recommended value for glass ionomer materials (0.5 mm/min) was applied [Bala et al., 2009; Mauro et al., 2009]. Based on the results of the present study, the Fuji VII EP exhibited the significantly higher shear bond strength scores when compared to the other materials. Although both the Fuji VII EP and Fuji Triage have similar components, it is thought that the addition of CPP-ACP (1–5%) to the content of the Fuji VII EP may affect the obtained results. This result is consistent with the findings presented in a study by Mazzaoui et al. [2003], which indicated that the incorporation of CPP-ACP into glass ionomer cements resulted in an increase in the microtensile bond strength of the materials.

In the present study, an in vitro model was used to compare the remineralisation efficacy of three different glass ionomer-based fissure sealant materials on initial caries lesions in human tooth enamel. The remineralisation potential of the tested materials on enamel subsurface lesions was evaluated using EDS analysis. EDS has been used for elemental analysis at the ultrastructural level. EDS is a microanalytical technique used in conjunction with SEM, wherein SEM is used for structural analysis, and EDS is used for elemental analysis [Hegde et al., 2007]. To the best our knowledge, no previous studies have investigated ion passage from the glass ionomer-based fissure sealant materials used in the study to the tooth structure with EDS. However, there are a few studies that have evaluated the fluoride released from GICs containing CPP-ACP at neutral and acidic pH values [Mazzaoui et al., 2003; Al Zaikat et al., 2011; Zalizniak et al., 2013]. For example, Mazzaoui et al. [2003] showed that GICs containing CPP-ACP released significantly more fluoride when compared with the control group in water, and no calcium release was detected in water from either material. Zalizniak et al. [2013] showed that the addition of CPP-ACP to GICs increased the release
of calcium ions, with no change in fluoride ions released. In addition, Zraikat et al. [2011] investigated the addition of CPP-ACP to GICs and found that significantly less fluoride was released in water when 5% CPP-ACP was incorporated into the GIC, and the calcium release from the GIC containing 5% CPP-ACP was small, but detectable in water. Zraikat et al. [2011] suggested that the differences between the results from these studies may be attributed to the formation and precipitation of CaF₂ following the release of both ions into the storage solution. When considering the results of the present study, while no significant differences were observed between the three materials and the different treatment times, in regards to the Ca/P ratio, for all treatment groups, a statistically significant difference in the fluoride content was observed when values after remineralisation were compared to demineralisation values. It is now widely accepted that remineralisation of incipient caries lesions is accelerated by trace amounts of fluoride. In addition, fluoride can inhibit demineralisation and plaque bacteria. The mineral formed during remineralisation is more resistant to acid than the original enamel or dentin mineral, especially if fluoride is present [Featherstone, 1999; Stoodley et al., 2008]. In light of these findings, the fissure sealant materials used in this study appear to be useful for reducing the incidence of caries due to microleakage or restoration failure.

**Conclusion**

Within the limitations of this study, the following conclusions can be drawn.

1. All fissure sealant materials used in the study seem sufficient as anti-caries agents.
2. In terms of microleakage, penetration ability and shear bond strength, Fuji Triage and Fuji VII EP gave compatible and satisfactory results.

**Acknowledgments**

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**References**